



## CHAPTER 19

### Thermophilic Microorganisms in Extraction of Metals from Ores

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To leach metals from porphyry copper samples in which chalcopyrite ( $\text{CuFeS}_2$ ) was the primary mineral, two thermophilic bacteria were used, *Sulfolobus acidocaldarius* and "ferrolobus." Ferrolobus has not been classified but is similar to *Sulfolobus acidocaldarius*. Both organisms are mixotrophic, thermophilic, and acidophilic, and oxidize sulfur and iron. Leaching was conducted in 250-ml Erlenmeyer flasks incubated at 60 C. In 60 days ferrolobus leached 51% of the copper from a -65 mesh chalcopyrite concentrate containing 29% copper; 8% of the copper was extracted from sterile controls. Leaching tests were conducted in 6 ft X 0.5 ft glass leach columns heated to 60 C. The percentages of copper, molybdenum, zinc, lead, and iron extracted from mineral samples inoculated with mixed cultures of *Sulfolobus acidocaldarius* and ferrolobus were compared with the extraction of these metals from samples maintained under identical conditions but sterilized with panacide. Mixed cultures of the thermophilic bacteria leached 38% of the copper from -3 + 100 mesh Duval Sierrita Corporation chalcopyrite ore (0.31% copper) in 161 days; 4% of the copper was extracted in the chemical control. Iron concn in solutions were variable over the leach period, with all iron present in the ferrous form. Molybdenum in solution increased with increasing iron concn. These studies indicate that thermophilic and acidophilic bacteria can extract copper from ores and mine wastes containing chalcopyrite.

#### INTRODUCTION

Bacterial leaching of metals from sulfide minerals has been studied extensively since the identification of *Thiobacillus ferrooxidans* (Colmer and Hinkle 1947). The primary role of *T. ferrooxidans* in extraction of metals has been established as the oxidation of ferrous iron in an acid environment. *T. thiooxidans*, which oxidizes reduced sulfur compounds, is also found in leaching environments. Several excellent reviews summarize the importance of the thiobacilli in extraction of metals (Corrans et al. 1972; Tuovinen and Kelly 1974). Chalcocite ( $\text{Cu}_2\text{S}$ ) is leached readily by the ferric iron and sulfuric acid generated by *T. ferrooxidans* and *T. thiooxidans*. However, chalcopyrite ( $\text{CuFeS}_2$ ) and molybdenite ( $\text{MoS}_2$ ) are very resistant to the action of these products at normal concn (Malouf 1972).

A microorganism that grows at high temperatures and low pH was isolated first from an acid thermal region of Yellowstone National Park (Brierley 1966). The organism, designated "ferrolobus," has not been classified but was characterized by Brierley and Brierley (1973). Ferrolobus is similar to *Sulfolobus acidocaldarius*, isolated and characterized by Brock et al. (1972). Ferrolobus and *S. acidocaldarius* oxidize reduced iron and sulfur compounds and grow on simple organic compounds and yeast extract. *S. acidocaldarius* has a temperature optimum of 70-75 C and a range of 55-80 C; the pH optimum is 2-3. The DNA base composition of *S. acidocaldarius* is 60-68% guanine plus cytosine. Ferrolobus has a temperature range of 45-70 C and a DNA base composition reported as  $57 \pm 3\%$  guanine plus cytosine. *Sulfolobus* and ferrolobus are spherical organisms with a diameter of 1  $\mu\text{m}$ ; both organisms lack a rigid cell envelope (Brock et al. 1972; Brierley and Brierley 1973). Characteristic lipids, presumably advantageous in stabilizing the bacterial membrane toward environmental stress,

