

FLOCCULATION OF FINE FLUORITE PARTICLES WITH *CORYNEBACTERIUM XEROSIS*

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ABSTRACT

The treatment of fine particles dispersed in liquids is common in several industries and especially important in mineral processing. The efficiency of settling operations can be substantially increased by flocculation. The aim of this work was to study the flocculation of fine fluorite particles by the bacterium *Corynebacterium xerosis*. Flocculation tests, microelectrophoresis measurements and optical microscopy were used to evaluate flocculation. The results showed that *C. xerosis* cells adhere to the fluorite surfaces promoting the aggregation of the particles. High quality flocs can be obtained rapidly at pH 7.0 using a cell concentration of 40 mg/l, considerably lower than previously reported in the literature. The results are discussed with reference to the surface characteristics of the mineral and of the microorganism.

Key words: adhesion, *Corynebacterium xerosis*, flocculation, fluorite.

INTRODUCTION

At various stages during the processing of minerals it is necessary to separate the aqueous suspensions into their component solid and liquid phases. Typical examples of this are thickening of flotation concentrates, recovery of pregnant leach liquors, and dewatering of tailings. In many cases, the mineral particles settle out of suspension very slowly, so that the liquid-solid separation is slow and incomplete.

The efficiency of such gravity settling operations can be substantially increased by aggregation of the mineral particles. Methods currently used for aggregation and settling of finely divided solids in slurries include pH adjustment, increasing ionic strength, introduction of inorganic coagulants such

as aluminum and ferric salts, and introduction of organic flocculants such as starch and long chain synthetic flocculants (10,15,16). These methods, in addition to increasing costs, can also lead to the release of polluting waste materials.

Finely divided mineral particles can also be flocculated by microorganisms. An isolate of *Arthrobacter* has been shown to promote the settling of fines in manganese ore washings (5). The bacterium *Mycobacterium phlei* (7,14) and the yeast *Candida parapsilosis* (12,13) improved settling rates in mineral suspensions, but flocculation was not as good as that obtained with commercial long chain flocculants. Most microorganisms adhere to solid surfaces if the charge and hydrophobic interactions between the cells and the solid surface are favorable, but do not always promote flocculation of mineral

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particles. The mechanism of this flocculation appears primarily to be cell adhesion and bridging between the particles.

Van Loosdrecht *et al.* (17,18) showed that cells of *M. phlei* and *Corynebacterium xerosis* were highly hydrophobic and negatively charged. Since previous work has shown that the former microorganism does not effectively promote flocculation of fluorite (8), we investigated the potential of the latter, *C. xerosis*, to flocculate fine fluorite particles. The effect of settling time, cell concentration, and pH of the suspension on solids settling was studied. The results are discussed in terms of the surface properties of the mineral and of the microorganism.

MATERIALS AND METHODS

Fluorite

Fluorite crystals (99% CaF₂) were obtained from a mining company (Criciúma - Santa Catarina, Brazil). The crystals were ground in a laboratory mill. The particle size distribution of the sample used for flocculation tests was determined by laser diffraction in a Malvern Particle Size Analyzer Model 3601 (Table 1).

Table 1. Particle size distribution of fluorite.

Size range	Weight (%)
larger than 40 µm	3
40 - 30 µm	9
30 - 20 µm	18
20 - 10 µm	29
10 - 5 µm	18
5 - 1 µm	21
smaller than 1 µm	2

Microorganisms

The bacterium *Corynebacterium xerosis*, from Caroline Biological Supply Company - USA, was grown for 72 hours in nutrient broth. Cultures were produced in 300 ml flasks, continuously shaken at 37°C. Cells were harvested by centrifugation, washed twice in distilled water and finally suspended in water to give a 2 g/l suspension. This stock cell solution was kept at 5°C and used in flocculation studies within 48 hours, either neat, or at various dilutions.

Flocculation tests

Flocculation studies were carried out using a standard "Jar Test" apparatus. Settling tests were

performed by preparing 1000 ml suspensions containing 10 g/l of fluorite. The pH of the medium was adjusted as required and the microorganisms added to the mineral suspension. The suspensions were mixed by stirring for 1 min at 250 rpm to promote a uniform distribution of the cells. The stirrer speed was reduced to 50 rpm for 2 min, to create low shear conditions and allow floc formation. Particles were then allowed to settle under gravity and samples were withdrawn at set time intervals from 10 cm below the water surface.

The flocculation efficiency was evaluated by the following parameters:

- naked eye and microscope observation of floc quality;
- residual turbidity in nephelometric units of turbidity (NTU);
- solids removal from water.

The solids concentration was determined gravimetrically. The solids removal percentage (or flocculation efficiency) was calculated using the relationship.

$$E(\%) = 100 \times \frac{(C_i - C_f)}{C_i}$$

where: C_i = initial concentration of solids

C_f = concentration of solids after time t

Zeta potential measurements

The electrokinetic properties were studied as a function of pH using a Rank Brothers Ltd microelectrophoresis apparatus. *C. xerosis* cells and fluorite particles were suspended in water containing 1x10⁻³ M NaNO₃ and the pH was adjusted by addition of NaOH and HNO₃. For each pH value 20 readings were taken and averaged. The zeta potential was calculated from electrophoretic mobility using the Smoluchowsky equation (9).

RESULTS

Fig. 1 shows photomicrographs demonstrating the flocculation of fluorite by *C. xerosis*. Fig. 1a shows dispersed fluorite particles in the absence of the microorganisms. In the presence of *C. xerosis* cells, particles are agglomerated (Fig. 1b). These fluorite/cell aggregates settle very rapidly in water allowing an almost complete solids removal and a high clarification of the water.

