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PROCEEDINGS OF THE THIRD ANNUAL GENERAL MEETING OF BIOMINET

FOREWORD

BIOMINET presents in this volume the invited technical papers (with the exception of Dr. J.W. Costerton's paper, which was unavailable) and the abstracts of the mini-presentations made by members of BIOMINET at the Third Annual Meeting held jointly with the Hydrometallurgy Section of the Canadian Institute of Mining in Toronto on August 20th, with the BIOMINET General Meeting and the mini-presentations being held on August 21st at the Howard Johnson Hotel in Oakville, Ontario.

The efforts of the Session Chairmen and of the many individuals involved in the arrangements for the Annual Meeting are greatly appreciated.

R.G.L. McCready
Editor

COMPTE RENDU DE LA TROISIÈME RÉUNION GÉNÉRALE ANNUELLE DE BIOMINET

AVANT-PROPOS

Dans ce volume, BIOMINET présente les exposés techniques sollicités (exception faite de l'exposé du Dr J.W. Costerton qui n'était pas disponible) et les résumés des mini présentations faites pas les membres de BIOMINET lors de la Troisième réunion annuelle tenue conjointement à Toronto le 20 août avec la Section d'hydrométallurgie de l'Institut canadien des mines. La réunion générale de BIOMINET ainsi que les mini présentations ont eu lieu le 21 août à l'hôtel Howard Johnson à Oakville en Ontario.

Nous apprécions grandement les efforts du Président de la Session et de toutes les personnes qui ont participé à l'organisation de la réunion annuelle.

R.G.L. McCready
Éditeur

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BIOHYDROMETALLURGY OF GOLD

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ABSTRACT

Biohydrometallurgical processes have recently gained impetus in metal extraction. This tendency is especially prevalent in the treatment of complex and low-grade (waste) materials. Currently, copper and uranium are extracted by bio-mediated heap, dump, and in-situ leach processes. However, this technology is ready to be employed in the ore processing of precious metals.

This paper reviews the literature on gold bio-extraction and provides information on some of NMIMT's related studies. Bioleaching of gold-bearing resources is discussed in terms of gold extraction from silicate-bearing ores by heterotrophic microorganisms; from solutions by biosorption; and from pyrite, arsenopyrite, and other metal sulphides by autotrophic bacteria. Where appropriate, experimental conditions and the economics of these processes are specified. The chemistry and thermodynamics of gold-ore leaching by microorganisms are summarized.

SESSION I: PRÉSENTATION 1

BIOHYDROMÉTALLURGIE DE L'OR

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RÉSUMÉ

Depuis quelques temps, les procédés biohydrométallurgiques sont de plus en plus répandus dans le domaine de l'extraction des métaux. Cette tendance vaut notamment pour le traitement des matériaux complexes et des matériaux de basse qualité (déchets). On extrait actuellement du cuivre et de l'uranium par des procédés de lixiviation biologique en tas, en terril et in situ. Or, cette technologie est applicable au traitement des minerais contenant des métaux précieux.

Le présent article passe en revue la documentation sur l'extraction biologique de l'or et renseigne sur certaines de nos études connexes. La biolixiviation des ressources aurifères regroupe l'extraction de l'or des minerais contenant des silicates par des microorganismes hétérotrophes, l'extraction de l'or de solutions par biosorption et l'extraction de l'or de la pyrite, de l'arséniopyrite et d'autres sulfures métalliques par des bactéries autotrophes. Lorsque cela convient, on spécifie les conditions expérimentales et les paramètres économiques de ces procédés. La chimie et la thermodynamique de la lixiviation des minerais aurifères par des microorganismes sont résumées.

BIOHYDROMETALLURGY OF GOLD

INTRODUCTION

Gold has been used traditionally as a coinage metal and high-quality jewellry material in the form of karat gold, or alloys of gold containing copper, silver, zinc, and platinum (1). In dentistry it is used in alloys containing 25 to 70% gold for inlays, crowns, and bridges. In the late twentieth century, gold has emerged as an essential industrial metal. For electronics (computer and space industries), gold is applied selectively to electrical contact areas. Gold alloys are also produced for solders. Tiny wires are made of gold for transistor connections. Other specialized gold compounds are made for diverse uses such as in medicine and glass industries.

The earth's crust and seawater have been estimated to contain approximately 4.8×10^{-3} g/t and 8×10^{-6} g/t gold (2), respectively. Nearly all gold-bearing deposits occur near acidic, igneous intrusions (3). The general deposit types include magmatic, contact metamorphic, replacement, and cavity-filling rocks. Gold often occurs in association with base metals and is often finely disseminated in metal sulphide and silicate matrices.

THE CONVENTIONAL TECHNOLOGY

The conventional technology of gold extraction from diverse resources is highly developed. Gold is recovered by chlorination, cyanidation, thiosulphate and thiourea leaching, amalgamation, flotation, gravity concentration, and smelting - or by a combination of these processes (4-16). These processes will not be discussed here, since the subject of the present article is bioleaching, which is a new technique for the extraction of gold from ores and other resources.

EXTRACTION AND RECOVERY OF GOLD BY HETEROTROPHIC MICROORGANISMS

It is well known and documented that microorganisms play an important role in the solubilization and migration of metal ions, as well as in the formation of ore deposits (17-19). In the 1960's, it was shown that heterotrophic bacteria could dissolve gold from lateritic minerals (20-29). In these studies, the maximum amount of gold did not exceed 1.5 mg dm^{-3} concentration. However, in some of the experiments final gold extraction was as high as 82% after 283 days of leaching. The leach mechanism was found to be complex and no final conclusions were made.

Certain kinds of fungi were found not only to extract gold from ores, but also to remove the dissolved gold from the leach solution by biosorption (30). In effect, fungi were found to remove gold from the leach solution by adsorbing it on their surface. These fungi cells were then filtered, dried, and roasted. A variety of active fungi were reported to remove up to 98% of gold from the leach solution in 15 to 20 h of contact. A portable and fully mechanized unit was developed (31) for the extraction of gold from ores. This unit was easily adapted to the existing field conditions. The unit, which consisted of a contacting rotary drum leach system, was able to process 1 m^3 h^{-1} of gold ore and was operated by a team of three people. Other laboratory biosorption experiments for the removal of gold from leach solution were carried

out as follows (32): fungal cells of *Aspergillus orzzae* were added to gold-containing solutions; in 10 days about 45% of the gold was found to be adsorbed by the fungi, and about 53% of the gold was precipitated in colloidal form and collected at the bottom of the glass flasks. The original solution contained 1.11 g dm^{-3} of gold, and the total yield of gold removal corresponded to approximately 98%. Bioaccumulation techniques, in which microorganisms are selectively absorbing precious metals from diverse industrial solutions, are being studied at Battelle Memorial Institute's Columbus laboratory (33). Similarly, another company, Advanced Mineral Technologies Incorporated, is working on the development of biosorption methods for the selective extraction of gold and silver from jewellry waste solution (33).

Dissolution of gold by microorganisms and their metabolites from ores was found to be most effective with strains of *Bacillus megaterium, Bacillus mesentericus niger, Pseudomonas liquefacienes,* and *Bacterium nitrificans*, which were isolated from gold mine drainage (34). The metabolites aspartic acid, histidine, serine, alamine, and glycine were found to play an important role in gold dissolution. Mutants synthesizing large amounts of these amino acids were obtained by ultraviolet irradiation and by the addition of ethylene imine to the leach solutions. The highest gold concentration in leach solutions varied from 10 to 35 mg dm^{-3} after 20 days of leaching. The effects of pure cultures of *Bacillus mesentericus 12*, as well as their amino acid and protein metabolite fractions, on solubilization of native gold powders were studied (35). In the presence of amino acid fractions, the concentration of gold was increased continuously, reaching a maximum value of 64.6 mg dm^{-3} in 360 days.

DISSOLUTION OF GOLD FROM SULPHIDE-BEARING SUBSTRATES BY MICROBIAL ACTION

If gold occurs finely disseminated within the host sulphide ore matrix, such as in pyrite, arsenopyrite, pyrrhotite, galena, and sphalerite, then the economic viability of many of the above-mentioned gold extraction processes is less than marginal. The leachant cannot penetrate into the interior of the solid ore to reach the enclosed gold submicron size particles. To liberate gold, these sulphide-bearing ores are often subjected to preoxidation. However, roasting is an energy-intensive process that is associated with environmental problems such as emission of volatile arsenic, lead, and sulphur - compounds that are toxic. An alternative approach to preoxidation by roasting is the application of bio-mediated reactions for the liberation of gold particles from sulphide ores.

Gold-Containing Pyrite Leaching

Leach experiments (36) were carried out with gold-bearing pyrite (150 g of gold/ton of ore) in Erlenmeyer flasks charged with 5 g of pyrite and 200 cm^3 of nutrient medium containing 200 mg of $(NH_4)_2SO_4$, 50 mg of KCl, 50 mg of K_2HPO_4, 500 mg of $MgSO_4$, and 10 mg of $Ca(NO_3)_2$ per litre of distilled water, and inoculated with 10 cm^3 of a pure strain of *Thiobacillus ferrooxidans* gave results as shown in Table 1. Experiments No. 1 were carried out without addition of bacteria to the leach solutions (sterile controls). Each figure in Table 1 was obtained after three weeks of leaching at room temperature. The efficiency of the bacteria in solubilization of gold from pyrite is evident, since from the stationary experiment no gold could be detected in the sterile controls, while in the shake flask experiments more than 6 times the amount of gold was solubilized in the inoculated runs than in the solution of sterile

experiments. During the bacterial oxidation of metal sulphides (pyrite, galena, and sphalerite), oxidized sulphur species (thiosulphates, polythionates, and sulphates) are formed (36). Most probably, gold is complexed by these anions and solubilized, for example, in the form of:

$$Au + 1/2O_2 + H_2O + 3S_2O_3^{2-} \longrightarrow Au(S_2O_3)_3^{5-} + 2OH^- \qquad \text{Eq 1}$$

Oxidation of pyrite results in the formation of sulphuric acid (37):

$$2FeS_2 + 7 1/2O_2 + H_2O \xrightarrow{\text{bacteria}} Fe_2(SO_4)_3 + H_2SO_4 \qquad \text{Eq 2}$$

The hydroxyl ions in Equation 1 will be neutralized with sulphuric acid available from Equation 2:

$$2\ OH^- + H_2SO_4 = 2\ H_2O + SO_4^{2-} \qquad \text{Eq 3}$$

Using crushed gold-containing pyrite, chalcopyrite, arsenopyrite, galena, and marcasite in inoculated (with *T. ferrooxidans*) and in sterile controls with individual or mixed sulphides (38), it was found that the gold extraction was not higher in the inoculated cultures than in the sterile controls. These findings contradict those of the previous investigators (36). It was concluded (38) from their laboratory experiments that *T. ferrooxidans* in dilute solution probably have no significant effect on the accumulation and migration of gold in the ground. Leaching experiments with tailings from the Front Range portion of the Colorado Mineral Belt (39) indicated that in the presence of bacteria and nutrients, the extraction of base metals was considerably improved compared to that in the sterile controls. However, the gold and silver extractions from these tailings were not improved in the presence of bacteria. The authors (39) suggested a two-step leach process to achieve the economic viability of their process. In the first step, a bacterial (acid) leaching would be carried out to remove parts of the base metals from the tailings and to liberate gold and silver. After this, the residue would be neutralized with lime. In the second step, a conventional cyanidation technique would be applied to extract gold and silver.

Using the above principle of two-step leaching (39), other investigators (40) demonstrated the applicability of this method in the extraction of gold and silver from refractory pyrite concentrates. They found that the gold extraction was directly proportional to the yield of bacterial pyrite preoxidation. For example, if no bacterial preoxidation were applied to the pyrite sample, the cyanidation resulted in approximately 25% gold extraction. When pyrite was preoxidized to 80%, the gold extraction by cyanidation exceeded 90%. The relatively low pH (less than 1) was found to be beneficial in keeping ferric ion in solution. The bacteria *T. ferrooxidans* are known to function best at around pH = 2.3 (41,42); pH values less than 1.5 usually inhibit bacterial activity. However, it was stated (40) that the bacteria were able to adapt well to pH values as low as about pH = 0.6. A similar observation was reported on the adaptability of *T. ferrooxidans* to very low pH values (43) during biotank-leaching of refractory ores. The gold extraction was studied from a zinc and lead flotation residue (containing 1.75 g/t of gold) by a double

leach process (44,45). In the first step, the premined and finely ground flotation was leached in an aerated and agitated leach tank by *T. ferrooxidans*, when parts of the zinc, iron, and copper contents were solubilized. After solid-liquid separation, the leach residue (acidic) was subjected to thiourea leaching to extract gold. It is believed that during the bacterial leaching of the sulphide-bearing flotation wastes, gold remained in its native form in the solid leach residue, but partly exposed and liberated as submicron size particles from the pyrite gangue. The extraction of gold with thiourea can be given by:

$$Au + Fe^{3+} + 2\ CS(NH_2)_2 = Au[CS(NH_2)_2]^+ + Fe^{2+} \qquad \text{Eq 4}$$

The ferric ion is provided from pyrite oxidation according to Equation 2. The advantage of using thiourea for the extraction of precious metals from the acidic leach residue of bacterial leaching, is that it requires no neutralization of the residue, which is a prerequisite for the cyanidation process. In addition, thiourea and its decomposition products are not environmentally hazardous compared to the cyanide leachant. Recent emphasis on the environmental effects and costs of conventional mining, plus success in accelerating bacterial action, suggest that biotechnology should be explored for gold ore processing (46). The costs of bioleaching of refractory sulphide gold ores have been estimated to decrease as the plant size increases (47). For example, the total costs including chemicals, salaries, maintenance, and capital costs, for a 50 t/day capacity plant are $83.80 per ton of concentrate treated; this figure decreases for a plant of 150 t/d capacity to $55.30 per ton of concentrate treated. The costs of producing the concentrates and the costs of cyanide leaching for the extraction of gold are additional expenses.

Extraction of Gold from Arsenopyrite-Bearing Resources

Bioleaching of arsenopyrite is well established (48,49). Investigation showed that the bacterium, *T. ferrooxidans*, is able to liberate gold from carbonaceous ore containing 6% of arsenic (50). The best leaching conditions were established to be 20% pulp density, pH = 1.5-2.0, and ore particle size 0.05 mm. Under these conditions, 90% of arsenic and 60% of iron were extracted from the ore in 10 days of leaching. The biochemistry arsenopyrite oxidation can be expressed by the following electrochemical reactions (51,52):

Anodic oxidation:

$$FeAsS \xrightarrow{\text{bacteria}} Fe^{2+} + As^{3+} + S + 7e \qquad \text{Eq 5}$$

Cathodic reduction:

$$31/2\ O_2 + 14\ H^+ + 7e \xrightarrow{\text{bacteria}} 7\ H_2O \qquad \text{Eq 6}$$

The As^{3+} ion liberated in Equation 5 will immediately go through a hydrolysis reaction to yield arsenious acid:

$$As^{3+} + 3\ H_2O \rightleftharpoons H_3AsO_3 + 3H^+ \qquad \text{Eq 7}$$

The arsenious acid (Eq 7), and Fe^{2+} and S (Eq 5), are furthermore oxidized by the bacteria:

$$H_3AsO_3 + 1/2\ O_2 \xrightarrow{\text{bacteria}} H_3AsO_4 \qquad \text{Eq 8}$$

$$Fe^{2+} \xrightarrow{\text{bacteria}} Fe^{3+} + e \qquad \text{Eq 9}$$

$$S^\circ + 11/2\ O_2 + H_2O \xrightarrow{\text{bacteria}} H_2SO_4 \qquad \text{Eq 10}$$

Ferric ion has high affinity to arsenic acid and ferric arsenate precipitate is formed:

$$Fe^{3+} + H_3AsO_4 \rightarrow FeAsO_4 + 3H^+ \qquad \text{Eq 11}$$

The sum reaction of Equations 5 through 11 is given by:

$$FeAsS + 31/2\ O_2 + H_2O \xrightarrow{\text{bacteria}} FeAsO_4 + H_2SO_4 \qquad \text{Eq 12}$$

Based on the above reactions of arsenopyrite oxidation, a pilot plant was established for the extraction of gold from arsenopyrite (51). In this process, when an arsenopyrite concentrate containing 10% arsenic was subjected to a double-stage bacterial leaching, approximately 90% of the arsenic content was extracted in 100 h of leaching. The leach conditions were: 20% pulp density, particle size 75% less than 0.074 mm, temperature 25-28°C, and pH = 1.75 to 2.1. After bacterial leaching and solid-liquid separation, the residue was neutralized with lime and leached with cyanide solution, resulting in a 92.5% gold extraction. The leach solution containing arsenate was neutralized with lime to pH = 3.5, the arsenate containing precipitate was discarded, and the solution was returned to the leaching step after adjusting for pH and nutrient concentrations.

Another study (53) revealed also that bacterial leaching methods are the most effective for releasing finely dispersed gold from arsenopyrite concentrates containing, for example, 8.4% arsenic, 24.1% iron, 26% sulphur and 290 g/t gold. The particle size of the concentrate was less than 0.074 mm. The leaching was carried out in a direct flow system in three reactors, each of 1.5 dm^3 working volume and containing 25% of solids. The pulps were inoculated with *T. ferrooxidans*, agitated mechanically, and aerated (in rates of 3 dm^3 of air per 1 dm^3 of pulp per minute). Typical leaching data are summarized in Table 2. The leaching was completed in 30 hours with total arsenic extraction of 95%. The leach residue was neutralized with lime and subjected to cyanidation in 20% pulp density suspensions containing 1% KCN. The gold extraction was in the range of 88 to 92%. If the gold-arsenopyrite concentrate were directly leached by KCN solution, the final extraction was only about 7 to 10%. If the gold-arsenopyrite were first calcined and then leached by KCN solutions, the gold extraction could not exceed 77%. This comparison of gold extraction data demonstrates the advantage and efficiency of the biohydrometallurgical approach.

The drainage of gold-bearing high-sulphide tailings abandoned in Alaska and the Yukon was found to be acidic, with a high concentration of arsenic as well as a large number of bacteria (*T. ferrooxidans*) (54-56). As a consequence, serious arsenic contamination of streams and groundwater occasionally occurs. Leach studies (56) indicated that, while more arsenic was released from tailing material by growing cultures of *T. ferrooxidans* than by abiotic controls, acid ferric sulphate solutions were most effective in solubilizing arsenic from the tailings.

The economic viability of a bacterially assisted leach process was addressed (57). In this process, bench-scale tests were carried out with arsenopyrite-pyrite mine waste (9.75 g Au/ton) and concentrate (161.0 g Au/ton). In the first step, bacteria oxidized the arsenopyrite-pyrite matrix to liberate gold and silver. After bacterial leaching, the leach residue was neutralized with lime and the precious metals were extracted by cyanidation. The overall gold recoveries from these substrates were 89.8% from the waste and 92.7% from the concentrate. On the basis of these results, the authors (57) recommended starting up a pilot-plant operation with a 300 kg of ore per day capacity.

A portion of the gold in many ores occurs as submicroscopic disseminations in sulphide minerals. This gold is generally unavailable for recovery by cyanidation process unless the ore is treated by fine grinding, roasting, pressure leaching, etc., which are expensive operations (58). Mintek (58) has evaluated the bacterial alternative for liberation of precious metal inclusions from pyritic ores and has developed mathematical modelling of this process. The model is based on both the propagating pore and the shrinking core concepts. It was also stated that the occurrence of gold in dislocations in auriferous pyrite has important metallurgical implications because these sites are preferred regions of bacterial corrosion.

The present study, a continuation of NMIMT's precious metal extraction studies (15,44,45), is an attempt to adapt bacteria (*T. ferrooxidans*) to increased concentrations of thiourea. In this case, extraction of gold from pyrite and arsenopyrite containing substrate could be simplified to a simple step leach process.

MATERIALS AND METHODS

BACTERIA

A culture of *Thiobacillus ferrooxidans* used in this investigation was originally isolated from acid mine drainage (59). It was adapted to ferrous iron and pyritic zinc sulphide flotation waste in a modified Silverman and Lundgren nutrient medium (60).

SUBSTRATE

Pyritic zinc sulphide flotation residue containing 4.25 g/ton of gold and 27.8 g/ton of silver, as well as 0.8% zinc, 0.6% lead, 10.8% iron, 5.2% magnesium, 0.6% copper, 10.9% total sulphur, 70.1% silica and other trace elements was homogenized by the four-quarter method. This substrate was a -200 mesh product.

BACTERIAL LEACH EXPERIMENTS

All bio-leaching experiments were carried out in 250 cm^3 Erlenmeyer flasks containing varying amounts of pyritic flotation tailings, 70 cm^3 nutrient basal salts medium (60) (with or without thiourea), with 5 cm^3 of a 2% solution of thymol in methanol added instead of inoculum. The flasks were incubated at a constant 250 rpm in a gyratory incubator shaker (Model 26, New Brunswick Sci. Co., Inc.) at 35°C. Experiments were carried out for varying durations from 1 to 20 days. Then, the leach suspensions were filtered and the filtrates analyzed for dissolved copper and zinc extractions on an atomic absorption spectrophotometer (Model 703, Perkin Elmer Co.).

The leach residues of bacterial leaching were placed in 500-cm^3 glass reactors (equipped with agitation, Eh, and pH measurements) to which 300 cm^3 of 0.5 mol dm^{-3} of thiourea solution (61), containing 0.0 to 8.0 g dm^{-3} of ozone as oxidant (62), were added. The pH of the leach suspensions was adjusted to 0.5 with 2 N sulphuric acid. These experiments were carried out at temperatures varying from 15 to 45°C at a constant 850 rpm agitation. Periodically, samples were removed from the leach suspensions and analyzed for gold and silver using a plasma furnace on an atomic absorption spectrophotometer (Model 703, Perkin Elmer Co.).

RESULTS AND DISCUSSION

During the bacterial leaching, the sulphide matrix of the flotation residue is partially reacted due to the following metabolic reactions (37,63-66):

$$ZnS + 2O_2 \xrightarrow{\text{bacteria}} Zn\,SO_4 \qquad \text{Eq 13}$$

$$2\,CuFeS_2 + 81/2O_2 + H_2SO_4 \xrightarrow{\text{bacteria}} Fe_2(SO_4)_3 + 2\,CuSO_4 + H_2O \qquad \text{Eq 14}$$

$$PbS + 2O_2 \xrightarrow{\text{bacteria}} PbSO_4 \qquad \text{Eq 15}$$

$$2FeS_2 + 71/2O_2 + H_2O \xrightarrow{\text{bacteria}} Fe_2(SO_4)_3 + H_2SO_4 \qquad \text{Eq 16}$$

In addition to reactions 13 through 16, chemical oxidation occurs especially by the action of ferric ion (67), for example:

$$ZnS + Fe_2(SO_4)_3 \longrightarrow ZnSO_4 + 2FeSO_4 + S \qquad \text{Eq 17}$$

The elemental sulphur and ferrous sulphate will be oxidized to sulphuric acid and ferric sulphate by the bacteria (68).

Typical results of bacterial leaching of pyritic flotation waste are given in Table 3. These figures represent the main value of duplicated measurements obtained with 25% pulp density suspensions after 5 days of leaching. The effect of microorganisms can be seen from the comparison of inoculated and sterile control data. The amount of lead in solution is very low since both lead sulphide and lead sulphate are insoluble in the sulphuric acid-containing leach media. Gold and silver, as expected, are not extracted during the bacterial leaching.

The efficiency of bacterial preoxidation on the extraction of gold from the pyritic flotation residue is demonstrated in Figure 1. After 2 h of leaching at 35°C with thiourea plus ozone, approximately 95% of the gold content was extracted (curve 1); when oxidant (ozone) was not present, the final extraction was only 62% (curve 2). When no bacterial preoxidation was carried out, only about 26% of gold was extracted with the thiourea plus ozone leach solution (curve 3); with thiourea solution alone, about 18% of the gold content of pyritic waste could be solubilized (curve 4). These data are in good agreement with the findings of other investigators (40,47,57) who used cyanide solution for gold extraction.

As shown in Equation 4, the solubilization of native gold by thiourea requires the presence of an oxidant. When no oxidant (ozone) was added to the leach solution, then possibly ferric ion remained within the bacterial residue was

the oxidant during the thiourea leaching. Since gold is known to form complexes with thiourea (69), it can be written:

$$[Au^+]_{tot} = Au^+ + Au(TU)^+ + Au(TU)_2^+ + Au(TU)_3^+ \qquad \text{Eq 18}$$

where TU = thiourea. Accordingly, the total dissolved gold concentration can be expressed by:

$$[Au^+]_{tot} = [Au^+] \sum_{i=i}^{3} \beta_i [TU]^i = [Au^+]Y_o \qquad \text{Eq 19}$$

where the β_i's are the coordination complex constants whose values are $\beta_1 = 1.0 \times 10^4$, $\beta_2 = 5.2 \times 10^4$, and $\beta_3 = 1.0 \times 10^5$, respectively. The Au^+-thiourea complex stability increases with increased amounts of thiourea added to the leach solution.

The effect of temperature on the initial rate of gold extraction by thiourea-containing ozone from the residue of bacterial preleach of the pyritic waste is shown in Figure 2. There is a continuous increase in the initial rate of gold extraction as the temperature is raised from 20 to 45°C. Higher temperatures than 45°C were not attempted because previous studies (44,45) with thiourea leaching resulted in a decrease in the rate of gold extraction above 50°C. From the Arrhenius presentation (Fig. 3), the activation energy and the frequency factor were determined to be $\Delta E_a = 39.2$ kJ and $A = 1.9 \times 10^{-3}$ mol $dm^{-3}s^{-1}$. The value of activation energy suggests that the gold extraction from the bacterial leach residue, with thiourea solutions containing ozone as oxidant, may be diffusion controlled.

To simplify the gold extraction from pyritic materials, a series of experiments was started to adapt *T. ferrooxidans* to increased concentrations of thiourea. If sufficiently high thiourea concentrations would be tolerated, then the gold-leaching process could probably be simplified to one step instead of two steps, which is the current practice. The first series of adaptation was carried out with 3K of ferrous ion-containing media (60) supplemented with 0 to 40 mg dm^{-3} of thiourea; the results are shown in Figure 4. All experiments were performed in duplicated runs and each point on the curves is the mean value of these measurements. These curves are almost identical and it can be concluded that the iron-oxidizing ability of *T. ferrooxidans* was not affected by the concentrations of thiourea up to the 40 mg dm^{-3} level. Therefore, a new series of experiments was carried out with bacteria grown on ferrous ion in the presence of 40 mg dm^3 of thiourea, but this time the thiourea concentration of the 3K of ferrous ion media was supplemented with thiourea up to 500 mg dm^{-3}. The results are summarized in Figure 5. There is a measurable effect of thiourea on the bacterial activity. For example, the iron oxidation ability of *T. ferrooxidans* was reduced by about 36% at 400 mg dm^{-3} of thiourea concentrations. The iron oxidation in the sterile controls, related to Figure 4 and 5 experiments, was less than one per cent, so these are not shown in the above figures. Despite the reduction in bacterial activity, the adaptation studies to thiourea are continuing and the results of these experiments will be reported elsewhere. For the present, it is believed that successive transfer of these bacteria to increased levels of thiourea may result in strains tolerant to thiourea concentrations high enough to enable the recovery of gold from sulphide substrates in one step.

SUMMARY

It is recognized that one of the most promising applications of biohydrometallurgy is the treatment of precious metals-containing resources (70). Most of the studies deal with the biological preoxidation of pyritic (40,41,71,72) and arsenic (51,54-57,73) ores and concentrates. The present study has demonstrated the potential for the development of a process whereby the biooxidation of sulphide-bearing gold ore is carried out in the presence of gold extractant, thiourea. This could simplify the precious metal extraction processes from refractory ores and, as a consequence, improve the economics of these processes. The preliminary data on adaptation of *T. ferrooxidans* to thiourea are encouraging.

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TABLES

Table 1 - Solubilization of gold from pyrite substrate by *Thiobacillus ferrooxidans*

Stationary experiments No.	Gold extraction g dm^{-3}	Shake flask experiments No.	Gold extraction g dm^{-3}
1	none	1	60
2	150	2	480
3	270	3	490
4	390	4	520

Table 2 - Leach parameters after 15 to 20 hours of leaching of gold arsenic concentrate by *Thiobacillus ferrooxidans*

pH	1.7
Eh	600 to 750 mV
Fe (total)	8.0 to 15.0 g dm^{-3}
As (total)	6.0 to 12.0 g dm^{-3}
Rate of Fe^{2+} oxidation	2.0 to 5.0 g cm^{-3} h^{-1}
Cell concentration	2.0 to 5.0 g dm^{-3} (wet weight)
Temperature	28 to 30°C

Table 3 - Typical leach results: Pyritic zinc sulphide waste leaching by *Thiobacillus ferrooxidans*

	Extractions (%)	
Elements	Inoculated	Sterile control
Cu	68.7	0.6
Zn	53.6	1.1
Fe	62.9	0.8
Pb	0.3	0.1
Au	0.4	0.0
Ag	0.1	0.0

FIGURES

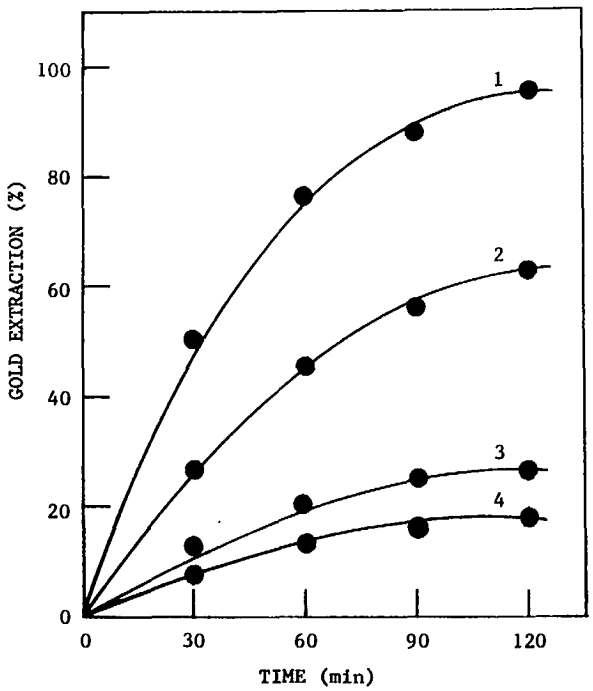

Fig. 1 - Extraction of gold from pyritic zinc sulphide flotation waste. 1 = bacterial leach residue plus thiourea and oxone; 2 = bacterial leach residue plus thiourea; 3 = as received pyritic zinc sulphide flotation waste plus thiourea and oxone; 4 = as received pyritic zinc sulphide flotation waste

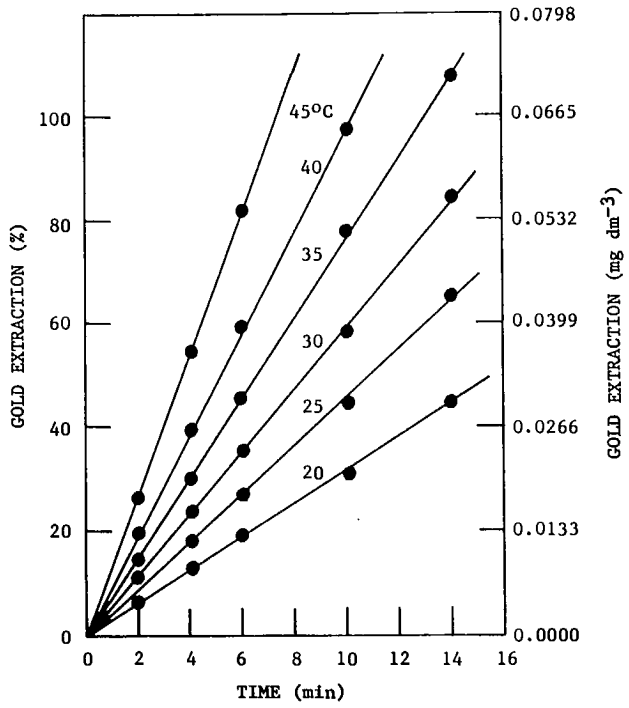

Fig. 2 - Effect of temperature on the initial rate of gold extraction

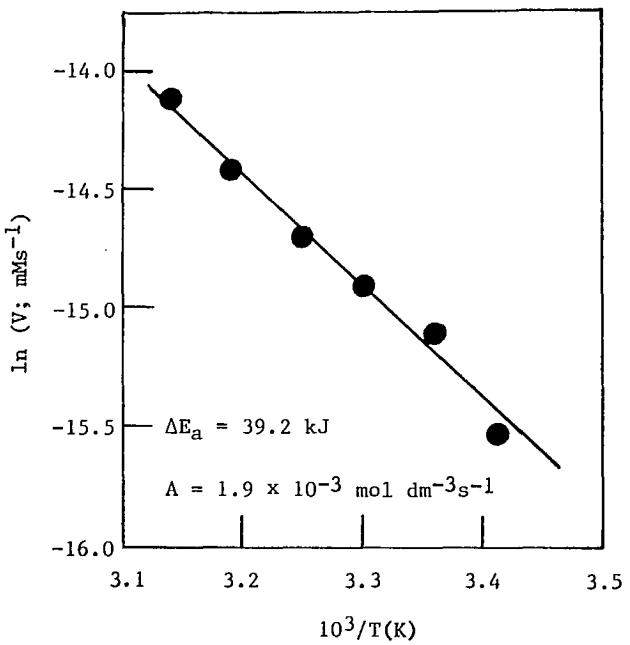

Fig. 3 - Arrhenius plot

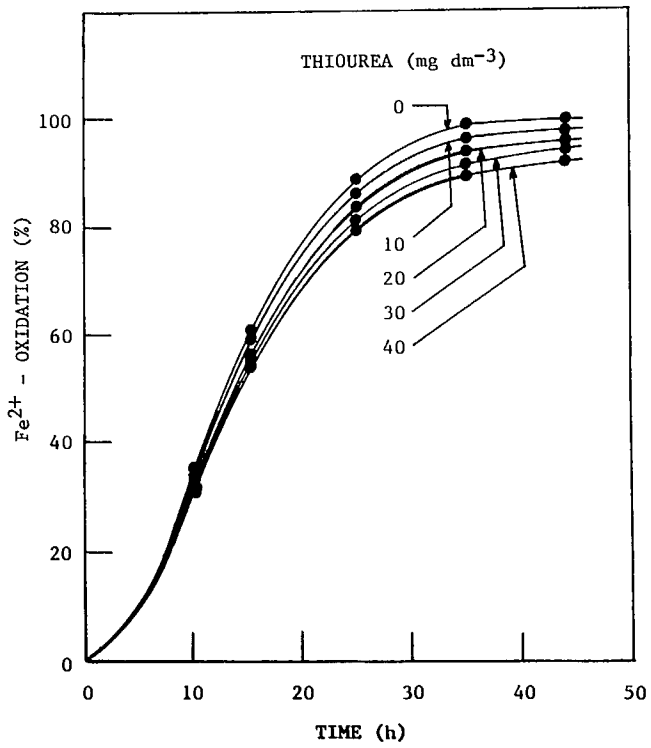

Fig. 4 - Adaptation of *Thiobacillus ferrooxidans* grown on ferrous ion to increased concentrations of thiourea

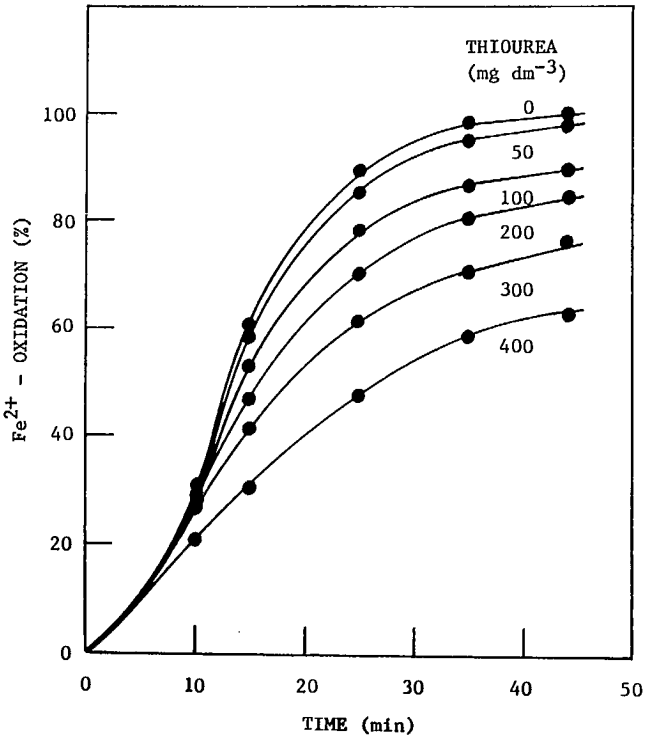

Fig. 5 - Effect of thiourea on *Thiobacillus ferrooxidans* grown on ferrous ion in the presence of 40 mg dm^{-3} thiourea

SESSION I: PAPER 2

DESTRUCTION OF CYANIDE FROM GOLD MILL EFFLUENTS USING ENZYMES

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ABSTRACT

A large number of anions and cations are present in gold mill effluents, and it is necessary to evaluate the potential for these ions to interfere with the analytical methods employed.

A number of metal ions were found to interfere with the analysis of both formamide and cyanide. The following metals and metal complexes were found to interfere with the analysis of cyanide: $K_3Fe(CN)_6$, $K_4Fe(CN)_6$, $FeCl_3$, $ZnCl_2$, $NiCl_2$, and $CuSO_4$. Formamide, $NaNO_3$, $(NH_4)_2SO_4$, and $NaNO_2$ at concentrations of 100 mM did not affect the determination of cyanide. The following compounds were found to interfere with the determination of formamide: $HgCl_2$, $CuSO_4$, $K_3Fe(CN)_6$, $K_4Fe(CN)_6$, and $FeCl_3$.

Preliminary results indicate that the ICI immobilized fungi may further metabolize formamide to NH_3 and CO_2. The ability of a number of cyanide-tolerant bacteria isolated from sewage effluent to degrade cyanide was found to be minimal. However, a *Bacillus* sp. isolated from soil was able to degrade 17.4 μ mol CN^-/mL/h. Preliminary results indicate that CANMET's isolate (*Bacillus* sp.) may have more promise than the ICI product for the degradation of cyanide.

SESSION I: PRÉSENTATION 2

ÉLIMINATION DES CYANURES CONTENUS DANS LES EFFLUENTS DES USINES DE CONCENTRATION DE L'OR AU MOYEN D'ENZYMES

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RÉSUMÉ

Les effluents des mines de concentration de l'or contiennent un grand nombre d'anions et de cations, et il est nécessaire d'évaluer les possibilités de perturbation par ces ions dans les méthodes analytiques employées.

On a trouvé que certains ions métalliques perturbent le dosage du formamide et des cyanures. On a trouvé que les métaux et les complexes métalliques suivants perturbent le dosage des cyanures: $K_3Fe(CN)_6$, $K_4Fe(CN)_6$, $FeCl_3$, $ZnCl_2$, $NiCl_2$ et $CuSO_4$. À des concentrations de 100 mM, le formamide, le $NaNO_3$, le $(NH_4)_2SO_4$ et le $NaNO_2$ ne perturbent pas le dosage des cyanures. Les composés suivants perturbent dans le dosage du formamide: $HgCl_2$, $CuSO_4$, $K_3Fe(CN)_6$, $K_4Fe(CN)_6$ et $FeCl_3$.

Les résultats préliminaires indiquent que les champignons immobilisés ICI pourraient métaboliser le formamide jusqu'en NH_3 et en CO_2. Certaines bactéries tolérant les cyanures, isolées dans des eaux d'égout, peuvent difficilement dégrader le CN^-. Cependant, une *Bacillus* sp., isolée dans le sol, a dégradé 17,4 ^{M}mol CN^-/mL/h. Les résultats préliminaires indiquent que notre isolat (*Bacillus* sp.) pourrait mieux dégrader le cyanure que le produit de l'ICI.

DESTRUCTION OF CYANIDE FROM GOLD MILL EFFLUENTS USING ENZYMES

INTRODUCTION

Cyanidation followed by zinc precipitation is the major process used for gold recovery in Canadian mills. The gold concentrate is treated with lime and sodium (or calcium) cyanide, and the mixture is then contacted in a series of countercurrent decantation cells. The mixture is then filtered and the supernatant is treated with zinc to precipitate the gold (1). Approximately 50% of the gold-free or barren solution is discharged from the mill to prevent the buildup of zinc and other metals in the circuit. The tailings slurry contains appreciable concentrations of cyanide. In gold mill effluents cyanide can exist as free cyanide, as metal-cyanide complexes, and as insoluble cyanide compounds. Barren solutions contain total cyanide levels of 40-750 ppm and tailings repulp streams contain 13-280 ppm of cyanide.

Both chemical and biological techniques have been used for the treatment of gold mill effluents (2). The best characterized technique for the treatment of cyanide-containing wastes is alkaline chlorination. The destruction of cyanide by alkaline chlorination may be accomplished by means of chlorine gas, calcium hypochlorite, or sodium hypochlorite. A process has been developed by Inco that involves the addition of SO_2 and air in the presence of a copper catalyst. Cyanide effluents can also be acidified to allow volatilization of HCN, followed by reneutralization. Various adsorption procedures such as ion exchange, ion flotation, and activated carbon have been considered. Most of the chemical techniques have a number of problems: (1) complex cyanides are not degraded; (2) residual cyanide levels can still be too high (>0.5 ppm); and (3) other toxic reaction products can be formed.

Biological treatment of cyanide-containing wastes has been considered as an alternative to chemical treatments. Laboratory and pilot-plant studies have been carried out with trickling filters (3), activated sludge digesters (4), extended aeration (5), fluidized-bed reactors (6), rotating biological contactors (7), and immobilized enzymes (8). At present, Homestake Mining Company is using rotating biological contactors for the detoxification of gold mill effluent (7). In their process, free and complex cyanides as well as thiocyanates are degraded to ammonia, which is subsequently biologically oxidized to nitrate. The process removes toxic metals by biosorption, and reduces the effluent cyanide concentration to very low levels (9-11). A mutant strain of *Pseudomonas paucimobilis* capable of degrading cyanide is an important feature of this process (12). The main disadvantages of this process are: (1) high capital cost; and (2) cyanide concentrations above 200 ppm are inhibitory to the organisms utilized.

Immobilized fungal cells have also been considered for the degradation of cyanide (8); ICI Ltd. is currently producing immobilized cells for this purpose. A number of organisms produce an enzyme (cyanide hydratase or formamide hydrolyase) (Fig. 1) that hydrolyzes cyanide to formamide (13,14). Subsequent metabolism of formamide to ammonia and CO_2 occurs very readily (14). ICI has selected a *Fusarium* sp. that has high cyanide hydratase activity and does not produce mycotoxins. The fungal biomass is extruded, pelletized, and irradiated to kill the fungal cells and any contaminating bacterial cells so that microbial decomposition of the fungal biomass is prevented. ICI proposed to

use a fluidized-bed reactor for treatment of large volumes of effluent. The advantages of this system are: (1) low capital cost; (2) initial cyanide concentrations of up to 8000 ppm can be detoxified; and (3) no toxic reaction products are formed. The disadvantages of this system are: (1) the immobilized cells have limited stability; (2) the enzymes will not hydrolyze metal-cyanide complexes; and (3) the technology has not been proven beyond laboratory-scale experiments. The objectives of this study are: (1) to evaluate the potential for a number of anions and cations to interfere with the analytical methods employed in this study; (2) to determine the effectiveness of the immobilized fungal cells produced by ICI for the degradation of cyanide, and to determine the degradation rate of cyanide by this preparation as well as its stability; and (3) to isolate other organisms and determine their ability to degrade cyanide.

MATERIALS AND METHODS

Cyanide was determined using sodium picrate and measured spectrophotometrically at 490 mm (14). Formamide was oxidized using alkaline hydroxylamine to hydrozamate, the concentration of which was measured spectrophotometrically at 540 mm as the ferric chloride hydroxamate complex (14). The effect of various inorganic ions on the determination of both cyanide and formamide was evaluated. The effect of various inorganic compounds on the determination of three levels of formamide (20, 40, and 100 ppm) and two levels of cyanide (5 and 12.5 ppm) was investigated.