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are found in the extremely thermophilic acidophiles (DeRosa et al. 1975). Studies of the ultrastructure of these organisms show each bounded by a plasma membrane and covered by a nonrigid extracellular coat (Millonig et al. 1975). In natural habitats (Brock and Darland 1970), hot springs at 75 C and pH 2-3, *Sulfolobus* was the predominant organism (Weiss 1973a). The occurrence of *Sulfolobus* in sulfur-rich, acid, hot springs and soils indicates they are important geochemical agents, contributing to the high acidity of these areas by oxidizing large amounts of elemental sulfur (Fliermans and Brock 1972; Mosser et al. 1973, 1974). Sulfur oxidation by *Sulfolobus* over the temperature range from 55 to 84 C is accompanied by the attachment of the organisms to the sulfur crystals (Shivvers and Brock 1973) by means of pili (Weiss 1973b). Weiss (1973b) reported that attachment is not a prerequisite for sulfur oxidation by the bacteria in culture, but attachment of cells to sulfur in flowing springs enables *Sulfolobus* to colonize these habitats. Direct observations, using the scanning electron microscope (SEM), revealed that ferrolobus formed microcolony structures on molybdenite ( $\text{MoS}_2$ ) (Brierley and Murr 1973; Brierley et al. 1973). Using the transmission electron microscope and a technique for microscopically examining the internal structure of a mineral section, Berry and Murr (1975) found no correlation between the disposition of attached *S. acidocaldarius* and ferrolobus and dislocations (structural defects) emerging on the surfaces of molybdenite platelets. The attachment of *S. acidocaldarius* and ferrolobus to molybdenite (Berry and Murr 1975) and chalcopyrite (Murr and Berry 1976) appeared adsorptive since no evidence of pili or other holdfasts were observed. Attachment of ferrolobus to pyritic and chalcopyritic phases of low-grade copper ores was observed using the SEM (Murr and Berry, unpubl.). Murr and Berry suggested that such observations provided strong support for the direct contact mechanism of bacterial oxidation and correlated bacterial attachment with release of copper and iron into solution. However, no quantitative data were provided, so the validity of their conclusions is contentious.

Rod-shaped, thermophilic, acidophilic organisms similar to thiobacilli have been reported (Schwartz and Schwartz 1965; Brierley 1966), and rod-shaped isolates are being studied as means of accelerating leaching reactions (N. W. LeRoux, pers. comm.). The activity of the spherical, thermophilic, acidophilic bacteria in natural leaching situations has not been thoroughly studied as they were only recently discovered. Preliminary studies show that over a 30-day period ferrolobus, supplemented with 0.02% yeast extract and 0.2% ferrous iron, extracted molybdenum from a molybdenite concentrate (58% Mo; 12-65  $\mu\text{m}$ ) 130 times faster than molybdenum dissolution from the chemical control. Ferrolobus tolerated 2,000 ppm hexavalent molybdenum (Brierley 1974), and molybdenum (VI) was reduced to molybdenum (V) when elemental sulfur was supplied to the organism in an aerobic or anaerobic nitrogen atmosphere. Under such conditions molybdenum (VI) may be serving as the terminal electron acceptor (Brierley and Brierley, unpubl. data), although this has not been demonstrated.

In the present study the thermophilic, acidophilic bacteria, *S. acidocaldarius* and ferrolobus, were used to leach metals from porphyry copper samples, with chalcopyrite as the primary mineral. Results obtained in the study and use of these organisms in extraction of metals from leach-resistant minerals are presented.

## MATERIALS AND METHODS

**Leach solution.** The leach solution for all column studies was 9K basal salts (Silverman and Lundgren 1959) diluted with distilled water rather than ferrous iron solution. All flask-leaching studies used Bryner and Anderson medium (1957); the pH was adjusted to 2.5 with sulfuric acid. When used as an energy source, elemental sulfur (flowers of sulfur) was sterilized

by intermittent steaming for 30 min on 3 consecutive days. A solution of 25 g  $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$  in 95 ml distilled water and 5 ml 1 N  $\text{H}_2\text{SO}_4$  was sterilized by autoclaving. When iron was used as an energy source, 4 ml of the sterilized solution were added to 100 ml of medium. Batch cultures and selected flask-leaching studies were supplemented with yeast extract (Difco) of 0.02% for *ferrolobus* and 0.2% for *S. acidocaldarius*. The yeast extract was sterilized with the medium.

*Preparation of cells.* Fernback flasks containing 1.5 liters of medium and 7.5 g sulfur were used for batch cultures of the organisms. The medium was supplemented with 0.02% yeast extract for *ferrolobus* and 0.2% yeast extract for *S. acidocaldarius*. The cells and sulfur were sedimented from solution at 27,000 g and resuspended in fresh medium. Cells were separated from sulfur by centrifuging the culture at 120 g for 10 min to sediment the sulfur. The number of cells was determined by most probable number (MPN) (Collins 1967).

*Assays.* Analyses for copper, total iron, zinc, lead, and nickel were completed on a Perkin-Elmer 303 atomic absorption spectrophotometer (Brandvold 1974). Ferrous and total iron were determined by dichromate titration (Blaedel and Meloche 1963) and molybdenum was analyzed by a modified thiocyanate colorimetric method (Meglen and Glaze 1973).

