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Oxidative dissolution of bornite by *Acidithiobacillus ferrooxidans*

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ABSTRACT

The oxidation of finely ground (−200 μm) bornite (Cu_5FeS_4) by *Acidithiobacillus ferrooxidans* was evaluated in oxygen uptake and shake flasks experiments. The oxidation was a net acid-consuming reaction. Residual bornite was not detected by X-fray diffraction in solids after 2 days of contact in acid leach solution, indicating that the chemical and biological oxidation of bornite was relatively fast. Virtually 100% of copper solubilization was achieved in *A. ferrooxidans* cultures with or without ferrous iron, while in abiotic controls the copper extraction was around 30%. Bornite was not oxidized by *Acidithiobacillus thiooxidans* in respirometric or shake flasks experiments. Covellite (CuS) was detected as a secondary phase under all experimental conditions. Sulfur and jarosite were formed only in the presence of *A. ferrooxidans*.

1. Introduction

Among Fe-bearing Cu-sulfides, bornite [Cu_5FeS_4] has a relatively widespread occurrence in diverse geological locations. Physical and chemical properties of this economically important Cu-sulfide have been characterized in previous studies [1,2]. Electron paramagnetic resonance studies suggest that copper occurs only in the Cu(I) valence in natural bornite [3]. Iron is in the Fe(III) state [3], as confirmed also by Mössbauer data [4]. Early valence changes upon bornite oxidation are complex and extremely difficult to track because of transient redox interconversions and perturbations and the S-bridge between Fe(III) and Cu(I) [5].

Bornite oxidation in acid solutions involves multiple oxidants in bioleaching reactions. Ferric iron-dependent oxidation in the absence of any other oxidants typically accumulates ferrous iron and elemental sulfur:

$$Cu_5FeS_4 + 12Fe^{3+} \rightarrow 5Cu^{2+} + 13Fe^{2+} + 4S^0 \quad (1)$$

Upon solubilization the monovalent Cu in bornite is oxidized very fast in the solution phase by Fe^{3+} as well as by dissolved O_2. Fe- and S-oxidizing bacteria, when present, can regenerate ferric iron and produce sulfuric acid from elemental sulfur:

$$4Fe^{2+} + O_2 + 4H+ \rightarrow 4Fe^{3+} + 2H_2O \quad (2)$$

$$2S^0 + 3O_2 + 2H_2O \rightarrow 2SO_4{}^{2-} + 4H^+ \quad (3)$$

Thus the concurrent ferric iron-dependent and bacterial oxidation can be approximated with the following net reaction:

$$4Cu_5FeS_4 + 37O_2 + 20H^+ \rightarrow 20Cu^{2+} + 4Fe^{3+} + 16SO_4{}^{2-} + 10H_2O \quad (4)$$

Because bornite contains structural Fe, the oxidation releases iron into solution which acts as a redox shuttle in the bioleaching. At pH values >1.5, ferric iron forms jarosite, an acid producing precipitation reaction with some monovalent and divalent cations in bioleaching solutions.

$$3Fe^{3+} + K^+ + 2SO_4{}^{2-} + 6H_2O \rightarrow KFe_3(SO_4)_2(OH)_6 + 6H^+ \quad (5)$$

Formulations of mineral salt solutions used for cultivating acidophiles usually include K^+ and $NH_4{}^+$ which readily incorporate into jarosite solid solution.

Acidophilic bacteria such as *Acidithiobacillus ferrooxidans* play a central role in the acid leaching of Cu-sulfides, but the efficiency of the bioleaching greatly varies with the mineral. In general, bornite is present as a minor sulfide mineral in many Cu ores and is relatively readily oxidized in bioleaching systems. However, only few studies have been reported for the bacterial oxidation of research-grade bornite. Previous electrochemical studies with

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bacteria and bornite electrodes have shown that bacterial oxidation can be described as an accelerated corrosion process [6–8]. However, the solid-phase products of bacterial oxidation of bornite have not been reported in the literature. The purpose of this was to assess the oxidative dissolution of research-grade bornite by *A. ferrooxidans* and *A. thiooxidans* in respirometric and shake flask experiments. The formation of liquid and solid phase products over time was also characterized.