In order to assay the enzyme activity of the ICI immobilized fungal cells, 50 mg of the fungus was suspended in 10 mL of 100 mM TRIS buffer (pH 8.5) at 4°C. After 20 min, the fungal material was ground in a glass tissue grinder and stored as a suspension at 4°C until used. Cyanide hydratase assays were carried out by adding 100 µL of the fungal suspension to 1 mL of substrate solution (100 mM KCN in 100 mM TRIS buffer, pH 8.5). The solutions were incubated for 10 to 20 min at room temperature, and then 200 µL of the reaction mixture was taken for formamide analysis. An additional 5-10 µL aliquot of the sample was used to determine residual cyanide.

To obtain cyanide-resistant organisms from various locations, two media containing the following levels of cyanide (0, 0.5, 1.0, 10.0, and 50 mM) were used. A minimal salts medium (15) was used for the isolation of cyanide-resistant bacteria and Sabourauds medium (containing sucrose instead of glucose supplemented with 20 units of penicillin and 20 µg of streptomycin per mL) was used for the isolation of cyanide-tolerant fungi. Two samples were taken from the primary treatment section of the Ottawa Municipal Sewage treatment plant and plated on the previously described media.

The cyanide-degrading ability of the following isolates was determined: 20 bacteria isolated from the sewage treatment plant (9 on 10 mM KCN plates and 11 on 50 mM KCN plates); a culture of *Pseudomonas paucimobilis* (ATCC #39204); and a culture of a *Bacillus* sp. To determine the cyanide-degrading ability of the cultures, 10 mL of log phase culture was centrifuged, resuspended in 1.0 mL of 10 mM phosphate buffer (pH 8.0), and 100 µL was added to the substrate solution. The samples were incubated for 2 h in the substrate solution (100 mM KCN, 100 mM phosphate buffer, pH 8.0), and then the residual cyanide concentrations were determined.

RESULTS

A number of metal ions interfere with the analysis of both formamide and cyanide. The following compounds at a concentration of 100 mM had no effect on the determination of formamide: $(NH_4)_2SO_4$, $ZnCl_2$, $NaNO_3$, $NaNO_2$, and KCN (Table 1). No interference was observed in the presence of $CuSO_4$ at 20 mM but interference was observed at 100 mM $CuSO_4$. The following compounds at various concentrations were found to interfere with the measurement of formamide: $k_3Fe(CN)_6$, $K_4Fe(CN)_6$, $NiCl_2$, $FeCl_3$, and $HgCl_2$ (Table 2). The following metals and metal complexes at a concentration of 20 mM were found to interfere with the analysis of cyanide: $K_3Fe(CN)_6$, $K_4Fe(CN)_6$, $FeCl_3$, $ZnCl_2$, $NiCl_2$, and $CuSO_4$ (Table 3). In contrast, formamide, $NaNO_3$, $(NH_4)_2SO_4$, and $NaNO_2$ at concentrations of 100 mM did not affect the determination of cyanide.

The rate of conversion of cyanide to formamide by the ICI immobilized fungal cells was initially quite high, but decreased with time (Fig. 2). The total formamide and cyanide remaining after hydrolysis was significantly lower than the cyanide present initially (Table 4). Bacteria were isolated on both the minimal salts medium and the Sabourauds agar. No fungi were isolated on either medium. Greater numbers of bacteria were isolated in the presence of cyanide on the Sabourauds medium than on the minimal salts medium (Table 5). The ability of 20 selected isolates to degrade cyanide was minimal for both induced (0.5 mM CM^- added mid-log phase) and non-induced cultures. The cyanide degradation rates varied from 0 to 7.6 μ mol CN/mL/h for the isolates. The ability of the *Pseudomonas paucimobilis* (ATCC #39204) isolate to degrade cyanide was also very poor. However, a strain of *Bacillus* sp. isolated from soil was able to degrade 17.4 μ mol CN^-/mL/h.

DISCUSSION

Gold mill effluents contain a large number of anions and cations, particularly metals and metal complexes. It was necessary to first determine the potential for these ions to interfere with the analytical procedures employed in this study. Most of the ions interfere with the cyanide and formamide determinations at concentrations above those encountered in gold mill effluents. Only $k_3Fe(CN)_6$ or $K_4Fe(CN)_6$ are likely to interfere with formamide or cyanide analysis. The incomplete recovery of cyanide and formamide during the hydrolysis of cyanide by the ICI immobilized fungal product suggests that cyanide has been converted to other products as well as formamide. Although the irradiation treatment has killed the fungal cells, there is likely sufficient amidase activity to degrade formamide to formate and ammonia. We have found the cyanide hydratase activity of the ICI immobilized cells to be unstable and sensitive to the enzyme assay technique (grinding, temperature, and length of storage time).

The decrease in formamide production with time (Fig. 2) also implies that the cyanide hydratase may be inactivated during the assay. In the opinion of the authors, the data obtained to date suggest that when the dried fungal preparation is rehydrated, proteolytic enzymes in the cells may be able to degrade the cyanide hydratase.

Because of the limited nutrient potential of the minimal salts medium, few cyanide-tolerant organisms were able to be isolated. In contrast, because of the rich organic nutrients in Sabourauds medium, a greater number of bacteria

were able to tolerate high cyanide levels. Although Sabourauds medium contains antibiotics to restrict the growth of bacteria, it is very likely that a large number of antibiotic-resistant bacteria will be found in primary sewage.

The addition of cyanide to a culture of *Gloeocercospora sorghi* resulted in an increase in the cyanide hydratase activity of the organisms (16). This phenomenon is known as enzyme induction. Many of the sewage isolates were able to tolerate high levels of cyanide but they were not able to readily metabolize it, even when the cultures were induced by the addition of cyanide. Thus, cyanide degradation does not appear to be a resistance mechanism for these bacteria. However, the *Bacillus* sp. degrades cyanide very readily, even when not induced, and certainly deserves further study.

In the authors' opinion, the ICI immobilized fungal product may not be suitable for industrial applications for the following reasons: the immobilized cells have a short half-life and would have to be frequently replaced; the cells do not degrade metal-cyanide complexes; and the fungal enzyme may be inhibited by metal ions. To date, *Bacillus* sp. appears to be the most effective organism for the degradation of cyanide. Future studies on the biological detoxification of cyanide will investigate two aspects: (1) The evaluation of the ICI product will be completed; and (2) further studies will be carried out on the *Bacillus* sp. with respect to its suitability for the degradation of cyanide, either in a rotating biological contactor or within a tailings pond.

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TABLES

Table 1 - Effect of various compounds on the spectrophotometric determination of formamide

Interfering compounds and concentrations	Added formamide concentration ppm	Measured formamide concentration ppm
$(NH_4)_2SO_4$ (100 mM)	20	19
	40	46
	100	106
$ZnCl_2$ (100 mM)	20	20
	40	38
	100	94
$NaNO_3$ (100 mM)	20	22
	40	42
	100	106
$NaNO_2$ (100 mM)	20	18
	40	35
	100	109
KCN (100 mM)	20	22
	40	44
	100	104
$PbCl_2$ (1.0 mM)	20	21
	40	42
	100	98

Table 2 - Effect of various compounds on the spectrophotometric determination of formamide

Interfering compounds and concentrations	Added formamide concentration ppm	Measured formamide concentration ppm
$K_3Fe(CN)_6$ (20 mM)	20	>300
	40	"
	100	"
$K_4Fe(CN)_6$ (20 mM)	20	"
	40	"
	100	"
$NiCl_2$ (20 mM)	20	19
	40	35
	100	81
$FeCl_3$ (20 mM)	20	21
	40	41
	100	89
$HgCl_2$ (2.0 mM)	20	144
	40	170
	100	243
$CuSO_4$ (20 mM)	20	27
	40	44
	100	96
	20	68
	40	103
	100	146

Table 3 - Effect of various compounds on the spectrophotometric determination of cyanide

Interfering compounds and concentrations	Added formamide concentration ppm	Measured formamide concentration ppm
Formamide (10 mM)	5.0	4.3
	12.5	14.3
$NaNO_3$ (100 mM)	5.0	4.8
	12.5	14.1
$NaNO_2$ (100 mM)	5.0	4.5
	12.5	14.4
$(NH_4)_2SO_4$ (20 mM)	5.0	4.6
	12.5	13.3
$(NH_4)_2SO_4$ (100 mM)	5.0	5.5
	12.5	16.6
$PbCl_2$ (20 mM)	5.0	4.2
	12.5	12.2
$PbCl_2$ (100 mM)	5.0	2.7
	12.5	11.8
$K_3Fe(CN)_6$ (20 mM)	5.0	1.1
	12.5	3.2
$K_4Fe(CN)_6$ (20 mM)	5.0	7.1
	12.5	16.5
$HgCl_2$ (2.0 mM)	5.0	0.7
	12.5	1.4
$FeCl_3$ (20 mM)	-	precipitate
$NiCl_2$ (20 mM)	-	"
$ZnCl_2$ (20 mM)	-	"
$CuSO_4$ (20 mM)	-	"

Table 4 - Formamide and cyanide concentrations subsequent to cyanide hydrolysis by the ICI immobilized fungi (initial cyanide concentration 100 & moles/mL, 2 h assay time)

Cyanide μ mol/mL	Formamide μ mol/mL	Cyanide + formamide μ mol/mL
62.4	12.9	75.3

Table 5 - Numbers of cyanide-tolerant bacteria isolated from primary sewage

Medium	KCN concentration nM	Colony forming Units/m sample 1	sample 2
Minimal salts	0	3.11×10^7	1.46×10^6
	0.5	1.80×10^7	2.64×10^5
	1.0	2.16×10^6	1.52×10^5
	10.0	6.80×10^4	3.51×10^4
	50.0	5.50×10	0
Sabourauds	0	2.30×10^6	5.16×10^4
	0.5	2.78×10^5	4.67×10^4
	1.0	3.22×10^5	4.01×10^4
	10.0	3.28×10^5	1.75×10^4
	50.0	3.32×10^3	7.70×10

FIGURES

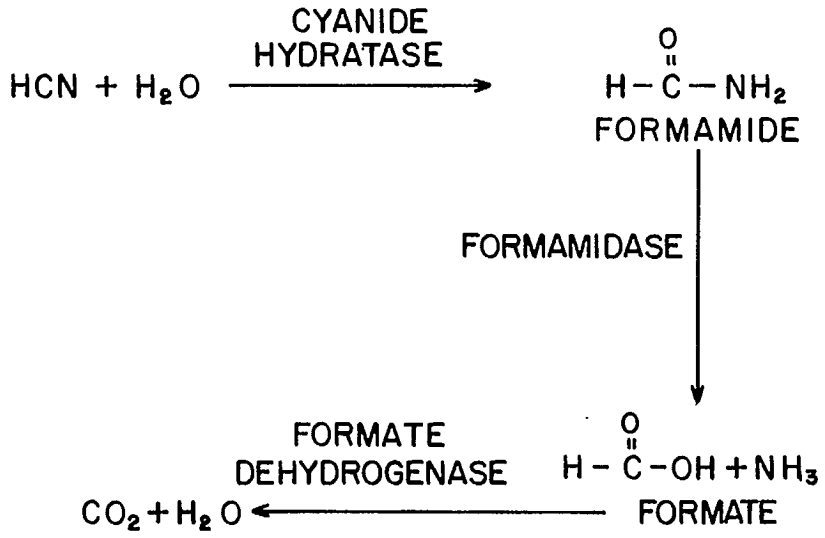

Fig. 1 - The pathway for the detoxification of cyanide by *Fusarium moniliforme*

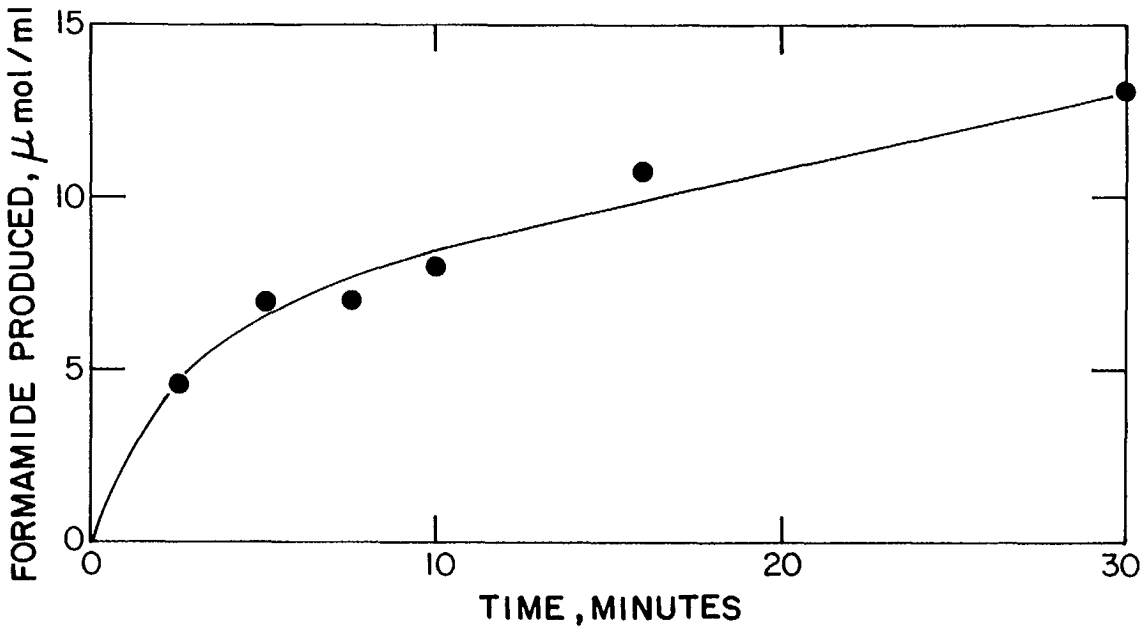

Fig. 2 – The rate of release of formamide from cyanide by the ICI immobilized enzyme preparation

SESSION I: PAPER 3

FLOWSHEET DESIGN, PROCESS CONTROL, AND OPERATING STRATEGIES IN THE BIOOXIDATION OF REFRACTORY GOLD ORES

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ABSTRACT

The use of biooxidation for the pretreatment of refractory sulphide gold ores and concentrates is now recognized as having immediate commercial potential. A wide range of feed materials, from low-grade tailings to high-grade concentrates with diverse mineralogies, have been shown to be amenable to the technology in laboratory batch testing.

This paper discusses the important considerations in the design of continuous pilot and commercial biooxidation plants and in the strategy for process control.

SESSION I: PRÉSENTATION 3

SCHÉMA DE FONCTIONNEMENT, CONTRÔLE ET STRATÉGIES DE FONCTIONNEMENT DU PROCÉDÉ DE BIOOXYDATION DES MINERAIS D'OR RÉFRACTAIRES

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RÉSUMÉ

La biooxydation comme procédé de pré-traitement des minerais et des concentrés d'or sulfurés réfractaires est maintenant reconnue comme ayant des possibilités commerciales immédiates. On a montré qu'une vaste gamme de matières premières allant de résidus de basse qualité à des concentrés de grande qualité ayant différentes compositions minéralogiques se prêtent au traitement discontinu en laboratoire par cette technologie.

La présente communication traite de facteurs importants dont il faut tenir compte dans la conception des usines pilotes et commerciales de biooxydation en continu et dans la stratégie de régulation du procédé.

FLOWSHEET DESIGN, PROCESS CONTROL, AND OPERATING STRATEGIES IN THE BIOOXIDATION OF REFRACTORY GOLD ORES

INTRODUCTION

Biooxidation for the pretreatment of refractory sulphide gold ores and concentrates has been extensively demonstrated on a laboratory bench scale. However, the design and operation of continuous bioleaching circuits in stirred reactors will require some innovative engineering for successful plant-scale application. It is the intent of this paper to indicate some of the important process design considerations and to identify critical bioleach operating parameters for continuous biooxidation of refractory gold-bearing ores and concentrates.

Flowsheet 1 (Fig. 1) indicates the conceptual location of the bioleach step in commercial operations, along with other sulphide oxidative pretreatments. The development of a bioleach system as well as the subsequent flowsheet design and control strategy will depend greatly on the feed source, mineralogy, and the degree of sulphide oxidation required.

Treatment of ores differs significantly from high-grade sulphide concentrate. A wide range of ore and concentrate types is amenable to biooxidation and can exhibit highly variable bioleaching characteristics. For example, feed materials for bioleach systems recently developed at Coastech, in which a greater than 30% increase in gold recovery over conventional processing was demonstrated, are shown in Table 1.

PROCESS DESIGN CONSIDERATIONS

Flowsheet 2 (Fig. 2) represents a generic bioleach-extraction process for the treatment of a refractory gold ore or concentrate. Discussions herein will be limited to the application of biotechnology in preoxidizing refractory sulphide hosts to enhance gold extraction by subsequent cyanidation of the bioleach residue. The discussions will also be limited to those applications using conventional stirred tank reactors. Flowsheet 2 can be modified considerably depending on actual bioleach and precious metals hydrometallurgy.

CRITICAL BIOLEACH PARAMETERS

Following baseline batch bioleach amenability testing and bacterial strain development, the critical bioleach parameters can be defined in continuous bench-scale bioleach reactors. For pilot and plant-scale design considerations, the critical bioleach variables listed in Table 2 must be defined.

ORE VERSUS CONCENTRATE

Where sulphide and/or gold concentration ratios are poor (<4:1) or gold recovery to sulphide concentrate is unacceptable by flotation and/or gravity methods, it is often necessary to treat the ore rather than a sulphide concentrate. Successful biooxidation of ore would require sufficient oxidizable sulphur to overcome the natural acid consumption of the ore. Otherwise, considerable makeup acid would be required to maintain the pH/Eh of the bioleach system. The additional expense to treat all of the ore can be easily justified if only small gold losses occur during preconcentration.

Treatment of ore offers some advantages and disadvantages; these are listed in Table 3.

Typically, biological pretreatment is considered for sulphide concentrates, mainly due to the economics of treating a smaller volume, assuming similar overall gold extraction when compared with treating the ore.

SULPHIDE SELECTIVITY

Where batch bioleach testing indicates that gold extraction as a function of sulphide oxidation is not linear, a high potential for selective sulphide oxidation is evident. Generally, fine gold is disseminated in refractory pyrite hosts along phase change boundaries. These phase change boundaries, where the Fe:S ratio alters slightly, are more readily oxidized. Biological oxidation takes advantage of this phenomenon resulting in, at times, highly selective sulphide oxidation to enhance precious metals recovery while leaving much of the pyrite host unchanged (1).

Selective oxidation may also occur if gold is associated with a particular sulphide mineral such as arsenopyrite. In this case, faster oxidation kinetics can result in the liberation of gold from arsenopyrite, while the often dominant sulphide pyrite is only partially oxidized.

Selective sulphide oxidation is advantageous in:

- reducing bioleach residence time;
- reducing aeration (oxygen) requirements;
- reducing acidic by-product for neutralization and disposal;
- reducing cooling water requirements;
- reducing or eliminating bioleachate solution exchange requirements to maintain high biooxidation rates.

SCALEUP AND PLANT-SCALE DESIGN CONSIDERATIONS

Recent pilot-plant studies at Equity Silver Mines indicated that the biohydrometallurgy and precious metals response of refractory sulphide concentrates scales up predictably from a bench-scale continuous apparatus to a 2 tpd continuous pilot circuit (2). The most critical effects of scale occurred in the design of pulp density, aeration rate, and residence time. These variables are highly interdependent and must be optimized on a reasonably large scale for proper plant-scale design.

Innovative reactor design is required in large reactors to make use of low pressure air (<70 kPa). Highly efficient air diffusion mechanisms must be employed. Effective mixing in the presence of large quantities of air must be addressed and a height:diameter ratio of the reactor <1.0 is recommended for this purpose.

Bioleaching and associated peripheral processing is conducted in an acid environment at a relatively high oxidation potential. Process equipment design must consider the corrosive nature of the bioleach system.

If a high degree of sulphide oxidation is required, significant pulp heating will occur due to sulphur oxidation. Consequently, a pulp temperature control

(cooling) method must be considered for proper reaction control. Although pilot testing (2) did not provide a meaningful indication of heat balance, recent design studies at Coastech have shown that even very large (>2000 m^3) leach tanks can be economically cooled in specific cases.

A high degree of sulphide oxidation will produce high levels of ferric iron and sulphate in solution. To maintain a reasonable biooxidation rate it might be necessary to remove leachate from the circuit for exchange with fresh solution. Continuous solution exchange is preferred, although intermittent exchange might be more practical on a large scale. Solution exchange can be defined during continuous piloting.

The overall water balance is important to define early in the design stages. Bioleaching can be inhibited by fouled water sources, especially cyanide solutions. It is essential to test a continuous bioleach circuit in locked cycle if process water is to be recycled. Sufficient cooling and dilution water must be available to satisfy the bioleach circuit. Internal solution recycle should be practised where possible.

PROCESS LIMITATIONS

Biological preoxidation offers significant economic advantages, and often metallurgical advantages, over roasting and pressure leaching for refractory gold processing. However, there are limitations to the technology. In the opinion of the authors, the most salient limitations are:

- ores that exhibit high acid consumption and the gold cannot be preconcentrated to a sulphide concentrate. It is difficult to maintain the bioleach reaction without significant makeup acid.

- gold and silver bioleachate loading. This is not a common problem and is apparently Eh/pH related. Precious metals can be recovered from the leachate by ion exchange columns, preferably using highly gold-selective resins.

- heat exchange is only a limitation in hot climates where the available cooling water temperature is close to the process temperature. In some cases, refrigeration might be necessary, especially if a high degree of sulphide oxidation is required.

- continuous solution exchange, if required, may be a problem for poor settling or filtering material.

- large volumes of exchange solution for treatment and recycle are generated for high-sulphide material at high-sulphide oxidation levels.

- large volumes of neutralization sludge may be produced from leachate treatment. The long-term stability of some sludges might be difficult to determine.

- air diffusion in finely ground slurries.

Biological pretreatment can generally be applied to most refractory sulphide materials. The most attractive application remains where selective sulphide oxidation can be exploited, such as that demonstrated with arsenopyrite-pyrite assemblages where the refractory gold is intimately associated with the arsenopyrite (2).

PROCESS CONTROL CONSIDERATIONS

BIOMASS MAINTENANCE

Maintenance of the bacterial biomass in the leach reactors is essential for continuous operation. There are a number of methods to do this. Flowsheet 3A (Fig. 3a) indicates a simple series of flow-through reactors. If the residence time of the first reactor is less than the biological doubling rate, then biomass 'washout' will occur. Flowsheet 3B (Fig. 3b) suggests a single large reactor followed by smaller cascading reactors, maintaining the biomass in the first. This might not be particularly practical for large-scale operations.

Flowsheets 3C and 3D (Fig. 3c and 3d) suggest more practical alternatives for biomass maintenance. Flowsheet 3C suggests a number of primary reactors employed in parallel with a distributed feed. This method has been employed successfully on a large pilot scale (2).

Flowsheet 3D suggests recycling the coarse material for further biooxidation as well as recycling some of the biomass. This method was suggested by Lawrence and Gunn (3). This strategy is attractive to maximize the reactor residence time and promotes accelerated bioleaching. However, it is not recommended where selective sulphide oxidation is the objective.

CONTINUOUS PROCESS CONTROL

Flowsheets 4A and 4B (Fig. 4a and 4b) suggest a generic process control strategy for continuous bioleach circuits. The strategy indicates the minimum process information and response required for automatic process control. The bioleachate exchange, if required, is shown separately in Flowsheet 4B for clarity.

The process control strategy suggested herein can be summarized:

Flowsheet 4A

- assumes constant feed rate and particle size;
- residence time is controlled by density and tank bypassing;
- bioleach rate is controlled by aeration rate, temperature, pH/ORP, nutrient addition rate.

Flowsheet 4B

- solution removal rate (by integral vacuum system on each reactor) is controlled by solution pH/ORP;

- treated solution recycle rate is controlled by pulp level in each reactor (note: makeup nutrient is metered to recycled leachate based on flowrate of recycle, Flowsheet 4A);

- excess recycle solution is bled to tailing or can be recycled elsewhere.

The control strategy suggested herein was based on continuous piloting of bioleach systems. Considerable modification of the strategy is expected as more operating experience is gained.

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TABLES

Table 1 - Feed materials for potential plant-scale bioleach systems

Sample	Ag (g/tonne)	Fe (%)	S (%)	As (%)	Sb (%)	Pb (%)	Cu (%)	Zn (%)
A	78.8	21.3	20.6	19.5	0.15	2.32	0.05	11.90
B	10.9	6.2	13.7	0.1	-	-	-	0.02
C	25.6	12.5	12.5	0.2	0.004	0.01	0.02	-
D	34.9	32.4	30.6	11.0	0.06	0.03	2.10	-
E	4.8	37.8	34.4	10.8	0.05	0.02	0.24	0.54
F	22.5	39.8	40.9	10.4	0.077	0.53	0.07	0.59
G	1295.8	33.6	-	0.8	-	-	-	-
H	13.5	19.0	14.0	19.8	-	-	0.08	-
I	112.4	35.8	41.9	7.4	0.07	1.90	1.20	-
J	6.0	14.9	18.0	8.3	0.20	Tr	0.10	-

N.B. Au assays omitted for client confidentiality.

Table 2 - Critical bioleach parameters

Bioleach parameter	Operating range*
- sulphur content	5-42%
- degree of sulphide oxidation	10-100%
- acid consumption of feed	variable
- temperature	30-45°C
- pulp density	10-30% solids
- aeration rate (O_2, CO_2)	0.05-0.50 m^3 air/min/m^3 pulp
- residence time (oxidation rate)	30-150 h
- pH	1.0-1.7
- redox potential	450-680 mV SCE
- nutrients (type and addition rate)	variable
- solution exchange ratio	0-4

*Range based on laboratory and/or pilot-scale continuous operations.

Table 3 - Treatment of ore versus concentrate

Advantage	Disadvantage
- higher overall gold recovery	- higher capital/operating costs for additional pulp volume
- higher operating density	- more difficult solid-liquid separation
- reduced or eliminated solution exchange	- potential acid consumption
- reduced cooling water	- difficulty in justifying fine grinding
- partial acid consumption during biooxidation	- decreased oxygen/CO_2 efficiency

FIGURES

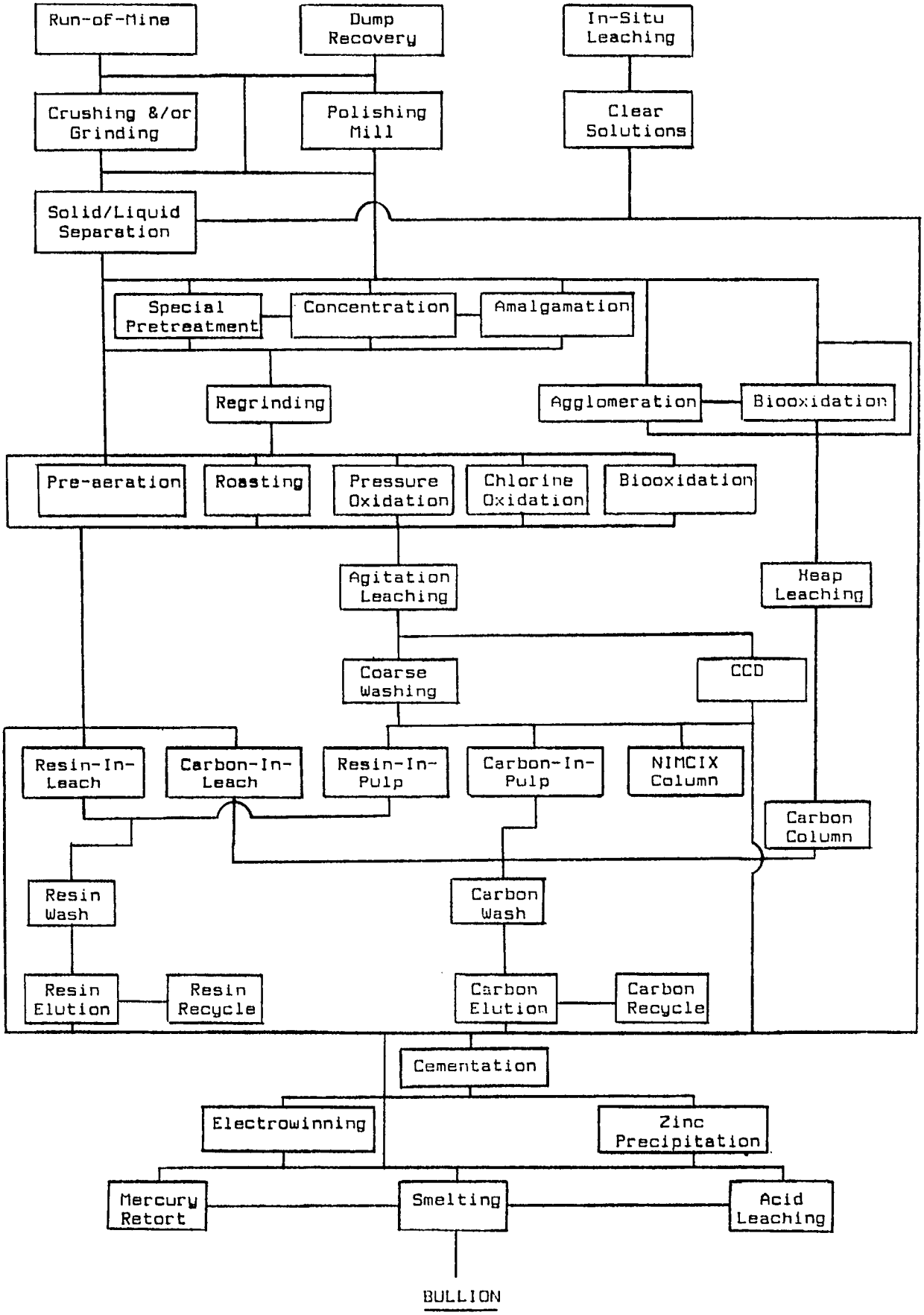

Fig. 1 - Flowsheet 1 - Gold/silver ore-processing alternatives

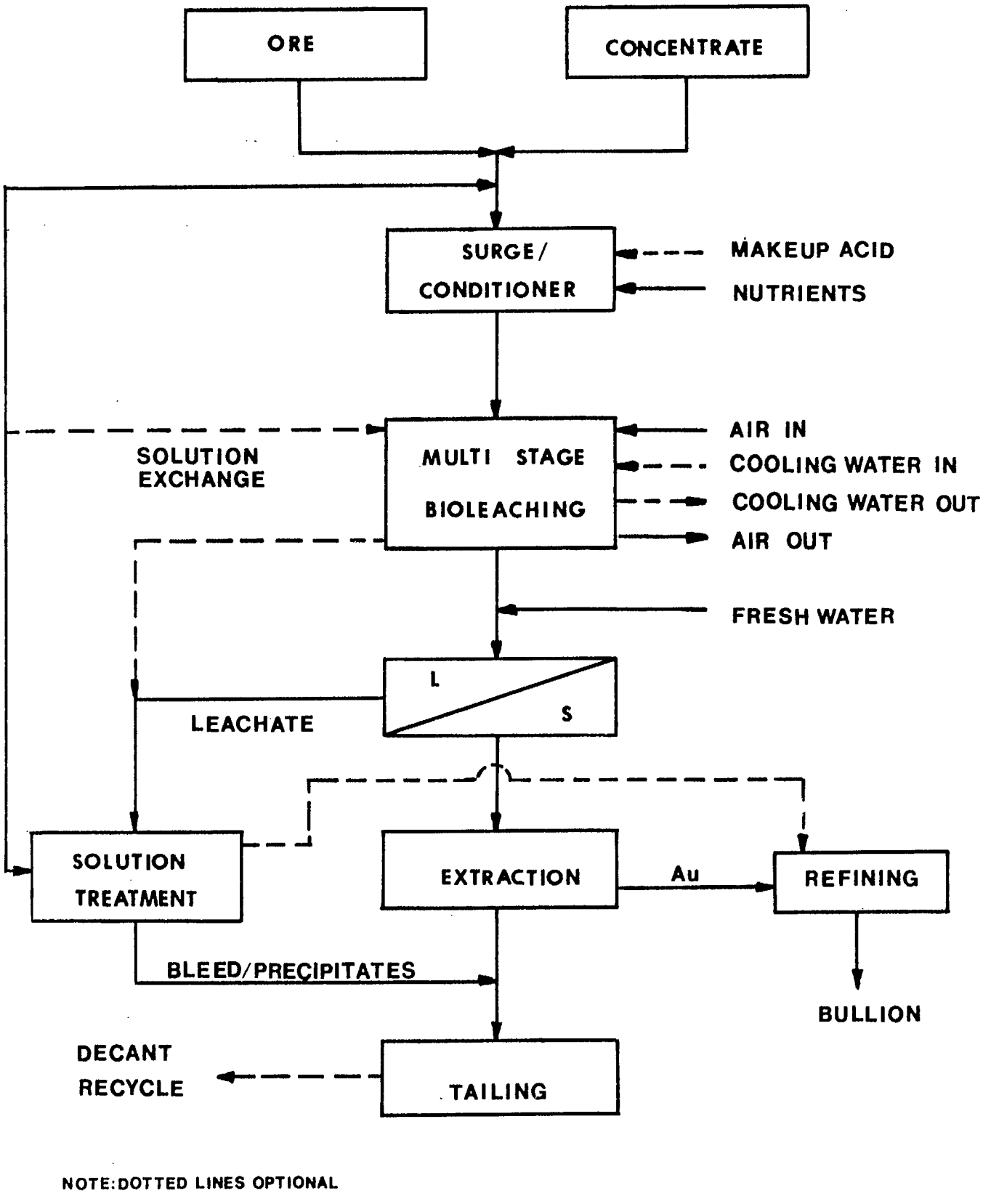

Fig. 2 - Flowsheet 2

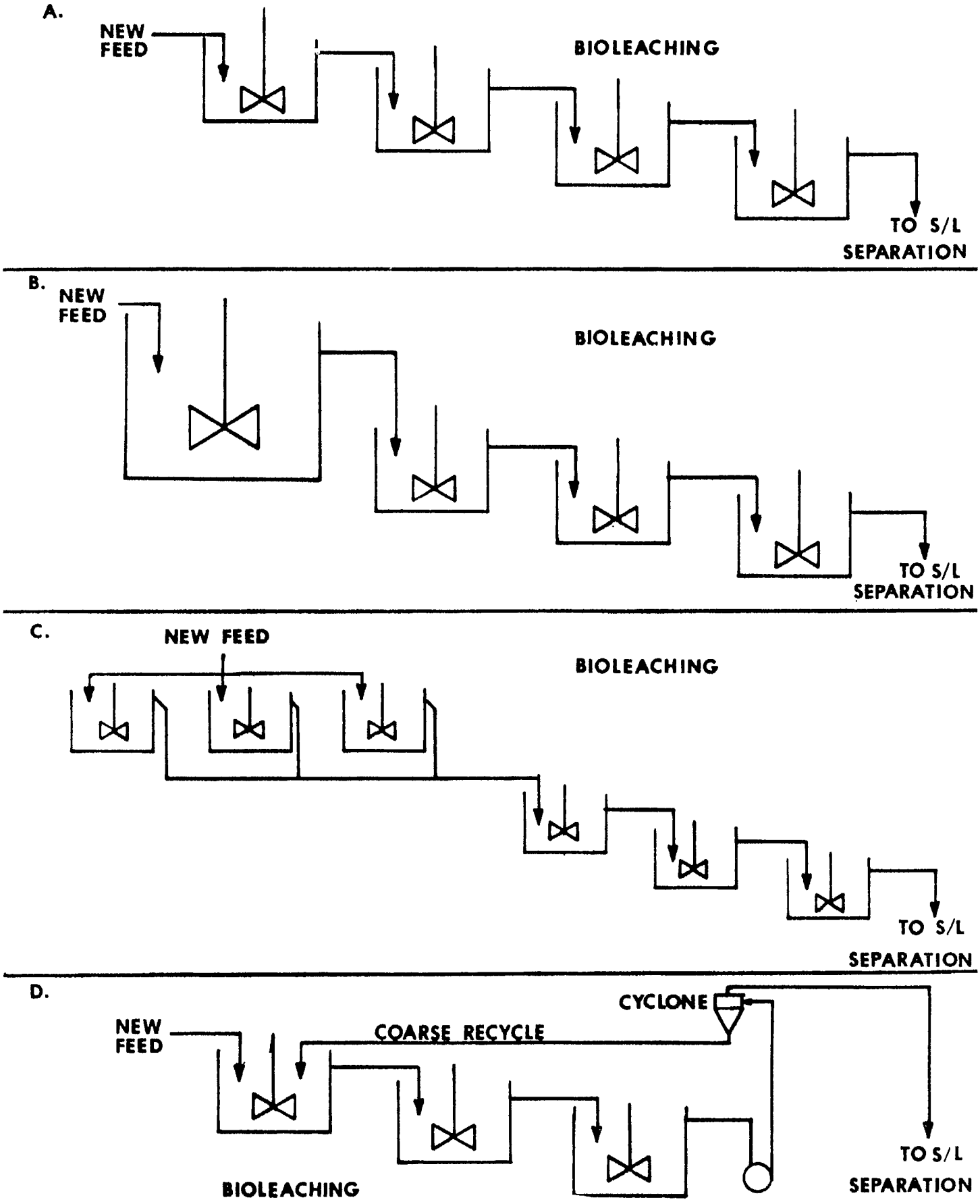

Fig. 3 - Flowsheet 3

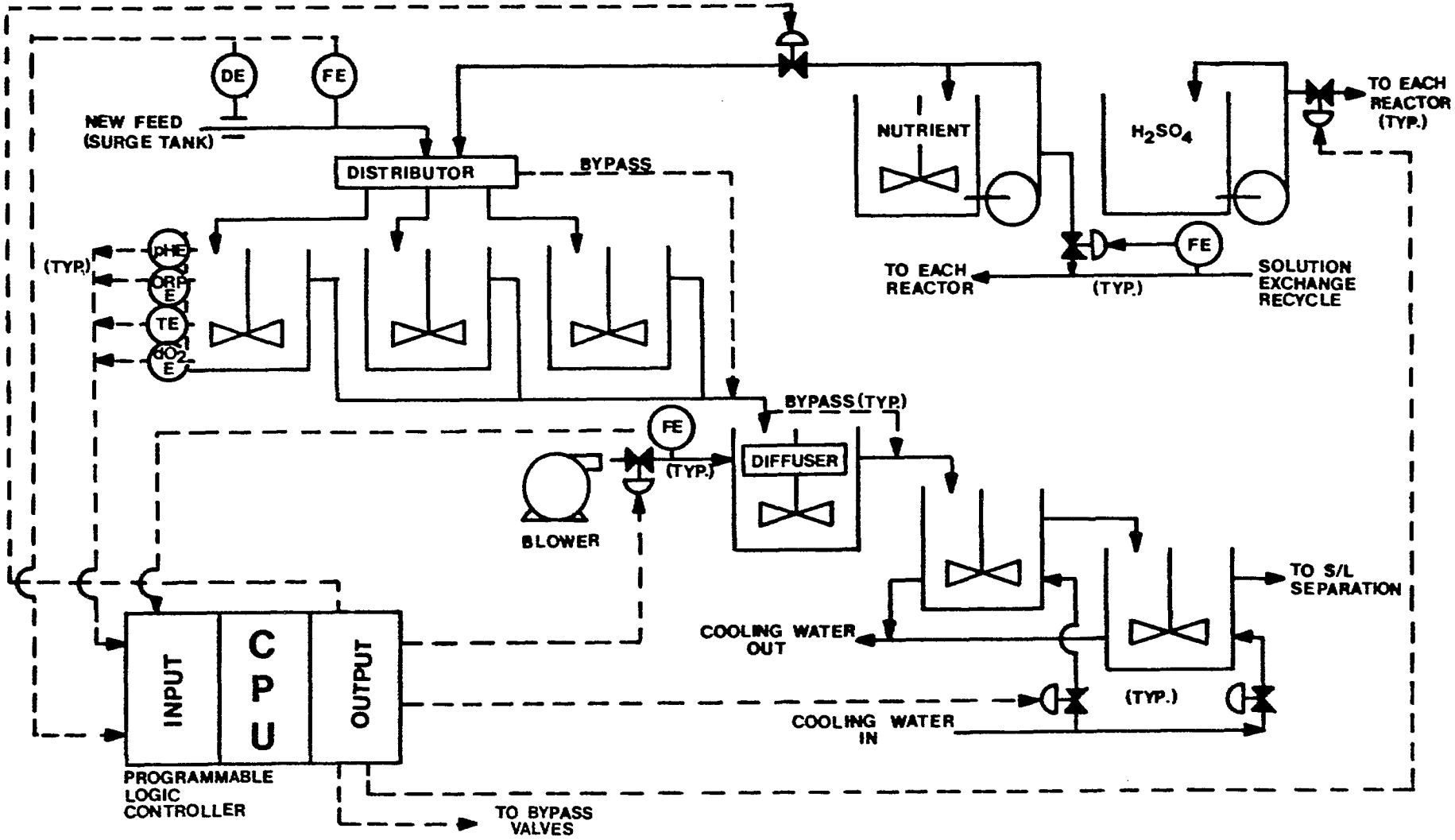

Fig. 4(a) - Flowsheet 4A

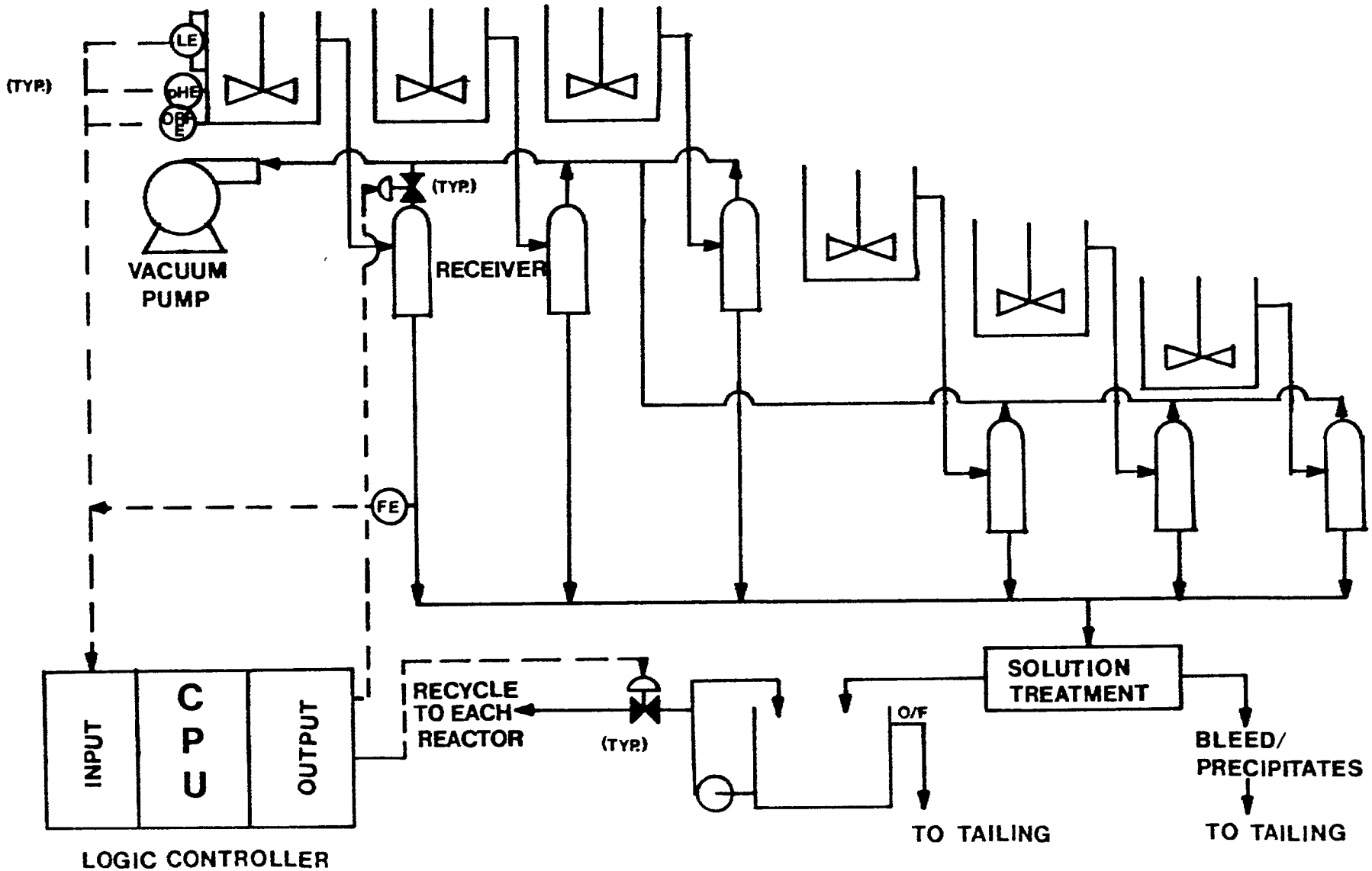

Fig. 4(b) - Flowsheet 4B (pulp flow not shown)

SESSION I: PAPER 4

COMPARATIVE ECONOMICS OF BACTERIAL LEACHING AND ROASTING FOR AN AURIFEROUS PYRITE CONCENTRATE

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ABSTRACT

Many ores are upgraded to produce auriferous sulphide concentrates (principally pyrite) that are not amenable to direct cyanidation. Often these refractory gold-bearing concentrates contain undesirable elements such as arsenic, antimony, etc., that present environmental and/or processing problems. The pretreatment techniques most often considered for processing these concentrates prior to cyanidation include roasting, pressure oxidation, chemical oxidation, preaeration, and recently, bacterial oxidation. In this paper, the technical performance and economics of bacterial preoxidation are compared to the classical roasting approach for treating a specific auriferous pyrite concentrate. The results provide flow sheets, preliminary operating and capital cost estimates, and rates of return for the two processes; they also demonstrate the potential advantages of bacterial preoxidation.