*Flask leaching.* A chalcopyrite concentrate (29% Cu) from the New Cornelia Branch, Phelps Dodge Corp., Ajo, AZ, was less than 65 mesh (212  $\mu\text{m}$ ). One gram of concentrate was added to 100 ml of medium in 250-ml Erlenmeyer flasks. Sterilization of leach solution and concentrate was by intermittent steaming for 30 min on 3 consecutive days. Experimental flasks were inoculated with 0.1 ml of a dense suspension of *ferrolobus*. Leach testing also included the addition of either 0.02% yeast extract or 0.2% ferrous iron or both. The flasks were incubated in stationary, covered water baths or water-saturated air incubators at  $60\text{ C} \pm 1\text{ C}$ . The pH and the copper concn of the leach solutions were monitored every 15 days for a 60-day period.

*Column leaching.* Two 6-ft columns were formed from insulated glass columns, 0.5 ft  $\times$  2.0 ft. These columns plus two insulated glass pipes, 0.5 ft  $\times$  6.0 ft, were heated to approx. 60 C with nichrome ribbon. Temperatures were regulated with Yellow Springs Instrument thermistor controllers (Brierley 1975). Leach solutions were pumped to the top of the columns in Tygon tubing and aerated with aspirators. Effluent solutions from the columns drained into reservoirs maintained at room temperature (Fig. 1).

Low-grade copper porphyry ores with chalcopyrite as the primary mineralization were obtained from Duval Sierrita Corp., AZ, and the Pinto Valley Mine, Cities Service Co., Miami Operations, AZ. The Sierrita ore was crushed to pass a 3 mesh (6.7 mm) screen and fines of -100 mesh (150  $\mu\text{m}$ ) were removed. The Pinto Valley material was crushed to 0.525 inches (13.3 mm) and fines of -48 mesh (300  $\mu\text{m}$ ) were removed. A size-analysis of each ore (heads) was made and each mesh size and split sample was analyzed for copper, molybdenum, zinc, and iron. Lead and nickel analyses were completed on the Sierrita ore. The ores were analyzed for opaque minerals by reflected light microscopy. Duplicate columns of each ore type were set up. The leach solution, 9K medium without iron, was dripped over the ore at the rate of 10 ml/min. One column of each set was inoculated with a mixed culture of *ferrolobus* and *S. acidocaldarius*; 50 ppm panacide (2, 2-methylenebis-4-chlorophenol), an antimicrobial agent, was initially added to each control column. Further additions of the sterilant were made during the leaching period.

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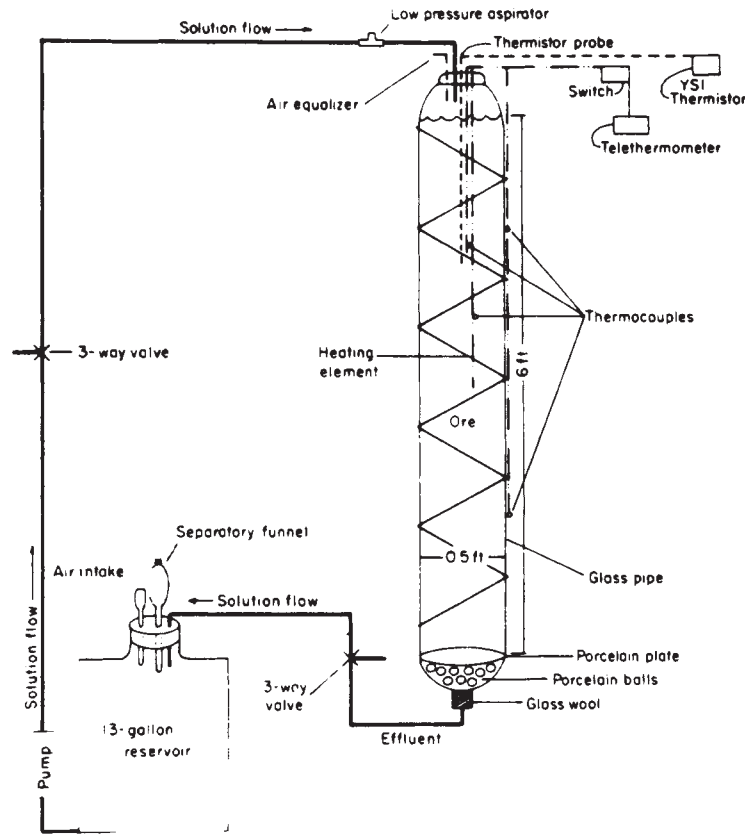


FIG. 1. Schematic view of column leaching system.