2. Materials and methods

A. ferrooxidans strain LR and *A. thiooxidans* strain FG-01, originally isolated from uranium mine effluents in Brazil [11] were used in this work. Ferrous sulfate-mineral salts medium [12] at pH 1.8 was used to maintain *A. ferrooxidans* LR and to grow cells for respirometric experiments. For *A. thiooxidans* FG-01, the same medium was used except that ferrous sulfate (6 g Fe^{2+} L^{-1}) was replaced by elemental sulfur (10 g L^{-1}) at initial pH 2.8 [12].

A research-grade bornite sample was obtained from Ward's Natural Science Establishment (Rochester, NY) and ground in a disc mill to 100% −200 μm size. The sample contained (w/w) 39.7% Cu, 19.7% Fe, and 29.4% S. X-ray diffraction (XRD) analysis revealed bornite as the major phase and minor amounts of accessory quartz (SiO_2), pyrite (FeS_2) and chalcopyrite ($CuFeS_2$) in this sample.

Respirometric experiments were performed using a Warburg apparatus with washed cells of *A. ferrooxidans* and *A. thiooxidans*, which were previously grown in the mineral salts medium. Cells were harvested by membrane filtration (0.45 μm pore size), washed twice, and resuspended in 0.01 M H_2SO_4. Cell suspensions were standardized by protein determination [13].

A. ferrooxidans cultures were initially adapted to grow with bornite through several subcultures in liquid medium through successive replacement of Fe^{2+} with bornite sample. Attempts to adapt *A. thiooxidans* to bornite by gradual replacement of elemental sulfur with the mineral were unsuccessful. The leaching experiments were carried out in 150 ml cultures in 250 ml flasks containing bornite (2.5%, w/v) in mineral salts solution. The flasks were sterilized by autoclaving (30 min, 120 °C) and inoculated with (5%, v/v) previously adapted *A. ferrooxidans* or inoculated with sulfur-grown *A. thiooxidans*. The cultures in shake flasks were incubated at 150 rpm and 30 ± 2 °C. In some experiments, the *A. ferrooxidans* cultures were supplemented with 30 mmol Fe^{2+} L^{-1} (as ferrous sulfate). The following formulations were used as uninoculated chemical controls: (i) mineral salts solution, (ii) mineral salts solution supplemented with 30 mmol Fe^{2+} L^{-1}, and (iii) mineral salts solution amended with filter-sterilized (0.45 μm pore size) spent medium from an *A. ferrooxidans* culture containing about 30 mmol Fe^{3+} L^{-1}.

Samples (15 mL) were periodically (days 2, 5, 7 and weekly thereafter) withdrawn from the flasks for measurements of pH and redox potential (an Ag^0/AgCl reference) and for chemical analyses of leach solutions. The concentrations of Fe^{2+} and total Fe in solution were determined titrimetrically with $K_2Cr_2O_7$ [14] and Fe^{3+} was calculated from the difference. Supernatants from centrifuged samples (10,000 × *g* for 15 min) were preserved in 1 M HCl for Cu analysis by inductively coupled plasma emission spectroscopy.

Solids were recovered by centrifugation on days 7, 28, and 56 and were air dried for XRD analyses. Dried residues were ground gently in an agate mortar and analyzed as top fill mounts in a Siemens D-5000 diffractometer, using graphite monochromated CuK radiation and 2 range from 10° to 70° in 0.02° 2 increments and a 2 s count time.

3. Results and dsiscussion

3.1. Oxygen uptake experiments with A. ferrooxidans and A. thiooxidans

Oxygen uptake coupled with bornite oxidation by resting cells of *A. ferrooxidans* and *A. thiooxidans* in manometric experiments is shown in Fig. 1. The oxygen uptake due to bornite oxidation by *A. ferrooxidans* was higher than that of *A. thiooxidans* and the abiotic control but only slightly faster than the chemical control in the 0.01 M H_2SO_4 medium (Fig. 1A). The oxygen consumption in the chemical control shows that bornite is abiotically oxidized in acid solution. However, in the pH 2.2 buffered-medium, the bacterial oxygen uptake was considerably higher than in both controls (Fig. 1B) and *A. thiooxidans*. It has previously been shown that glycine–H_2SO_4 buffer has no stimulatory or inhibitory effect on *A. ferrooxidans* [15]. As shown in reaction (2), bornite oxidation is net acid consuming and the pH increases during the incubation. The slow bornite-dependent oxygen uptake by *A. ferrooxidans* in