SESSION I: PRÉSENTATION 4

ANALYSE ÉCONOMIQUE COMPARATIVE DE LA LIXIVIATION BACTÉRIENNE ET DU GRILLAGE D'UN CONCENTRÉ DE PYRITE AURIFÈRE

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RÉSUMÉ

Un grand nombre de minerais sont transformés en concentrés sulfurés aurifères (surtout la pyrite) qui ne peuvent être soumis à la cyanuration directe. Il arrive souvent que ces concentrés aurifères réfractaires contiennent des éléments indésirables tels que l'arsenic, l'antimoine, etc., qui posent des problèmes environnementaux ou de traitement. Les techniques les plus utilisées pour le pré-traitement de ces concentrés avant la cyanuration comprennent le grillage, l'oxydation sous pression, l'oxydation chimique, la pré-aération et, plus récemment, l'oxydation bactérienne. Dans la présente communication, nous comparons l'efficacité technique et la rentabilité de la pré-oxydation bactérienne et celles du grillage classique dans le traitement d'un concentré de pyrite aurifère particulier. Les résultats comprennent des schémas de fonctionnement, des estimations préliminaires des coûts d'exploitation et du coût en capital ainsi que des taux de rendement pour les deux procédés, et ils font ressortir les avantages potentiels de la pré-oxydation bactérienne.

COMPARATIVE ECONOMICS OF BACTERIAL LEACHING AND ROASTING FOR AN AURIFEROUS PYRITE CONCENTRATE

INTRODUCTION

The increased attention that gold has received in the last 10 years has prompted mining companies to look for lower grade ore bodies. Many of these low-grade ores do not respond well to conventional direct cyanidation approaches. As free-milling ores decline, gold is sought in refractory environments and this, ultimately, drives up the treatment costs.

There are a number of reasons for gold ores to be termed refractory, most of which can be identified by thorough mineralogical examination. The mineralogical occurrence of gold can, to a great extent, impact the method by which the ore is treated and, consequently, the capital and operating costs. Gold in a refractory ore can often occur as discrete particles locked in a host of silicate or carbonate gangue, or associated with sulphides or sulphosalts. Depending on its particle size, gold can be liberated and recovered by mechanical size reduction such as regrinding. This approach is most effective for gold in the 10-50 micron range. Gold can also be associated with clay, iron hydroxides, or other minerals that tend to coat the gold surface and inhibit the penetration of the cyanide solution, resulting in poor plant recovery. Because of their resistance to cyanide dissolution, gold tellurides and other refractory gold minerals are also responsible for losses to the tailings. For these reasons, it is paramount that the mineralogical occurrence of the gold be thoroughly and accurately determined before selecting a final process flowsheet.

Frequently, gold occurs as finely disseminated, submicroscopic particles in a refractory sulphide host such as pyrite or arsenopyrite, and is often mistaken for being in solid solution with the host sulphide. Because of its finely disseminated nature, gold in this category cannot be ground to liberation, nor is direct cyanidation a pratical means for extracting the precious metal values. Attempts to extract gold by these methods typically result in recoveries in the 20-60% range and in high cyanide consumptions. There are, however, means available for pretreating pyritic gold ores to render them amenable to cyanidation.

Of the methods now in use for preoxidation of sulphide concentrates, the most prominent are autoclave, chemical, preaeration, and roasting. In recent years, bacterial preoxidation has been emerging as an alternative means of treatment for sulphide ores. This paper focuses on two of the treatment alternatives, i.e., roasting and bacterial preoxidation.

The investigation was done on a selected pyrite concentrate that was generated by flotation and with the chemical analysis given in Table 1. A screen distribution of the pyrite showed that the concentrate was 95% minus 74 μ (200 mesh), and almost 80% of the gold and silver was contained in the minus 37 μ (400 mesh) fraction, which accounted for 68% of the sample weight. Another characteristic of this concentrate is that a portion of the gold is locked in silicious gangue, which is unaffected by either roasting or bacterial leaching.

In this paper, the potential of treating a by-product or scavenger concentrate that would otherwise report to the tailings at an existing facility is evaluated. For this reason, mining, milling, disposal, gold recovery, and plant infrastructure are not included. This approach allows a more direct comparison between the two preoxidation methods. In addition to the roasting and bacterial circuits, pyrite flotation and cyanide destruction circuits have been added to each case. All monetary values are given in U.S. dollars.

ROASTING

BACKGROUND

The theory and practice of roasting pyrites as a pretreatment to enhance the recovery of gold by cyanide leaching has been extensively reported and will not be repeated here. The principal objectives of pyrite roasting, whether the roasting is conducted in a rotary kiln, an Edwards horizontal hearth roaster, a Herreschoff-type multiple-hearth roaster, or a fluid-bed reactor, are to:

- liberate the gold by oxidizing the iron sulphides to oxides; typical reactions for pyrite roasting are summarized below.

$$FeS_2 \longrightarrow FeS + S$$
$$S + O_2 \longrightarrow SO_2$$
$$FeS_2 + O_2 \longrightarrow FeS + SO_2$$
$$4\ FeS_2 + 11\ O_2 \longrightarrow 2\ Fe_2O_3 + 8\ SO_2$$
$$3\ FeS_2 + 8\ O_2 \longrightarrow Fe_3O_4 + 6\ SO_2$$
$$3\ FeS + 5\ O_2 \longrightarrow Fe_3O_4 + 3\ SO_2$$
$$4\ FeS + 7\ O_2 \longrightarrow 2\ Fe_2O_3 + 4\ SO_2$$
$$4\ Fe_3O_4 + O_2 \longrightarrow 6\ Fe_2O_3$$
$$2\ SO_2 + O_2 \longrightarrow 2\ SO_3$$

- produce a porous calcine product that is easily attacked by the cyanide solution to permit high extraction of the liberated gold.

A secondary objective in many commercial facilities is to remove or destroy any contaminants that would interfere with the cyanide leach circuit.

Roasting has historically been the technology selected for most pyritic ores that proved to be refractory to standard leaching by cyanides. The attractiveness of roasting as a pyrite pretreatment was substantially increased by the introduction of the fluid-bed reactor, with its inherent advantage of lower cost, better control, greater reliability, and more flexibility for most applications. The flexibility of roasting has been demonstrated by the range of operating conditions (temperature, oxidation level, number of stages, etc.) that have been successfully utilized at various mines to cope with the particular characteristics of the feed. The success of roasting is indicated by the partial list of plants currently operating with a cyanide leach of roasted calcines (Table 2).

Roasters, particularly fluid-bed reactors, are somewhat simple, low-cost devices. However, an overall roaster circuit is a complex and costly facility as the following list of necessary auxiliary operations suggests:

- feed preparation circuit that usually consists of a filter and dryer or slurry system to provide a consistent feed;

- compressed air circuit;

- calcine discharge circuit involving seals, coolers, and transfer of hot, abrasive solids;

- gas discharge circuit that often includes:
  - cyclones
  - ESP's
  - baghouses
  - fans
  - wet scrubbers
  - acid plants.

Obviously, the main contributor to the cost of roasting, both capital and annual operating, is the treatment of the gaseous exhaust. Often hazardous fumes and dusts must be collected, and most countries require the removal of SO_2 prior to gas discharge. The high cost of these control devices has limited the application of pyrite roasting to those mines with sufficient gold to justify the effort or to areas where a substantial by-product credit for sulphuric acid is available.

The flowsheet depicted in Figure 1 was selected for evaluation. The process is fairly standard except perhaps for the inclusions of an acid leach circuit. Early studies had shown that sufficient copper was present in the feed (≈1%) to cause excessive cyanide consumption, if the copper were not removed from the calcine.

EXPERIMENTAL PROGRAM

Roasting

Representative pyrite samples were roasted to produce calcines in a 4-inch diameter fluidized-bed reactor at 600°C, 650°C, and 700°C; 6000 gm of pyrite were treated under essentially steady-state conditions at each temperature. Because of the fineness of the pyrite, the air rate was limited to 1.45 SCFM (1 atm, 70°C) to produce a space velocity of 0.8-0.9 ft/s in the roaster bed. The roaster off-gas varied between 7-8% SO_2, with N_2 and unreacted O_2 providing the balance. The product calcine (bed overflow, cyclone product, and scrubber solids) was collected and blended.

All calcines were generally deep red, indicating nearly complete conversion to hematite. Weight loss and calcine assays were essentially unaffected by the change in temperature. A composite assay for the three runs is presented in Table 3.

Copper Leaching

Approximately 1500 gm of calcine from each fluid-bed test was slurried and leached in H_2SO_4 for 3 h at 15% solids in an agitated, baffled vessel to remove soluble copper. Leaching was conducted at 70°C and at a pH of 2. The final leach slurry was filtered, with the filter solids being washed thoroughly to remove soluble elements, particularly copper.

Although most of the copper in the calcine is water soluble, a hot acid leach was selected to ensure the copper removal and to minimize cyanide consumption. Dissolution of the copper required an average of 22 lb H_2SO_4 per ton of calcine. Copper extraction ranged from 77.6% to 85.9%, with a definite trend toward increased solubility at the lower roasting temperatures (Table 4).

Cyanide Leaching

Three cyanide levels, 2.5 g/L, 0.5 g/L, and 0.25 g/L, were selected for testing. A sample of unroasted pyrite was run at the same time to provide a comparison (Table 5). Each sample was leached in a 24-h bottle test at pH 10.5 (adjusted by adding lime).

The extractions of both gold and silver increased, as expected, with increasing cyanide level. Variations in roasting temperature, however, did not seem to impact the dissolution of gold to the extent originally anticipated. The added gold and silver extractions at the 2.5 g/L cyanide level are seen to be accomplished with a three-fold increase in cyanide consumption for all calcines tested. There appears to be a clear metallurgical advantage in using approximately 0.5 g/L NaCN. Lime (CaO) consumption varied somewhat erratically from 5.3 to 9.1 kg/t for the roasted calcines. The average for the entire data set was 7.3 kg CaO/t.

ECONOMIC CONSIDERATIONS

Capital and operating costs were estimated for treating 150 MTPD of the pyrite defined in Table 1. The cost estimates were prepared assuming a 0.5 g/L cyanide level in solution, and the average recoveries and reagent consumptions reported for the test program (Table 6).

Capital Cost

The roasting experiments suggested that a single 31-ft diameter fluid-bed roaster would be sufficient for 150 MTPD of the pyrite. The roaster was sized for the anticipated 0.3 m/s space velocity. An equipment list was prepared for the cost estimate including the roaster, boiler, cyclones, ESP, and packaged Acid Plant. A total capital cost was computed via a factored estimate based on internal experience with a similar facility and site (Table 7).

Operating Costs

The main contributors to the operating costs of the plant are reagents, energy, labour, and maintenance, with reagent costs being dominant. The reagents include sodium cyanide and lime for gold leaching, chloride for cyanide destruction, and scrap iron for copper precipitation. An operating cost summary, using unit costs for the mine where the pyrite was obtained, is presented in Table 8. The energy, labour, and maintenance costs were estimated from a similar operation.

Revenues

Revenue credits were taken for gold, silver, copper, and any steam produced in the waste heat boilers. No credit was taken for the sulphuric acid sales since the local market barely covers the cost for shipping. Note, however, that no reagent charge for the acid used to leach copper was applied.

Metal payments are based on a 95% payment for the recovered gold and silver, and a 75% payment for recovered copper. The selling prices for the metals were assessed to be $350/tr oz, $5/tr oz,, and $0.60/lb for gold, silver, and copper, respectively. Note that the gold revenue was based on an average mine life grade of 12 g/t.

A credit for steam production was taken for producing 200 million Btu/day of steam. The credit was based on replacing fuel oil at $3.60/L used in the generation of steam. The credit at 150 MTPD is approximately $500 000 per year. A revenue summary is presented in Table 9.

BIOOXIDATION

BACKGROUND

Bacterial leaching has long been a component in processing uranium and copper ores, principally because of its relatively low capital and operating costs and minimal environmental concerns. Kennecott pioneered the large-scale application of bioleaching of copper at Bingham Canyon in the 1960's and was followed by Duval, Ranchers Exploration and Development, Baghdad, and numerous other mining companies throughout the world. Current applications include heap, in-situ, and dump-leaching operations.

Until recently, most bioleaching efforts were directed toward copper. With the worldwide interest in low-grade auriferous sulphide ores, bacterial leaching is now competing to find its place in precious metals' mining. Several mining companies are actively pursuing this 'new' technology.

Bacterial leaching is fundamentally a natural process that results from a variety of bacteria (most notably *Thiobacillus ferrooxidans*) oxidizing sulphide host minerals to liberate metal values. In the case of pyrite, the following equations describe the bacterially "catalyzed" reactions:

$$2\ FeS_2 + 7\ O_2 + 2H_2O \longrightarrow 2\ Fe(SO_4) + 2\ H_2SO_4$$
$$4\ FeSO_4 + O_2 + 2\ H_2SO_4 \longrightarrow 2\ Fe_2(SO_4)_3 + 2\ H_2O$$
$$FeS_2 + Fe_2(SO_4)_3 \longrightarrow 3\ FeSO_4 + 2\ S^\circ$$
$$2\ S^\circ + 2\ H_2O + 3\ O_2 \longrightarrow 2\ H_2SO_4$$
$$Fe_2(SO_4)_3 + 6\ H_2O \longrightarrow 2\ Fe(OH)_3 + 3\ H_2SO_4$$
$$Fe(OH)_3 + H_2SO_4 \longrightarrow Fe(OH)(SO_4) + 2\ H_2O$$

With gold, unlike copper, the dissolution of the host sulphide mineral does not put the metal of interest into solution for subsequent recovery; rather, it liberates the metal for further treatment (e.g., cyanidation). Bacterial leaching can be applied to the recovery of gold in at least two major ways. The first is in a heap leach where crushed low-grade ore is stacked and allowed to oxidize with the aid of bacteria over a period of months. Bacterial preoxidation offers a low-cost means to treat this unconcentrated ore directly, and may require only minimal capital beyond that needed to process the oxide portion of the deposit.

The second application is in the treatment of an upgraded product. In general, roasting and the other methods of preoxidation require that the ore be

upgraded by a processing step such as flotation in order to produce a concentrate that is economical to treat. Bacterial leaching in a stirred tank reactor can be applied in much the same way as roasting. The focus of this paper is on bacterial preoxidation of an auriferous concentrate.

The flowsheet given in Figure 2 represents a conceptual diagram of what we anticipate a full-scale operation to look like. It should be kept in mind that unlike roasting, there was not a broad database of operating plants from which to draw design parameters. The objective of this test program was to identify the potential of bacterial leaching as a low-cost alternative to roasting.

EXPERIMENTAL PROGRAM

Biooxidation

Laboratory tests that served as a basis for the economic evaluation were carried out in bench-scale reactors with accompanying ancillary equipment. Prior to beginning the tank tests, bacteria were adapted to the pyrite defined in Table 1. This adapted material served as a resource for innoculating the agitated 5-L batch reactors to which nutrients and an air/CO_2 mixture were added. Temperatures were maintained between 30°C and 40°C. Many of the typical operating conditions that were followed have been well documented by others in the literature. Follow-up work using continuous reactors of a comparable scale confirmed the batch results. Bacteria were allowed to react with the pyrite until either 40% (low level) or 60% (high level) of the pyrite was digested as determined by iron concentration of the solution and solids. Following bacterial preoxidation, the solids were washed to remove soluble iron, copper, and residual acid in order to minimize reagent consumption during cyanidation.

Once the bacteria had adapted to the pyrite sample, leaching occurred within a few days. One of the advantages of applying bacterial leaching to this concentrate is that several of the copper minerals present (enargite, covellite, chalcocite) were solubilized along with the pyrite. As a result, 50-60% of the copper was extracted without the use of additional acid. This copper otherwise would have acted as a cyanide consumer during cyanidation. Effluent solutions contained 10-20 gpl of iron and 100-250 gpl of copper.

Another of the advantages of bacterial leaching is its ability to selectively attack different minerals at different rates. Also, the way bacteria attack a given mineral permits liberation of the contained gold values without completely destroying the host matrix. Figure 3 gives an indication of the selective attack of the bacteria on a pyrite particle. The bacteria attack along crystallographic planes and grain boundaries to liberate or expose gold to cyanidation. This accounts for the fact that with jut 60% pyrite oxidation, up to 85% of the gold can be extracted.

Cyanidation

Residues from the bacterial preoxidation step were leached with sodium cyanide in bottle tests for gold recovery as described in the preceeding roasting section. Metal extractions during cyanidation are summarized in Table 10 for the low and high bacterial oxidation levels.

Cyanide consumptions for the respective low and high cases were 5.2 kg/t and 3.0 kg/t. These cyanide consumptions reflect the fact that not all of the copper minerals were leached during bacterial leaching. Optimization of the preoxidation step could result in a saving of cyanide. The lime consumptions for the two cases were 3.7 kg/t and 2.8 kg/t.

Increased gold and silver extractions may be possible if the pyrite were oxidized to a greater extent. It must be remembered, however, that a portions of the gold and pyrite are locked within the silicious gange and are not liberated during leaching. To this extent, gold extractions approaching 100% are not possible using this preoxidation approach.

Economics

The economics for using a bacterial-leaching step were based on the high level of bacterial oxidation. The equipment for the circuit was sized to treat 150 tpd of pyrite, using materials of construction suitable for the corrosive environment. The total equipment cost is the result of a detailed list of the major equipment required to implement the flowsheet shown in Figure 2.

With the exception of flotation equipment to produce a pyrite concentrate and a cyanide destruction circuit, no equipment was considered outside the direct needs generated by the bacterial leaching circuit. The capital costs for the conceptual circuit shown in Figure 2 were factored from the equipment cost and are given in Table 11.

The relative simplicity of the bacterial-leaching circuit permits a low-to-moderate capital investment for treating the 150 MTPD by-product pyrite concentrate. The purchased equipment, installation, and simplified buildings and structures all contribute to a reduced capital outlay in comparison to roasting.

The operating costs were determined in a similar fashion and are shown in Table 12. The main contributors to the operating costs are reagents and expenses related to the specific site selected for constructing the preoxidation plant.

Revenues

The revenue credits were taken for gold, silver, and copper. Since this process does not involve a waste heat boiler, no credit was taken for heat generation. Payments are based on the same metals prices and contract terms specified under the roasting section. The total revenue from gold, silver, and copper recovery following bacterial preoxidation is $5 663 000 annually.

CONCLUSIONS

The results from this comparative study for a 150 tpd pyrite treatment facility indicate that bacterial leaching offers a positive alternative to the conventional roasting approach. In Table 13, the economic highlights show that while the two methods generate roughly the same total revenue, the other factors favor bacterial oxidation. Roasting provides a significant operating profit but it is also a very capital-intensive process. The end result for the roasting case is a negative net present value (at 12%) and only a 2.3% internal rate of return over the 10-year life of the project.

Although bacterial leaching does not show an exciting rate of return for this low-gold concentrate and a $350/oz gold price, it does demonstrate that bio-oxidation can provide an economic edge over roasting. When gold prices again reach $400/oz, the financial picture becomes rather attractive. The internal rate of return for biooxidation climbs to 14.7% with a $949 000 net present value. The capital in this case can be paid back in just over 5 years. A slight improvement in the gold tenor of the concentrate from 12 up to 14 g Au/t would also produce similar results.

Certainly, this economic review does not provide the answer to all applications. Each feed material must be evaluated on its own merit. Considerations such as the location of the mine, transportation, government restrictions, sulphuric acid market, and metallurgical response can all have an impact on the outcome of a specific project.

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TABLES

Table 1 - Pyrite analysis

Au, g/T	13.4
Ag, g/T	91.1
Cu, %	1.0
As, %	0.4
SiO_2, %	17.4
Fe, %	34.7
S, %	39.7

Table 2 - Roasting operations

Mine	Rate	Roaster type	Bed temperature
Cochenour Willans Gold Mine	12-17 tpd	1 Stage FBR	640°C
Dickenson Mines Ltd.	12-15 tpd	1 Stage FBR	700°C
Campbell Red Lake Mines Ltd.	48-60 tpd	2 Stage FBR	540°C 700°C
Giant Yellowknife	180 tpd	2 Stage FBR	538°C 565°C
Dalny Mine	34-60 tpd	2 Stage FBR (1 Stage)	580°C 580°C
Kalgoolie Mining Assoc.	84 tpd	1 Stage FBR	620°-650°C
North Kalgurlie Mines Ltd.	36-84 tpd	1 Stage FBR	600°-650°C
Harmony Mines	240 tpd	1 Stage FBR	825°C

Table 3 - Chemical characteristics of roasted calcines

Measured	Weight loss 20%
Au	13.8 ppm
Ag	106 ppm
Cu	1.0%
S	3.6%
SiO_2	18.5%

Table 4 - Copper dissolution

Roasting temperature	% Cu ext.
600°C	85.9
650°C	83.5
700°C	77.6

Table 5 - Cyanide leach results

Roasting temp.	g/L NaCN level	% Au ext.	% Ag ext.	kg/t NaCN	kg/t lime consumption
600	2.5	86.5	75.4	4.5	7.8
600	0.5	75.7	43.0	1.4	7.8
600	0.25	70.2	14.5	0.9	7.4
650	2.5	88.7	72.4	5.7	5.3
650	0.5	72.3	47.3	2.0	6.9
650	0.25	64.0	4.0	1.5	6.4
700	2.5	90.5	72.4	1.7	7.7
700	0.5	89.5	65.6	0.7	9.1
700	0.25	66.7	3.2	0.6	8.0
NR	2.5	66.4	16.7	15.9	2.9
NR	0.5	48.9	2.1	5.0	3.1
NR	0.25	43.6	0.9	2.0	4.2

Table 6 - Metal recoveries and reagent usages assumed

Recoveries	
Gold	79%
Silver	52%
Copper	82%
Reagent consumption	
NaCN, kg/t	1.2
Lime, kg/t	6.2
H_2SO_4, kg/t	7.0
Scrap iron, kg/t	8.3
Chlorine, kg/t	9.9

Table 7 - Capital cost estimate

Purchased equipment	$ 3 497 000
Installation	1 048 000
Piping	1 398 000
Insulation	175 000
Instrumentation	700 000
Electrical	875 000
Building	3 273 000
Total direct cost	$10 966 000
Engineering	$ 1 097 000
Construction expenses	1 097 000
Contractor's fee	658 000
Contingency	1 383 000
Site-related expenses	2 282 000
Total installed cost	$17 483 000

Table 8 - Operating cost summary

Reagents	$ 1 464 000
Energy	276 000
Labour	656 000
Maintenance	525 000
Contingency	292 000
Total operating cost	$ 3 213 000

Table 9 - Revenue summary

Gold	$ 4 550 896
Silver	324 515
Copper	365 310
Steam	500 000
Total	$ 5 740 721

Table 10 - Metal extractions

	Low	High
Au %	71	85
Ag %	27	80
Cu %	4.5	11
Fe %	0.11	0.29

Table 11 - Capital costs for bacterial preoxidation ($000)

Purchased equipment	$2 613
Installation	784
Piping	784
Insulation	183
Instrumentation	523
Electrical	392
Building	157
Total direct costs	$5 436
Engineering	$ 435
Construction	544
Contractor's fee	272
Contingency	667
Site-related expenses	1 020
Total installed costs	$8 374

Table 12 - Operating costs for bacterial preoxidation ($000)

Reagents	$2 509
Energy	424
Labour	417
Maintenance	251
Contingency	252
Total operating costs	$3 853

Table 13 - Comparative economic summary

	Roast	Bio
Total revenue ($000)	$ 5 741	$ 5 663
Operating costs ($000)	3 213	3 853
Capital costs ($000)	17 483	8 374
Operating profit ($000)	2 528	1 810
NPV @ 12% ($000)	(6 293)	(1 186)
Simple payback (yrs.)	9.0	6.5
IRR (%)	2.3	8.4

FIGURES

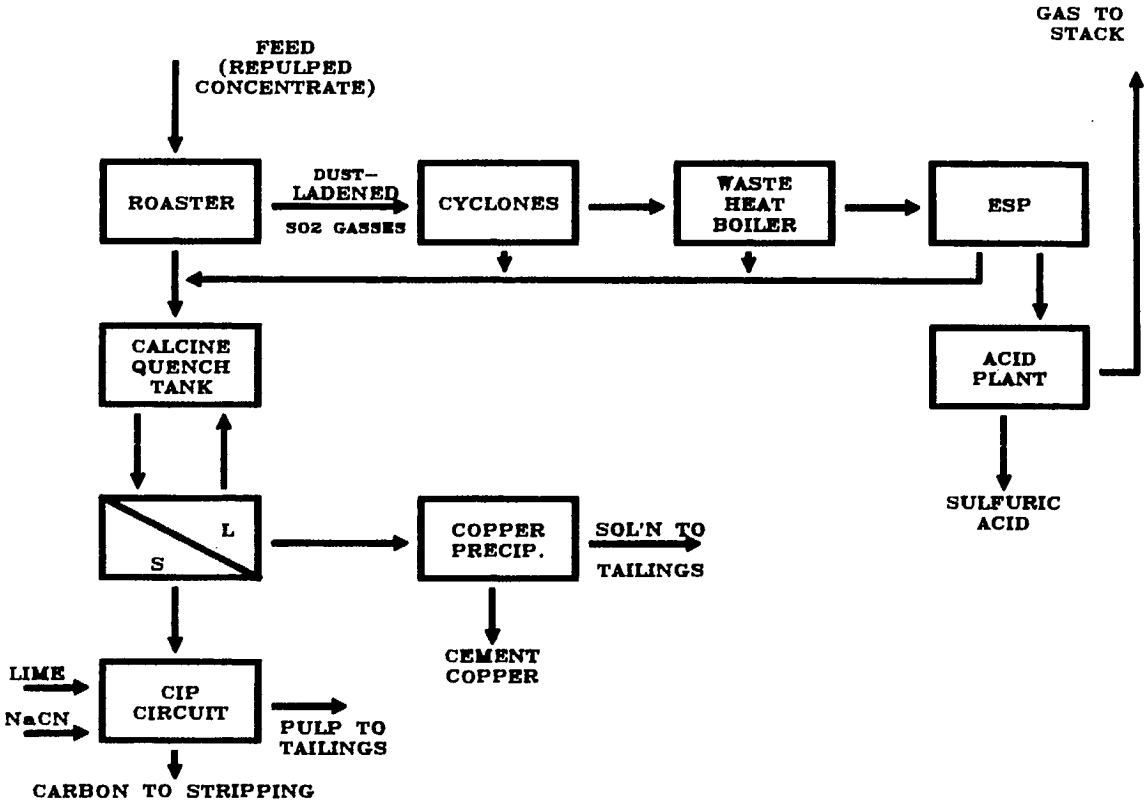

Fig. 1 - Fluid-bed roasting

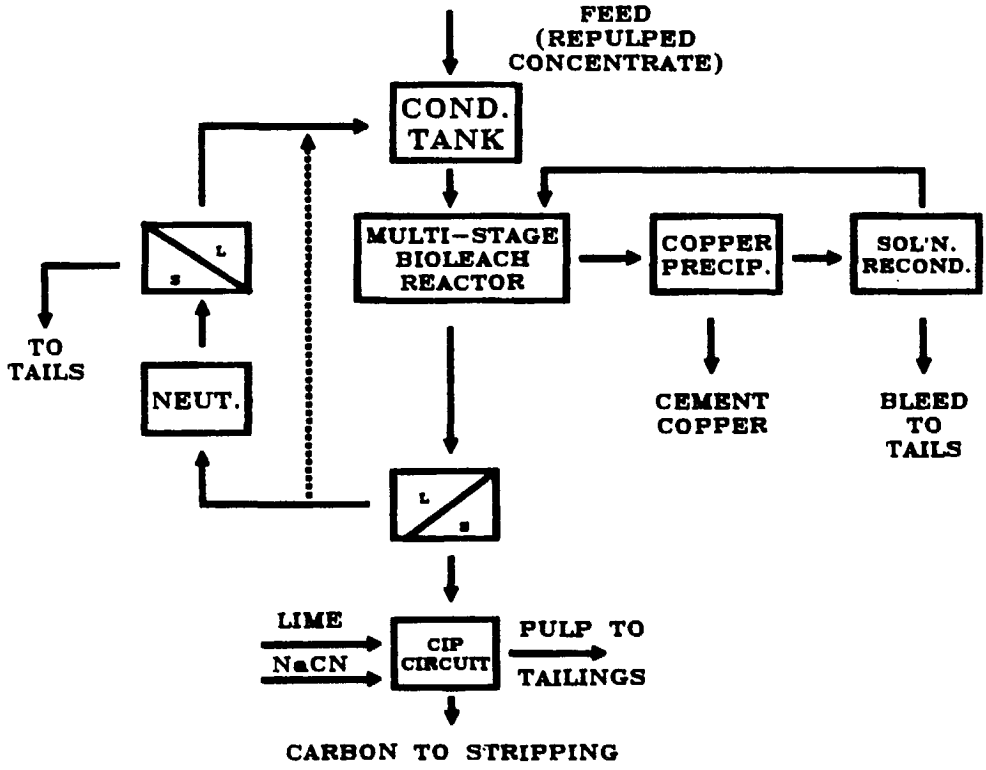

Fig. 2 - Bioleach flowsheet

Fig. 3 - Photomicrograph of selective bacterial attack on pyrite matrix. Scale: 1 cm = 100 μ

SESSION I: PAPER 5

A NEW BIOTECH PROCESS FOR REFRACTORY GOLD-SILVER CONCENTRATES

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ABSTRACT

Giant Bay Biotech has completed a two-year program to develop a biotechnological process for recovery of gold and silver from refractory sulphidic concentrates. The results of detailed laboratory and pilot-plant tests will be presented.

An engineering study has demonstrated the technical merits of the process and shown it to be less expensive than roasting or pressure leaching. As a result, Giant Bay is proceeding with plans to construct and operate a commercial demonstration plant to prove the process to the mining industry.

SESSION I: PRÉSENTATION 5

NOUVEAU PROCÉDÉ DE TRAITEMENT BIOTECHNIQUE DES CONCENTRÉS AURIFÈRES/ARGENTIFÈRES RÉFRACTAIRES

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RÉSUMÉ

Giant Bay Biotech vient de terminer un programme de deux ans visant à mettre au point un procédé biotechnique de récupération de l'or et de l'argent contenus dans des concentrés sulfurés réfractaires. Les résultats des essais détaillés menés en laboratoire et en usine pilote seront présentés.

Une étude a fait ressortir les avantages techniques du procédé et a montré que ce dernier est moins onéreux que le grillage ou la lixiviation sous pression. Par conséquent, Giant Bay envisage de construire et d'exploiter une usine de démonstration commerciale en vue de convaincre l'industrie minière de l'efficacité du procédé.

A NEW BIOTECH PROCESS FOR REFRACTORY GOLD-SILVER CONCENTRATES

INTRODUCTION

Biotechnology is gaining acceptance in the mining industry as a new and exciting tool for the processing of sulphide ores and concentrates. Present applications include biological leaching of copper from low-grade ores and waste materials (1), and biological leaching of pyritic uranium ores (2).

In 1984, Giant Bay Biotech (formerly PM Mineral Leaching Technologies) embarked on an intensive research and development program to perfect a biological leaching process for the treatment of refractory precious metal ores and concentrates. The program culminated in the development of the BIOTANKLEACH process for concentrates and the BIOHEAPLEACH process for ores.

This paper describes the development of the BIOTANKLEACH process, from initial laboratory bench-scale tests through to the 16-week pilot-plant testing program carried out to provide the necessary data for an engineering feasibility study. The results of this feasibility study, prepared by Wright Engineers Limited of Vancouver, are summarized in this paper.

PROCESS CHEMISTRY

THE LEACH

The BIOTANKLEACH process uses specially adapted strains of bacteria such as *Thiobacillus ferrooxidans* to rapidly leach sulphide concentrates, containing minerals such as pyrite and arsenopyrite, at essentially ambient temperature and pressure. The desulphurized residue produced by the process contains all of the now-liberated gold and silver, which is recovered by conventional cyanidation.

The biochemical reactions that occur during the leaching of pyrite-arsenopyrite mixtures are quite complex, but can be summarized by the equations shown below:

$$2FeS_2 + 15/2O_2 + H_2O \rightarrow Fe_2(SO_4)_3 + H_2SO_4 \quad \Delta H°298$$

$$= -369 \text{ kcal/mole} \qquad \text{Eq 1}$$

$$2FeAsS + 7O_2 + H_2SO_4 + 2H_2O \rightarrow Fe_2(SO_4)_3 + 2H_3AsO_4 \quad \Delta H°298$$

$$= -350 \text{ kcal/mole} \qquad \text{Eq 2}$$

Iron and arsenic are ultimately oxidized through to the Fe^{3+} and As^{5+} states. By careful culturing and adaptation of the bacteria, strains can be developed that will thrive in the acidic, concentrated solutions produced by the process. For mixed pyrite-arsenopyrite concentrates, a strain has been developed to withstand iron and arsenic concentrations of 50 and 20 g/L, respectively, at a pH of 0.5.

Oxygen from the air supplies the oxidant required for the leach reactions. Pyrite oxidation requires 1.88 moles O_2 per mole S^{2-} oxidized, while arsenopyrite oxidation uses 3.50 moles per mole S^{2-} oxidized. Bioleach reactors must therefore be designed to provide extremely efficient and rapid oxygen uptake, as well as an effective means to remove the heat generated by the leach reactions.

Considerable evidence exists which suggests that the bacteria attack sulphide mineral particles preferentially at crystal imperfections or dislocations - weak spots where the gold is often located (3). As a result, partial sulphide oxidation is frequently sufficient to liberate virtually all of the gold. Lower sulphide oxidation requirements impact directly on process economics, as leach retention times are reduced, and aeration, cooling, and neutralization requirements are lowered.

SOLUTION NEUTRALIZATION

Slurry exiting the bioleach stage undergoes liquid/solid separation; the solution is then neutralized to precipitate iron, arsenic, and sulphate as a mixture of jarosite, ferric arsenate, and gypsum. Similarly, oxidized solids are neutralized prior to cyanidation. Limestone and lime are both effective neutralizing agents. The neutralization reactions that take place are shown below.

Free Acid Neutralization (pH 0.5-2.0):

$$H_2SO_4 + CaCO_3 + H_2O \rightarrow CaSO_4 \cdot 2H_2O + CO_2 \qquad \text{Eq 3}$$

Iron and Arsenic Precipitation (pH 2.0-3.5):

$$2H_3AsO_4 + Fe_2(SO_4)_3 + 3CaCO_3 + 7H_2O \rightarrow 2FeAsO_4 \cdot 2H_2O + 3CaSO_4 \cdot 2H_2O + 3CO_2 \qquad \text{Eq 4}$$

$$3Fe_2(SO_4)_3 + 5CaCO_3 + 19H_2O \rightarrow 2H_3OFe_3(SO_4)_2(OH)_6 + 5CaSO_4 \cdot 2H_2O + 5CO_2 \qquad \text{Eq 5}$$

Miscellaneous Metals Precipitation (pH 3.5-7 or 10):

$$MSO_4 + Ca(OH)_2 + 2H_2O \rightarrow M(OH)_2 + CaSO_4 \cdot 2H_2O \qquad \text{Eq 6}$$

M = Mg, Cu, Ni, Zn, etc.

Careful examination of Equations 1 to 5 shows that, depending on the relative proportions of FeS_2 and FeAsS in the concentrate, limestone-lime requirements are about 0.67-1 mole per mole of sulphide oxidized. Thus, for every 1% sulphide oxidized, the theoretical limestone consumption is 21-31 kg/t, or hydrated lime consumption is 15-23 kg/t. Because limestone is reactive up to a pH of 4 and is less expensive than lime, it is the reagent of choice for the bulk of the neutralization. Lime would then be required to raise the pH from 4 to 7-10. Reagent costs can be reduced drastically if a source of alkaline tailings is readily available for neutralization. The natural alkalinity of most concentrates will also lower actual limestone consumption.

BENCH-SCALE PROGRAM

CONCENTRATE SAMPLES

Over the past two years, approximately 30 different concentrate samples from North America and Australia have undergone preliminary testing for their response to biooxidation. The samples varied widely in mineralogy and precious metal content. Bacterial cultures were developed capable of oxidizing a wide assortment of sulphide minerals, including pyrite, arsenopyrite, chalcopyrite, sphalerite, tetrahedrite, realgar, orpiment, and stibnite. In addition, several samples containing preg-robbing carbon were tested to determine whether bioleaching could produce sufficiently strong oxidizing conditions to deactivate carbon. In all but two cases, precious metal recovery was enhanced significantly by biooxidation.

CONTINUOUS BENCH-SCALE EVALUATION

Three of the refractory concentrates tested were chosen for a more detailed, continuous bench-scale test program. Two of the concentrates contained pyrite and arsenopyrite, while the third concentrate contained pyrite and preg-robbing carbon. Assay data for these concentrates is presented in Table 1.

The continuously operating system consisted of a feed tank, three 5-L capacity air-sparged, agitated leach tanks connected in series, and a product receiver. Temperature in the leach tanks was controlled at 35°C. Provision was made to recycle biomass and biologically produced acid, if desired.

Product slurry exiting the last leach tank was periodically filtered, the solids washed and saved for cyanidation. Leach and wash solutions were neutralized with limestone to precipitate iron, arsenic, and sulphate. The tailings slurry was then filtered and the tailings water recycled back to the feed tank.

Continuous operation was maintained for a 2-8 month period (depending on the concentrate) to gain information on the following items:

- extent of sulphide oxidation required to maximize precious metal recovery;
- benefits of re-grinding the concentrate prior to leaching;
- effect of pulp density and retention time on oxidation rates in each leach stage;
- potential build-up of impurities harmful to the bacteria.

Figure 1 portrays gold recovery vs % sulphide oxidation for the three concentrates. All samples required only partial oxidation to liberate essentially all of the gold. Samples 1 and 2 (the two FeS_2/FeAsS concentrates) required 75% and 84% sulphide oxidation, respectively, whereas sample 3 (the FeS_2/C concentrate) required only 51% sulphide oxidation. In all cases, gold recovery was improved dramatically to the 97-98% range.

The ideal continuous operating conditions that produced the required degree of sulphide oxidation are summarized in Table 2.

Re-grinding the concentrate was found to significantly improve leach rates in two out of three cases. Initial pulp density of 200 g solids per litre of pulp was found to give a combination of fast leach rates and high oxidation without any impairment in bacterial activity. Operation at even higher pulp densities is possible on a bench-scale, but on a commercial scale the required oxygen uptake may be beyond the capabilities of existing agitation equipment. Retention time required to achieve adequate sulphide oxidation varied from a single-stage 36-h leach for the FeS_2/C concentrate, to a two-stage 100-h leach for one of the FeS_2/FeAsS concentrates.

Other interesting discoveries from the continuous bench-scale program were:

- arsenopyrite leaches preferentially to pyrite;
- bioleach rates achieved during continuous leaching were improved dramatically over leach rates obtained during the rapid-leaching phase in batch tests. This was attributed to continuous leaching conditions being more ideal for optimum bacterial growth, and to continued bacterial adaptation with time;
- there was no accumulation of any impurities deleterious to biological activity.

PILOT-PLANT PROGRAM

Concentrate 1, a mixed pyrite-arsenopyrite concentrate produced in eastern Canada, was used as feed for the pilot-plant program. Objectives of the pilot study were to:

- confirm and improve operating conditions established during the bench-scale evaluation;
- gain information on the oxygen utilization possible during leaching;
- establish thickening and filtering rates for downstream products;
- test long-term stability of tailings produced by the process;
- obtain detailed mass and flow balances on the overall process.

EQUIPMENT DESCRIPTION AND OPERATION

The pilot-plant campaign was carried out from August to December, 1985 on 3 t of concentrate. A schematic diagram of the pilot-plant layout is depicted in Figure 2. The leach circuit consisted of a fiberglass feed tank and three plexiglass bioleach tanks. Feed tank dimensions were 0.76 m (30 in.) diameter by 1.52 m (50 in.) high with a capacity of 600 L. The feed tank was baffled and agitated with an overhead stirrer equipped with two stainless steel axial flow impellers. Pulp from the feed tank, consisting of concentrate, recycled process water, make-up water and bacterial nutrients, was pumped continuously at a controlled rate to the first leach tank.

The three leach tanks were connected in series, with pulp passing from one tank to the next by gravity overflow. Tank dimensions were 0.61 m (24 in.) diameter by 0.91 m (36 in.) high. Each tank held 167 L of pulp, with the liquid level to tank diameter ratio fixed at one. The leach tanks were baffled and agitated with overhead stirrers equipped with stainless steel disk-type radial flow impellers. Air was blown into each tank near the bottom, directly underneath the impeller.

Each tank was equipped with temperature controllers that delivered hot or cold water through stainless steel cooling coils situated in the tanks. The tanks were heated initially to 35°C, but once bioleaching commenced constant cooling was required due to the exothermic nature of the reactions. Temperatures in the tanks were generally controlled at 35°C, but occasionally were as high as 43°C with no harmful effect on leaching. The bacteria may be able to tolerate even higher temperatures with further adaptation. Periodic examination of the cooling coils over the duration of the 16-week run revealed no evidence of scale formation.

All leach tanks were provided with control equipment for pH, air sparging rate and stirrer rpm. Because of excellent bacterial adaptation, pH control in the leach circuit was not required. Oxygen utilizations were determined by measuring the oxygen content of the off-gas, calculating the rate of oxygen consumption, and expressing it as a percentage of the oxygen added from the sparged air.

Product slurry exiting the last leach tank was flocculated to facilitate solids settling, and then pumped to a thickener. Thickener underflow, containing all the gold and silver, unreacted sulphides and gangue, was either washed, re-thickened and stored, or stored unwashed for cyanidation testing. Thickener overflow, containing dissolved iron, arsenic, sulphur, and other trace metals, was continuously neutralized in a tank, using slurried limestone, to a pH of 3.5 to 4. Provision was made to recycle some of the biomass and acid to the feed tank if desired. Neutralization caused the precipitation of virtually all of the arsenic, iron, and sulphate as a mixture of gypsum, jarosite, and ferric arsenate. These tailings were then thickened and stored in drums, with the thickener overflow solution recycled back to the leach step.