Samples were collected weekly and analyzed for pH, Eh, ferrous iron, total iron, copper, molybdenum, and zinc. The leach solution from the Sierrita ore also was analyzed for lead and nickel. Sulfuric acid was added to maintain a pH of 2-3. When a concn of approx. 2 g of copper per liter was reached in the leach solution, copper was precipitated by adding iron filings to the leach solution. At the conclusion of the leach tests, samples from the top, middle, and bottom sections of each column were collected and analyzed for metals remaining and were cultured for the presence of *ferrolobus*, *S. acidocaldarius*, and *T. ferrooxidans*. The bacterial activity of the mesophilic and thermophilic bacteria was determined as  $^{14}\text{CO}_2$  uptake in counts/min/g dry weight of ore (CPM/g) (Smith et al. 1972). Split samples of the leached ore (tailings) were assayed. The tailings were sized, and each mesh size was analyzed chemically. Chemical analyses of tailings more accurately quantified the percentage of metals extracted than did analysis of the leach solution.

## RESULTS

### *Flask Leaching*

The average values for 10 experiments of leaching condition under which Ajo concentrate was tested appear in Table 1. A study of this table yields the following conclusions: when leaching the concentrate with *ferrolobus* without addition of iron or yeast extract, the copper extrac-

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TABLE 1. Percent copper extracted from Ajo chalcopyrite concentrate in flask-leaching studies

Flask Contents	Percent Copper Extracted			
	30 Days		60 Days	
	Control	Inoculated	Control	Inoculated
9K + concentrate	5	8	8	51
9K + yeast extract + concentrate	4	29	4	82
9K + Fe(II) + concentrate	30	43	36	59
9K + yeast extract + Fe(II) + concentrate	19	40	43	90

tion was only 8% in 30 days, but greatly increased to 51% at 60 days. Copper extracted from controls was low at both samplings. Leaching was probably greatest during the last 30 days either because of the larger number of bacteria present or because of the increased concn of iron in solution released from the chalcopyrite. Yeast extract increased the percentage of copper extracted in the inoculated flasks. The yeast extract probably enhanced the growth of the organisms which could directly attack chalcopyrite or acted as a wetting agent which enhanced bacterial attachment and promoted a direct attack mechanism. Adding ferrous iron to the leach solution enhanced copper extraction. At 60 C, ferrous iron is oxidized rapidly to ferric iron which solubilized copper. However, the addition of ferrous iron to inoculated flasks of leach solution with chalcopyrite did not result in as great a copper extraction as when *Ferrolobus* was supplemented with yeast extract. *Ferrolobus* probably prefers ferrous iron to chalcopyrite as an energy source; the iron oxidized rapidly, but chalcopyrite was not attacked biologically, indicating that direct bacterial attack by *ferrolobus* results in greater copper extraction than the reaction of ferric iron with chalcopyrite. Providing *ferrolobus* with both ferrous iron and yeast extract yielded extraction data similar to that for iron supplementation only, at 30 days, but extraction was greatly enhanced at 60 days. Yeast extract and ferrous iron probably increased the number of bacterial cells resulting in increased iron oxidation. Undoubtedly, both the bacteria and the ferric iron yielded increased copper production.

Clearly from these data, *ferrolobus* enhanced the extraction of copper from chalcopyrite. Although the largest copper extraction occurred when yeast extract and iron were added to the culture, the relative importance of ferric iron and direct bacterial attack on chalcopyrite leaching cannot be determined. When a large bacterial population has developed and leaching activity is at a maximum, sufficient iron for leaching would probably be provided from the breakdown of chalcopyrite. The mechanism of direct bacterial attack on chalcopyrite has not been defined.

#### Column Leaching

The extraction rates of metals from two, low-grade, porphyry copper ores inoculated with *ferrolobus* and *S. acidocaldarius* and leached in heated columns were compared with the metal extraction rates of the low-grade ores sterilized with panacide and maintained under identical conditions.

*Duval Sierrita ore (columns A and B)*. The primary mineralization of mill feed from Duval Sierrita Corp. was chalcopyrite, with some digenite ( $\text{Cu}_9\text{S}_5$ ) and covellite ( $\text{CuS}$ ). A split

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sample of the ore assayed 0.31% Cu, 0.05% Mo, 0.02% Zn, 5.76% Fe, 0.01% Pb, and 0.01% Ni. To each column (A and B) 105 lb of -3 + 100 mesh (6.7 mm - 150  $\mu$ m) ore were added. Eighteen liters of 9K medium were recycled continuously over the ore. The pH was maintained at approx. 2.5 by addition to each column of 1050 ml concentrated sulfuric acid over the 160-day leach period. Column A was inoculated twice with a ferrolobus-*Sulfolobus* suspension for a total of  $6.8 \times 10^8$  bacteria; panacide was added initially to column B with further additions on days 15, 113, and 148. Culturing of column A reservoir solution showed increasing numbers of *T. ferrooxidans* in excess of 1800 per 100 ml on day 147 of the leach period. The reservoir was heated to about 70 C for several hours to reduce this number. A sample from column B reservoir on day 147 was positive for *T. ferrooxidans*. Fungal growth in the reservoir and Tygon tubing of the panacide control column was observed to be massive throughout the leach cycle.