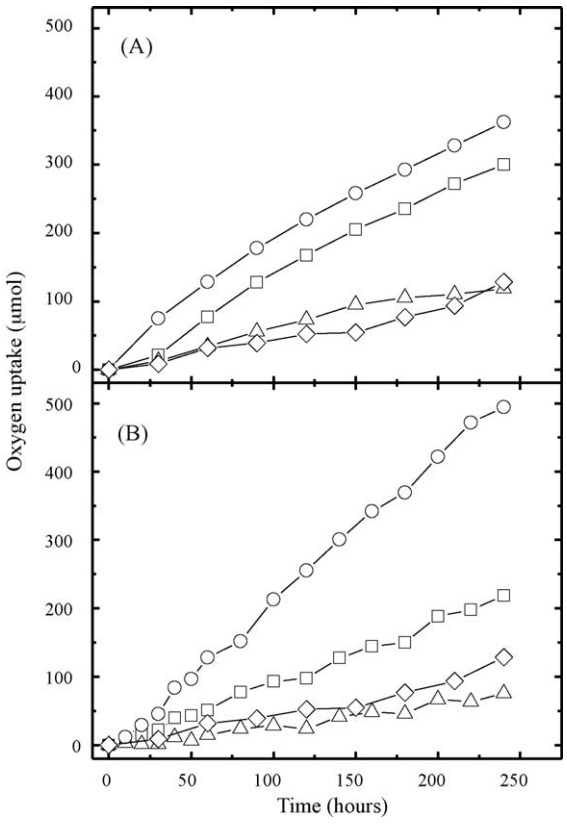

Fig. 1. Oxygen uptake by *A. ferrooxidans* and *A. thiooxidans* in manometric experiments using bornite (100 mg–200 μmol) as substrate. Cell suspensions (150 μg protein/manometric flask) were incubated in (A) acid water–H_2SO_4, pH 1.8, and (B), glycine–H_2SO_4 buffer, pH 2.2. *A. ferrooxidans* (○); *A. thiooxidans* (◇); abiotic control (□). Dead cells (△) were used in both experiments to account for non-specific O_2 uptake. Oxygen uptake activity was not monitored to the completion.

unbuffered medium may be the result of the increase of pH to 4.5, which is outside of the pH range at which *A. ferrooxidans* is normally active [16]. The abiotic control involving inactivated cells showed slightly less oxygen consumption compared to the chemical control. Bornite was not oxidized actively by *A. thiooxidans* cell suspension and the oxygen consumption was similar to that of abiotic control.

3.2. Bornite leaching of experiments with A. ferrooxidans

In bornite bioleaching experiments with *A. ferrooxidans*, the pH reached 3.5 within the first days of the experiment (Fig. 2). The concurrent Fe^{2+} oxidation by *A. ferrooxidans* (Fig. 3A and B) was also acid-consuming (Eq. (2)). After this initial period a one-time pH adjustment was necessary to maintain a permissive pH range for bacterial growth. In the chemical controls, the redox potential was relatively stable at ∼360 mV (Fig. 2A). Iron oxidation was also apparent from the redox potential of 550 mV (Fig. 2). The concentration of Fe^{3+} reached about 20 mmol L^{-1} in 20 days and kept constant until the end of the experiment. Ferric iron was not detected in the abiotic control during the experiment (Fig. 3A).

In the Fe^{2+}-amended *A. ferrooxidans* culture, Fe^{2+} was oxidized and the Fe^{3+} concentration increased to about 10 mmol L^{-1} in 7 days. In the abiotic control dissolved iron remained in the reduced form (Fig. 3B).

Spent culture filtrate of *A. ferrooxidans*, containing about 30 mmol Fe^{3+} L^{-1}, was also used as a sterile, chemical lixiviant (Figs. 2C and 3C). The redox potential decreased to comparable