Leach parameters that varied during the testing program were retention time, pulp density, agitation, and air-sparging rate. Settling and filtering tests were performed on the biooxidized product and on the tailings generated by the process. Detailed cyanide leach tests on the biooxidized product were performed to determine retention time and the cyanide strength required. Complete mass balances for iron, arsenic, sulphur, gold, and silver were periodically performed.

TEST RESULTS

Continuous leaching on the pilot-plant scale confirmed the essential operating conditions established during bench-scale testing. Figure 3 presents Tank 1 leach results obtained during a 41-day period when the following near-optimum conditions prevailed: solids particle size of 90% -400 mesh, pulp density of 200 g/L (17.5% solids), and aeration rate of 0.066L/L-min. As the retention

time was gradually decreased from 84 h to 40 h, both iron and arsenic oxidation rates increased gradually to reach 660 mg/L-h, respectively, while iron and arsenic oxidation remained fairly constant throughout at 55-65% and 80-90%, respectively. Oxygen utilization also increased steadily and was measured at 54.5% during the 47-h retention time phase.

These results seem to indicate that, under the conditions tested, first-stage leaching was controlled by the availability of substrate for the bacteria. The bacteria appeared to have no difficulty adjusting to changes in retention time. A reasonably efficient utilization of oxygen from the air was achieved.

A complete material balance carried out on the 34th day of this continuous test (47-h retention time in each tank) gave cumulative sulphide oxidations of 62% after stage 1, 78% after stage 2, and 94% after stage 3. Bioleaching produced an overall solids-weight reduction of about 30%. Approximately 10% of the oxidized iron and arsenic remained with the solids as precipitated jarosite and ferric arsenate.

Figure 4 shows the typical kinetics of gold and silver extraction and cyanide consumption from thickened, unwashed product obtained by a batch cyanide leach test in a stirred tank. Both gold and silver extraction peaked at 98% and 68%, respectively, after only 8 h of cyanidation. By comparison, cyanidation of roaster calcine takes 5 to 7 days to achieve maximum extraction. Cyanide consumption after 8 h of leaching was approximately 4 kg/t original concentrate.

Thorough washing of the biooxidized product did not improve gold or silver recovery or reduce cyanide consumption. Incomplete extraction of silver was probably due to the lock-up of some silver with jarosite. Process modifications are being tested to reduce jarosite formation, which should result in improved silver recovery. High cyanide consumption is due mainly to the formation of thiocyanate by side reactions; further research to reduce cyanide consumption is continuing.

Previous investigators have claimed that bioleaching can cause partial dissolution of gold and silver, which is then difficult to recover. Periodic, careful analysis of product thickener overflow by the MIBK extraction method never revealed gold concentrations higher than 0.1 ppb, or silver concentrations higher than 0.1 ppm. Thus, precious metal loss into bioleachate was not a problem with this concentrate.

Settling and filtering tests on the biooxidized product and tailings were carried out with the help of personnel from Eimco Process Equipment Inc. These tests were instrumental in aiding in the design of the overall commercial process. Settling tests were performed in 2-L graduated cylinders equipped with a raking mechanism. Filtering tests were performed using standard leaf test procedures.

The oxidized product required flocculation before adequate settling rates were achieved. A number of different flocculants were tried, with the best performance obtained by using a guar gum-based cationic flocculant called Guartec CD, supplied by Henkel Corporation of Tucson, Arizona. The optimum dosage of flocculant was 144 g/t of oxidized solids (100 g/t original concentrate). A thickener underflow of 50% solids was produced, which separated 84% of the

total leach solution from the solids. Computer analysis of the settling data by Eimco, using the Wilhelm-Naide method, gave a thickener unit area of 0.52 m^2-d/t.

To minimize water balance problems in the cyanidation circuit, the product solids can be further de-watered by filtering prior to cyanidation. Acidic slurry direct from bioleaching did not vacuum filter easily, but solids neutralized to pH 10.5 with limestone and lime filtered extremely well. Thus, product solids would be first thickened, and the thickener underflow then neutralized to pH 10.5 before vacuum filtering. Leaf tests on neutralized thickener underflow indicated a filter cake containing 30% moisture could be readily produced.

Tailings produced by the process settled reasonably well. Attempts to accelerate settling by flocculation failed to turn up an effective flocculant during the time available, but further research should produce a suitable flocculant. Without flocculating, Eimco calculated a thickener unit area of 0.96 m^2-d/t to produce a 52.5% solids underflow.

The tailings produced by the process contained mainly gypsum, jarosite, and ferric arsenate, along with hydroxides of metals such as copper, cobalt, nickel, and zinc. Some experts have recently expressed concern that ferric arsenate can eventually break down due to reaction with carbon dioxide in the air, causing re-dissolution of arsenic (4). To test this theory, a tank test was set up in which tailings were re-pulped to about 20% solids, stirred, and sparged with air enriched to 5% carbon dioxide. This test was carried out for 24 h a day for a 6-month period. During that time, the concentration of arsenic in solution never exceeded 0.1 ppm, showing that virtually no redissolution of arsenic occurred. This test demonstrated that the tailings are very stable over long periods of time and should pose no environmental problems.

ENGINEERING FEASIBILITY STUDY

The results of the pilot-plant testing program were used by Wright Engineers Limited of Vancouver to carry out a feasibility study for the BIOTANKLEACH process, and to compare its capital and operating costs with those of the alternative treatment processes of roasting and pressure leaching.

Wright Engineers used a northern Ontario location for the plant. It was assumed the plant would be erected at an existing mine site so that no charges for infrastructure would be included. The costing for a plant with a capacity of 100 tonnes of concentrate per day was carried out in detail, and the costs for a 50- and 200-tonne-per-day plant were factored.

Figure 5 shows a conceptual flowsheet for a 100-tonne-per-day plant.

FEED PREPARATION

For the design of the plant, a concentrate thickener underflow feed of 55% solids was used. This material, approximately 60% passing 400 mesh, will be fed to a small ball mill operating in closed circuit with a cyclone. Cyclone overflow at 90% passing 400 mesh and 20% solids will be stored in a surge tank ahead of the bioleach circuit. The degree of concentrate regrinding will vary between concentrates, with some not requiring any regrinding. A power requirement of five kWh per tonne of concentrate was selected as being typical.

BIOLEACH CIRCUIT

Eight agitated tanks, each 6.4 m diameter by 6.4 m high, will be used for the bioleach circuit. The tanks will be arranged as shown schematically in Figure 6.

The first stage of leaching will use four tanks and have a retention time of 48 h. The second and third stages will each use two tanks and have a retention time of 24 h each, for a total retention time of 96 h. The tanks have been arranged in 2 parallel lines, but can be operated as 4 stages of 24-h residence time each if desired.

Air will be sparged to the tanks at a total rate of 174 Nm^3/min (6160 CFM) supplied by an 83 kPa compressor. For design purposes, a conservative estimate of 40% oxygen utilization was used. The bacteria need carbon dioxide for a carbon source, but enough CO_2 is available from the air and from carbonates in the concentrate to ensure rapid bioleaching rates. Minimal amounts of nutrients will be supplied by use of a commercial-grade fertilizer.

Agitators were sized by Dr. J. Oldshue of Mixing Equipment Company Inc., who is considered one of the foremost experts in the scaling up of agitated reactor systems. To ensure adequate oxygen uptake, first-stage reactors would require 75 kW (100 hp) agitators, second-stage reactors would require 56 kW (75 hp) agitators, and third-stage reactors would require 37 kW (50 hp) agitators.

Since the oxidation reactions are exothermic, cooling of the pulp will be required, particularly in the first leach stage where more than 60% of the heat will be produced. Each tank will be equipped with stainless steel cooling coils, the water flowing in a closed loop with a cooling tower.

TREATMENT OF LEACH PRODUCT

The leach residue (73 t/d) leaving the last leach stage will be thickened to 50% solids in a 7.3-m diameter thickener. The thickened solids will first be neutralized with a ground limestone slurry to approximately pH 4, and subsequently with a lime slurry to pH 10.5. The neutralization process will be carried out in 4 tanks in series, providing a total residence time of 10 h. Neutralized product will be dewatered using a 3-m diameter by 3.7 m long drum belt filter, and then directed to cyanidation.

PRODUCT THICKENER OVERFLOW NEUTRALIZATION

The product thickener overflow will be neutralized in two stages to precipitate ferric arsenate, jarosite, and gypsum. Neutralization will be carried out in four tanks in series, with a total residence time of 10 h. Slurry from Tank 4 will be recirculated to Tank 1 to produce a coarser precipitate with better thickening characteristics.

Tailings (89 t/d) will be thickened to 52.5% solids in an 11-m diameter thickener. Thickener underflow will be pumped to the concentrator tailings pumpbox and discharged to the tailings dam. Thickener overflow will be pumped to the feed preparation circuit for grinding and to the lime and limestone preparation circuits.

HEAT BALANCES

Heat balances have been prepared using the summer and winter conditions listed in Table 3.

Cooling water from the leach circuit would enter a cooling tower at a temperature of 31°C and exit at 26°C. Detailed calculations gave the heat balances listed in Table 4.

Cooling requirements will increase only marginally if the pulp density of the leaching slurry is increased. However, an increase in sulphide content from 16% to 20% will increase the cooling water requirements in summer conditions to 21 047 m^3 per day, while a decrease in sulphide content to 10% will reduce cooling water requirments to 9830 m^3 per day.

If the plant were operated in a tropical climate, where atmospheric conditions were 35°C and 100% relative humidity (assumed worst conditions), once-through cooling using sea water or river water at an assumed temperature of 22°C could be used. Also, the leach could be operated at a higher temperature, say 43°C.

CAPITAL COST ESTIMATES

Capital and operating costs have been prepared for the BIOTANKLEACH process at operating rates of 50, 100, and 200 t per day. Comparative costs have been estimated for the alternative processes of roasting and pressure oxidation at the rate of 100 t per day. Costs for the 50- and 200-tonne-per-day alternatives were estimated by applying appropriate factors to the 100-tonne-per-day base case. A summary of these costs is shown in Table 5.

OPERATING COST ESTIMATES

Direct costs have been estimated for operation and maintenance of the bacterial leach plant as shown in Table 6.

The reagent costs at the Ontario location are high because of excessive transportation costs. These costs may be significantly less at other locations.

These costs do not include capital amortization and working capital/inventory carrying charges. In addition, the following costs are not included and have been assumed to be part of the mine infrastructure:

- tailings and sewage disposal systems
- water, steam, instrument, and plant air supply
- environmental monitoring and reporting
- administrative charges.

COMPARISON OF ALTERNATIVE OXIDATION PROCESSES

The BIOTANKLEACH capital and operating costs for the 100-tonne-per-day case were compared to the alternative processes of roasting and pressure leaching. The cost comparison is summarized in Table 7.

The overall cost of the BIOTANKLEACH process is significantly less than both roasting and pressure oxidation. Capital and operating costs of roasting are

excessive because of the high cost of cleaning the sulphur dioxide flue gas. The capital cost of pressure oxidation is considerably higher than both alternatives, because the cost for an oxygen plant - $3 426 000 - is included.

CONCLUSIONS

The BIOTANKLEACH process for refractory precious metal concentrates offers a viable alternative to conventional processes. Advantages of bioleaching include: operation at room temperature and pressure; efficient use of oxygen from air as the oxidant; and disposal of iron, arsenic, and sulphur as environmentally safe waste products.

The results of an engineering feasibility study have indicated that the BIOTANKLEACH process is more economical than roasting and pressure leaching. As a result, Giant Bay is proceeding with plans to construct and operate a 10-25 tpd commercial plant to prove the process to the mining industry.

The process has been designed to incorporate equipment commonly used and accepted in the mining industry today. However, innovative methods of agitation, aeration, and cooling are being investigated in an on-going effort to further reduce costs and improve the efficiency of the process.

Patents on the process have been filed in the major gold-producing countries in the western world and, at the time of writing, have been accepted in at least one country, South Africa. Giant Bay is interested in acquiring properties that can benefit from the process, or will provide the technology to interested parties on a royalty or license fee basis.

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TABLES

Table 1 - Concentrate assays

Sample	% Fe	% As	% S	% C	Au (g/t)	Ag (g/t)
No. 1 pyrite-arsenopyrite	21-25	5-7	15-18		240-340	30-50
No. 2 pyrite-arsenopyrite	20-24	6-8	16-21		61-77	18-27
No. 3 pyrite-carbon	14-16	0.3	15-17	7	110-150	160-200

Table 2 - Bioleach operating conditions for concentrate samples

		Concentrate 1 FeS_2/FeAsS	Concentrate 2 FeS_2/FeAsS	Concentrate 3 FeS_2/C
Concentrate regrind?		Yes	Yes	No
Pulp density, g/L		200	200	200
Number of leach stages		2	2	1
Total retention time, h		94	100	36
% Sulphide oxidized		77.6	88.7	49.3
Sulphide oxidation rate, mg/L-h	Stage 1	415	353	458
	Stage 2	104	179	
	Average	260	266	458

Table 3 - Summer and winter conditions

Conditions	Summer	Winter
Design air temperature (°C)	25	-25
Humidity of the air (%)	50	0
Operating temperature of leach tanks (°C)	35	35
Operating temperature of cyclone overflow (°C)	25	15

Table 4 - Heat balances

Conditions	Summer	Winter
Heat input (kcal/day)	-92 004 670	-92 004 670
Heat losses (kcal/day)	10 422 137	29 739 675
Net heat input (kcal/day)	-81 582 533	-62 264 995
Cooling water flow (m^3/day)	16 415	12 528
Airflow (m^3/min)	174	174
Total number of cooling coils	58	58

Table 5 - Capital costs (US$)

	Plant size		
	50 tpd	100 tpd	200 tpd
Total direct cost	2 081 000	3 121 000	4 712 000
EPCM	250 000	375 000	556 000
Contingency	188 000	282 000	436 000
Total capital cost	2 519 000	3 778 000	5 704 000

Table 6 - Summary of operating costs (US$)

	Plant size		
	50 tpd	100 tpd	200 tpd
Reagents	20.81	20.81	20.81
Labour	28.32	14.16	7.08
Spares	2.12	1.80	1.33
Consumables	0.50	0.37	0.27
Power	8.91	5.94	3.96
Total operating cost $ per tonne concentrate treated	60.66	43.08	33.45

Table 7 - Cost comparison (US$)

	Capital	Operating ($/t)
Roasting	4 889 000	55.90
Pressure oxidation	7 481 000	42.79
BIOTANKLEACH	3 778 000	43.08

FIGURES

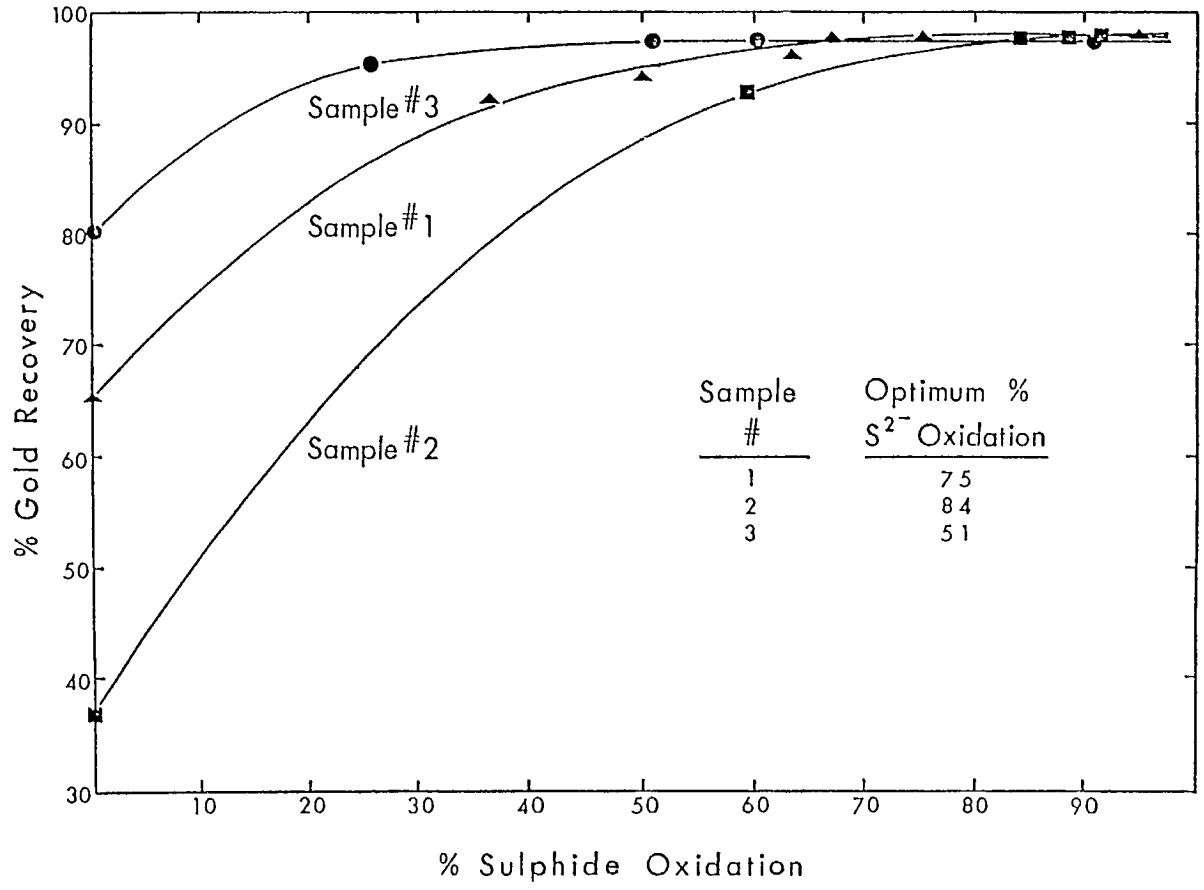

Fig. 1 - Gold recovery vs % sulphide oxidation

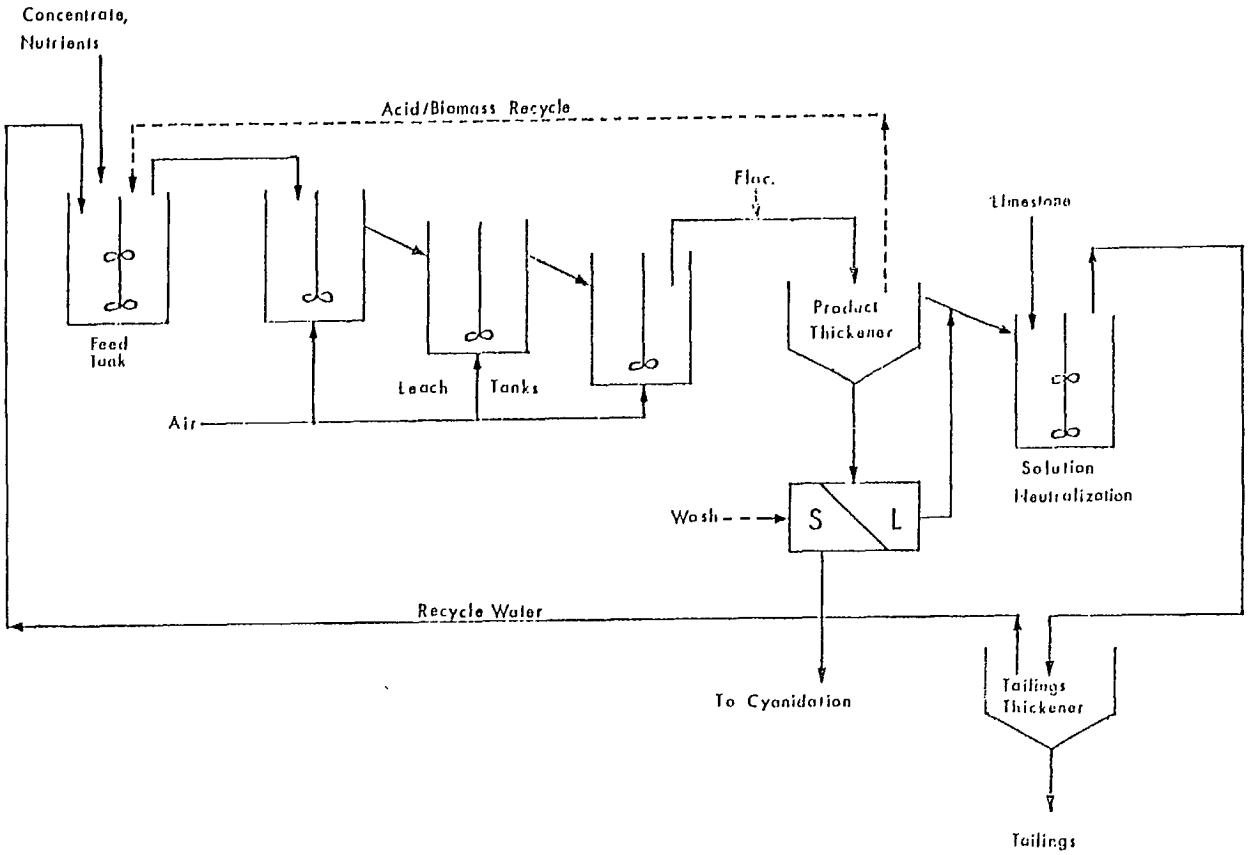

Fig. 2 - Pilot-plant flowsheet

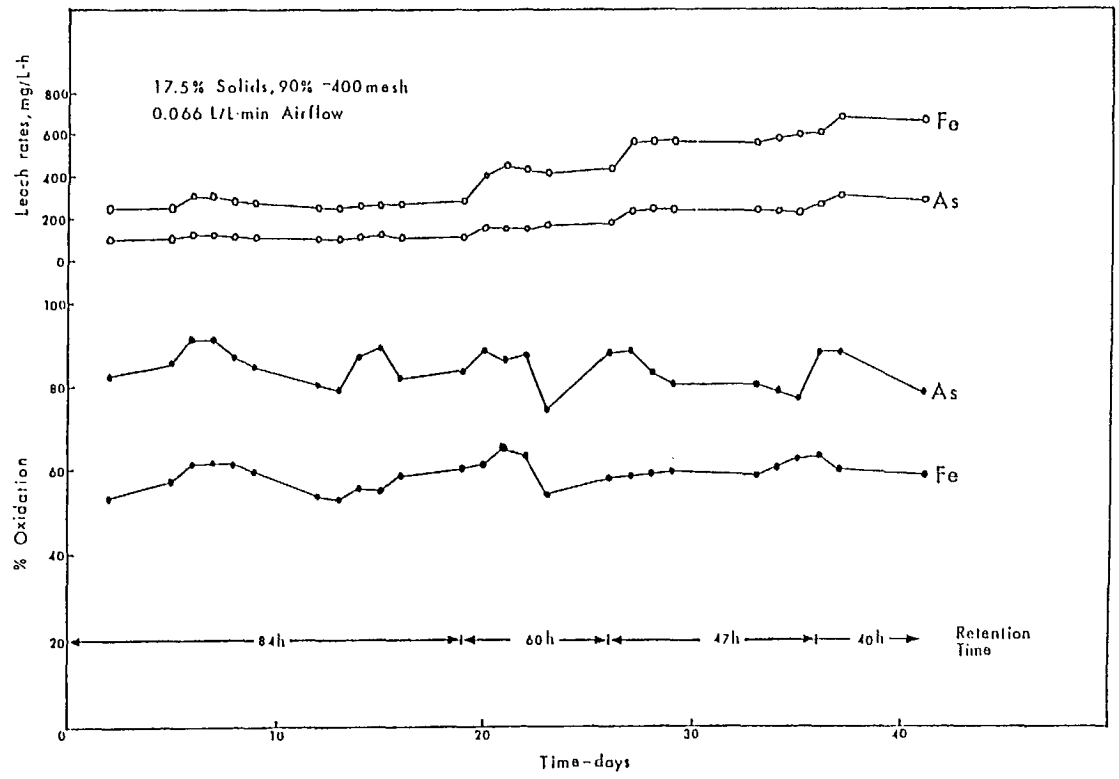

Fig. 3 - Pilot-plant tank 1 leach results

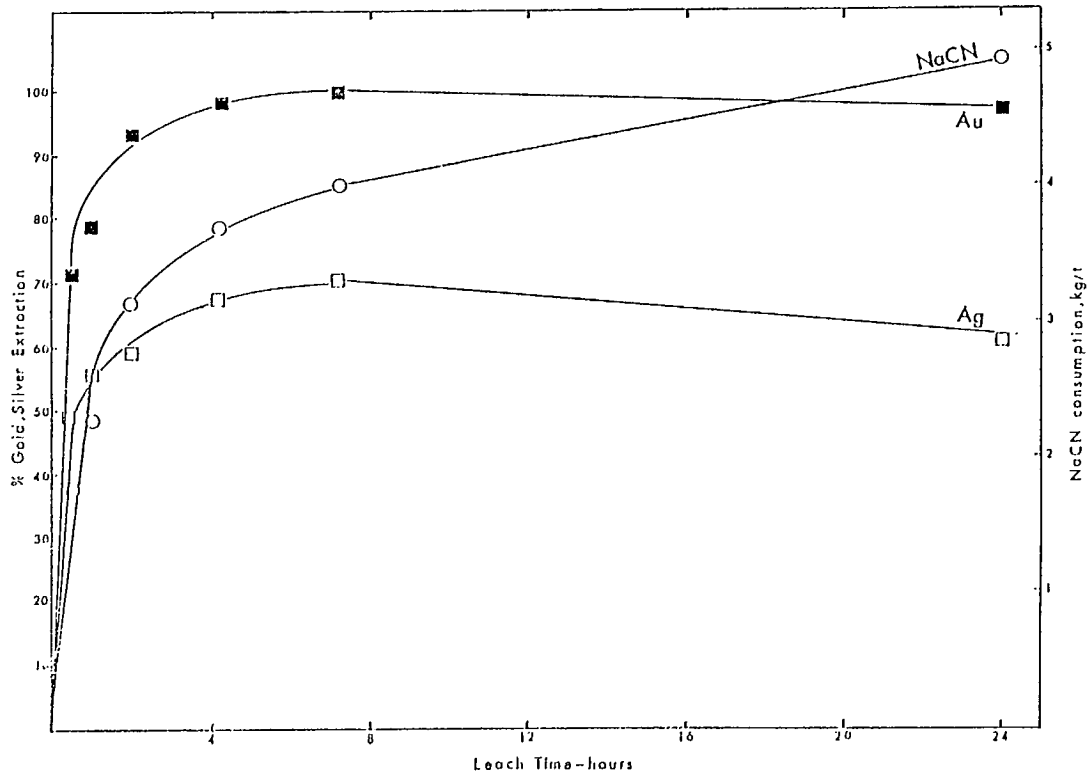

Fig. 4 - Kinetics of gold and silver dissolution and cyanide consumption from biooxidized concentrate

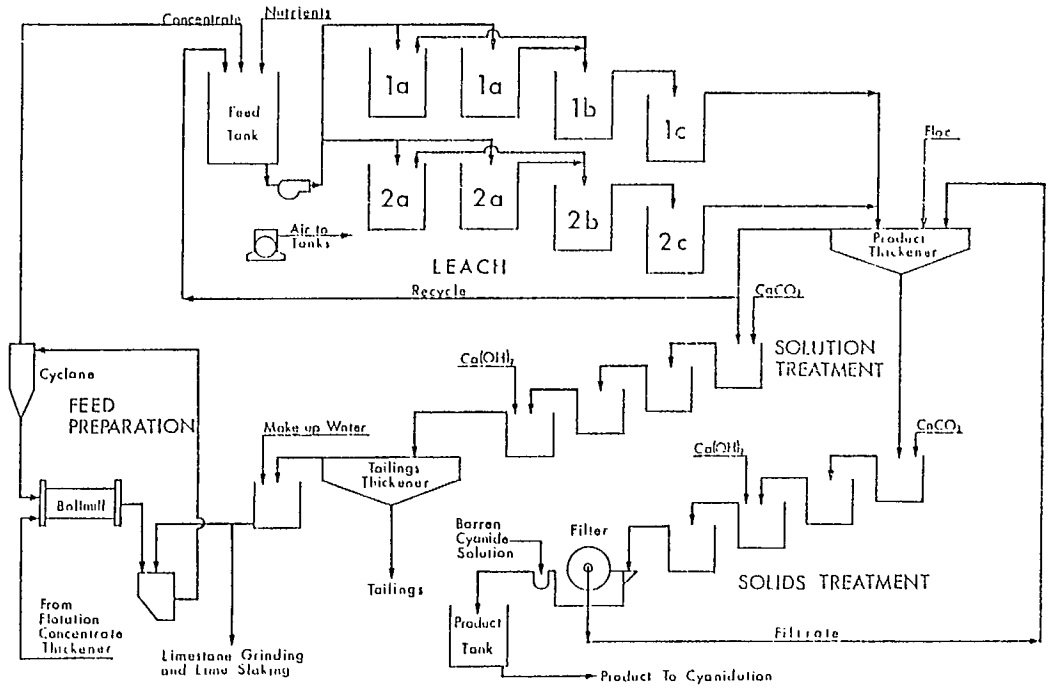

Fig. 5 - BIOTANKLEACH conceptual flowsheet

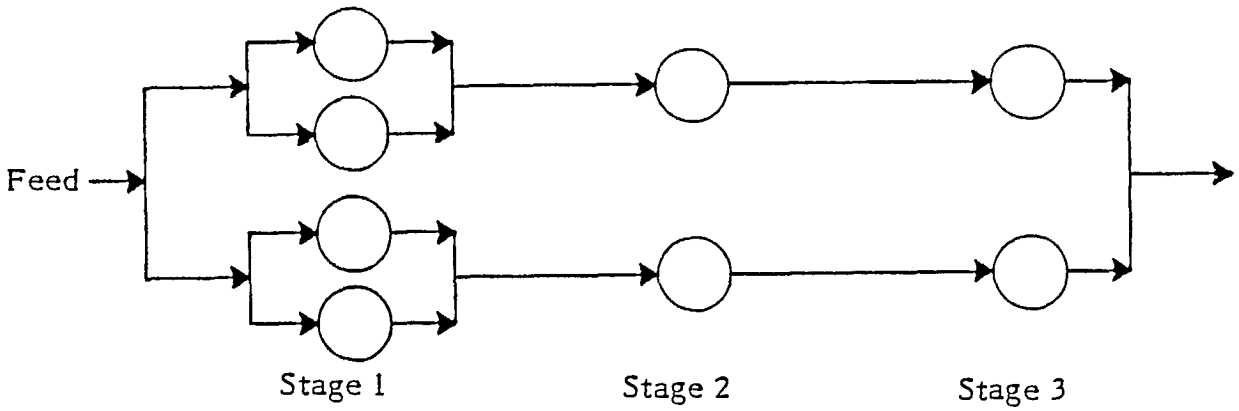

Fig. 6 - Bioleach circuit

SESSION II

POTENTIAL MICROBIAL PROCESSES FOR MINING

SESSION II: PAPER 6

BIOADSORPTION OF URANIUM BY FUNGAL STOPE ISOLATES

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ABSTRACT

The ability of certain fungal, yeast, algal, and actinomycete species to adsorb uranium from dilute solutions has been well documented in the scientific literature. This "bioadsorption" process is generally reversible; thus, the adsorbed uranium can be recovered and the biomass regenerated for further use by mild treatments such as elution with sodium carbonate solution. However, from the standpoint of developing a technically and economically viable process, much of the published laboratory data are considered to be inadequate since testwork has generally been conducted on synthetic uranium solutions. The performance of biomass in actual leach solutions is relatively unknown.

Denison Mines is currently conducting bioleaching of uranium underground. The leachates produced during this process are substantial in volume and low in concentration, resulting in high pumping costs.

Two acidophilic fungi and one neutrophilic actinomycete have been isolated from the Denison Mines stopes. In this paper, the performance of these biomasses in adsorbing uranium and yttrium from Denison minewater under various conditions is discussed. The results are also compared to those obtained using an organism not found in the Denison stopes, *Rhizopus arrhizus*.

SESSION II: PRÉSENTATION 6

BIOADSORPTION DE L'URANIUM AU MOYEN D'ORGANISMES FONGIQUES ISOLÉS DANS LES CHANTIERS D'ABATTAGE

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RÉSUMÉ

Le pouvoir de certaines espèces de champignons, de levure, d'algues et d'actinomycètes d'adsorber de l'uranium à partir de solutions diluées est un sujet bien documenté dans la littérature scientifique. Le procédé de "bioadsorption" est généralement réversible; par conséquent, l'uranium adsorbé peut être récupéré et la biomasse peut également être régénérée pour un usage ultérieur par des traitements modérés, comme l'élution avec une solution de carbonate de sodium. Toutefois, au point de vue de l'élaboration d'un procédé viable tant au niveau technique qu'économique, la plupart des données publiées sur les travaux effectués en laboratoire sont considérées comme inadéquates puisque les travaux d'essais sont généralement exécutés avec des solutions uranifères synthétiques. Le comportement de la biomasse dans les solutions de lixiviation réelles est à toutes fins utiles inconnu.

La Denison mines effectue en ce moment la biolixiviation souterraine de l'uranium. Ce procédé toutefois, entraîne une hausse des coûts de pompage en raison de l'important volume et de la faible concentration des produits de lixiviation obtenus.

Deux champignons acidophiles et un actinomycète neutrophile ont été extraits des chantiers de la Denison Mines. La présente communication traite du comportement de ces biomasses dans différentes conditions au cours de l'adsorption de l'uranium et de l'yttrium des eaux d'exhaure de la Denison. Enfin, les résultats d'essais sont comparés aux résultats obtenus avec un organisme non présent dans les chantiers de la Denison, soit le *Rhizopus arrhizus*.

BIOADSORPTION OF URANIUM BY FUNGAL STOPE ISOLATES

INTRODUCTION

The use of microorganisms to recover uranium from dilute solutions is well documented. However, most of the studies to date have been conducted using synthetic uranium solutions, thus limiting the data to academic applications. The performance of biomass in actual process streams is relatively unknown.

In a study conducted at the Ontario Research Foundation in Mississauga, Ontario, the biosorption performance of three isolates from Denison Mines bioleaching stopes and an organism not found at the mine site were studied. The recovery of both yttrium and uranium from Denison minewater was investigated. Consequently, a proposed biosorption flowsheet was developed for one of the organisms including preliminary costs, operating parameters, and design criteria.

Details of the study and the results obtained during the testwork are summarized in this paper.

BACKGROUND

At Denison Mines in Elliot Lake, underground biologically assisted leaching of low-grade uranium deposits is being conducted. Although this in-stope leaching practice is economically attractive, substantial volumes of low-concentration leachate are produced underground and thus must be pumped to surface. This minewater solution has been identified as a potential application for biological recovery techniques.

In a previous study conducted by Senes Consultants Inc. under contract with the Department of Supply and Services, the uranium adsorption characteristics of *Rhizopus arrhizus*, *Rhizopus oligosporous*, *Streptomyces levoris*, *Saccharomyces cerevisiae*, *Chlorella vulgarus*, immobilized cells and a mixed bacterial culture were quantified and compared. It was concluded that the *R. arrhizus*, *R. oligosporus*, *S. levoris*, and the mixed bacterial culture were effective biosorbents for uranium.

During this study, a number of observations relevant to the development of a viable biosorption process were noted. In particular, uranium uptake by the biomasses was significantly reduced at lower pH values (e.g., 2.0) or in the presence of other metallic cations (particularly ferric ion).

Since bioleach liquors are generated in high volumes, the pH adjustment of such solutions to 3.5-4.0 would represent a significant cost factor. Thus, desirable characteristics for a candidate biosorbent would be a higher adsorption capacity at lower pH values or, failing that, a biosorbent that is more physically robust for manipulation at pH values less than 3.0.

With such criteria in mind, the project outlined in this paper was developed to study organisms demonstrating the ability to survive and grow in the low pH conditions experienced within the Denison leach stopes.

An acidophilic *Penicillium*, an acidophilic *Rhizopus*-like organism, and a neutrophilic *Actinomycete* species were hence isolated at Denison Mines in Elliot Lake for study in the program. Their performance was compared to that of *Rhizopus arrhizus*, an organism not found in the leach stopes.

PRODUCTION OF BIOMASSES

The four biomasses (three Denison Mines isolates and the *Rhizopus arrhizus*) were received as Agar cultures in sealed petri dishes. These were inoculated into dilute nutrient broth media, pH 4.5, and incubated at room temperature. The nutrient broth medium found to produce good biomass growth in all cases consisted of:

Dehydrated nutrient	(Difco or Gibco)	- 0.8 g
Glucose	(BDH)	- 5 g
Distilled water		- 1000 mL

An alternate medium composed of the dilute nutrient broth described above, supplemented with different concentrations of tryptone (0.5-2.0% w/v), was also investigated for the culturing of the *Actinomycete*. It was found that the biomass yield was higher than with the dilute nutrient broth medium alone. However, the gelatinous nature of the resultant biomass made it impossible to recover it easily by filtration, particularly at the highest tryptone concentrations. It was therefore preferable to use the nutrient broth/glucose medium throughout the study.

The effects of temperature on biomass growth were investigated by inoculating growth from each culture into 100-mL aliquots containing pH 4.5 nutrient broth medium. These were incubated with shaking at room temperature (23°C), 27°C, and 32°C. Room temperature was found to be the preferred incubation temperature, since the optical densities of the cultures were observed to decrease significantly as the incubation temperature was increased.

In all cases, biomass production was found to be most rapid at pH 4.5. Decreasing the pH significantly slowed the growth process. No growth at all was noted for the *Rhizopus*-like organism at low pH values (2.3). Thus, large-scale biomass production was conducted at pH 4.5.

Large-scale production of the biomasses for subsequent adsorption testwork was conducted in 4.0 x 7.5 L microferm fermenters (New Brunswick Scientific). Initially, mechanical agitation with aeration was employed. However, operation without mechanical agitation, i.e., as an airlift fermenter, did not show any detrimental effects and, in fact, promoted favourable "pelletized" growth of the *Penicillium* and *R. arrhizus*. Hence, all fermenters were subsequently operated in this "airlift" mode, with the stirrers only used immediately before sampling to dislodge any biomass attached to the surfaces.

The neutrophilic *Actinomycete* exhibited typical mycelial growth. The hyphae were not septate but did display branching and spore production. This biomass grew in submerged culture as fibrous clumps that tended to attach to the fermenter surfaces. As the *Actinomycete* cultures aged, pigments leached from the biomass into the culture supernatant, producing a pink hue. However, this pigmentation did not appear to influence the production of biomass. Typical productivities for this species were 0.6 g dry solids per litre fermenter

capacity per day. Harvesting of the *Actinomycete* was difficult because filtration rates were poor due to the "gluey" nature of this biomass.

The culture identified as the acidophilic *Rhizopus* organism was not considered typical. It appeared to be a bacterium and displayed rods, sometimes in chains of 3-5 rods in length. Some cellular inclusions were very visible in the phase contrast microscope. Growth rates during large-scale production were poor. It was subsequently identified that culture supplied to ORF was that of a contaminant organism. Bulk culturing of this organism was hence terminated.

Under microscopic examination the *Penicillium* species showed branching, septate hyphae with many single conidia that were not always terminally located. The organism formed well-developed pellets (4 to 7 mm diam) when cultivated in submerged culture with gentle agitation and aeration.

Typical productivities for the *Penicillium* species were approximately 0.15 g dry solids per litre fermenter capacity per day.

The *R. arrhizus* showed the typical rhizopus mycelium and spore production under the phase contrast microscope. In submerged culture the biomass formed large, fluffy pellets (10-15 mm diam) which "deflated" upon filtration and did not return to the pelletized nature when re-introduced to water.

The growth rate of the *R. arrhizus* culture was not measured but, based on the yields of wet biomass, was similar to that of the *Penicillium* species.

Based on observations from the biomass production studies, the *Penicillium* species is considered to have the most attractive physical attributes of the three biomasses produced in bulk. It is very resilient and disperses well in a mixed reactor. The pellets are also sufficiently dense as to settle rapidly on standing.

BIOSORPTION STUDIES

BATCH ADSORPTION TESTS

Batch uranium and yttrium biosorption studies were conducted on the three biomasses produced in bulk using Denison minewater, a detailed analysis of which is given in Table 1. Initially, the aim of the testwork was to establish biomass performance under varied conditions, such as pH, metal cation concentration, temperature, and other potentially interfering situations. However, during the program it was quickly discovered that uranium loading onto the biomasses is significantly affected by solution pH alone, regardless of the other factors. Hence, further investigations on the effects of changes in process parameters other than solution pH were not conducted.

Uranium loading rate curves for the three biomasses using pH 2.0 and 4.0 minewater are shown in Figures 1 and 2, respectively. Biomasses used for these studies were inactivated prior to testing by heating in a sterile saline solution (1% w/v) to 80-85°C for 20 min. The tests were conducted in 250-mL Erlenmeyer flasks using a Burrell wrist-action shaker.

Yttrium loadings are not presented because adsorption of this element by all of the biomasses tested was negligible, even at the higher pH values.

The substantial increase in biomass uranium loading performance at higher solution pH values is evident from the results shown in Figures 1 and 2. Biomass final loadings at pH 4.0 were between 8.5 and 12.5 mg U/g dry biomass, while at pH 2.0 they were less than 4 mg U/g dry biomass.

The rate curves at pH 4.0 indicate that *Penicillium* has both the highest loading capacity and the fastest adsorption rate of three biomasses tested.

Equilibrium adsorption studies conducted at pH 4.0 to determine maximum loading capacities gave the following ranking of biomasses:

Biomass	Ranking	U-adsorption (mg/g)
Penicillium	1	21
Actinomycete	1	21
R. arrhizus	2	10

These results would seem to contradict those shown in Figure 2. However, since it appears that none of the biomasses reached equilibrium during the rate tests, the values determined from equilibrium testing would be more accurate.

Biomasses used for all the studies summarized to this point were heat inactivated prior to testing. A study was consequently conducted to compare the performance of viable and heat shocked *Penicillium* and *Actinomycete* in minewater as-received (pH 2.0). The results, shown in Figure 3, indicate that substantially better performance was obtained for both biomasses after heat inactivation. Adsorption rate, and hence apparent adsorption capacity, is increased by heat treatment. This is as indicated in the Senes studies, where the reaction rate constants increased by a factor of 2 or more in cultures inactivated by heat treatment. It appears that heat inactivation induces physical changes in the cell aggregates, causing an increased rate of uranium uptake. However, biomass external physical appearance is not affected by this operation.

BATCH DESORPTION TESTS

Batch desorption studies were conducted on the three biomasses using procedures identical to the batch-loading tests. Eluants used in the study were: H_2SO_4, Na_2CO_3, and NaCl. These were selected based on their effective performance in previous studies by Senes, on their environmental acceptability, and on their compatibility with current uranium milling techniques. Biomass for these tests was loaded using pH 4.0 Denison minewater.

The results are given in Table 2. Na_2CO_3 was shown to be the most effective eluant of the three tested, resulting in 100% desorption on both *Actinomycete* and *Penicillium* after 2 h. Subsequent rate tests conducted on the biomasses indicated that almost complete desorption was affected on the *Penicillium* within 5 min using Na_2CO_3. The *Actinomycete*, however, showed much slower elution with 100% removal achieved only after contacting for the full 2 h.

Elution of the *Rhizopus* was very poor, with a maximum efficiency of only 39.6% (obtained using 0.1N Na_2CO_3).

In general, the results are similar to those obtained during the Senes study, as Na_2CO_3 was found to be the most effective eluant. The *Rhizopus* results obtained here, however, are in disagreement with the Senes program, which reported up to 80% uranium elution from this biomass using sodium bicarbonate. The variance between the two results could be due to different biomass growth procedures, resulting in an alteration of the cell structure.

CONTINUOUS COLUMN TESTS

Continuous column tests were conducted to determine adsorption and elution performance over repeated cycles. *Penicillium* was chosen as the test species because of its good adsorption and elution performance, and its superior physical characteristics.

A 1.5 cm ID burette with a constant head tank feed was used for the tests. The biomass was placed into the column within layers of glass wool and demineralized sand to prevent the escape of cells. The solution flowrate was maintained by adjusting the outlet stopcock position. Adsorption was conducted using pH 4.0 minewater at a flowrate of approximately 6 BV/h. Elution was carried out with 0.1N Na_2CO_3 at 3 BV/h.

Four adsorption/elution cycles were conducted. Prior to the final cycle, 5 bed volumes of 0.02 M EDTA dissodium salt was run through the column to determine the regenerative effects of this chelating agent on the biomass.