The copper concn obtained during the leaching of Duval Sierrita ore are shown in Fig. 2. The 14-day lag at the beginning of the experiment may have resulted from the small initial inoculum of 540 organisms. A stationary phase in copper extraction was noted in both columns between days 90 and 112. A shutdown of heat and solution flow may have affected

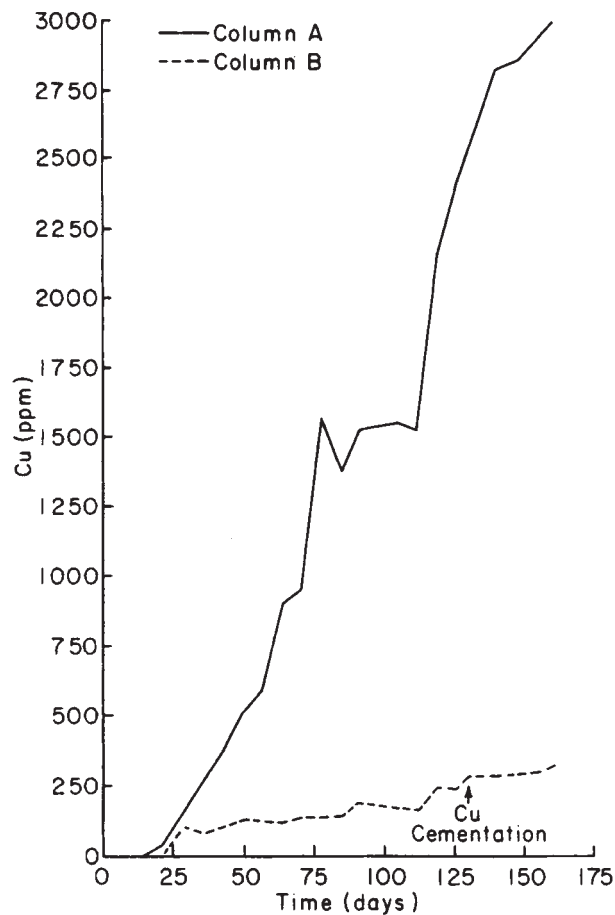


FIG. 2. Copper concn in leach solutions of columns A (inoculated) and B (control).

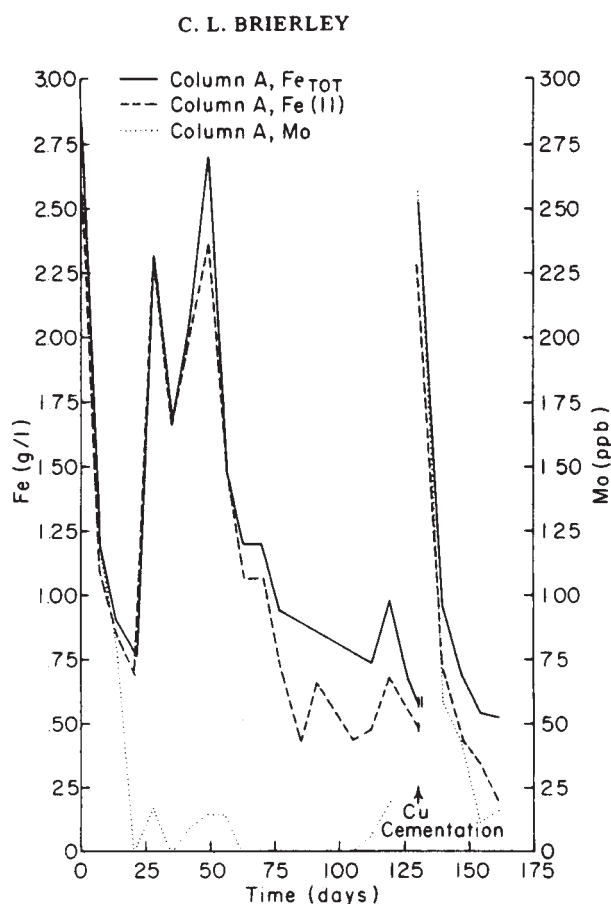


FIG. 3. Ferrous iron, total iron, and molybdenum in leach solution of column A (inoculated).

adversely copper extraction for a 21-day period. Copper extraction from the inoculated column (A) was 38%, as calculated from solution assays, and proceeded at an average rate of 21 mg/liter/day; the average rate of extraction for the control was 1.9 mg copper/liter/day for 4% extraction. Copper concn of samples collected throughout column A were as follows: top, 0.24%; middle, 0.16%; and bottom, 0.18%, suggesting copper leaching to be greatest at the middle and bottom of the column. Copper concn in ore samples throughout column B remained nearly the same as in the original ore sample, ranging from 0.32% to 0.28%. By analysis of tailings 42% and 0% copper were extracted from columns A and B, respectively. Very little ore decrepitation occurred in either column.

Figures 3 and 4 show the concn of total iron, ferrous iron, and molybdenum in solution for columns A and B. Nearly all iron in solution was ferrous since ferric iron is insoluble at 60 C at pH 2.5. Concentration of iron in solution was less in column A than in column B, resulting from greater bacterial activity in A oxidizing the iron which increased the iron precipitation. Iron concn were variable over the leach period. During copper precipitation, a large concn of iron was added to the solution in A. This concn rapidly decreased probably because of bacterial oxidation and subsequent precipitation. Molybdenum concn increased as iron concn increased. Probably molybdenum and iron attained certain values forming insoluble ferri-molybdate ( $\text{Fe}_2(\text{MoO}_4)_3 \cdot 8\text{H}_2\text{O}(\text{?})$ ) or other complexes, both iron and molybdenum were removed from solution.

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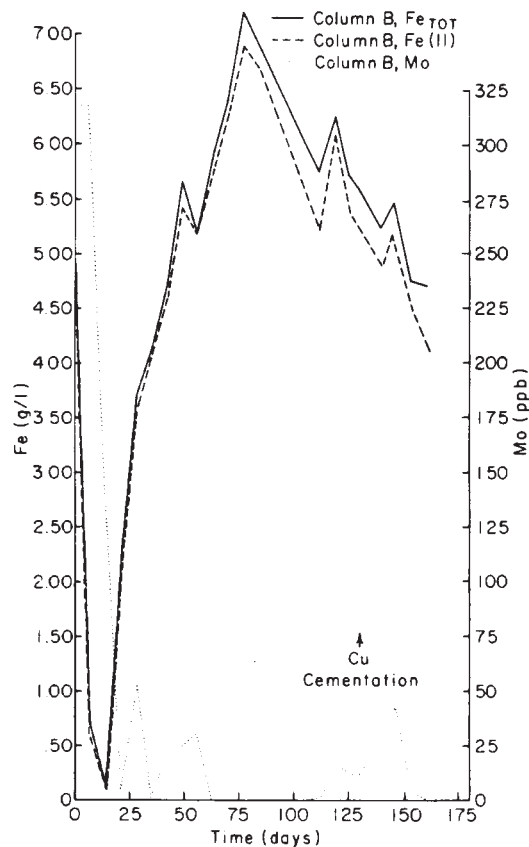


FIG. 4. Ferrous iron, total iron, and molybdenum in leach solution of column B (control).

After 160 days, zinc extraction in columns A and B was 17% and 12%, respectively. The thermophilic bacteria increased only slightly the extraction of zinc from this ore. Lead concn in the leach solutions of columns A and B attained a maximum of 2.6 ppm, but the values varied over the leach period. The inability to extract lead was probably due to the formation of lead sulfate which is only slightly soluble in a sulfuric acid solution. After 160 days, 7% of the nickel was leached from A and 6% from B, suggesting some chemical extraction of nickel.

Biological analysis results of Duval Sierrita tailings are shown in Table 2. The greatest numbers of ferrolobus-*Sulfolobus* were found in the top and middle sections of column A and more than 1800 *T. ferrooxidans* per gram of ore were cultured from the three sections. Coexistence of ferrolobus-*Sulfolobus* and *T. ferrooxidans* would not be expected due to differing temperature requirements. However, column A consisted of 2-ft sections of pipes, each having a portal to the outside; cooling of the ore around these portals probably resulted in niches having temperatures of about 45 C which is compatible for growing both strains. Tailings from column B indicated *T. ferrooxidans* contamination of the top section; no ferrolobus-*Sulfolobus* were noted. Thermophilic and mesophilic bacterial activity also is shown in Table 2. Activity for the thermophilic bacteria is very low. Since this assay depends on incorporation of  $^{14}\text{CO}_2$ , low activity in the presence of high bacterial counts may result from either bacterial utilization of other carbon sources such as organic carbon or a very low metabolism rate of ferrolobus and *Sulfolobus*. Activity measurements of *T. ferrooxidans* correlate well with the approximate numbers of these organisms observed.