Results of the study are given in Table 3. Initially, the *Penicillium* loading was similar to that obtained during batch testing (22.5 mg U/g biomass). However, after repeated cycles the quantity of uranium on the biomass was built up, so that after the third cycle the capacity was up to 36.1 mg/g. Net uranium loading, though, steadily decreased with repeated use. During the third cycle only 15.2 mg/g uranium was adsorbed(35.1% efficiency) compared to the initial 22.5 mg/g (40.6% efficiency). This is probably due more to the inefficiencies in desorption rather than to adsorption limitations.

Unlike the efficient elution shown during batch testing, *Penicillium* desorption performance during column testing was very poor. Initially, only 44.3% of the loaded uranium was removed. This fell to 13.6% after four cycles. This phenomenon is probably due to the uptake of uranium during extended periods of adsorption, in a form that is not readily leachable. Since continuous column testing most closely approaches actual operating conditions, it is imperative that the *Penicillium* elution performance be improved before the biomass can be used effectively in full-scale processes. Continuing testwork should focus on establishing a better understanding of the adsorption and elution mechanisms, with the goal of improving biomass desorption during repeated cycles of use.

EDTA was not found to elute uranium or regenerate the *Penicillium* to its original loading capacity, as shown by the cycle 4 results. Only 13 mg/g loading (30.4% efficiency) was achieved after EDTA washing.

EVALUATION OF BIOMASS

The results of the testwork indicate that the acidophilic *Penicillium* species had the best overall chemical performance and physical characteristics of the three biomasses studied.

The process for the recovery of uranium using biomass is based on the following technical requirements:

- efficient biomass preparation;
- efficient loading of the biomass;
- physical separation of biomass and leach solution;
- elution of the biomass to produce a high concentration eluate;
- physical separation of the biomass and eluate;
- (possible) regeneration of the biomass.

In view of these requirements, as well as the physical and chemical properties of *Penicillium*, a flowsheet was developed based on continuous column operation for biomass loading and elution. This is shown in Figure 4.

The use of columns for loading and elution of the biomass is ideal as the material is retained with the column, and hence losses of biomass due to agitation and transportation do not occur. The challenge in using columns is to obtain an acceptable flowrate through the biomass bed. Acceptable solution flowrates were obtained in the testwork due to the pelletized nature of the *Penicillium* species. Another advantage of this system is that the equipment used is similar to the ion exchange systems widely used for uranium recovery, particularly by Denison Mines and Rio Algom in Elliot Lake.

Since the biomass loading at low pH values was found to be inadequate, adjustment of the minewater to approximately pH 4.0 using lime or, alternatively, magnesia is required. The pH adjustment is carried out in two agitated tanks in series, with the discharge from the second tank sent to the iron precipitate thickener. The underflow from the thickener is sent to vacuum drum filters, where the iron precipitate is filtered and washed before being discharged from the circuit. The drum filtrate is recycled to the thickener. The thickener overflow is then stored in the pregnant solution reservoir ready for feeding to biomass loading.

The biomass loading and elution section consists of six banks of columns, each bank having four columns in series. Each bank operates independently of the others and has its own automatic sequence controller.

A split elution technique has been selected to increase the concentration of pregnant eluate. In this method, half of the eluate from a cycle is stored for reuse in a second cycle, while the other half is fed to yellowcake precipitation. Sodium carbonate has been selected as the eluant.

Yellowcake precipitation is carried out by the addition of sodium hydroxide. The precipitates are thickened and sent to the conventional mill circuit. Overflow is carbonated in flotation cells using carbon dioxide from burner off-gases prior to reuse in the elution circuit.

Biomass production is conducted in two aerobic batch fermenters operating at ambient temperature and pressure, with draft tube agitation. Heat inactivation is also carried out in the fermenters.

All the equipment in the flowsheet can be located underground close to the bacterial leaching areas, with the exception of the yellowcake precipitation circuit, which would be located on surface in the mill.

The preliminary design criteria have been developed for *Penicillium* and are shown in Table 4. The criteria have been selected from the test results and typical uranium milling practice.

Based on the productivities of *Penicillium* determined during the study (0.45 g dry biomass/L fermenter capacity/80 h incubation period) and on current medium costs, the cost of biomass production would be $0.217/dry gram.

This price should be regarded as the "worst possible" value since cheaper industrial sources of glucose, peptone, trace nutrients, etc. should be investigated. At present, since these alternatives have not been evaluated as nutrients, no attempt has been made to base the cost calculation on the current quoted costs for potential industrial substitutes.

No exceptional scale-up problems are envisioned for this flowsheet. However, the operation of the fermentation step under conditions experienced in the mining industry may present some potential difficulties since contamination must be carefully guarded against. This can be avoided by instituting special operating procedures.

The major technical problems related to the conceptual flowsheet deal with (i) poor *Penicillium* elution performance in the columns, and (ii) the ability of *Penicillium* to withstand the conditions experienced in the circuit. More extensive testwork, particularly on a larger scale, will be required to resolve these problems.

CONCLUSIONS

In general, uranium adsorption and elution from Denison minewater was achieved for the three biomasses studied. Of the three organisms, *Penicillium* showed the best adsorption/elution performance and the most attractive physical characteristics.

Commercial-scale application of *Penicillium* using columns for loading and elution has been selected as the most feasible technique, with no exceptional scale-up problems. However, further work on improving the elution characteristics of this biomass and on determining its performance in a large-scale operation are necessary.

Losses of ion exchange resin are very low and resin life is long. Therefore, for biosorption to have any chance of replacing conventional ion exchange, it must have satisfactory performance in these areas and superior performance in other areas, such as loading and elution.

ACKNOWLEDGEMENTS

The authors would like to thank Dr. John Christison of the Ontario Research Foundation for his work in producing the biomasses and Mr. Andrew Marchbank of Denison Mines Limited for his assistance throughout the project.

TABLES

Table 1 - Head analysis of Denison minewater

Element	Concentration mg/L	Element	Concentration mg/L
Co	3.8	Pb	<0.02
Zn	2.3	Ni	2.0
Cd	0.02	Cr	0.11
B	0.44	Na	9.0
Bi	0.16	Th	71
P	8.5	Y	10
Be	0.005	As	0.84
Si	17	U	90
Fe	1380	SO_4^{2-}	5400
Mn	0.88	TSS	6
Ca	101	Cl^-	18
Mg	5.3	Fe^{2+}	19
Cu	2.2	TOC	6.2
Al	28	NO_3^-	92
V	<0.01	Fe^{3+}	1361
Mo	<0.05	PO_4	26

Table 2 - Uranium equilibrium desorption (120 min contact)

Desorbing agent	*Actinomycete*	*Penicillium*	*R. arrhizus*
0.1N H_2SO_4			
Initial bio loading	4.025	12.0	25.0
% desorbed	20.5%	72.5%	34.0%
0.1N Na_2CO_3			
Initial bio loading	4.025	13.0	25.0
% desorbed	100.0%	100.0%	39.6%
0.1N NaCl			
Initial bio loading	4.025	16.4	13.7
% desorbed	12.7%	98.1%	3.5%

Table 3 - Cyclic column adsorption/desorption studies - penicillium

Cycle 1		
Uranium adsorbed	=	22.5 mg/g
Volume minewater through	=	35 BV
Adsorption efficiency	=	40.6%
Uranium desorbed	=	10.0 mg/g
Volume eluant through	=	3.5 BV
Desorption efficiency	=	44.3%
Final biomass loading	=	12.5 mg/g
Concentration factor*	=	1.8
Cycle 2		
Uranium adsorbed	=	19 mg/g
Volume minewater through	=	22 BV
Adsorption efficiency	=	56.1%
Uranium desorbed	=	10.6 mg/g
Volume eluant through	=	3 BV
Desorption efficiency	=	33.6%
Final biomass loading	=	20.9 mg/g
Concentration factor	=	3.1
Cycle 3		
Uranium adsorbed	=	15.2 mg/g
Volume minewater through	=	45.5 B
Adsorption efficiency	=	35.1%
Uranium desorbed	=	7.5 mg/g
Volume eluant through	=	3 BV
Desorption efficiency	=	20.8%
Final biomass loading	=	28.6 mg/g
Concentration factor	=	2.6
EDTA Wash		
Volume through	=	5 BV
No uranium eluted		
Cycle 4		
Uranium adsorbed	=	13 mg/g
Volume minewater through	=	48 BV
Adsorption efficiency	=	30.4%
Uranium desorbed	=	5.6 mg/g
Volume eluant through	=	5 BV
Desorption efficiency	=	13.6%
Final biomass loading	=	36.0 mg/g
Concentration factor	=	3.1

*concentration factor = $\frac{\text{Conc of U in adsorbate}}{\text{conc of U in desorbate}}$

Table 4 - Preliminary design criteria for penicillium

Area 01 - Fermentation	
No of fermenters	2
Fermenter type	Aerobic/airlift
Method of operation	Batch
Retention time	80 h
Solids content (glucose)	5 g/L
Operating temperature	Ambient
Operating pressure	Atmospheric
Biomass loss in the adsorption-desorption process	10%
Air addition rate	2 L/min x fermenter volume
Inactivation temperature	80°C
Biomass yield	0.45 g (dry) biomass/L fermenter capacity
Area 02 - Biomass filtration	
Type of filter	Belt filter or equivalent
Area 03 - pH adjustment	
Leach solution feed rate	100 m^3/h
Precipitation method	Lime addition [$Mg(OH)_2$ preferred]
pH of leach solution feed	2
pH in precipitation vessel	4
Operating temperature	Ambient
No. of stages	2
Retention time per stage	30 min
Liquid/solid separation	Thickener/clarifier + drum filter
Area 04 - Biomass loading	
Leach solution feed rate	100 m^3/h
Leach solution assay	100 mg U/L
Net biomass loading	25 mg U/g dry biomass
Method of loading	Columns
No. of stages	6 parallel stages with 4 columns in series per stage
Leach solution:biomass ratio	30:1 (by volume)
Area 05 - Biomass elution	
Type of system	Common to biomass loading
Eluant	Sodium carbonate
Retention time/stage	15 min
% uranium recovery in elution	95% (based on batch tests) 40% (based on column tests)
Elution technique	Split elution

Table 4 (cont'd)

Area 06 - Yellowcake precipitation	
Method	Caustic soda
No. of stages	2
Retention time/stage	6 h
Area 07 - Yellowcake dewatering	
Type of system	Thickener
Underflow solids density	30% solids
Area 09 - Carbonation	
Method	Flue gases from combustion
Type of system	Flotation cells

FIGURES

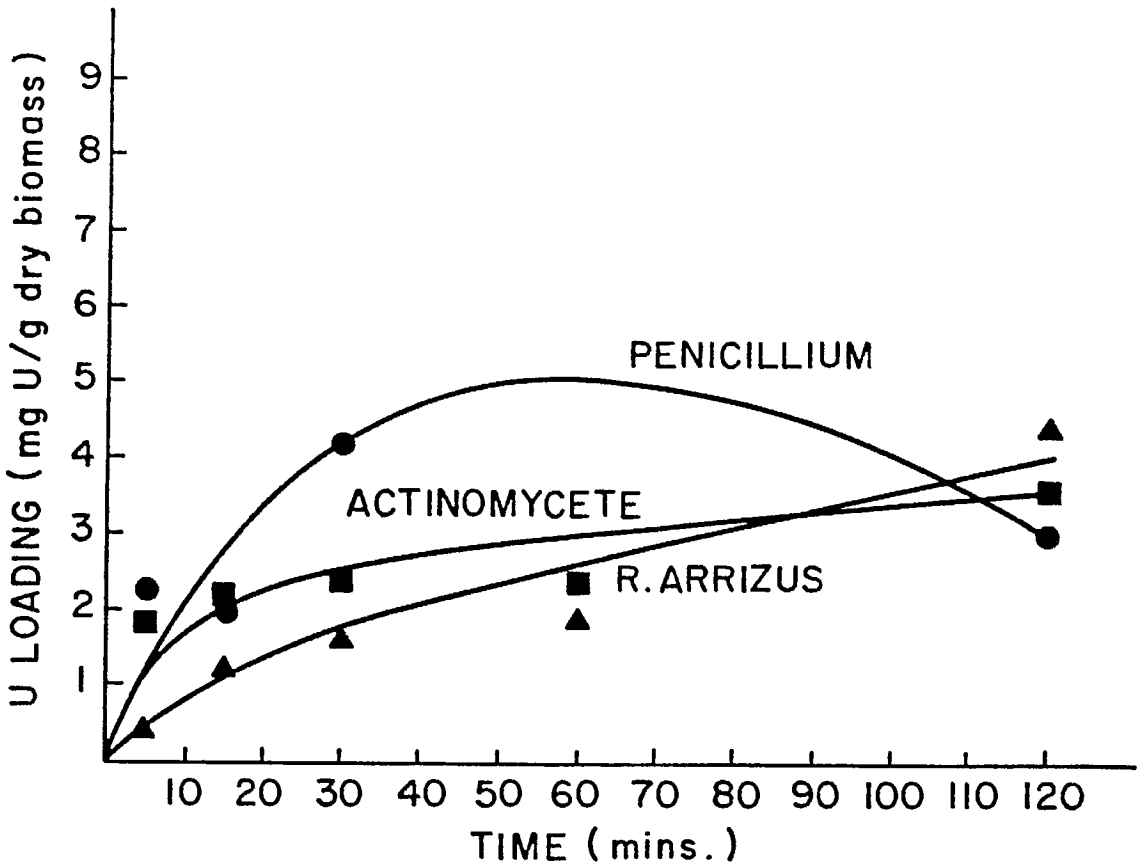

Fig. 1 - Biomass uranium adsorption rate minewater as received (pH 2.0) 0.5 g biomass/100 mL solution

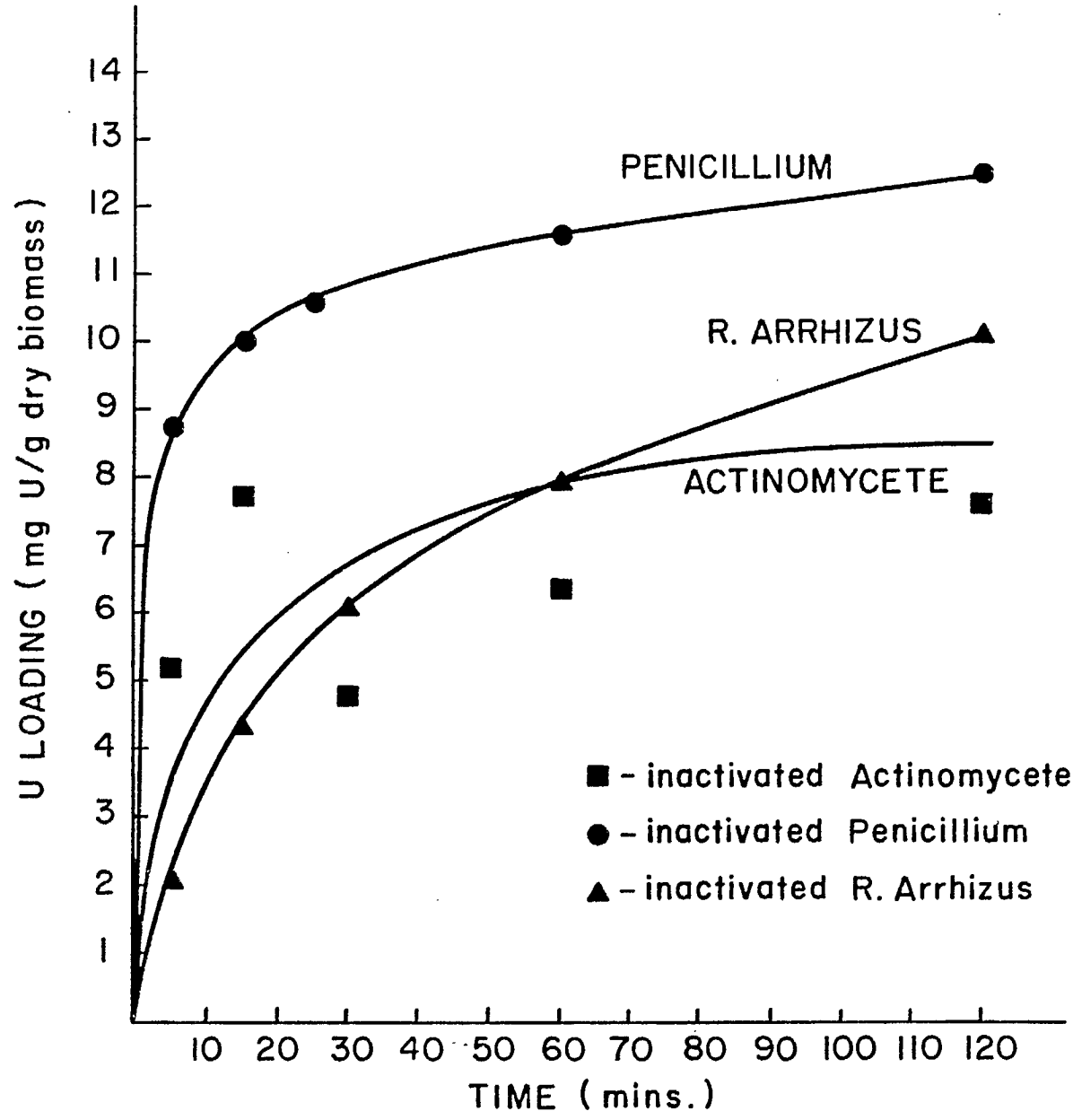

Fig. 2 - Biomass uranium adsorption rate minewater adjusted to pH 4.0 0.5 g biomass/100 mL solution

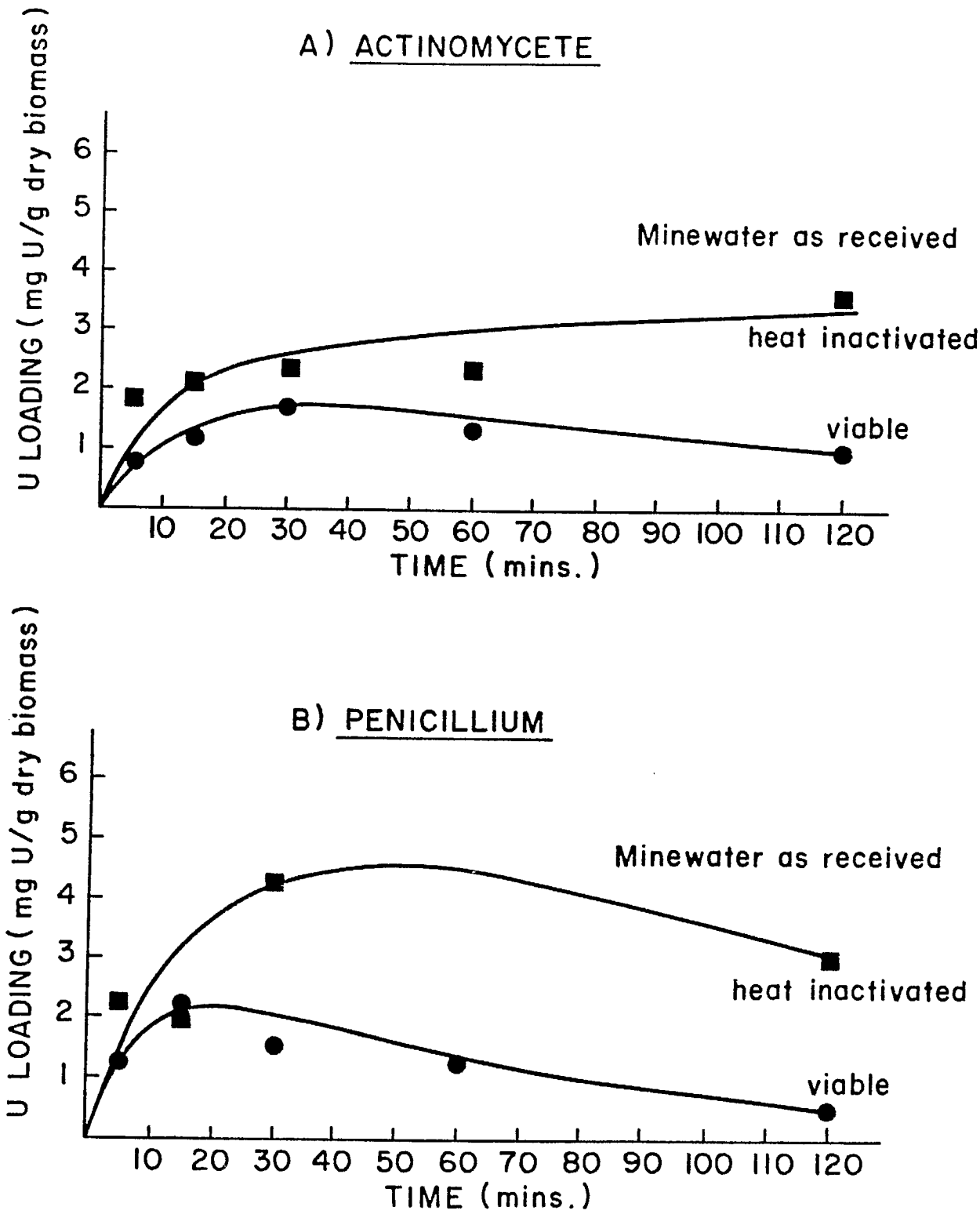

Fig. 3 - Effect of heat inactivation on uranium adsorption rate

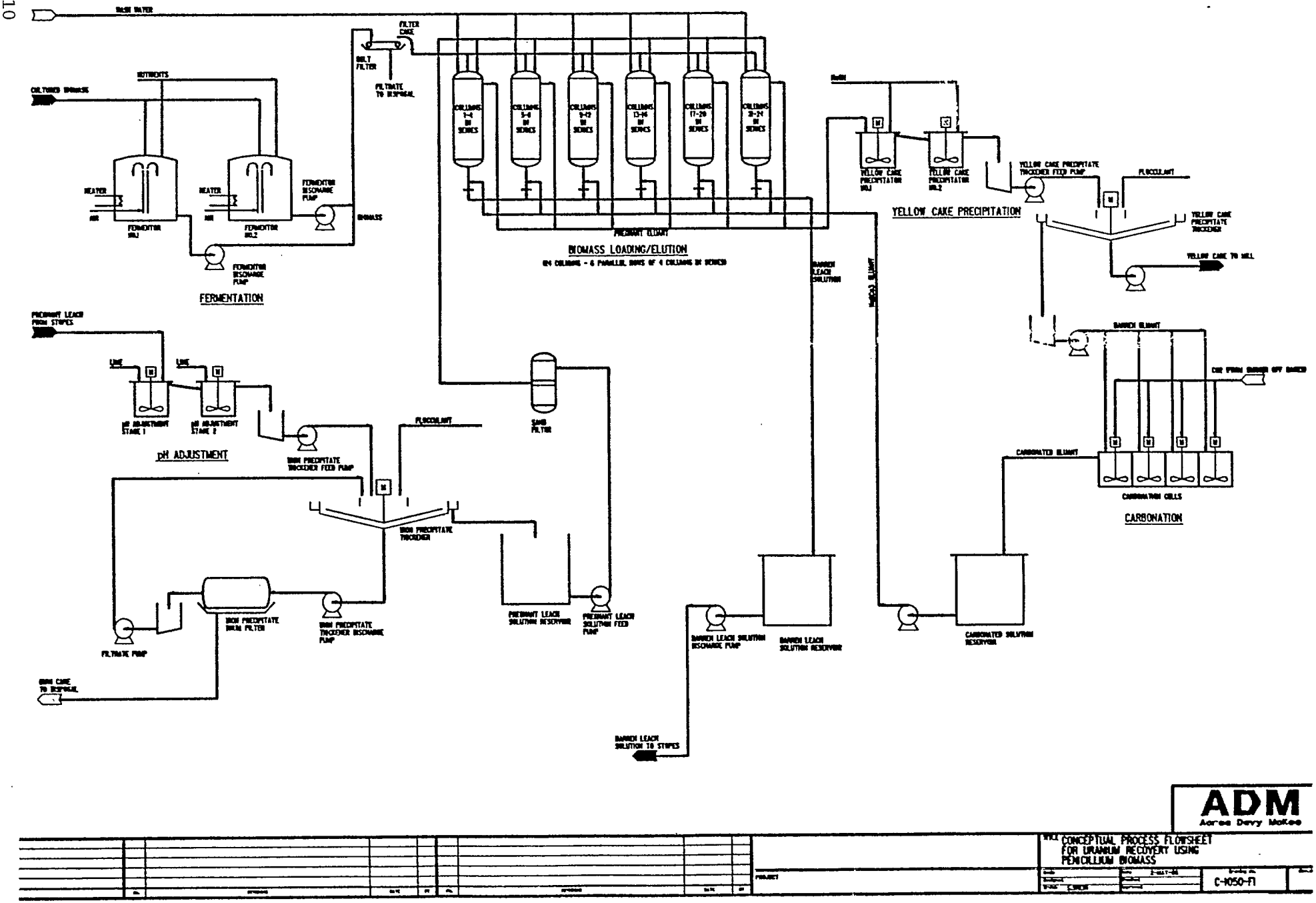

Fig. 4 - Proposed flowsheet for bioadsorption and recovery of uranium

SESSION II: PAPER 7

RECOVERY OF COBALT BY A NEW BIOSORBENT

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ABSTRACT

Extensive screening revealed a seaweed (alga) biomass type that in its non-living state possesses a very high cobalt sequestering capacity, comparable to that of activated carbon and the ion-exchange resin currently used in the cobalt production processes. Performance of the new biosorbent material was studied by examining its equilibrium metal uptake isotherms generated under different solution conditions such as initial cobalt concentration, pH, temperature, and the presence of selected cations and anions. The rate of cobalt uptake, a crucial parameter for recovery process design, was determined. Almost all cobalt sorbed can also be desorbed with a solution of $CaCl_2$. The biosorbent material can be used repeatedly in the sorption-desorption cobalt concentration process.

SESSION II: PRÉSENTATION 7

RÉCUPÉRATION DU COBALT AU MOYEN D'UN NOUVEAU BIOSORBANT

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RÉSUMÉ

Après une sélection poussée, on a découvert un type d'algue marine qui, à l'état non vivant, a pour le cobalt un pouvoir séquestrant aussi remarquable que le charbon activé et la résine échangeuse d'ions qu'on utilise actuellement dans les procédés de production de cobalt. On a étudié le comportement de cette nouvelle substance biosorbante en examinant ses isothermes de sorption des métaux à l'équilibre dans différentes conditions de solution: concentration initiale de cobalt, pH, température et présence de certains cations et anions. La vitesse de sorption du cobalt qui est un paramètre critique dans la conception des procédés de récupération a été déterminée. Presque tout le cobalt sorbé peut aussi être désorbé à l'aide d'une solution de $CaCl_2$. La substance biosorbante peut être réutilisée plusieurs fois dans le procédé de concentration du cobalt par sorption-désorption.

RECOVERY OF COBALT BY A NEW BIOSORBENT

INTRODUCTION

It has been well documented that certain types of microbial biomass, living or non-living, possess a high potential to sequester and accumulate inorganic ions present in aqueous solutions (1-4). Metal concentration by microbial biomass that may be readily available, sometimes as a by-product of fermentation processes, is termed biosorption. This phenomenon is quite complex and includes adsorption of metals onto the surface of a microorganism and/or into the cellular structure in combination with ion-exchange, complexation, and/or microprecipitation. Biosorption is generally considered to be a rapid physical-chemical process. The cell wall structure is porous and allows metal ions, as well as large organic nutrient molecules, to pass freely through (4). Polysaccharides of the cell wall could provide binding amino, carboxyl, phosphate, and sulphate groups (5). In addition to these functional binding groups, polysaccharides often have ion-exchange properties (6). Proteins in the cell wall could offer functional groups and also offer peptide bonds to bind ions. Sometimes, bivalent cations such as Ca^{2+} or Mg^{2+} help the formation of ionic bond between the negatively charged cell surfaces and cations in the solution. Interactions between metal and the cell wall components are responsible for the sequestering of metals by biosorption. Since many metals feature a complex solution chemistry (7), it is not always possible to determine what ionic species are actually present in the solution. The equilibria involved are dependent upon pH concentration, the anions present, and other factors. The transport system for the ions is sometimes dependent on both temperature and energy (8). Biosorbent materials could also be regenerated for multiple reuse (9).

The extraordinary usage of cobalt in many important industrial applications, and the main occurrence of cobalt resources in the politically unstable areas of the world, make it one of the most important and strategic metals. The current and foreseen requirements for cobalt provide an impetus for development of new, feasible recovery processes, as well as for establishment of practical secondary recovery procedures applied in conjunction with environmental concerns for decontamination of, e.g., electroplating solutions. Newly discovered metal sequestering properties of certain types of biomass presented herein have resulted from studies focussing on investigation and development of a new line of sorbent materials for removal and recovery of cobalt from solutions.

MATERIALS AND METHODS

Samples of biosorbents based on microbial biomass originating from natural sources have been collected and processed. About 20 different types of biomass, including fungi and marine algae, were made available and are listed in Table 1. Anion exchange resin IRA-400 (Rohm-Haas Co), cation exchange resin Duolite-C20 (Rohm-Haas Co), and activated carbon were also examined for comparison. The equilibrium biosorptive uptake capacity (q) has been determined as a function of cobalt equilibrium (residual) concentration, using the standard procedure for determination of activated carbon adsorption isotherms.

Cobalt solutions were contacted with the biomass samples in 500-mL flasks. The suspension volume was always 200 mL unless otherwise specified. Solutions were mixed at 180 rpm on a rotary shaker for 16 h at room temperature.

Initial cobalt solution concentrations ranged from 10 mg/L to 1000 mg/L. Cobalt biosorption isotherms were determined with the range of initial concentrations at pH 4.0.

Separation of the biomass from the solution was achieved by vacuum filtration using sartorius membrane filters of 0.45 μm average pore diameter; these filters present the least washable total organic carbon (2).

Cobalt solutions of desired concentration were prepared by dissolving [$CoCl_2 \cdot 6H_2O$] in distilled deionized water. Atomic absorption spectrometry was used for the chemical analysis of cobalt in the solution.

In order to confirm the retention of cobalt on the biomass following the biosorption contact, the metal mass balance was closed by analyzing the cobalt-laden biomass for cobalt content. For this purpose, the exposed biosorbent was digested with the mixture of percholaric acid (1 part) and nitric acid (2.5 parts). An instrumental analysis of the exposed biomass was also carried out by means of X-ray EDA mounted on a JEOL-JEMCX electron microscope. Such analysis provided information relative to the cobalt content of the biomass before and after the biosorption experiments.

RESULTS

All examined biomass samples exhibited a certain degree of biosorption of cobalt, with the maximum equilibrium uptake values for different biomass types ranging from a few mg/g up to 170 mg/g (initial concentration was 1000 mg/L). As shown in Figure 1, the most efficient biosorbent was characterized by a desirable steep biosorption isotherm and was capable of reducing the equilibrium cobalt concentration to less than 2 mg/L at approximately q = 16 mg/g. It exhibited a very high cobalt uptake capacity, outperforming ion exchange resin Duolite-C20 used for the removal of cobalt.

Observed differences were not significant for temperature changes from 23°C to 40°C and from 4°C to 40°C (Fig. 2). A significant negative temperature effect was observed at 80°C and over.

The effect of pH on the biosorptive uptake of cobalt was significant. In general, the optimum pH for all biosorbent types, except one, was around pH 4.5 (Table 1). Lower cobalt uptake was observed at pH 2 than at around pH 4 (Fig. 2). The initial cobalt concentration did not have an appreciable effect on biosorption isotherms.

In the preliminary determination of the contact time adequate for the given biosorption experiments, different time intervals ranging from 2 min to 24 h were examined. As seen from the biosorption kinetic curve in Figure 3, cobalt uptake was relatively fast. It was established that more than 90% of the cobalt uptake safely takes place within less than 10 min of contact with the biosorbent material.

The effect on biosorptive cobalt uptake of other ions in the solution was examined. K, Ca, Cu, Fe, Ni, Zn, Cr, Pb, and U as cations, and NO_3^-, CO_3^{2-}, SO_4^{2-}, and PO_4^{3-} as anions were added to the cobalt-containing solution and the effect on the biosorption isotherm of these co-ions was determined for a selected biosorbent. Examinations were carried out at pH values of pH 2 and

pH 4 for different concentrations of co-ions. At pH 4 many cations except K, Ca, and Fe had an appreciable effect on the Co-uptake capacity of biomass, regardless of the initial concentration of co-ion (Fig. 4). At pH 2, a very low co-ion effect, except for Ca and Fe, was observed as was low cobalt uptake (Fig. 5). While the presence of PO_4^{3-} and SO_4^{2-} anions in the cobalt solutions at pH 4.0 did not exhibit any effect, NO_3^- proved to be the strongest inhibitor of the biosorptive cobalt uptake regardless of the solution pH and the initial concentration.

Almost all cobalt sorbed can also be desorbed with a solution of 0.1 M $CaCl_2$ while the liquid-solid ratio was 10. The sorption-desorption cycle of cobalt was repeated 5 times (Fig. 6).

DISCUSSION

Biosorbent material is mainly composed of organic components of biological origin such as protein, polysaccharides (algin in case of algae), lipids, and some inorganic components occurring naturally such as salts of metal cations as Na, K, Ca, and Mg (Fig. 7 and 8). Generally, they become a negatively charged anionic polymer, in which $-COO^-$ groups form as a result of mild acid hydrolysis (10). The hydrolysis products of Co^{2+} from $CoOH^+$, Co_2OH^{3+}, $Co_4(OH)_4^{4+}$ to $Co(OH)_4^{2-}$ have all been well established (7), depending on the pH of the solution. Perhaps, an electrostatic interaction occurs between the negatively charged surface of biomass and positively charged cobaltous ions, resulting in their binding at around pH 4.5.

It is known that the effect of temperature on ion exchange equilibria is very small, and that adsorption and ion exchange are considerably exothermic and rapid phenomena (2). Experimental results could imply that cobaltous ions could possibly exchange with some cations of biosorbent.

A recent study by our coworkers demonstrated the relationship between the ionic size and sorption of metal ions into the sorbent material (11). The uptake mechanism might depend on the charge and could be correlated with the ionic radius or the hydrated radius of the ion, including electrostatic attraction. The knowledge of the solution chemistry of metals is essential for the interpretation of biosorption results. To overcome co-ion effect on the biosorption of cobalt, more study is required. A process might be developed that could apply to either the sorption stage or to the desorption stage. It might also serve as a basis for elution of the sequestered metal, opening the welcomed possibility of regeneration and multiple re-use of the biosorbent in many subsequent application cycles.

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TABLE

Table 1 - Comparative adsorption of cobalt by natural adsorbants and selected resins

Species of biomass	Maximum uptake (q) $\frac{\text{mg Co}}{\text{g biomass}}$	Optimum pH
Fun A	95	4.0
Fun R	82	4.0
MaL D	47	4.0
MaL P	30	4.0
MaL I	45	4.0
MaL S	63	4.5-5.5
MaL U	100	4.5
MaL A	176	4.5-5.5
MaL H	83	7.0
MaL G	51	4.0
MaL K	125	4.5-5.5
MaL L	92	4.5
IRA 400	28	5.5
Duolite C20	88	4.5
Activated carbon	75	4.0

FIGURES

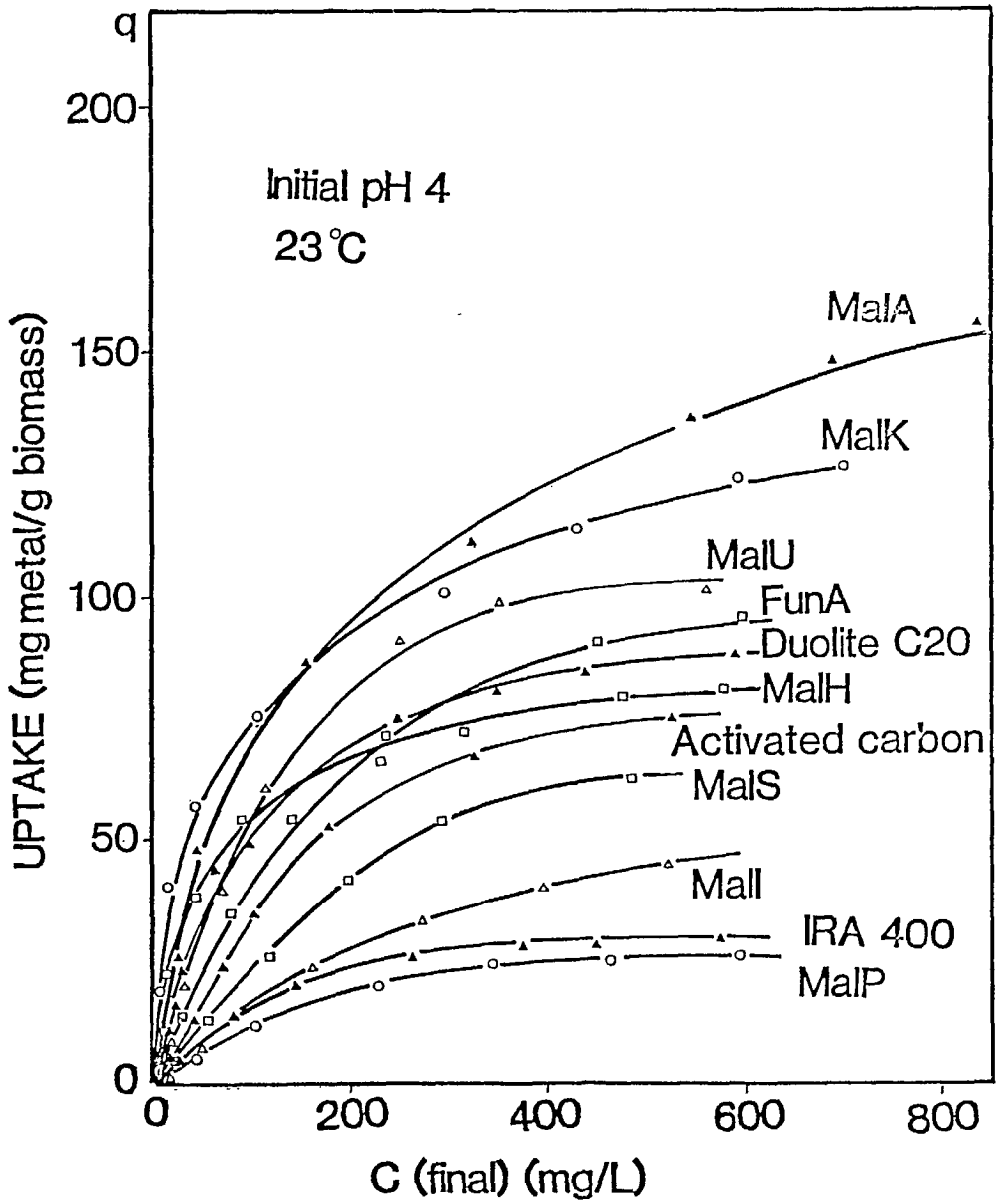

Fig. 1 - Sorption of cobalt by examined biosorbents

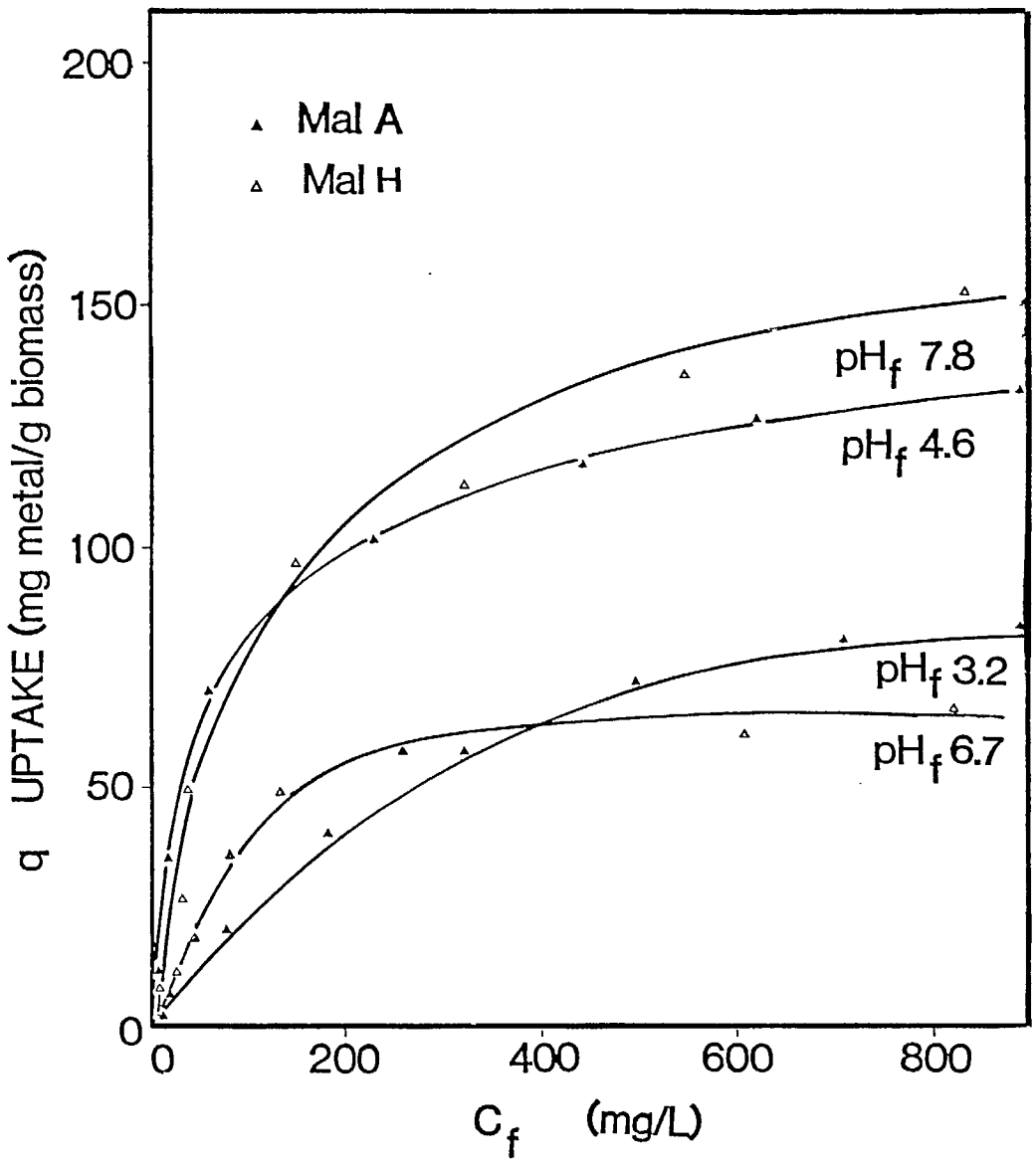

Fig. 2a - The pH effect on the biosorption of cobalt by biosorbent MalA

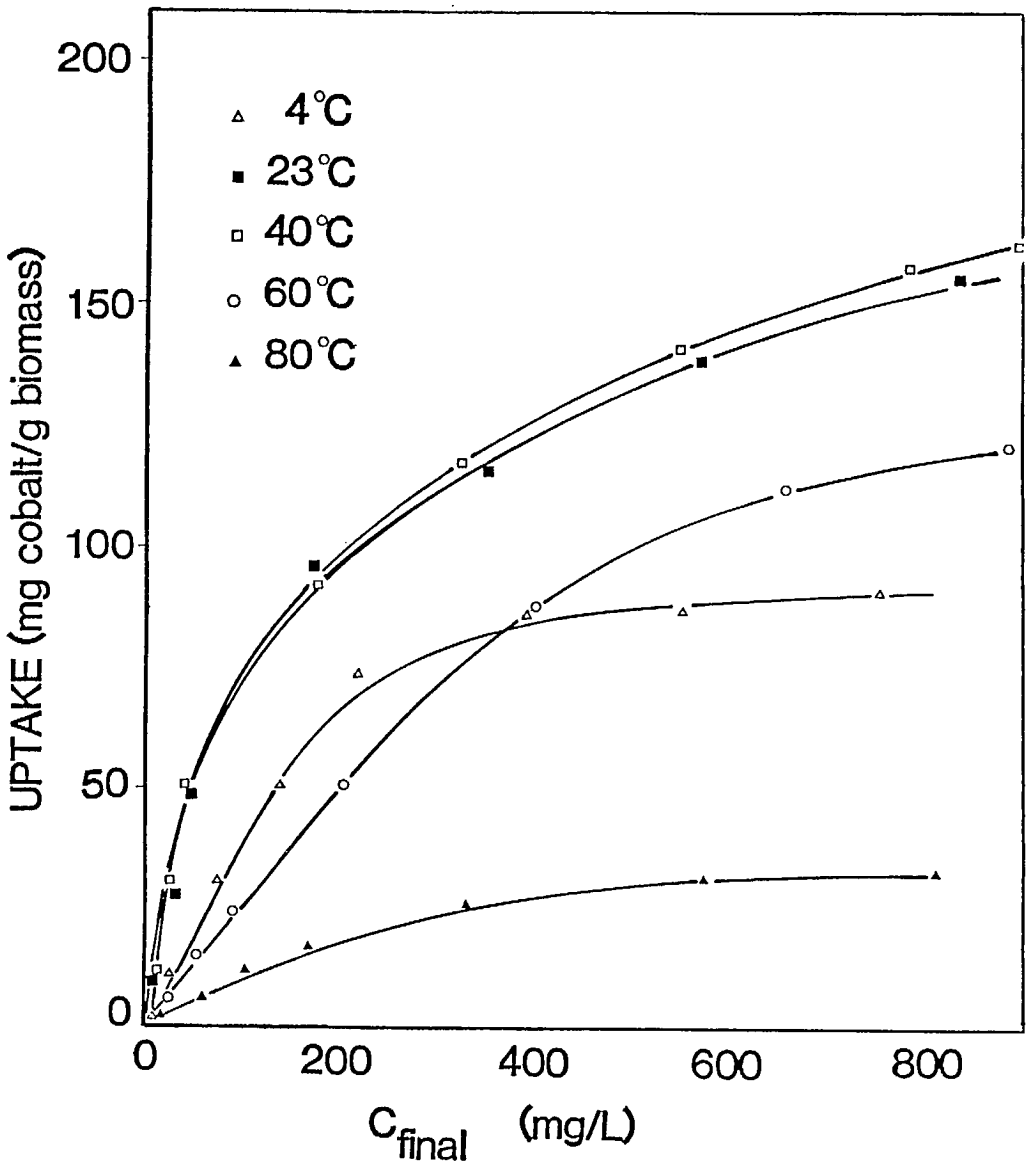

Fig. 2b - The effect of temperature on the biosorption of cobalt by biosorbent MalA