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TABLE 2. *Thermophilic and mesophilic bacterial numbers and activity in leached ores*

Column	Column Section	Thermophilic Bacteria		Mesophilic Bacteria	
		Number/g	Activity (CPM/g)	Number <i>T. ferrooxidans</i> /g	Activity (CPM/g)
A	Top	> 1800	0.8	> 1800	88.6
	Middle	1600	6.4	> 1800	243.6
	Bottom	200	1.8	> 1800	380.4
B	Top	0	5.1	+	262.4
	Middle	0	0	0	42.3
	Bottom	0	0	0	47.9
C	Top	> 19000	48.2	> 19000	14.4
	Middle	9600	6.3	0	16.6
	Bottom	> 18700	2.0	0	14.3
D	Top	+	-	0	2.4
	Middle	+	13.5	0	3.1
	Bottom	+	3.2	0	25.7

Legend: 0 = no growth; + = good growth; - = no data; > = in excess of the number recorded.

*Pinto Valley low-grade ore (columns C and D).* Pyrite ( $\text{FeS}_2$ ) was the most abundant sulfide mineral, with chalcopyrite ( $\text{CuFeS}_2$ ) being the next most abundant. Some covellite ( $\text{CuS}$ ) and chalcocite ( $\text{Cu}_2\text{S}$ ) were present. Light minerals included quartz and feldspar, with small amounts of biotite, magnetite, and limonite. Chemical analysis of a split sample of ore (0.525 inch + 48 mesh) had 0.23% Cu, 0.02% Zn, 2.17% Fe, and 0.01% Mo. Each column had 109 lb of ore added, and 24 liters of 9K leach solution were recycled continuously. The pH of each system was maintained at approx. 2.5, with total addition of 250 ml concentrated sulfuric acid. Column C was inoculated with a mixed culture of  $8.4 \times 10^4$  ferrolobus-*Sulfolobus* and, after 115 days of leaching, reinoculated with an additional  $3.5 \times 10^4$  of these bacteria. Periodic culturing showed contamination of reservoir C with *T. ferrooxidans*, and temporary heating of the reservoir was employed to reduce the number. At the conclusion of the experiment,  $1.6 \times 10^5$  *T. ferrooxidans* per 100 ml were present in the reservoir solution. Panacide was added initially to column D; further additions were made later in the leach cycle.

Results of biological analyses of the Pinto Valley tailings are shown in Table 2. Grab samples from the top and bottom sections of column C showed extensive growth of ferrolobus-*Sulfolobus*, with somewhat less growth in the middle section. Activity results for the thermophilic bacteria were low, suggesting depressed metabolic rates or use of other carbon sources by the organisms. The top section of column C was contaminated with *T. ferrooxidans*; however, no growth of this organism was observed in the middle or bottom sections. All  $^{14}\text{CO}_2$  activity values were low for the mesophilic bacteria in C, suggesting that the contaminants in the top section were metabolizing at a low rate. Although counts for ferrolobus-*Sulfolobus* were not made of the tailings from column D, culturing indicated excellent growth of these organisms throughout. Activity measurements were again low, as observed in previous experiments. *T. ferrooxidans* were not detected in column D.

The average rate of copper extraction was 1.7 times greater for column C than D. After 202 days of leaching, 49% of the copper was extracted from C and 27% from D, as measured by accumulation of copper in the leach solution. Analysis of the tailings showed 30% copper extracted from C and 13% from D. Figure 5 shows that copper was extracted at nearly the

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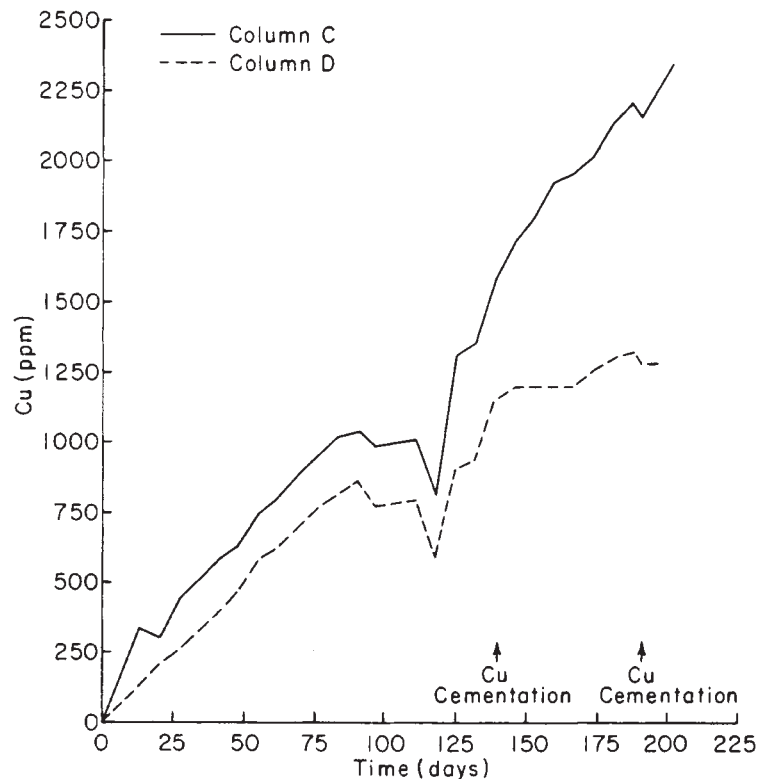