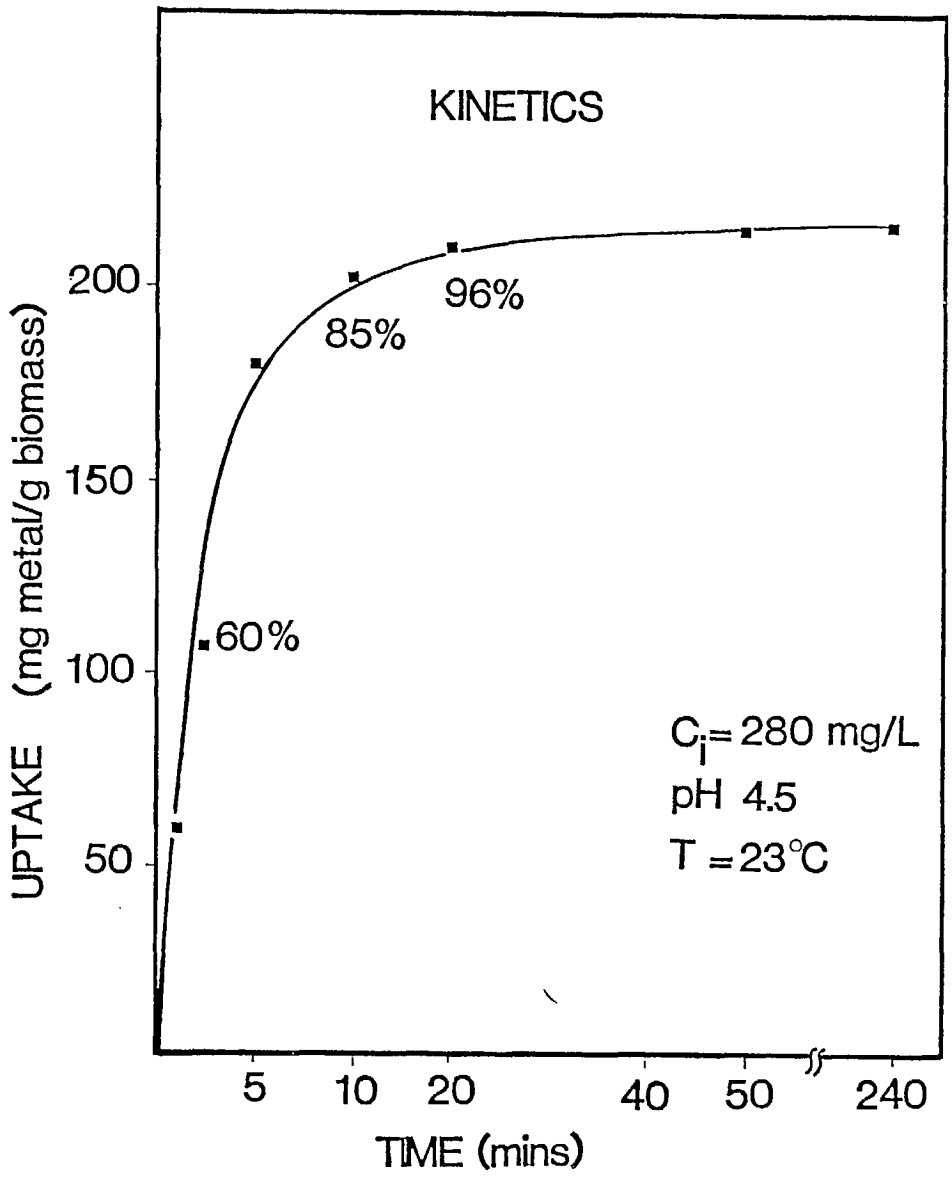

Fig. 3 - Kinetics of biosorption of cobalt by biosorbent MalA

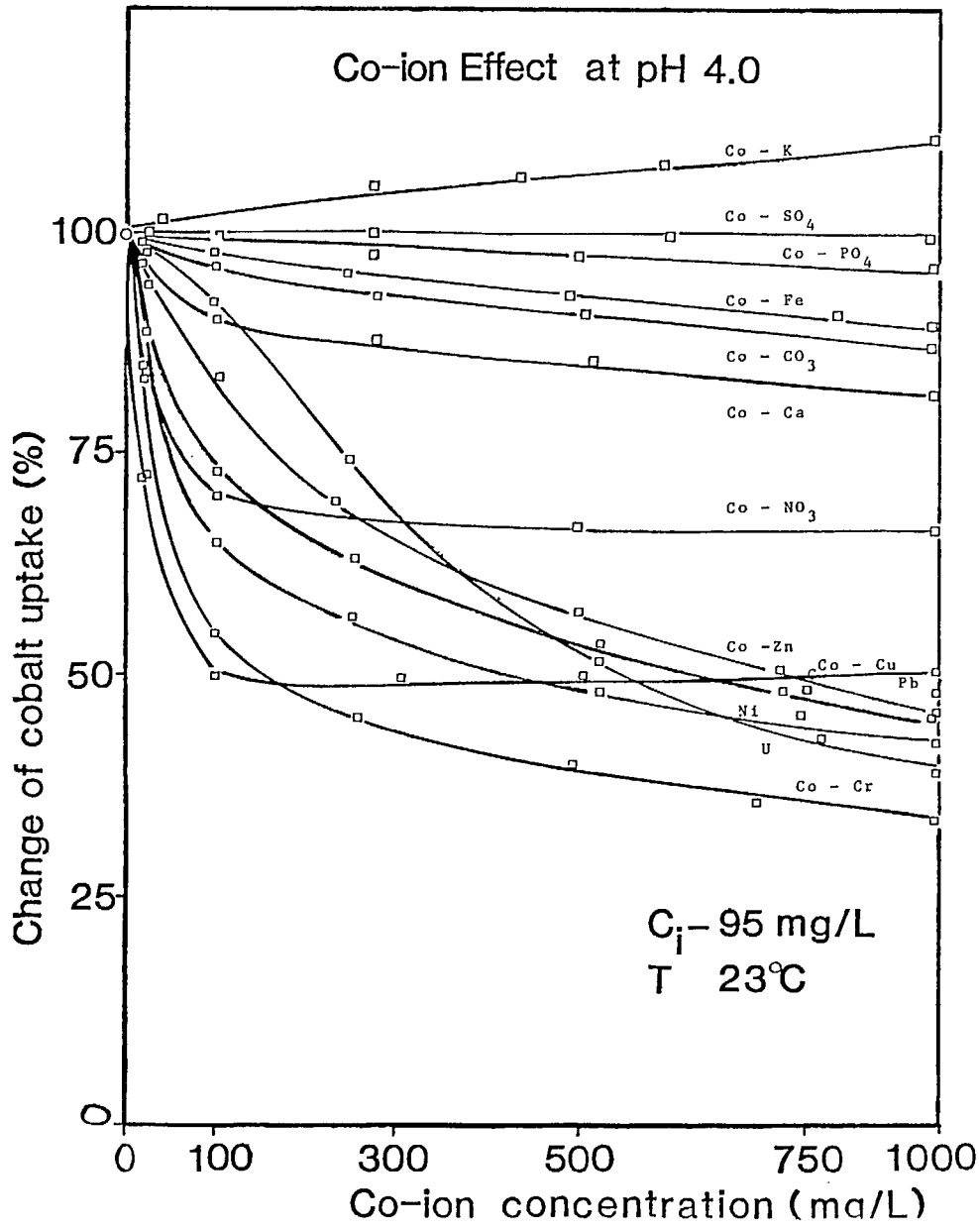

Fig. 4 - Effect of co-ions on the biosorption of cobalt at pH 4

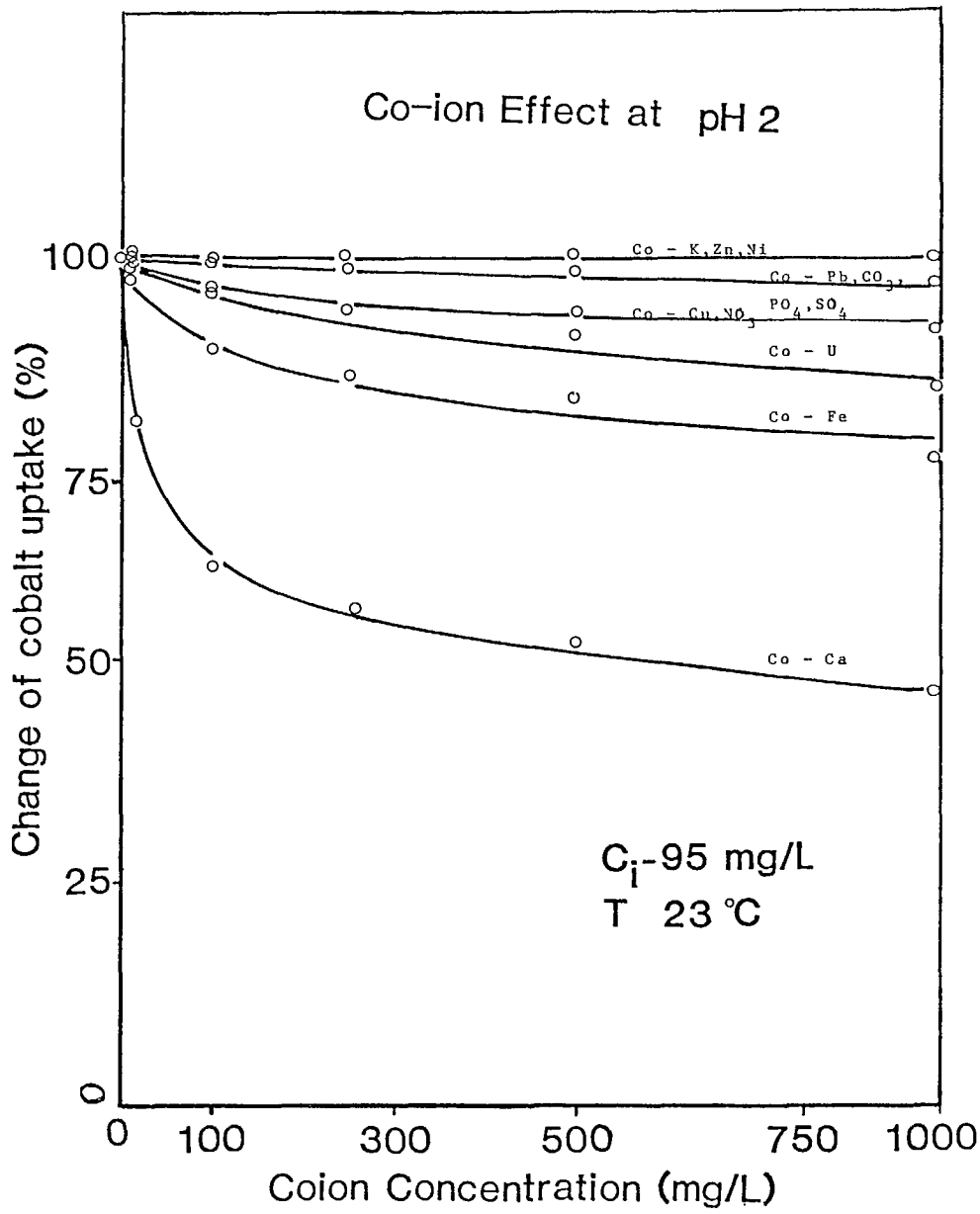

Fig. 5 - Effect of co-ions on the biosorption of cobalt at pH 2

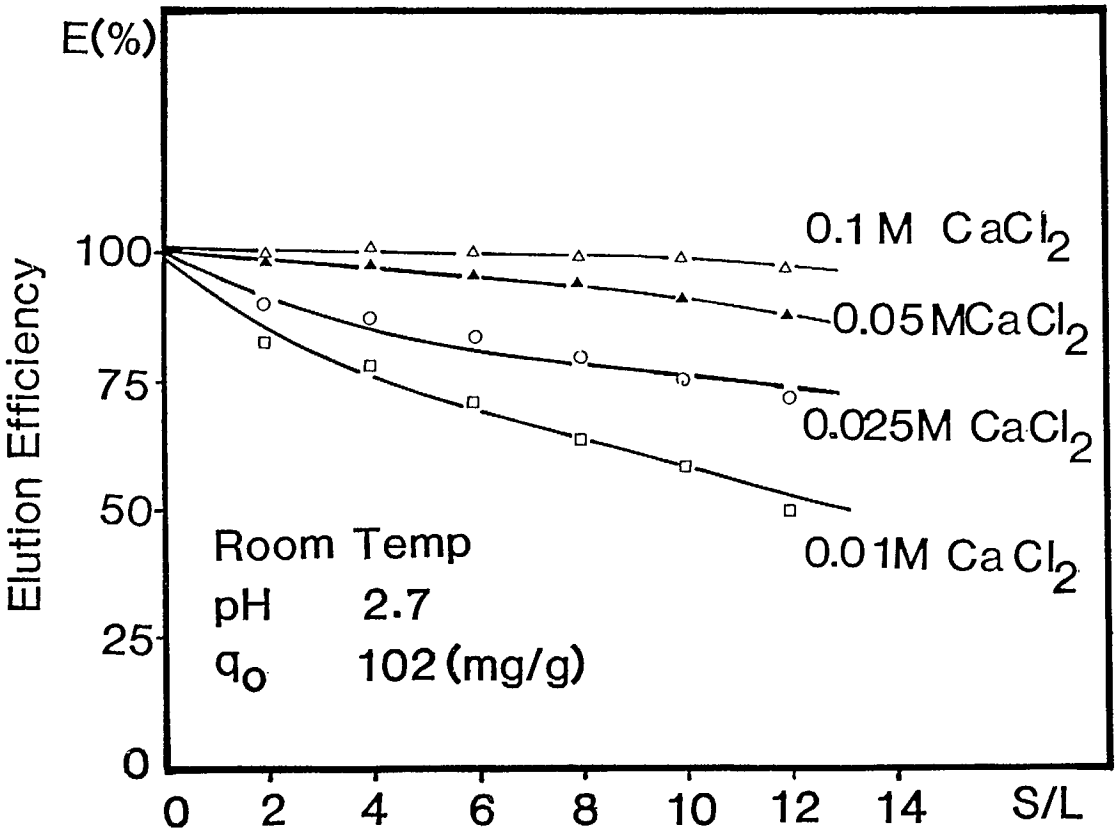

Fig. 6 - Desorption of cobalt from biosorbent MalA using $CaCl_2$ solution

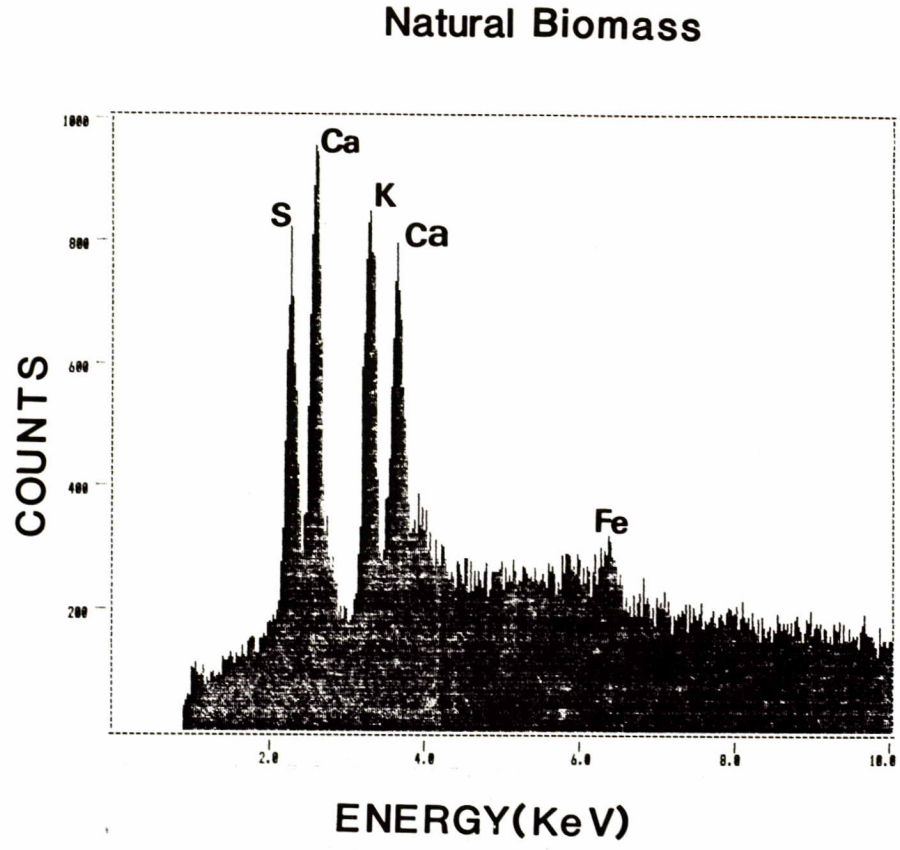

Fig. 7 - EDAX analysis of native MalA

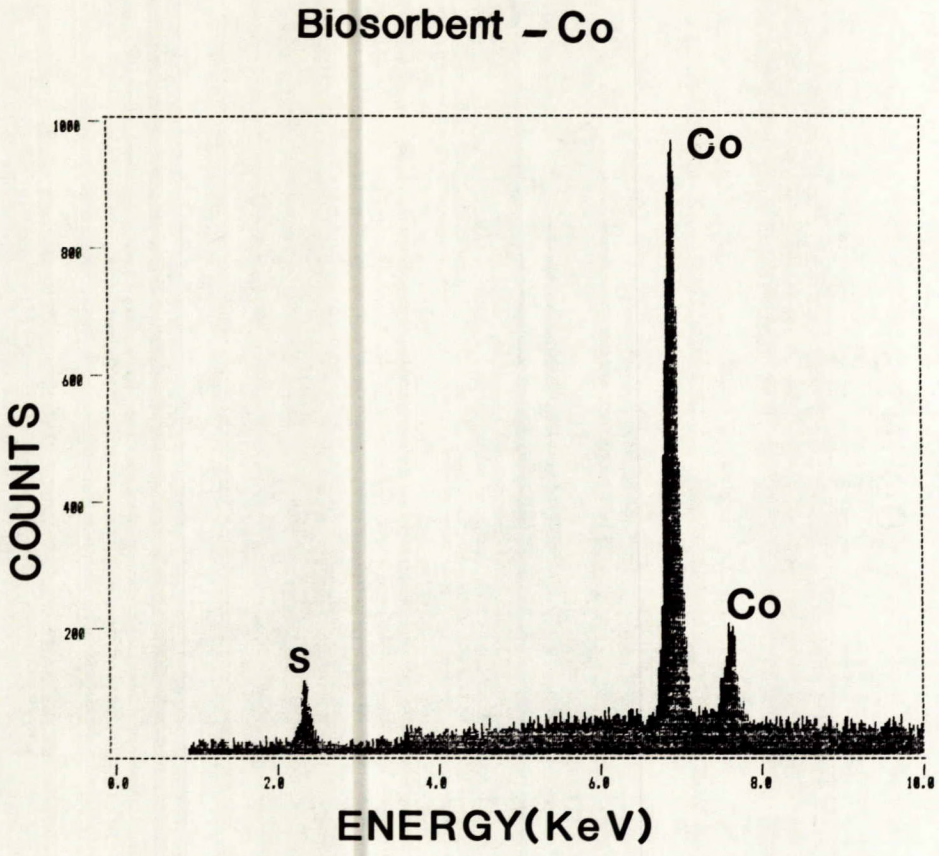

Fig. 8 - EDAX analysis of cobalt-laden MalA

SESSION II: PAPER 8

BACTERIAL LEACHING OF A COMPLEX SULPHIDE ORE

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ABSTRACT

Bacterial leaching of a sulphide ore containing chalcopyrite, sphalerite, and pyrite as major components was studied with *Thiobacillus ferrooxidans* and *Thiobacillus thiooxidans* isolated from the mine sites. Six *T. ferrooxidans* and two *T. thiooxidans* strains were isolated. Shake flask leaching experiments of powdered ore showed effective leaching of Cu and Zn by *T. ferrooxidans* or *T. thiooxidans* strains alone, and also by combinations of both. Ore-adapted bacteria generally performed better than non-adapted ones. *T. thiooxidans* was better in maintaining the acid pH of the medium and in increasing the Cu-Zn extraction ratio. In mixed culture experiments, *T. thiooxidans* seemed to predominate in the low-phosphate (0.2 mM K_2HPO_4) medium, while *T. ferrooxidans* predominated in high-phosphate (0.6 mM K_2HPO_4) medium. In column-leaching experiments with crushed ore, ore plus sand, ore plus slag, and ore plus tailings, effective leaching of Cu and Zn was achieved in all cases except in the tailings column. The Cu-Zn extraction ratio was much higher in the column leaching than in the shake flask leaching, especially after introduction of *T. thiooxidans* in addition to *T. ferrooxidans*. The phosphate concentration of 0.6 mM instead of 0.2 mM K_2HPO_4 was beneficial in maintaining the acid pH during leaching experiments, both in shake flask experiments and in column experiments.

SESSION II: PRÉSENTATION 8

LIXIVIATION BACTÉRIENNE D'UN MINERAI SULFURÉ COMPLEXE

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RÉSUMÉ

L'étude a porté sur la lixiviation bactérienne d'un minerai sulfuré contenant principalement de la chalcopyrite, de la sphalérite et de la pyrite, au moyen de *Thiobacillus ferrooxidans* et de *T. thiooxidans* isolés dans des chantiers miniers. Six souches de *T. ferrooxidans* et deux souches de *T. thiooxidans* ont été isolées. Des expériences de lixiviation en fiole d'un minerai en poudre ont montré que les souches *T. ferrooxidans* ou *T. thiooxidans* et certaines combinaisons des deux lixivient efficacement le Cu et le Zn. Les bactéries adaptées au minerai donnent en général de meilleurs résultats que les bactéries non adaptées. *T. thiooxidans* a été le plus efficace à maintenir le pH acide du milieu et à augmenter le taux d'extraction de Cu/Zn. Dans les expériences sur des cultures mixtes, *T. thiooxidans* a semblé dominé dans le milieu pauvre en phosphate (0,2 mM K_2HPO_4), tandis que *T. ferrooxidans* a semblé dominé dans le milieu riche en phosphate (0,6 mM K_2HPO_4). Dans des expériences de lixiviation en colonne de minerai broyé, de minerai additionné de sable, de minerai additionné de laitier et de minerai additionné de résidus, on a obtenu une lixiviation efficace du Cu et du Zn dans tous les cas sauf dans la colonne des résidus. Le taux d'extraction de Cu/Zn a été de beaucoup supérieur dans la lixiviation en colonne que dans la lixiviation en fiole, particulièrement après l'introduction de *T. thiooxidans* en plus de *T. ferrooxidans*. La concentration de phosphate de 0,6 mM au lieu de 0,2 mM K_2HPO_4, a permis de maintenir le pH acide autant pendant les expériences de lixiviation en fiole que pendant les expériences de lixiviation en colonne.

BACTERIAL LEACHING OF A COMPLEX SULPHIDE ORE

INTRODUCTION

Bacterial leaching of metals from sulphide ores depends on the activity of *Thiobacillus ferrooxidans* to oxidize ferrous iron to ferric iron, and of *T. ferrooxidans* and *Thiobacillus thiooxidans* to oxidize sulphide to sulphate: $2Fe^{2+} + 1/2\ O_2 + 2H^+ \rightarrow 2Fe^{3+} + H_2O$ and $S^{2-} + 2O_2 + 2H^+ \rightarrow H_2SO_4 \rightarrow 2H^+ + SO_4^{2-}$. The precipitation of ferric iron as ferric hydroxide: $2Fe^{3+} + 6H_2O \rightarrow 2Fe(OH)_3 + 6H^+$ or jarosite: $3Fe(OH)_3 + 2SO_4^{2-} + 4H^+ \rightarrow HFe_3(SO_4)_2(OH)_6 + 3H_2O$ makes the overall ferrous iron oxidation acid-generating, which is essential in maintaining the low pH required for the growth and activity of these bacteria. The ferric iron formed can oxidize sulphide minerals: $MS + 2Fe^{3+} \rightarrow M^{2+} + S^o + 2Fe^{2+}$, where MS is a sulphide mineral and M^{2+} is a solubilized metal. Elemental sulphur, S^o, is oxidized by *T. ferrooxidans* or *T. thiooxidans*: $S^o + 1\ 1/2\ O_2 + H_2O \rightarrow H_2SO_4$. These reactions have been discussed in various recent reviews (1-3).

We have undertaken a pre-feasibility study of bacterial leaching of copper and zinc from a sulphide ore at the Flin Flon Mine of the Hudson Bay Mining and Smelting Co., Ltd., Manitoba Canada (HBM&S) with a contract from HBM&S, under the Canada-Manitoba Mineral Development Agreement. This paper reports the initial phase of the work, including the isolation of bacteria and shake flask and column-leaching experiments.

MATERIALS AND METHODS

ORE

A sulphide ore used was obtained from the HBM&S Flin Flon Mine. The ore contained 6-7% chalcopyrite, approximately 10% sphalerite, 60-70% pyrite plus approximately 5% pyrrhotite, 4% calcite, and some diorite. The elemental analysis was 4.9% Cu, 12.5% Zn, 30% Fe, and 37.5% S. The low Cu ore used for one column experiment has 0.81% Cu, 8% Zn, 30.5% Fe and 43.3% S. The ore was ground to 200 mesh (0.074 mm) for shake flask experiments, and was crushed and screened 1/2 to 1 in. (12.5-25 mm) for column experiments.

MEDIA

Media used for the growth of *T. ferrooxidans* were the 9K medium (4): 3 g $(NH_4)_2SO_4$, 0.1 g KCl, 0.5 g K_2HPO_4, 0.5 g $MgSO_4 \cdot 7H_2O$, 0.01 g $Ca(NO_3)_2$, 1 mL 5M H_2SO_4, and 44.2 g $FeSO_4 \cdot 7H_2O$ per litre; and a modified 9K medium (5); (M9K): 0.4 g $(NH_4)_2SO_4$, 0.1 g K_2HPO_4, 0.4 g $MgSO_4 \cdot 7H_2O$, and 33.3 g $FeSO_4 \cdot 7H_2O$ per litre and adjusted to pH 2.3 with H_2SO_4.

The medium for *T. thiooxidans* was Starkey's medium (6): 0.3 g ammonium sulphate, 3.5 g KH_2PO_4, 0.5 g $MgSO_4 \cdot 7H_2O$, 0.25 g $CaCl_2$, and 18 mg $FeSO_4 \cdot 7H_2O$ per litre. Powdered sulphur (50 g/L) was spread evenly on the surface after inoculation, as described by Suzuki (7).

For leaching experiments, either the M9K medium without ferrous sulphate (HP or high-phosphate medium) or a low-phosphate medium (LP): 66 mg $(NH_4)_2SO_4$, 35 mg K_2HPO_4, and 123 mg $MgSO_4 \cdot 7H_2O$ per litre, and adjusted to pH 2.3 with H_2SO_4 (5) was used.

GROWTH OF BACTERIA

Isolation or growth of *T. ferrooxidans* was carried out in 250-mL Erlenmeyer flasks containing 90 mL of the M9K medium with 10-mL portions of inoculum (either water sample or previous culture). The flasks were incubated at 25°C on a rotary shaker at 150 rpm for several days. *T. thiooxidans* was grown at 25°C in 250-mL Erlenmeyer flasks with 90 mL of Starkey's medium plus 10 mL of water sample for isolation, or in 2-L Fernbach flasks with 900 mL of Starkey's medium plus 100 mL of previous culture for growth of the organism.

SHAKE FLASK LEACHING

Shake flask leaching experiments were carried out in 250-mL Erlenmeyer flasks containing 90 mL of the HP medium or the LP medium, plus 10 g of 200-mesh ore and 10 mL of inoculum, *T. ferrooxidans*, *T. thiooxidans*, or a combination of both. The flasks were incubated at 25°C on a rotary shaker at 150 rpm. Five-millilitre samples were removed periodically, and after filtration and centrifugation were analyzed for Cu, Zn, and Fe by atomic absorption spectrophotometry.

COLUMN LEACHING

A column-leaching study was carried out following, in general, the methods of McCready (5) at 25°C. A clear acrylic column (7.5 cm x 59 cm inside) had a perforated acrylic base 3 cm from the bottom to hold the crushed ore and a bottom liquid reservoir, from which liquid was led by gravity to a culture reservoir (1-L Erlenmeyer flask). The culture was aerated through a glass sparger with a moist air and circulated onto the top of the column at a rate of 2 mL/min by a Gilson Miniplus 2 peristaltic pump. The column top had a liquid inlet and an air outlet. The volume of the ore bed was 2.5 L and that of the bottom reservoir was 0.15 L. The samples used for column experiments were: ore and slag crushed to 1/2 to 1 in. (12.5-25 mm), sand, and tailings (finely ground). Four columns contained ore alone (4 kg), 1.5 kg of ore plus 1.5 kg of sand, 2 kg of ore plus 2 kg of slag, and 1.5 kg of ore plus 1.5 kg of tailings. Each sample was mixed with 4 mL of 95% sulphuric acid, by tumbling in a container before packing in the column. Glass wool was placed on the top surface of the sample. The column was then washed with several bed volumes of distilled water until the pH of effluent became approximately 2. Five hundred millilitres of the low-phosphate (LP) medium was placed in the culture flask and the peristaltic pump was activated to circulate the medium through the column. A culture of iron-grown inoculum in the M9K medium was added to the culture flask to start the experiment. The pH of the flask was measured and adjusted daily with 5 M H_2SO_4 to maintain it at pH 2.3. Every 20 days, 100 mL of fresh medium was added to the flask in Experiment I. Cu, Zn, and Fe concentrations were determined by atomic absorption spectrophotometry as described above.

RESULTS

ISOLATION OF BACTERIA

Seven water samples from various seepage and drainage at different locations in the mine area were examined for the presence of bacteria growing on ferrous iron (*T. ferrooxidans*) or sulphur (*T. thiooxidans* and *T. ferrooxidans*). A

total of eight strains (SM-1 to SM-8) were isolated, six *T. ferrooxidans* (SM-1 to 5 and SM-8) and two *T. thiooxidans* (SM-6 and 7). Although SM-8 was isolated in the sulphur medium, it was not stable in the medium after repeated transfer and was not used in these experiments.

SHAKE FLASK EXPERIMENTS

The isolated *T. ferrooxidans* and *T. thiooxidans* strains, as well as our laboratory strains of *T. ferrooxidans* (ATCC 13661 and 19859, designated as Tf-1 and Tf-2, respectively) and *T. thiooxidans* (ATCC 8085, designated as Tt-1), were studied for their leaching effectiveness.

IRON-GROWN CELLS

T. ferrooxidans grown on Fe^{2+} in the M9K medium failed to leach metals from the ore in the LP medium without the addition of sulphuric acid for the first 4 days. In this low-phosphate (0.2 mM) medium, a 10% 100-mesh ore raised the pH to 5.0 from 2.3 during shaking, probably because of calcium carbonate present as calcite.

The pH rise was less drastic in the HP medium (0.6 mM K_2HPO_4) because of the increased buffering capacity of phosphate, and successful leaching was achieved with a 10% inoculum of Fe^{2+} grown isolates (Table 1 and Fig. 1). Only four of the isolates (SM-1, 2, 4, and 5) were capable of growing on the ore-leaching metals. They also maintained the acid pH. Control (no bacterial inoculation), SM-3, and two laboratory strains of *T. ferrooxidans* (Tf-1 and Tf-2) failed either to leach metals or to maintain the acid pH. In Figure 1 the time-course of a leaching experiment with SM-4 strain is shown, but the results with SM-1, 2, and 5 were similar except that the length of lag period varied before the initiation of leaching by each strain.

SULPHUR-GROWN CELLS

Leaching of the ore by the sulphur-grown bacterial strains is shown in Table 2. SM-6 and SM-7 leached metals effectively and also maintained the acid pH. The laboratory strain of *T. thiooxidans* (Tt-1) was poor in the leaching activity and did not maintain the acid pH. The control flask without bacterial inoculation showed little leaching. Although in this experiment an additional phosphate was introduced by the 10% inoculum (Starkey's medium), SM-6 and SM-7 were capable of producing acid fast enough to maintain the acid pH in the low-phosphate (LP) medium without additional phosphate or sulphuric acid, presumably due to the efficient oxidation of sulphide to sulphuric acid. The leaching of iron was poor and little ferric iron was produced.

ORE-ADAPTED CELLS

Four *T. ferrooxidans* isolates (SM-1, 2, 4, and 5) and one *T. thiooxidans* (SM-6) isolate were selected for a study of ore adaptation to the leaching of metals. The results are shown in Table 3 and Figures 2 and 3. The ore-adapted *T. ferrooxidans* started the leaching of Cu and Zn without a lag period (Fig. 2 and 3), which was present in iron-grown cell experiments (Fig. 1). Both *T. ferrooxidans* and *T. thiooxidans* produced better leaching results in the HP medium than in the LP medium. *T. ferrooxidans* in the LP medium required acid for the first four days to maintain acid pH. *T. thiooxidans*

leaching showed a higher Cu-Zn ratio than *T. ferrooxidans*. SM-2 culture was not stable and apparently died during the experiment.

MIXED CULTURES

In the next set of experiments, a combination of *T. ferrooxidans* (SM-1, 2, 4, and 5) and *T. thiooxidans* (SM-6) was studied. The results are presented in Table 4 and Figure 4. It was observed that the mixed cultures attained the appearance of *T. thiooxidans* (SM-6) in the LP medium: a great deal of metallic sheen on the liquid surface, very little ferric iron production, low Fe leaching, and generally high Cu-Zn ratios. In the HP medium, on the other hand, the cultures attained the appearance of *T. ferrooxidans* (iron grown): no metallic sheen on the liquid surface, a great deal of ferric iron production (yellow colour in the filtrate), high Fe leaching, and generally lower Cu-Zn ratios. It is possible that there was some competition between the two organisms present in each flask, leading to these results. A biphasic plot observed, for example, in the leaching of Cu in the LP medium (Fig. 4) may be due to the leaching of Cu initially by SM-4, followed later by SM-6 (Fig. 2).

In general, the mixed cultures were not significantly better than individual cultures alone in metal leaching under these experimental conditions. An interesting observation was that ore-adapted SM-2, which when used alone was unstable in the ore-leaching experiment (Table 3), proved to be quite stable in the mixed culture experiment with SM-6.

Based on these results, it is clear that no isolate performs best under all circumstances in the leaching of metals; rather, each isolate is best suited for a particular experiment or purpose. SM-6 was selected as *T. thiooxidans* strain and SM-4 as *T. ferrooxidans* strain to be used for column-leaching studies.

COLUMN-LEACHING EXPERIMENTS

Experiment I

Four columns were packed with ore alone (column 1), ore plus sand (column 2), ore plus slag (column 3), and ore plus tailings (column 4) as described in Materials and Methods; leaching experiments were conducted for 47 days with iron-grown SM-4 inocula. The results are shown in Figures 5, 6, and 7a and Table 5. Leaching of Cu and Zn progressed very well in columns 1 to 3, with much higher Cu-Zn ratios than in shake culture experiments. In column 4 (ore plus tailing) no leaching occurred. It was necessary to add almost daily 0.5 mL of 5M H_2SO_4 to maintain the flask pH to 2.3. Total acid volumes required during the 47 days were 18.5 mL, 20 mL, 18.5 mL, and 20 mL, for columns 1 to 4. Column effluent had a pH of 3.5, except in the ore-tailings column where the effluent came out as pH 5.3. This high column pH was obviously detrimental to the growth of bacteria in this column, resulting in no metal leaching from the ore in the tailings column.

Experiment II

Since the shake flask experiments showed that *T. thiooxidans* isolates maintained the acid pH of LP medium without addition of sulphuric acid, the effect

of inoculation of the columns with sulphur-grown SM-6 was studied. The results are shown in Figures 5, 6, and 7b and Table 6. A major effect of *T. thiooxidans'* addition was to increase the extraction of Cu and to decrease that of Zn, approaching equal percentage extractions of Cu and Zn. The effect was particularly striking in the ore-plus-sand column (column 2). In column 4 (ore plus tailings), there was still no leaching except for a trace of Zn. It was still necessary to add acid to maintain the acid pH of the medium. Total volumes of 5 M H_2SO_4 added during the 30-day experiment were 15 mL, 16 mL, 17 mL, and 16 mL for columns 1 to 4.

Experiment III

In the shake culture experiments, the use of a high-phosphate medium (0.6 mM instead of 0.2 mM) had a beneficial effect on the maintenance of acid pH. The phosphate concentration was therefore increased to 0.6 mM in Experiment III. The results are shown in Figures 5, 6, and 7b and Table 7. As far as the extraction of metals is concerned, there was no marked difference from that in Experiment II. The Cu extraction decreased slightly in columns 1 and 2, lowering the Cu-Zn ratio. The extraction of both Cu and Zn increased in column 3, but the Cu-Zn % extraction ratio decreased. A dramatic effect was seen in the maintenance of acid pH in columns 1 and 2. After 20 days, these columns required no daily acid addition to remain at pH 2.1. Thus, total volumes of 5 M H_2SO_4 added during the 30-day experiment were 10 mL, 10 mL, 14.5 mL, and 14.5 mL for columns 1 to 4. There was still very little leaching in column 4 except for a small amount of Zn.

DISCUSSION

Six strains of *T. ferrooxidans* and two strains of *T. thiooxidans* were isolated from mine water samples. They were shown to be more effective in leaching metals from the ore than the laboratory strains of these bacteria (Tables 1 and 2), probably because of their adaptation to, or selection by, the natural habitat in the mine.

Both iron-grown *T. ferrooxidans* and sulphur-grown *T. thiooxidans* isolates were capable of leaching Cu and Zn from the ore effectively in shake cultures (Tables 1 and 2, Fig. 1), but the leaching efficiency was improved with ore-adapted bacteria (Table 3 and Fig. 3). In the case of *T. ferrooxidans*, the lag period was abolished by ore-adaptation. *T. thiooxidans* had a higher Cu-Zn extraction ratio, particularly in the low-phosphate (LP) medium, produced very little ferric iron (no ability to oxidize ferrous iron), and required no acid addition to maintain the acid pH. *T. ferrooxidans*, on the other hand, had a low Cu-Zn extraction ratio, produced a large amount of ferric iron, and required acid addition for the first 4 days in the LP medium. Mixed cultures of *T. ferrooxidans* and *T. thiooxidans* were also effective in leaching (Table 4 and Fig. 4), but they were not significantly better than individual organisms. Interestingly, *T. thiooxidans* seemed to predominate in the low-phosphate (0.2 mM) medium, while *T. ferrooxidans* predominated in high (0.6 mM) phosphate medium. This was probably due to a competition between the two bacterial species and should be interesting if confirmed by the change in bacterial cell populations.

Column-leaching studies demonstrated a successful Cu and Zn leaching by *T. ferrooxidans* and a mixed culture of *T. ferrooxidans* and *T. thiooxidans* (Tables 5-7 and Fig. 5-7) of crushed ores in columns with ore alone, ore plus sand, and ore plus slag. Tailings inhibited the leaching probably because of raised pH in the column (effluent pH of 5.3), which did not allow the growth of these acidophilic bacteria. These bacteria, nevertheless, were alive in the culture reservoir, although smaller in numbers than in the reservoirs of other columns. The microscopic counting of the number of bacterial cells, as well as the estimation of activities for Fe^{2+} oxidation, sulphur oxidation, and ore oxidation (O_2 consumption on a Gilson Oxygraph using a Clark electrode) in 9K medium at pH 3.3, were carried out at the end of each experiment after collecting cells by centrifugation. These data are only preliminary and do not include bacterial cells attached to ore particles in the columns. They indicate, however, that active bacteria were present in the reservoirs throughout the experiments.

An important result of the column experiments was a much higher Cu-Zn extraction ratio obtained than in shake flasks, particularly in the presence of *T. thiooxidans* (Experiment II). In column 2 (ore plus sand) of Experiment II, nearly equal percentages of Cu and Zn were extracted. This is in contrast to our shake flask results and a recent report of a column-leaching study with a similar sulphide ore (8) where Zn was extracted faster than Cu, resulting in much lower Cu-Zn ratios.

Both the concentration of Cu accumulated (Fig. 5-7), which reached 3.6 g/L in Experiment III (Fig. 5), and the % extraction/30d (Tables 5-7), which reached the values of 1.30 in column 2 (ore plus sand) in Experiment II and 1.35 in column 3 (low Cu ore plus sand) in Experiment III, are excellent results in terms of Cu recovery by bacterial leaching, which was our initial objective.

The lower acid consumption observed in Experiment III suggests a possibility of eliminating or reducing the acid requirement by the use of both *T. ferrooxidans* and *T. thiooxidans* in a high (0.6 mM) phosphate medium.

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TABLES

Table 1 - Leaching by iron-grown isolates

Bacterial strains	Extraction achieved (%) Cu	Zn	Fe	Final pH
SM-1	7.8	76.2	20.7	2.1
SM-2	7.3	67.7	22.0	2.1
SM-3	0.8	13.8	0.0	4.4
SM-4	10.0	68.8	19.8	2.0
SM-5	7.8	72.6	10.3	2.4
Tf-1	0.9	13.4	0.0	4.6
Tf-2	0.9	10.7	0.0	4.7
Control	0.0	8.9	0.0	4.8

Shake flask leaching of 200-mesh ore (10 g/100 mL of HP medium) at 25°C with a 10% inoculum (except control) of iron-grown bacteria.

Table 2 - Leaching by sulphur-grown isolates

Bacterial strains	Extraction achieved (%) Cu	Zn	Fe	Final pH
SM-6	3.3	14.3	1.8	2.3
SM-7	2.6	11.4	1.2	2.4
Tt-1	0.9	8.8	0.0	4.5
Control	0.0	3.2	0.0	5.2

Shake flask leaching of 200-mesh ore (10 g/100 mL of LP medium) for 9 days at 25°C with a 10% inoculum (except control) of sulphur-grown bacteria.

Table 3 - Leaching by ore-adapted isolates

Bacterial strains	Extraction achieved (%) Cu	Zn	Fe	Final pH
(a) in LP medium				
SM-1	7.1	97.9	8.9	3.1
SM-4	6.3	99.8	10.3	2.7
SM-5	5.9	77.1	13.3	2.9
SM-6	8.3	25.0	4.1	2.8
(b) in HP medium				
SM-1	7.6	82.4	18.9	2.1
SM-2	2.2	22.1	0.7	3.9
SM-4	8.3	99.2	20.0	2.1
SM-5	8.8	77.4	23.2	2.0
SM-6	10.4	82.6	12.7	1.9

Shake flask leaching of 200-mesh ore (10 g/100 mL of LP medium or HP medium) for 16 days at 25°C with a 10% inoculum from ore-grown cells (previous leaching experiment flasks in HP medium, after removal of ores by filtration). In the low-phosphate (LP) medium, it was necessary to add to each flask 70 µL of 5 M H_2SO_4 at inoculation time and every 24 h for three more days except SM-6.

Table 4 - Leaching by mixed cultures

Bacterial strains	Extraction achieved (%) Cu	Zn	Fe	Final pH
(a) in LP medium				
SM-1 + SM-6	10.0	64.0	9.7	1.9
SM-2 + SM-6	8.8	96.3	12.9	2.1
SM-4 + SM-6	8.2	41.6	8.4	2.3
SM-5 + SM-6	10.6	50.6	9.2	2.1
(b) in HP medium				
SM-1 + SM-6	9.8	76.8	26.5	1.9
SM-2 + SM-6	8.4	76.8	23.1	2.0
SM-4 + SM-6	9.4	86.7	22.4	2.1
SM-5 + SM-6	10.2	88.6	22.7	1.9

Shake flask leaching of 200-mesh ore (10 g/100 mL of LP medium or HP medium) for 18 days at 25°C with 5% each of *T. ferrooxidans* and *T. thiooxidans* inoculum from ore-grown cells (previous leaching experiment flask in the respective medium, after removal of ores by filtration). In the low-phosphate (LP) medium, it was necessary to add to each flask 70 µL of 5 M H_2SO_4 at inoculation and every 24 h for three more days.

Table 5 - Leaching rates for Cu and Zn in columns of Experiment I

	Columns			
	1	2	3	4
	Ore alone	Ore + sand	Ore + slag	Ore + tailings
Ore (kg) in column	4	1.5	2	1.5
Cu (g) in column	196	74	98	74
Zn (g) in column	500	188	250	188
Cu ppm/47 d	2300	1000	2780	0
Zn ppm/47 d	8900	7880	9640	0
Cu% extraction/47 d	0.82	0.95	2.00	0.00
(30 d)	(0.52)	(0.61)	(1.27)	
Zn% extraction/47 d	1.25	2.93	2.70	0.00
(30 d)	(0.80)	(1.87)	(1.72)	

Column-leaching experiments were carried out as described in Figure 5 with iron-grown SM-4 for 47 days in LP medium. The total liquid for Experiment 1 was 700 mL.

Table 6 - Leaching rates for Cu and Zn in Columns of Experiment II

	Columns			
	1	2	3 (Low Cu)	4
	Ore alone	Ore + sand	Ore + sand	Ore + tailings
Ore (kg) in column	4	1.5	1.5	1.5
Cu (g) in column	194	73	12.2	74
Zn (g) in column	494	182	120	188
Cu ppm/30 d	2190	1890	264	0
Zn ppm/30 d	7080	5180	3310	68
Cu% extraction/30 d	0.56	1.30	1.09	0.00
Zn% extraction/30 d	0.72	1.42	1.38	0.05

Column-leaching experiments were carried out as described in Figures 5 and 7b. All columns except column 3 had been subjected to leaching with iron-grown SM-4 for 47 days (Experiment I) prior to the replacement of the medium and reinoculation with sulphur-grown SM-6 culture to initiate Experiment II. Column 3 (low Cu ore plus sand) was newly set up and inoculated with sulphur-grown SM-6 followed by iron-grown SM-5 cultures. The experiment was continued for 30 days in LP medium and the total liquid was 500 mL.

Table 7 - Leaching rates for Cu and Zn in columns of Experiment III

	Columns			
	1	2	3 (Low Cu)	4
	Ore alone	Ore + sand	Ore + sand	Ore + tailings
Ore (kg) in column	4	1.5	1.5	1.5
Cu (g) in column	193	72	12.1	74
Zn (g) in column	490	179	118	188
Cu ppm/28 d	1880	1440	304	0
Zn ppm/28 d	6260	5160	5320	564
Cu% extraction/28 d	0.49	1.00	1.26	0.00
(30 d)	(0.52)	(1.07)	(1.35)	
Zn% extraction/28 d	0.64	1.44	2.26	0.15
(30 d)	(0.68)	(1.54)	(2.42)	

Column-leaching experiments were carried out as described in Figures 5 and 7b. The columns had been subjected to leaching in Experiment II with inocula of both iron-grown and sulphur-grown cultures in LP medium, prior to the replacement of medium with LP medium containing 0.6 mM K_2HPO_4 (high phosphate) to initiate Experiment III. Although the experiment was continued for 30 days, the initial analysis of metals was done after 2 days. The total liquid was 500 mL.

FIGURES

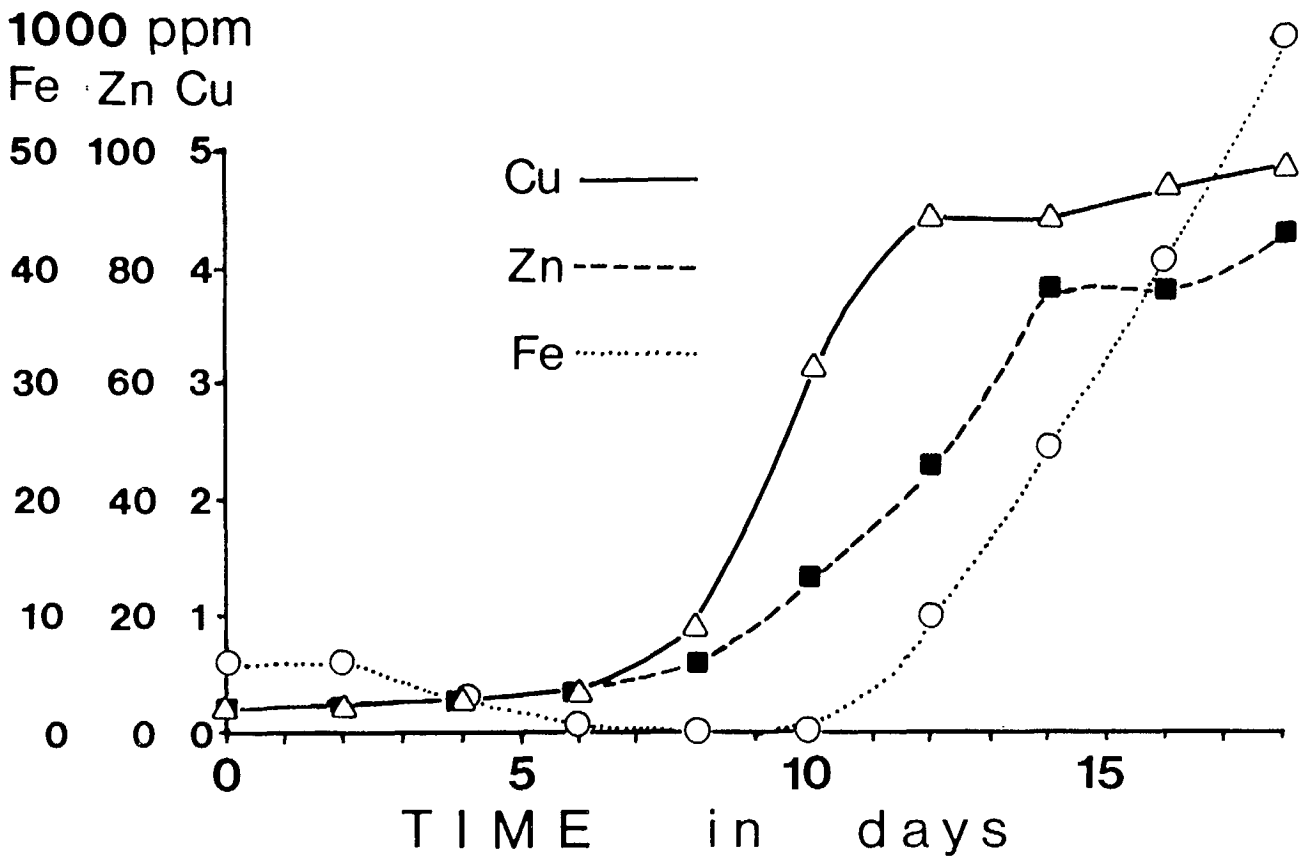

Fig. 1 - Time course of shake flask leaching by iron-grown SM-4 in the high-phosphate (HP) medium. Conditions are described in Table 1

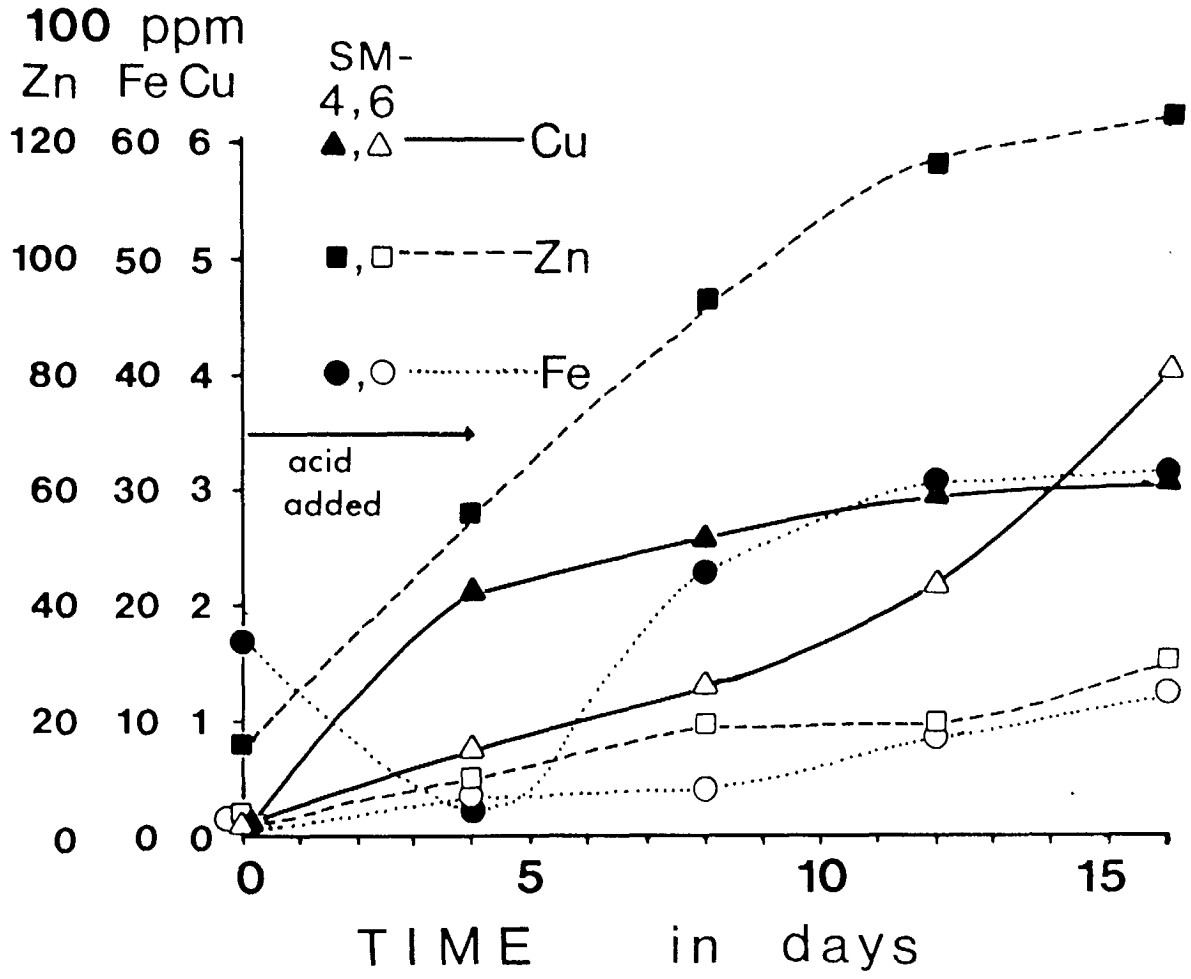

Fig. 2 - Time course of shake-flask leaching by ore-adapted SM-4 and SM-6 in the low-phosphate (LP) medium. Conditions are described in Table 3

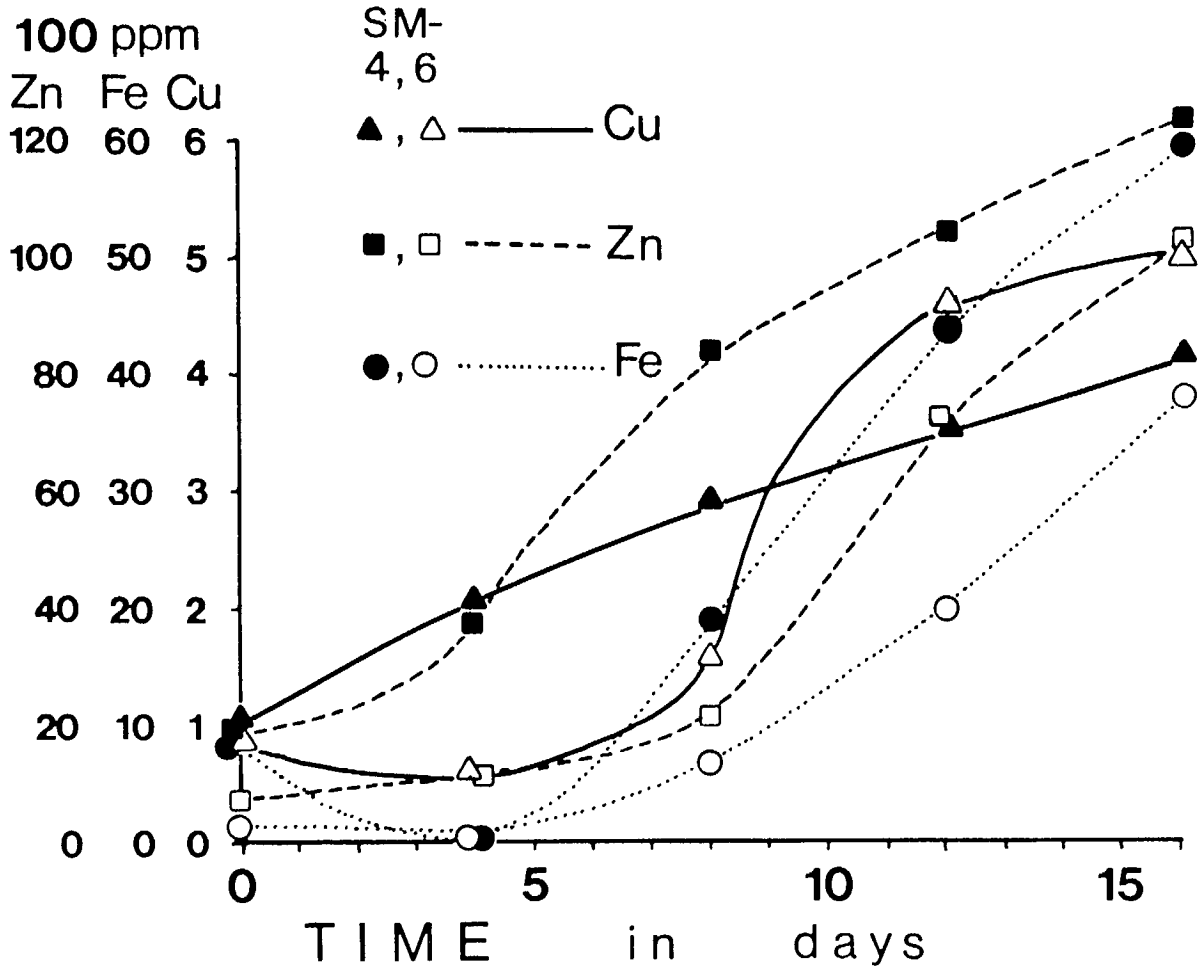

Fig. 3 - Time course of shake-flask leaching by ore-adapted SM-4 and SM-6 in the high-phosphate medium. Conditions are described in Table 3

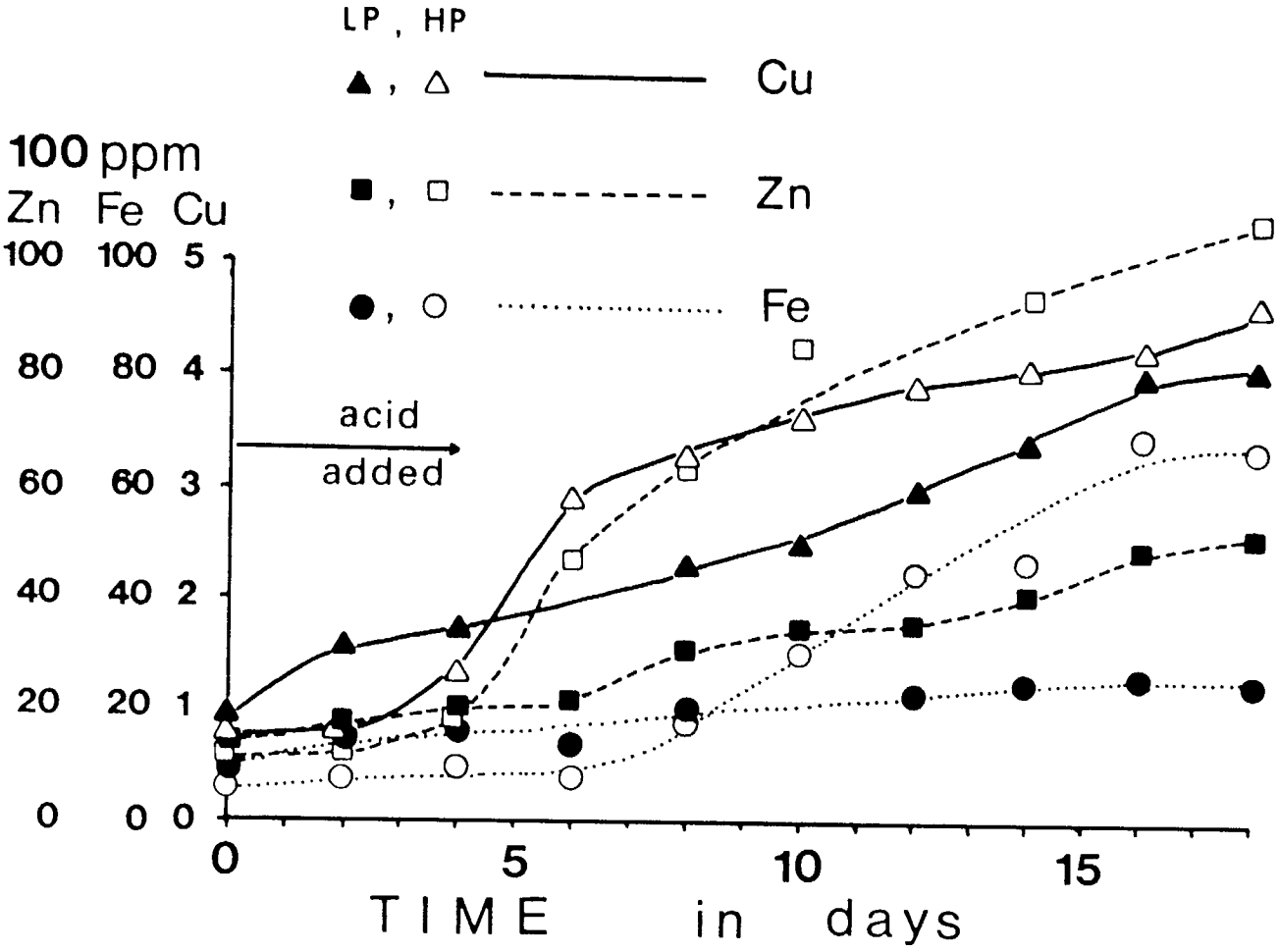

Fig. 4 - Time coarse of shake-flask leaching by a mixed culture of SM-4 and SM-6 in LP or HP medium. Conditions are described in Table 4

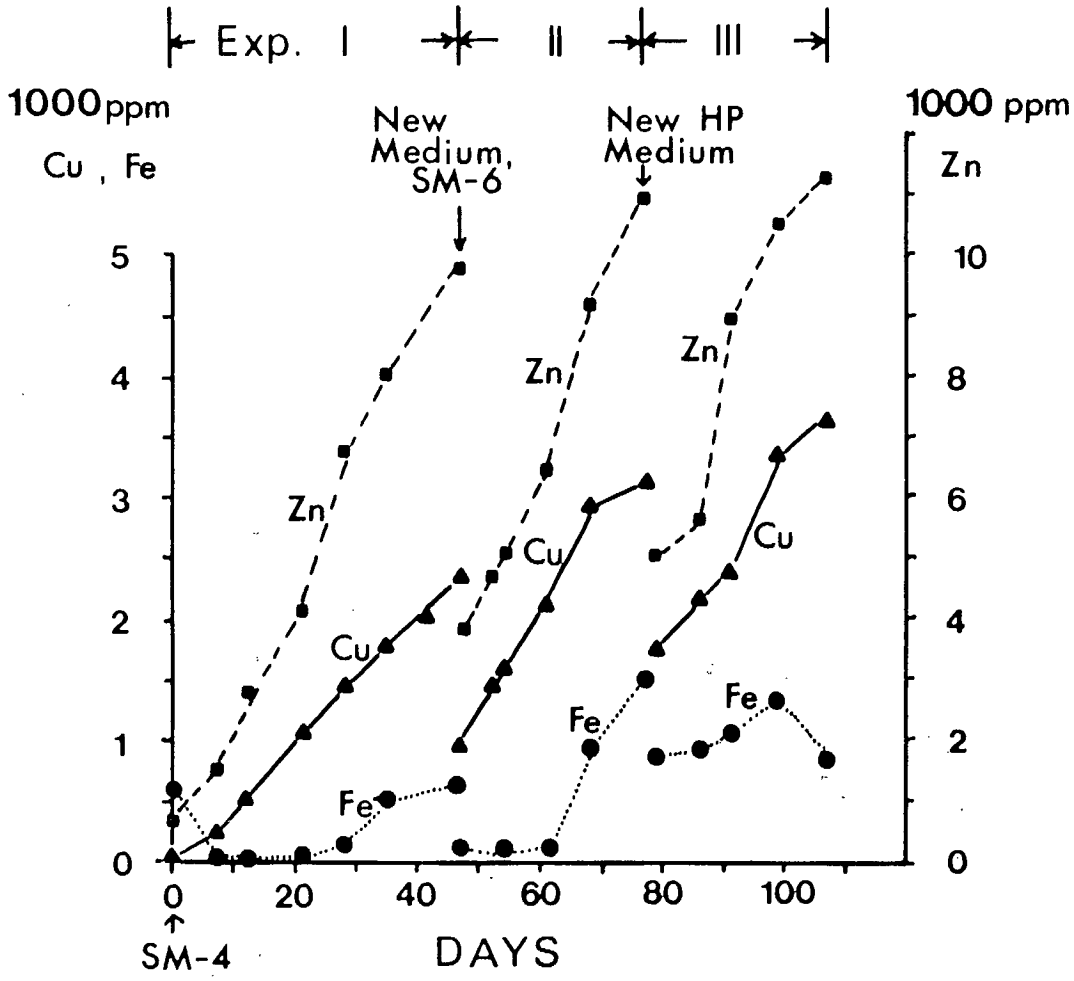

Fig. 5 - Time course of ore column leaching. Ore (4 kg) was packed in a column as described in Materials and Methods and was percolated (2 mL/min) with a medium (500 mL LP medium) inoculated with iron-grown SM-4 culture from a culture flask at 25°C (Experiment I). After 47 days, the medium in the flask was replaced with 500 mL of fresh LP medium and reinoculated with 50 mL of SM-6 culture (sulphur-grown from an ore-adapted inoculum). Leaching was continued for another 30 days (Experiment II). In Experiment III, the medium was again replaced with 500 mL of LP medium containing 0.6 mM K_2HPO_4 (high phosphate) and leaching was continued. In all the experiments, the pH was maintained at 2.3 by daily addition of 5 M H_2SO_4 to the culture flask

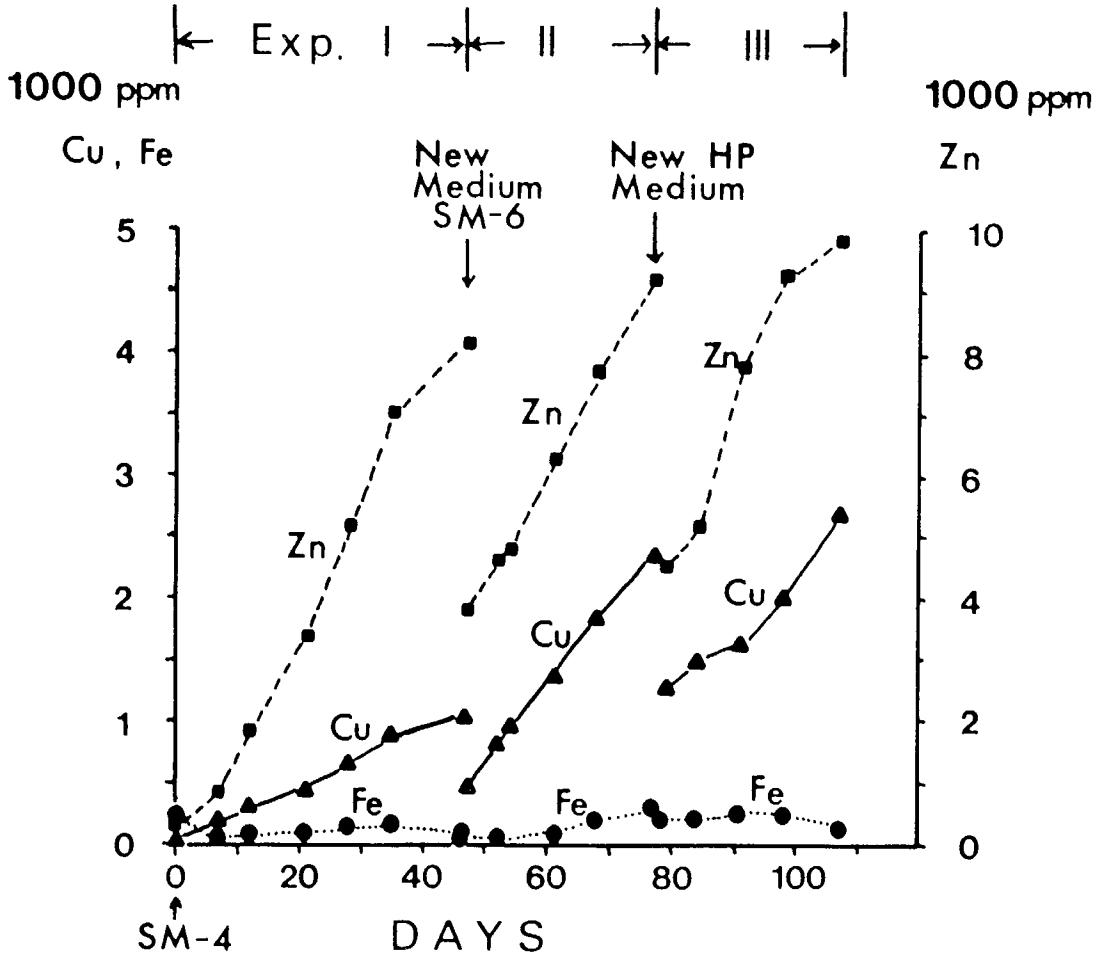

Fig. 6 - Time course of ore-sand column leaching. Ore (1.5 kg) plus sand (1.5 kg) were packed in a column, and the experiments were carried out as described in the legend for Figure 5

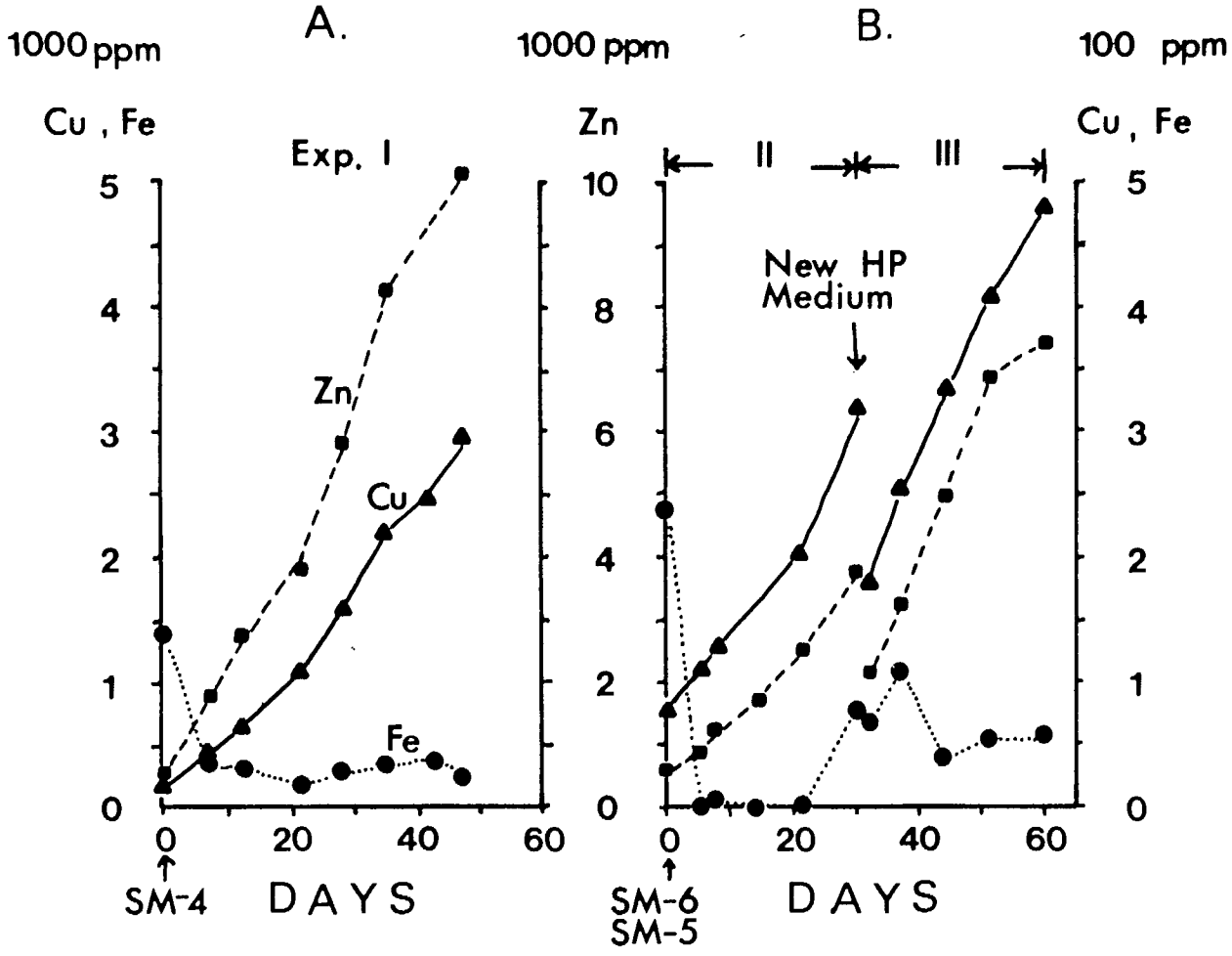

Fig. 7 - Time course of ore-slag column leaching (a) and low Cu ore-sand column leaching with a mixed culture (b).

(a) Ore (2 kg) plus slag (2 kg) were packed in a column. Experimental conditions were the same as in Experiment I of Figure 5

(b) Low Cu ore (1.5 kg) plus sand (1.5 kg) were packed in a column. The leaching experiments were similar to Figure 5 with 500 mL of LP medium, except that 50 mL of SM-6 sulphur-grown culture was added as the initial inoculum 2 days before the second inoculation with 50 mL of SM-5 culture (iron-grown) to start the experiment. After 30 days (Experiment II), the medium was replaced with 500 mL of LP medium containing 0.6 mM K_2HPO_4 (high phosphate) and leaching was continued for another 30 days (Experiment III)

SESSION II: PAPER 9

MICROBIAL DEGRADATION OF CARBONACEOUS MATERIAL IN A COPPER MOLYBDENUM CONCENTRATE

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ABSTRACT

Extensive carbonaceous mineralization has been encountered by a major copper molybdenum producer in Canada. The mining operation currently experiences this mineralization in various zones of the ore body. The carbonaceous mineral has been identified primarily as gilsonite. The mineral resembles a petroleum or bitumen-like substance rather than a coal-like material. Gilsonite and molybdenum respond the same metallurgically during flotation, gilsonite thus contaminating the molybdenum concentrate.

Past conventional metallurgical work has been unsuccessful in developing a flowsheet to economically separate the hydrocarbon from the molybdenum by mineral-processing methods. Roasting has been investigated but is not economical. Biotechnology has thus been examined as a possible alternative. This paper describes the research strategy employed in developing a suitable microbiological process to degrade the carbonaceous material in the concentrate so that a gravity separation technique will be more efficient.

A mixed population of bacteria, containing predominantly the organism *Pseudomonas*, has been found to actively degrade the asphaltene fraction of the carbonaceous material. Studies completed to date include acclimation of the bacterial strains to the ore concentrate samples, optimization of hydrocarbon removal, and pilot cyclone testing to separate the degraded hydrocarbon from the molybdenum concentrate. Semi-continuous leach degradation testing has indicated that the solid carbonaceous material remained suspended due to the production of a surfactant by the bacteria.

Bacterial and metallurgical data are presented and discussed from the bench and pilot-scale test programs.

SESSION II: PRÉSENTATION 9

DÉCOMPOSITION MICROBIENNE DES MATÉRIAUX CHARBONNEUX D'UN CONCENTRÉ DE CUIVRE MOLYBDÈNE

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RÉSUMÉ

Une vaste minéralisation charbonneuse a été découverte par un important producteur canadien de cuivre molybdène. Cette minéralisation se trouve actuellement dans différentes zones du corps minéralisé exploité. Le matériau charbonneux a été identifié comme étant principalement de la gilsonite. Le matériau ressemble plutôt à une substance pétrolifère ou bitumineuse qu'à un matériau charbonneux. Au cours du traitement métallurgique, la gilsonite et le molybdène ont répondu de la même manière à la flottation; par conséquent, le concentré de molybdène a été contaminé par la gilsonite.

Au cours des travaux métallurgiques classiques, on n'a pas réussi à mettre au point un schéma de fonctionnement permettant de séparer les hydrocarbures du molybdène de façon rentable par des procédés de traitement des minéraux. On a étudié, entre autres, le procédé de grillage mais ce dernier ne s'est pas avéré rentable. Par conséquent, la biotechnologie a été examinée en tant qu'alternative. La présente communication décrit la stratégie de recherche utilisée pour mettre au point un procédé microbiologique approprié à la décomposition des matériaux charbonneux contenus dans un concentré, afin qu'une méthode de séparation par gravité soit plus efficace.

On a trouvé une population mixte de bactéries, contenant surtout le genre bactérien Pseudomonas, capable de décomposer activement la fraction d'asphaltène du matériau charbonneux. Les études réalisées à ce jour, comprennent l'acclimatation des souches bactériennes aux échantillons de concentré de minerais, l'optimisation du procédé d'extraction des hydrocarbures, et l'essai cyclone pilote visant à séparer les hydrocarbures décomposés du concentré de molydène. Les résultats d'essais de décomposition par lixiviation discontinue ont démontré que la production d'un agent tensioactif par la bactérie a eu pour effet de mettre en suspension le matériau charbonneux solide.

On présentera et examinera, de plus, les données bactériennes et métallurgiques obtenues à partir des résultats des programmes d'essais à l'échelle de laboratoire et pilote.

MICROBIAL DEGRADATION OF CARBONACEOUS MATERIAL IN A COPPER MOLYBDENUM CONCENTRATE

PROJECT BACKGROUND

Hydrocarbon mineralization, gilsonite, which contaminates approximately 10% of a copper molybdenum deposit on Vancouver Island, British Columbia, has caused numerous problems since plant start-up in 1971. This hydrocarbon adversely affects the bulk and molybdenum flotation circuits, decreasing the grade of the final molybdenum concentrate. An example of the grades of the bulk and final molybdenum concentrate is shown below:

	Concentrate grades			
	% Mo	% C	% Cu	% Fe
Bulk concentrate	2.55	2.69	24.15	7.81
Molybdenum final concentrate	19.22	24.32	1.13	3.93

There have been extensive metallurgical studies to alleviate the hydrocarbon problem, but with little success to date. No separation method has been found to separate the hydrocarbon from the concentrate. Gravity separation by the use of one-inch diameter cyclones showed some promise. Cycloning the hydrocarbon from the molybdenum concentrate proved practical at low contamination levels. For high levels of hydrocarbon contamination, the cyclone process could not meet grade or recovery targets. Consequently, cycloning was not implemented on a routine basis.

Conventional metallurgical methods of treating the ore to separate the hydrocarbon have been exhausted so other methods were sought to address the problem.

The use of microorganisms to treat the hydrocarbon-rich ore has been evaluated as an adjunct to the present flowsheet. The "sticky" asphalt-like hydrocarbon in the copper molybdenum concentrate may cause fine concentrate particles to aggregate, thus preventing the separation of hydrocarbon from molybdenite. The hydrophobic nature of the hydrocarbon results in a naturally floatable ore fraction. Figure 1 illustrates an association of molybdenite and gilsonite from the molybdenum final concentrate. This observation suggests that mineral separation problems may be due to non-selective "sticking" or coating of the hydrocarbon on the mineral surfaces. Organic solvents, such as benzene, have been used to determine the relative abundances of extractable and non-extractable hydrocarbon. The relative abundances are shown below for both the bulk copper-molybdenum concentrate and the final molybdenum concentrate.

	Benzene Extraction of Hydrocarbon		
	% C Before extraction	% C After extraction	% C Organic extractable
Bulk copper-molybdenum conc	2.78	2.37	14.75
Final molybdenum conc	18.09	15.08	16.64

The benzene extracted material was found to occur in two forms:

1. An "asphalt-like" substance, very viscous and sticky, and completely insoluble in water.

2. A solid extractable hydrocarbon that could be precipitated from the benzene-extracted asphalt fraction. This fraction was attained by redissolving the extract in heptane, and filtering it off through a 0.45 millipore filter.

By using microorganisms to treat the hydrocarbon-rich concentrate, it was possible to degrade the "sticky" asphalt-like hydrocarbons present in the concentrate. This rendered the bulk of the hydrocarbon contained in the concentrate less viscous and sticky. It was anticipated that conventional gravity separation methods (i.e., cycloning, tabling, or heavy liquid separation) could subsequently be used to enhance the separation of hydrocarbon from molybdenite, producing a higher grade of molybdenite concentrate.

The main objectives of this research programme were to:

- isolate microorganisms having the ability to alter the hydrocarbon in the mine's concentrates;

- optimize bacterial growth parameters for maximum degradation of hydrocarbon;

- try conventional metallurgical methods to separate the remaining refractory carbonaceous material from the molybdenite.

MICROBIOLOGICAL BACKGROUND

Hydrocarbon-degrading microorganisms encompass many types of microorganisms including algae, fungi, yeasts, bacteria, and protozoa. However, there are a limited number of genera. Each of the individual organisms has unique specificities for hydrocarbon degradation. An organism may degrade only methane, or C_3-C_5 hydrocarbons, while another microorganism may degrade only high molecular weight paraffinic substances. In addition, the individual physiology of the particular organism will determine how the hydrocarbon is degraded. It may be oxidized to an alcohol, ketone, or carboxylic acid with no carbon chain degradation. Further degradation may take place to oxidize these substances to carbon dioxide and water by other strains. For this study, the petroleum hydrocarbons can be classified into the following fractions:

- saturated hydrocarbons (alkanes, branched alkanes, cyclic alkanes, and polycyclic alkanes);

- aromatic hydrocarbons (benzene, toluene, naphthalene);

- polar hydrocarbons (alcohols, ketones, aldehydes);

- asphaltenes (high molecular weight hydrocarbons, insoluble in most organic solvents) (1).

The above represents four individual substrate groups that are degraded in similar ways by individual organisms. The ability of a microorganism to degrade hydrocarbons can thus be specifically determined by analyzing the degradation products.

Bacteria are the most studied group of microorganisms capable of degrading and/or oxidizing hydrocarbons. Of the hydrocarbon-oxidizing bacteria, the genus *Pseudomonas* is the most predominant (2-4). Other important bacteria include *Corynebacterium*, *Mycobacterium*, *Rhodococcus*, *Micrococcus*, *Acinetobacter*, and *Nocardia*. All are facultative hydrocarbon degraders requiring oxygen to metabolize the hydrocarbon.

All organisms need nutrients to survive. If the growth medium for the organisms is less than optional, slow or no growth will result. The elements required are C, H, N, O, P, and S. Carbon and hydrogen come from the hydrocarbon material. Nitrogen is required in the form of ammonia (NH_3) or nitrate (NO_3^-), phosphorous as phosphate (PO_4^{3-}), and sulphur as sulphate (SO_4^{2-}). Ions and trace elements are also required.

Oxygen is essential for the metabolism of hydrocarbon-oxidizing bacteria, since the oxidation process utilizes O_2 from air rather than from water (5,6). Iron-containing monooxygenases are the enzymes used to catalyze the hydrocarbon oxidation reactions. Dissolved oxygen concentration directly affects hydrocarbon-oxidizing capacity. Thus, for an effective hydrocarbon-oxidizing system to be established, aeration of the medium is essential.

It is also necessary to maintain pH values (i.e., 6.5-7.5) and temperatures in a suitable range (20-40°C).

Many studies of hydrocarbon-degrading microorganisms have dealt with a single microorganism degrading a single hydrocarbon. However, studies show that some compounds resistant to degradation by a single microorganism can be effectively degraded by a mixed population (7). Mixed cultures harbour many advantages over growth in pure cultures, namely:

- higher growth rates
- stability of cultures
- resistance to contamination
- nutrient cross-over (8).

The development of an effective hydrocarbon oxidation-degradation system utilizing microorganisms depends upon a wise choice of organisms. These microorganisms must attack the insoluble high molecular weight hydrocarbons entrained in the copper molybdenum concentrates.

BENCH-SCALE STUDIES

Hydrocarbon-degrading microorganisms were obtained from the following sources:

- the waters and soils of the mining operation adjacent to areas high in hydrocarbon content;

- activated sludge microorganisms from waste petroleum degradation plants at Esso, Sarnia and Petro Canada, Clarkson.

Microbial strains, from the above sources, were selected naturally by subculturing weekly in aerated 3-L flasks at 15% solids of each of the rougher and final ore concentrates. The nutrient formula is presented in Table 1. Parameters including pH, redox potential, bacterial numbers, and available phosphorous were monitored daily.

From each of the subcultured bacterial sources, 0.1-mL aliquots of 100-fold dilutions were plated on nutrient salts agar and allowed to dry. Solvent-extracted hydrocarbon from the concentrate was suspended in a minimal volume of methylene chloride, and sprayed on the plates until a uniform layer of hydrocarbon was deposited on the surface of the plate. After sufficient incubation (about 7 days) at 30°C, individual colonies were selected by observing them under a microscope at low power (100X). Colonies that showed large zones of clearing ("haloes") of hydrocarbon were selected. Pure cultures of these bacteria were then propagated on conventional media (plate count agar).