FIG. 5. Copper concn in leach solutions of columns C (inoculated) and D (control).

same rate from both columns for the first 125 days. Since the control column was contaminated with ferrolobus-*Sulfolobus*, this would explain the nearly equal copper-extraction rates observed. At day 118 of the experiment, panacide was added to D and successive additions were made on days 140, 154, and 188. Such additions may have decreased the numbers of the thermophilic bacteria and thus reduced the rate of copper extraction as observed in the last 70 days of the experiment. Such evidence suggests that copper extraction was controlled biologically. The greatest copper extraction was from the smallest sieve sizes (-16+48 mesh; 1.00 mm-300  $\mu$ m), which agrees with other studies (Ehrlich and Fox 1967) that particles with greater surface areas leach faster.

Ferrous and total iron values noted in C and D leach solutions were similar to those observed for column A. Iron values were variable, and nearly all iron in solution was present as ferrous due to the insolubility of ferric iron. High concn of ferrous iron added during copper removal were removed rapidly from solution by precipitation after biological oxidation. Molybdenum concn varied with the iron; the greater the iron concn, the greater amount of molybdenum in solution. Recovery of molybdenum from these systems would be difficult due to the extreme variability in concn.

Zinc was extracted at nearly the same rate from both columns (C and D) of Pinto Valley ore. The rate of zinc extraction for 189 days was 0.37 mg/liter/day for approx. 60% extraction. Since both columns contained the ferrolobus-*Sulfolobus* strains, establishing whether extraction was biological or chemical was not possible. Some decrepitation of the -4 + 6 mesh ore size in column C was observed, but generally the ore was resistant to break down.

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## DISCUSSION

The results obtained from flask-leaching tests of chalcopyrite concentrate suggest that copper is extracted by ferrolobus. Supplementing ferrolobus with yeast extract and ferrous iron enhanced copper extraction. The addition of these supplements probably increases the bacterial population and increases the ferric iron concn. Experimentation on the numbers of bacteria, bacterial attachment, and iron and copper dissolution would establish the mechanism(s) of attack on chalcopyrite.

Although copper extraction from Duval Sierrita chalcopyrite ore inoculated with ferrolobus-*Sulfolobus* was approx. 10 times greater than extraction of copper from the control column, assessing the contribution of thermophilic bacteria to the leaching process is difficult since the inoculated column was contaminated with *T. ferrooxidans*. This problem also existed in column C. The flask studies showed that ferrolobus readily extracted copper from chalcopyrite, and some studies (Malouf 1972) indicate that chalcopyrite is quite resistant to leaching by *T. ferrooxidans*. This evidence, therefore, suggests that the thermophilic bacteria were primarily responsible for the copper extraction.

Column D was contaminated with the thermophilic bacteria, but not *T. ferrooxidans*. Leaching proceeded in system D at a rate comparable to C until panacide was added at regular intervals. This suggests that the thermophilic bacteria were primarily responsible for copper extraction.

Low  $^{14}\text{CO}_2$  uptake values, recorded for thermophilic bacterial activity on grab samples from tailings containing large populations of these bacteria, suggest that other methods for measuring the activity should be developed. ATP spectrophotometric methods are being examined by J. A. Brierley (pers. comm.).

Comparison of size analyses of heads and tails indicated only slight decrepitation of the ores. This suggests that large pieces of ore would be difficult to leach since large surface areas would not be made available by breakdown of the ore. Although the dissolution rates of several metals from ores containing chalcopyrite as the primary mineral were examined, only copper was extracted at an accelerated rate by the thermophilic, acidophilic bacteria. The results indicate that these bacteria would be useful to *in-situ* mining where temperatures could be maintained for optimum growth of the organisms, and to vat leaching systems and dump leaching operations in which exothermic chemical reactions increase temperature sufficiently for thermophilic bacterial growth.

## ACKNOWLEDGMENTS

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