Bacterial hydrocarbon degradation studies were performed on benzene-extracted hydrocarbon from molybdenum final concentrate reprecipitated on silica sand to a level of 5% (w/w). The hydrocarbon was homogeneously coated on the sand particles by adding a minimal amount of benzene, then mixing and allowing the benzene to evaporate in a fume hood.

A 10-g sample of "tar-sand" was placed in a 250-mL flask and 100 mL of nutrient solution was added. Seven flasks were prepared. Various diluted mixed bacterial cultures in 1-mL aliquots were added to each flask. Seven cultures were screened for visible stripping of tar from sand. The flasks were placed on a reciprocal shaker for 27 days at 20°C at 120 strokes per minute. Bacterial colony-forming units/mL were enumerated daily on plate count agar by the standard plate assay using tenfold dilutions. The pH values were also monitored daily. Control flasks were also placed on the shaker.

Nutrient solutions in the shaking flask studies were replaced on a 48-h basis by allowing the solids to settle, and decanting the supernate. In some cases, the nutrient solutions contained residual suspended hydrocarbon.

The three most successful cultures were selected for bacterial identification and further studies. Bacterial populations commonly peaked at the third or fourth day of growth and gradually levelled to constant populations. The nutrient medium was highly buffered to maintain a neutral pH, although pH values stabilized at slightly acid levels. In the absence of buffer and bacteria, a control flask demonstrated that natural oxidation lowered the pH to 2.8 over 27 days. Figure 2 shows a photograph of an uninoculated unshaken tar sand sample in water. Free sand particles were not observed.

Figure 3 shows a photograph of an uninoculated tar sand sample suspended in mineral salts solution and shaken for 8 days. Some sand particles have been freed due to mechanical shaking, but the majority were in contact with the hydrocarbon material.

Figure 4 shows a photograph of a bacterially inoculated flask in mineral salts solution shaken for 8 days. A comparison of Figure 4 to Figure 3 shows a

definite enhancement of the separation of tar from the sand particles in the bacterially inoculated flask. However, some sand remained in contact with the tar. Similarly, Figure 5 displays a bacterially treated flask. The silica sand exhibited increased separation from the tar in the treated flask than in the control flask. When the solutions from the bacterially treated flasks were examined, stringy pieces of tar-like material were observed floating in the nutrient solution. This demonstrates the partial emulsification of the hydrocarbon. It was found to be less "sticky" than the originally extracted hydrocarbon.

Figure 6 shows an unshaken, uninoculated tar sand sample. Figure 7 shows an uninoculated, shaken, control sample. In both cases, few free sand particles can be seen. Conversely, the bacterially treated tar-sand sample shown in Figure 8 illustrates that the sand particles have been liberated from the hydrocarbon to a substantially greater extent than the control.

Following completion of the experiment, the hydrocarbon from each of the flasks was thoroughly washed with water and stripped from the sand by washing the sand with benzene. The benzene was removed by evaporation and the residual hydrocarbon weighed. Table 2 shows the percentage of hydrocarbon removed from the tar sand compared to the control sample. A greater amount of hydrocarbon was removed by bacterial treatment. There was some loss of hydrocarbon in the bacterially treated flasks due to the decantation procedure.

PILOT-SCALE STUDIES

A pilot-scale batch bacterial treatment was performed on 78 kg of blended molybdenum concentrate. A 10-L bacterial seed culture was prepared as inoculum for the pilot-scale test. The seed culture was prepared by growing the bacteria in 3-L batches on the concentrate at 8% solids in nutrient medium for 5 days, filtering the solids, and saving the remaining bacterial solution. This process was continued until 10 L of solution was prepared.

A pilot-scale reactor was constructed to treat the molybdenum concentrate (Fig. 9). Warm tap water was added to the reactor to a volume of 900 L. The 78 kg of concentrate was then slowly added to the reactor with the agitator running to ensure proper wetting. This resulted in a pulp density of 8% solids. The nutrient formula was then added. Sodium hydroxide was used to raise the pH to 7.5. Finally, 10 L of the seed bacterial solution was added to the reactor to start the treatment process. The bacterial population at the start of the treatment was 4.1×10^5 colony-forming units (CFU) per mL.

The reactor was aerated to provide oxygen to the system. It was also mechanically agitated to ensure adequate mixing of the reactor contents and to improve the dispersion of air bubbles. Molybdenite is naturally floatable and thus a novel means had to be devised to limit the froth build-up on top of the reactor. This was done by setting up a slurry pump to recirculate reactor solution through two spray jets directed at the reactor surface, thus killing the froth. Bacterial counts, pH, temperature, and redox potential readings were taken daily to follow the concentrate treatment process.

Following 10 days of treatment, the solids were allowed to settle, three-quarters of the bacterial solution was discarded, and the solution replaced with fresh nutrient. This was to allow the bacteria to remain in the logarithmic

phase of growth. Solution replacement was performed every 48 h until the treatment was terminated on day 15.

During the decantation procedure, it was noted that approximately 2% of the solids in the reactor did not settle after 24 h. Settling did occur in the uninoculated control flasks, and thus this suspension phenomenon was attributed to the formation of a bacterial extracellular polysaccharide or surfactant.

The data obtained from the pilot treatment are shown in Table 3. Bacterial counts in the reactor remained relatively constant throughout the treatment. Bacterial counts around 10^8 CFU/mL were common. The pH remained relatively constant, within the range 7.18-7.67. Temperature values in the reactor increased from 19.5°C to 29.3°C throughout the 8-day oxidation process. This was probably due to the heat generated by bacterial metabolism. Accordingly, the redox potential values rose from relatively low oxidizing environments (+28 mV) to higher oxidizing potentials (+180 mV) over the 7-day period.

A one-stage cyclone process was used to separate carbon from the bacterially treated molybdenum concentrate. Figure 10 illustrates the equipment layout.

The cycloning studies were carried out on slurry decanted from the treatment vessel. Slurries of pulp density of 3, 7.3, 9.3, and 12% solids were prepared for each cyclone run. Cyclone pressures were adjusted to 25, 30, 35, or 40 psi for each run by using the backflow valve. Timed runs for 20 or 30 s were performed on the prepared slurries at the different pump pressures. Cyclone underflow and overflow samples were assayed for car on, Mo, Cu, and Fe.

There were two distinct trends in the cyclone data:

1. Carbon rejections increased as slurry densities increased, and

2. Molybdenum recovery decreased with slurry density but increased with cyclone-operating pressure.

Figure 11 compares carbon rejections obtained from laboratory cyclone runs with those obtained from the plant cyclone trials. This indicates that carbon rejections were considerably higher with the bacterially treated concentrate. Molybdenum recoveries, however, were lower (Fig. 12).

SEMI-CONTINUOUS TREATMENT OF CONCENTRATE

In the batch bacterial treatment of the molybdenum concentrate, there was minimal loss of carbon due to bacterial treatment. This indicated that degradation results would be enhanced if the bacterial solution were decanted to remove biomass and suspended particulate carbon. Consequently, a semi-continuous concentrate treatment system was used to assess the removal of carbon by bacterial oxidation and subsequent surfactant production. Table 4 presents the test results.

Following 24 h of exposure, a surfactant was produced by the bacterial culture in the treatment flask. Assays of the suspended particles and the assay of the total reactor contents were determined. The results, shown in Table 5,

indicate that a considerable amount of carbon was concentrated in the suspended fraction. In a control flask (no bacteria), no noticeable surfactant was produced. All of the solids in this control test settled readily.

DISCUSSION

The results of this study demonstrated that hydrocarbon degrading microorganisms have readily acclimated to a natural hydrocarbon contaminant present in the copper molybdenum concentrate.

The mixed population of microorganisms survived several stages of subculturing to remove undesirable microorganisms and inefficient hydrocarbon degraders. The bacteria were isolated from activated sludge from a waste petroleum processing plant. The mixed population, composed of 5 strains, degraded the asphalt and light oil fractions of the molybdenum concentrate very effectively in a slurry of up to 15% solids. The slurry was supplied with nitrogen, phosphorous, sulphur, magnesium, iron, and yeast extract to nourish the bacteria. The mixed population was composed of two *Pseudomonas*, one *Flavobacterium*, one *Mycobacterium*, and one *Arthrobacter*. It was not known whether these strains could survive in the absence of one another on the hydrocarbon substrate. The stability of this mixed culture indicates that a complex enzymatic system may have developed, and that synergistic and co-oxidational roles are present.

It was shown that about 15% of the hydrocarbon in the bulk and final concentrates was extractable with organic solvents. The benzene soluble fraction contains the aliphatic hydrocarbons, the aromatic hydrocarbons, and the asphaltene hydrocarbons. The remaining hydrocarbons were presumed to be insoluble solid hydrocarbons such as graphitic carbon and/or carbonate carbon.

The "sticky" benzene-soluble fraction is considered the cause of the hydrocarbon-molybdenum separation problem. It is believed that the majority of the bacterially degradable carbon originated from this fraction.

The results obtained from the tar-sand studies demonstrated that the bacteria effectively stripped the tar from the synthetic tar-sand. This stripping can be due to three possible mechanisms: (i) mechanical separation due to shaking; (ii) bacterial hydrocarbon degradation; and (iii) droplet dispersion by a bacterial surface-active agent.

The mechanical stripping of tar from tar-sand is considered minor relative to bacterial action. More than likely, bacterial hydrocarbon degradation leads to the breakdown of large hydrocarbon chains into smaller ones. This may lower the viscosity of the hydrocarbon material, rendering it easier to separate from the sand particles via surfactant or mechanical mechanisms. The production of surface-active agents by the mixed bacterial culture is believed to emulsify the hydrocarbon material, a very important factor in tar-sand separation. It is suggested that both mechanical shaking and bacterial surfactants are necessary in the separation process. Emulsification also enhances the rate of degradation by providing greater hydrocarbon surface area.

Bacterial surfactant production was most noticeable when the bacterial culture was propagated on the final molybdenum concentrate. It is likely that more than one surfactant was produced by the different bacterial species, since *Pseudomonas* and *Arthrobacter* are both known to produce surfactants.

The one-stage cyclone testing of the bacterially treated final molybdenum concentrate showed increased rates of carbon rejection compared to plant results on untreated material. However, molybdenum recoveries were not as high. With a two-stage cyclone process, molybdenum recoveries may be increased to acceptable levels.

During the semi-continuous bacterial treatment of the molybdenum concentrate, surfactant production was clearly evident. The carbon assays of suspended particles and total reactor contents were the key process-monitoring parameters. For future work, it is suggested that bacterial surfactant production be examined in conjunction with separating carbon from the concentrate by aqueous decantation.

CONCLUSIONS

Hydrocarbon-oxidizing microorganisms are readily obtained and can be adapted to degrade hydrocarbons from metallurgically produced concentrates. The hydrocarbon material is converted into biomass, with the secondary production of surfactants and organic acids. However, some of the hydrocarbon is refractory and difficult to degrade completely. Surfactant production by the microorganisms results in the suspension of particulate carbon. This phenomenon might be used on the basis of a physical separation process. Other operating mines with hydrocarbon contamination problems may be able to use similar concepts to remove hydrocarbon.

Based on these results, it is recommended that research and development be continued on the application of hydrocarbon-degrading bacteria in the metallurgical industry. It has been shown that bacterial hydrocarbon treatment can play a part in the removal of carbon from problem ores. The aqueous bacterial system can be readily interfaced with conventional mineral-processing flowsheets.

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TABLES

Table 1 - Bacterial nutrient formulation

	g/L	g/900 L
Na_2SO_4	2	1800
$MgSO_4$	0.2	180
KH_2PO_4	0.5	450
NH_4Cl	1	900
KNO_3	2	1800
$FeSO_4$	trace	0.4
Yeast extract	1	900

Yeast extract was solubilized in a minimal volume of water prior to addition in the reactor. pH was adjusted to 7.5 using NaOH.

Table 2 - Residual hydrocarbon determination from degraded and undegraded "tar sand" residues

Flask No.	Initial weight of hydrocarbon	Weight of hydrocarbon after (g)	Weight lost (%)
1. Inoculated	0.50	0.4468	10.64
2. Inoculated	0.50	0.3822	23.56
3. Inoculated	0.50	0.3533	29.34
4. Control	0.50	0.4763	4.74

Table 3 - Bacterial numbers (as colony-forming units/mL), pH, temperature, and Eh measurements from a pilot-scale bacterial treatment of 78 kg of blended molybdenum concentrate at 8% solids

Day	CFU/mL	pH	Temperature (°C)	Eh (mV)	Notes
0	4.1×10^5	7.49	19.5	+28	
1	6.9×10^7	7.40	22.0	+66	
2	8.2×10^7	7.54	24.0	+54	
3	8.9×10^7	7.62	25.0	+42	
6	1.0×10^8	7.53	25.0	+148	
7	6.2×10^7	7.67	26.5	+180	
8	5.2×10^7	7.43	29.5	+150	aeration off
9	1.5×10^8	7.18	29.5	+57	
10	5.4×10^8	7.48	29.5	+125	solution replacement
13	6.4×10^8	7.30	25.0	+95	solution replacement
14	6.2×10^7	7.18	25.5	+125	
15	3.5 x 108	7.21	24.5	+120	end of treatment

Table 4 - pH, bacterial counts (CFU/mL), carbon assays, and Eh measurements from a semi-continuous treatment of molybdenum concentrate. Nutrient solution was replaced every 24 h

Day	pH	Eh	CFU/mL	%C	Notes
0	7.1	-	3.0×10^6	22.7	
1	7.48	-63 mV	1.0×10^9	-	
2	7.26	-	6.1×10^8	-	
4	7.28	-54 mV	1.2×10^8	21.7	
5	7.46	-55 mV	2.3×10^8	17.2	
6	7.33	-27 mV	8.7×10^7	19.8	
7	7.22	+40 mV	-	18.3	Protozoa present
8	7.35	+45 mV	-	17.6	
11	7.41	+47 mV	8.2×10^8	17.6	
12	7.38	+43 mV	3.3×10^8	17.2	
13	7.53	+52 mV	-	17.0	
14	7.50	+47 mV	-	16.8	

Table 5 - Assays of carbon and molybdenum from particles remaining in the semi-continuous treatment flask after 1 h of settling and head assay when agitated

	%C	%Mo
Head assay	18.8	12.5
Assay of suspended particles	40.5	3.5

FIGURES

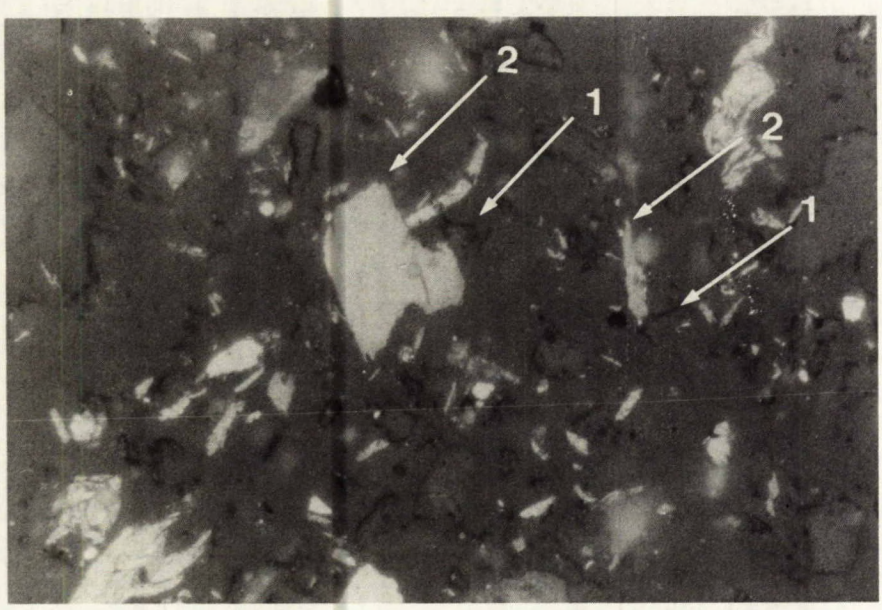

Fig. 1 - Mineral associations between gilsonite (1) and molybdenite (2)

Fig. 2 - Unshaken, undegraded tar-sand mixture (control) in water

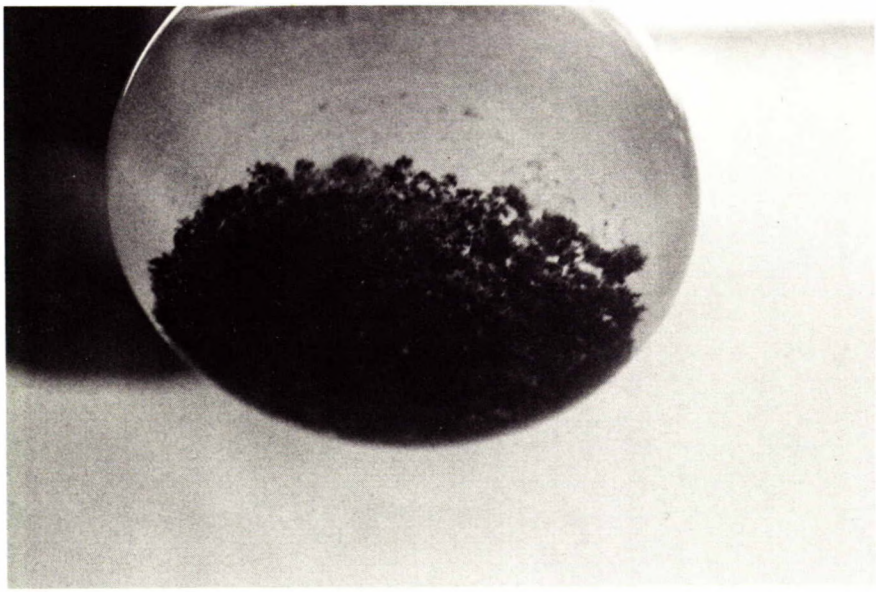

Fig. 3 - Uninoculated, shaken tar-sand mixture in mineral salts solution (control) - 8 days shaking

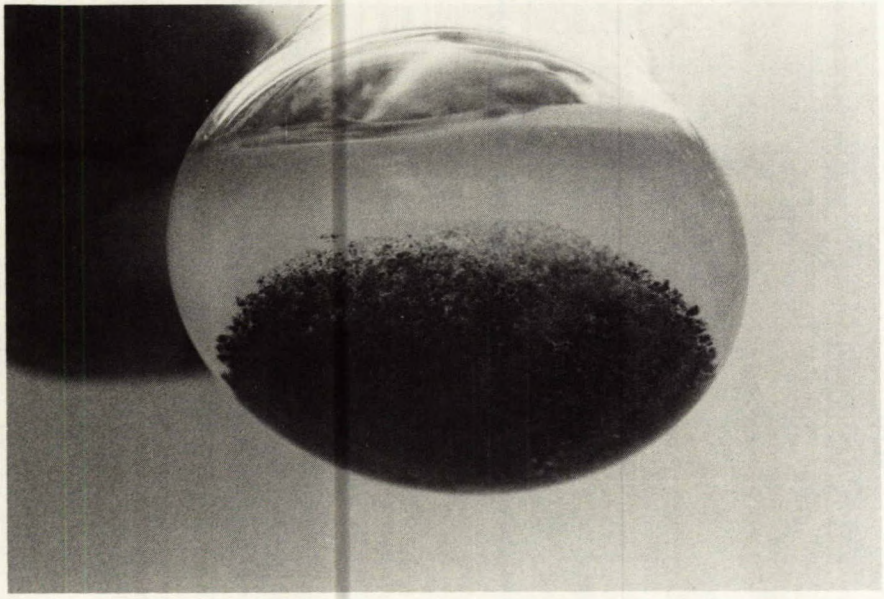

Fig. 4 - Inoculated tar-sand mixture in mineral salts solution. Flask No. 1 - shaken 8 days. Notice how mixture is "free flowing" and not agglomerated together

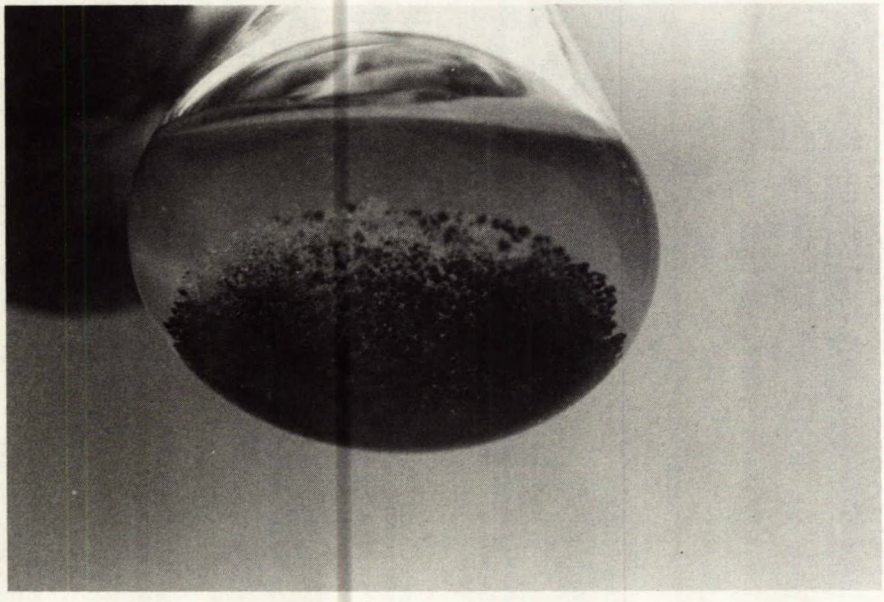

Fig. 5 - Inoculated tar-sand mixture in mineral salts solution. Flask No. 3 - shaken 8 days. Notice free grains of silica present

Fig. 6 - Uninoculated tar-sand sample. Flask No. 8 shaken 8 days. Still sticky and asphalt-like

Fig. 7 - Uninoculated tar-sand sample - no shaking

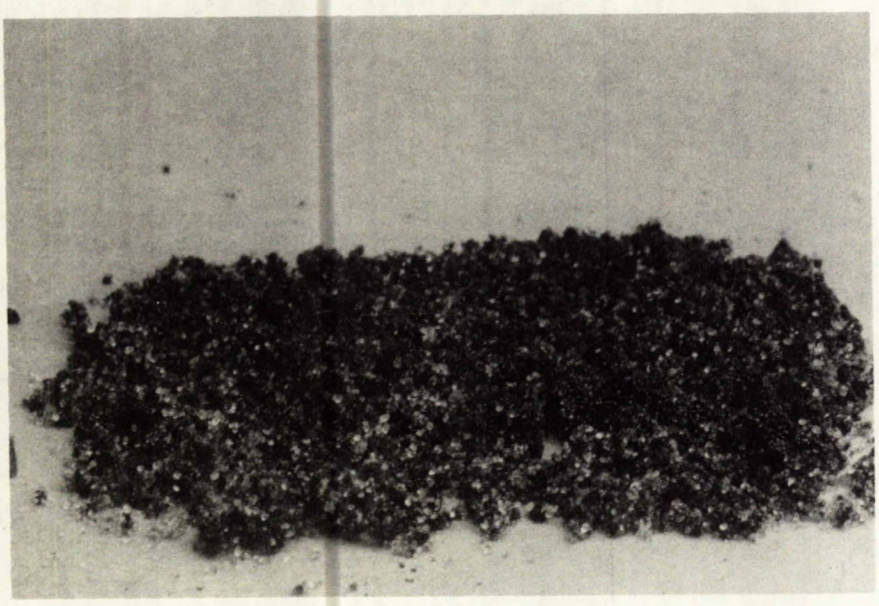

Fig. 8 - Inoculated tar-sand sample. Flask No. 1 - shaken 8 days. Notice grains of silica where all hydrocarbon has been degraded

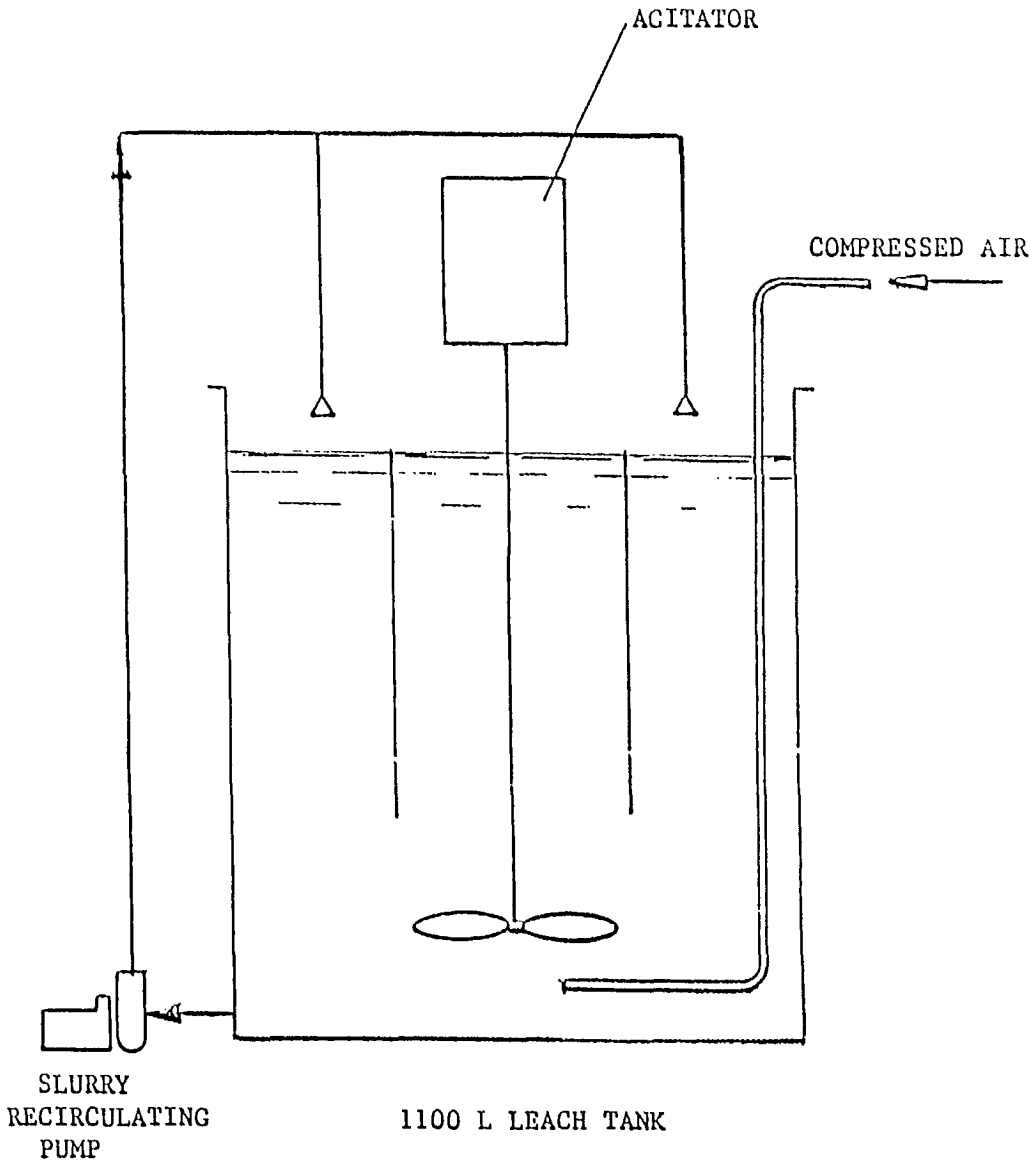

Fig. 9 - Reactor system employed for pilot-scale treatment of 78 kg of molybdenum ore concentrate

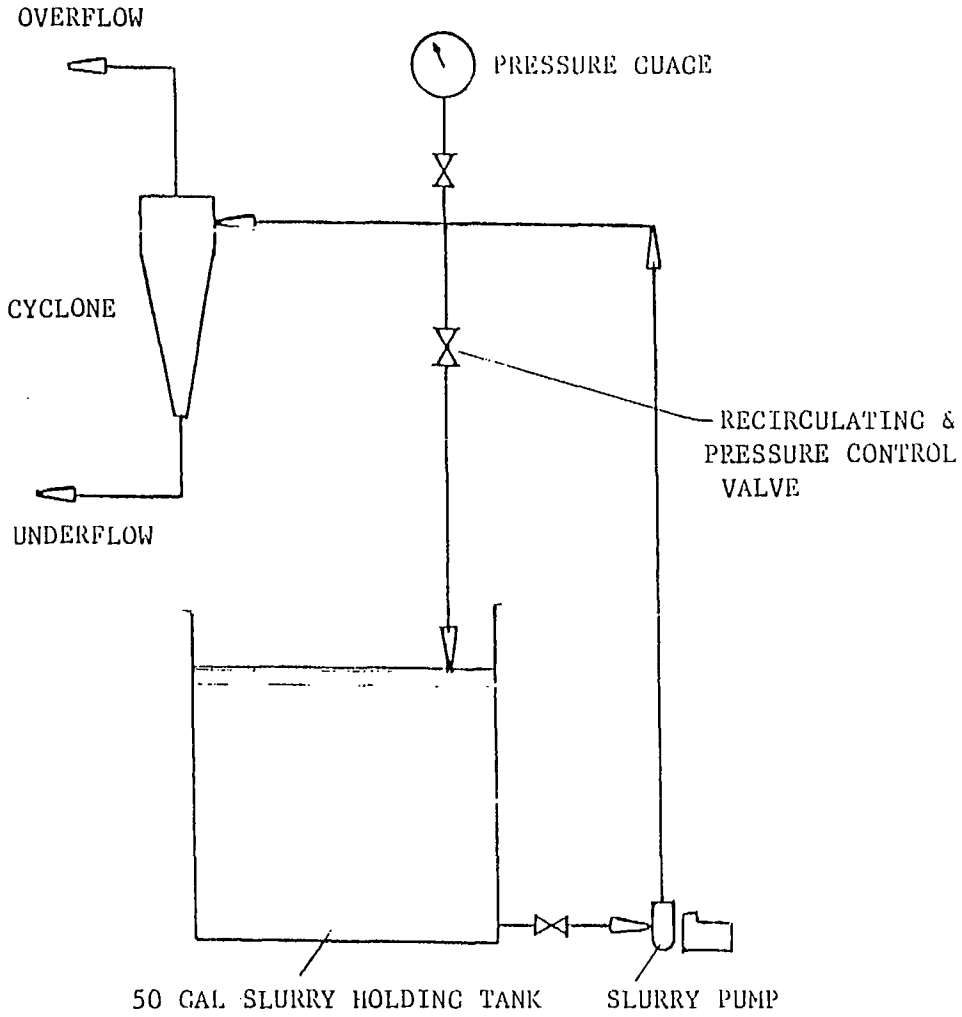

Fig. 10 - Single-stage cyclone setup for testing metal recoveries from a bacterially treated molybdenum ore concentrate

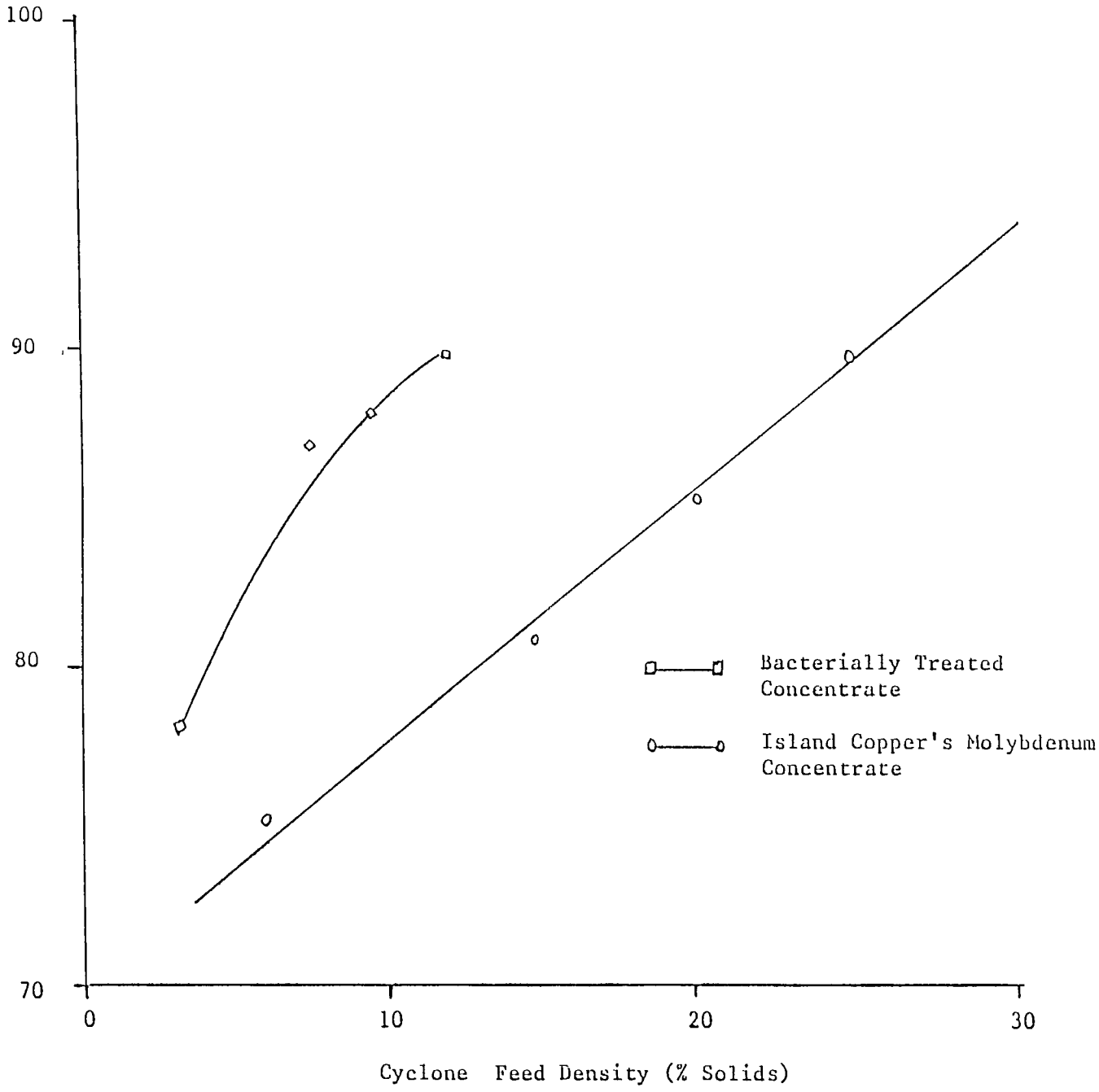

Fig. 11 - Carbon rejections obtained from cyclone runs from:
(i) a bacterially treated molybdenum ore concentrate and
(ii) Island Copper's cycloning data

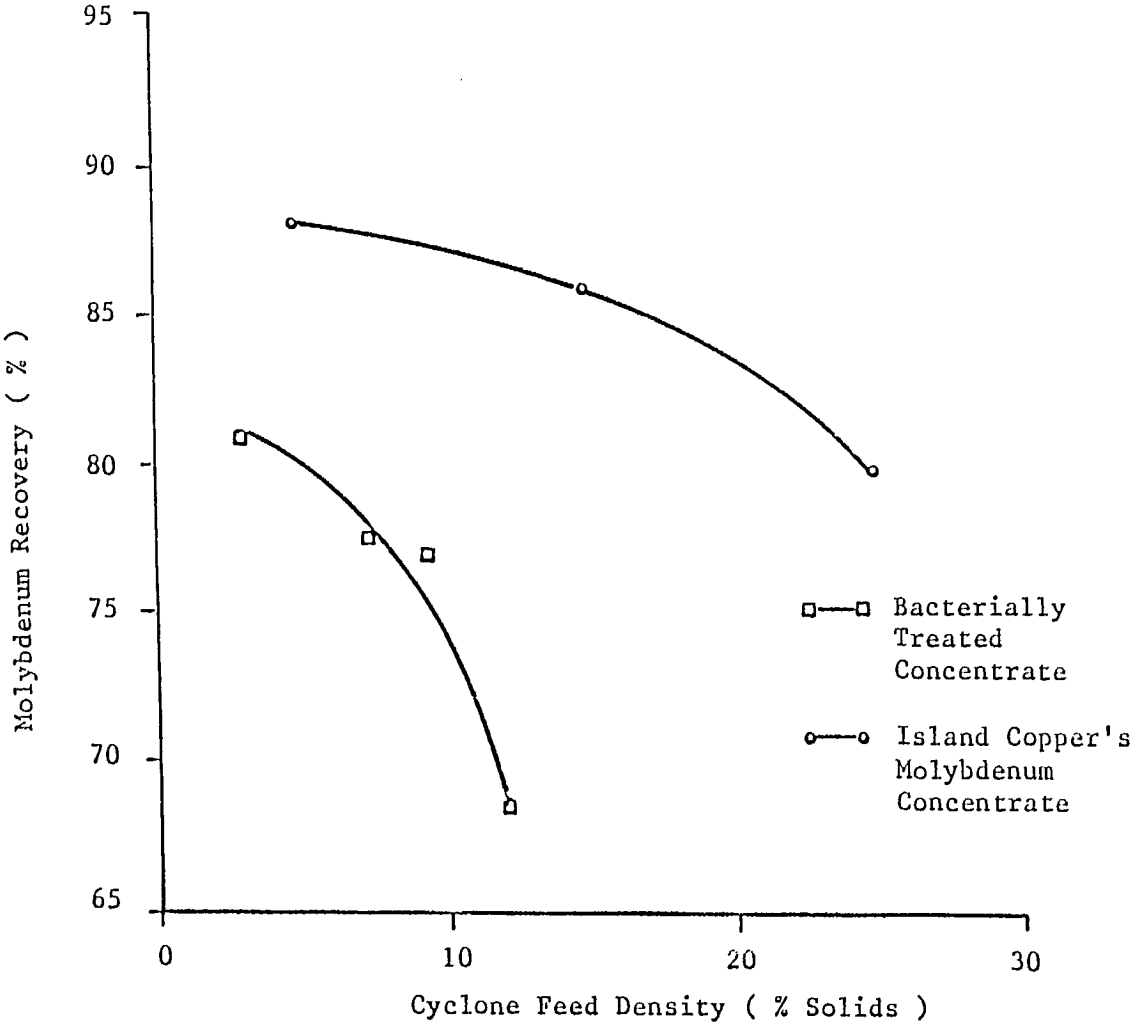

Fig. 12 - Molybdenum recoveries obtained from cyclone runs from:
(i) a bacterially treated molybdenum ore concentrate and
(ii) Island Copper's cycloning data

SESSION III

MINI-PRESENTATIONS

SESSION III: PAPER 10

RECOVERY OF GOLD BY BIOSORPTION

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ABSTRACT

The ability of different types of microbial biomass to remove gold from aqueous solutions has been examined. Extensive screening revealed a microbial biomass type that in its non-living state possesses a very high gold uptake capacity, comparable to that of activated carbon and the ion-exchange resin currently used in the gold production processes. Biosorption equilibrium uptake isotherms have been used for evaluation of the biosorptive uptake capacity of the biomass and served for comparing different types of sorbents. The extent of gold removal from aqueous solutions and the rate of its sequestering were assessed under different environmental (process) conditions such as initial concentration of the gold solution, pH, temperature, and the presence of selected anions and cations. Gold biosorption, as indicated by uptake isotherms, was independent of the initial gold solution concentration. Solution pH affected significantly the exhibited uptake, which was maximum at a pH lower than pH 3 and at room temperature. No discernible differences in uptake were observed between pH 2.5 and pH 1. Gold uptake by biosorption decreased with decreasing temperature.

SESSION III: PRÉSENTATION 10

RÉCUPÉRATION DE L'OR PAR BIOSORPTION

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RÉSUMÉ

On a examiné la capacité de différents types de biomasse microbienne à extraire l'or de solutions aqueuses. Après une sélection poussée, on a trouvé un type de biomasse microbienne qui, à l'état non vivant, a une capacité de sorber l'or aussi remarquable que celle du charbon activé et de la résine échangeuse d'ions qu'on utilise actuellement dans les procédés de production d'or. Les isothermes de biosorption à l'équilibre ont servi à évaluer la capacité de biosorption de la biomasse et à comparer différents types de sorbant. La quantité d'or extraite de solutions aqueuses et le taux de séquestration ont été évalués dans différentes conditions environnementales (du procédé) telles que la concentration initiale de la solution d'or, le pH, la température et la présence de certains anions et cations. La quantité d'or biosorbé, telle qu'indiquée par les isothermes de sorption, est indépendante de la concentration initiale de la solution d'or. Le pH de la solution a influé considérablement sur la biosorption observée qui a été maximale aux pH inférieurs à 3 et à la température ambiante. Aucune différence discernable de biosorption n'a été observée entre pH 2,5 et pH 1. La biosorption d'or diminue avec la température.

SESSION III: PAPER 11

BIOLOGICAL POLISHING OF ACIDIC SEEPAGE CREEKS

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ABSTRACT

Ecological engineering methods are being developed on a pyrrhotite-covered tailings area as part of an Inco/Reactive acid tailings (CANMET) research program started in 1985. This presentation summarizes the chemical, physical, and biological analysis of a seepage creek emerging from a tailings dam, with the aim of developing a self-sustaining seepage treatment system for close-out.

The objective in the first year was to describe in detail the chemical, physical, and biological characteristics of a seepage creek, as well as to attempt a synthesis of ongoing chemical and physical processes to develop concepts on long-term seepage treatments. Nine stations at 50-m intervals along the creek were monitored for chemical and physical data (July, August, and September), and the biology of the stream was described.

It was found that organic matter (living and dead) in the seepage creek appears to provide a catalytic surface for precipitation of iron hydroxide from the water. Steady iron removal occurs over the upper 250 m of the creek. The hydraulic retention time of the creek water increases as the water enters a broad expanse (=pond) of brome grass. A 6.5-fold reduction in total soluble iron occurs across this area (to a level of 0.2 ppm) along with a concomittant drop in pH of 2 full units. The latter acidification resulted in a re-solution of metals.

The results indicate that a self-maintaining treatment system for seepages has to consist of a two-step biological polishing system. At the head of the seepage, an alkaline or neutral ecosystem has to remove iron and this should be followed by an acidic ecosystem that will remove dissolved heavy metals. Experimental channels in which an acidic ecosystem can be established are being developed.

SESSION III: PRÉSENTATION 11

TRAITEMENT BIOLOGIQUE FINAL DES RUISSEAUX DE DRAINAGE ACIDES

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RÉSUMÉ

Dans le cadre d'un programme de recherche INCO/CANMET sur les résidus acides réactifs, entrepris en 1985, on met au point des méthodes de traitement écologique d'une zone de résidus recouverte de pyrrhotine. Cette présentation résume l'analyse chimique, physique et biologique d'un ruisseau de drainage s'écoulant d'un bassin à résidus, dans le but de mettre au point un système autonome de traitement des eaux de drainage destiné à être utilisé lorsque la mine sera fermée.

L'objectif au cours de la première année était de décrire en détail les caractéristiques chimiques, physiques et biologiques d'un ruisseau de drainage et d'essayer de faire la synthèse des processus chimiques et physiques qui s'y produisent, afin d'établir des principes de traitement à long terme des eaux de drainage. On a recueilli (juillet, août et septembre) des données sur des paramètres chimiques et physiques dans neuf stations espacées de 50 m le long du ruisseau, et on a décrit la biologie du ruisseau.

On a trouvé que la matière organique (vivante et morte) dans les eaux de drainage semble constituer une surface catalytique propice à la précipitation de l'hydroxyde de fer contenu dans l'eau. Le fer est systématiquement éliminé sur les 250 premiers mètres du ruisseau. Le temps de rétention hydraulique de l'eau du ruisseau augmente là où ce dernier débouche sur une large étendue (= étang) de brome. La concentration de fer soluble total est réduite par un facteur de 6,5, à 0,2 ppm, dans cette zone, tandis que le pH y subit une chute non concomitante de 2 unités entières. Cette acidification provoque la remise en solution des métaux.

Les résultats indiquent qu'un système autonome de traitement de finition des eaux de drainage doit comporter deux étapes de traitement biologique. Dans la partie amont des eaux, un écosystème alcalin ou neutre doit éliminer le fer, tandis qu'en aval, un écosystème acide doit éliminer les métaux lourds dissous. Des canaux expérimentaux dans lesquels on peut établir un écosystème acide sont en voie d'aménagement.

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