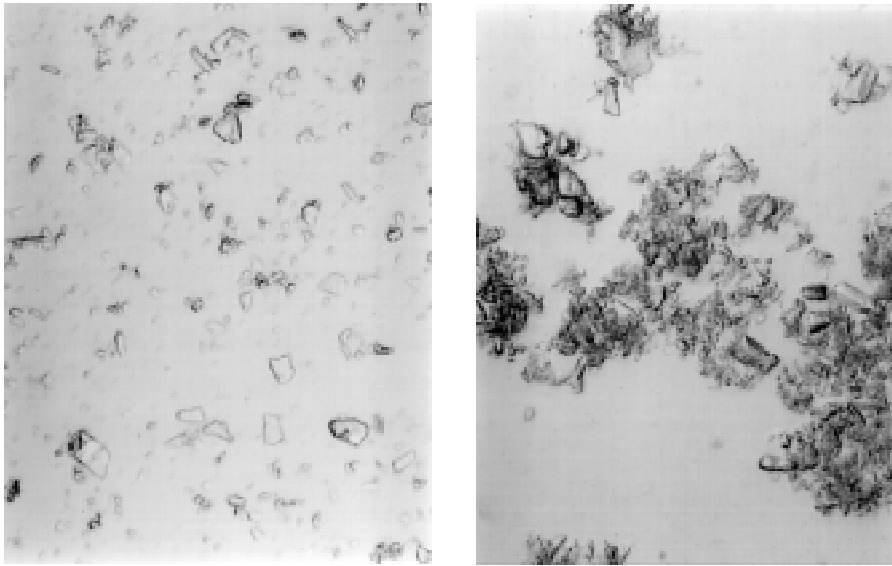


Figure 1. Photomicrographs of fluorite particles suspended in water (a) and aggregated in the presence of *Corynebacterium xerosis* (b). Magnification 100x.

The effect of settling time on flocculation efficiency in the presence of 40 mg/l *C. xerosis* cells at pH 7.0 is shown in Fig. 2. The result is compared to a plot obtained in the absence of flocculant. In the presence of bacteria, near maximum flocculation is obtained within 30 s after the beginning of settling. The same test procedure was used to evaluate the effect of settling time on residual turbidity, which

was decreased from 130 NTU to 20 NTU in approximately 5 min in the presence of *C. xerosis* (Fig. 3).

Fig. 4 shows the flocculation results as a function of cell concentration. Good flocculation is obtained at a microorganism dosage of 20 mg/l and maximum flocculation at 40 mg/l. About 96 % of the solids present in the suspension were removed

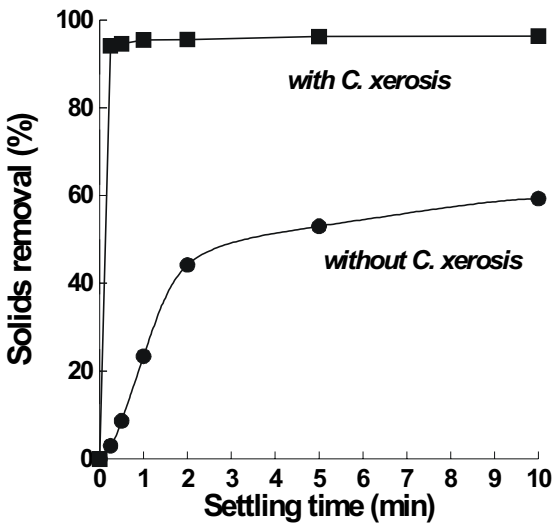


Figure 2. Solids removal efficiency of fluorite suspensions as a function of settling time in the presence or absence of 40 mg/l of *Corynebacterium xerosis* (pH 7.0 +/- 0.2).

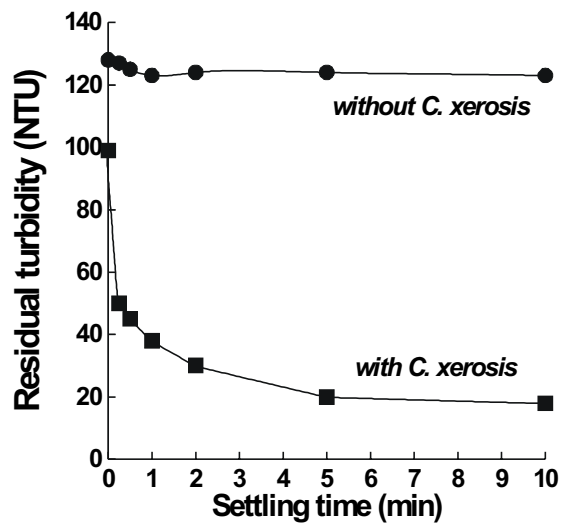


Figure 3. Residual turbidity of fluorite suspension as a function of settling time in the presence or absence of 40 mg/l of *Corynebacterium xerosis* (pH 7.0 +/- 0.2).

in a settling time of 1 min. Higher concentrations than 40 mg/l did not produce any improvement in floc quality.

Fig. 5 represents flocculation efficiency as a function of pH. Also shown are data for fluorite flocculation in the absence of microorganisms. At the bacterial cell concentration used, the suspensions were readily flocculated at pH values of 4.0 - 9.0, with an optimum pH of around 7.0.

Fig. 6 displays the zeta potential of *C. xerosis* cells and of fluorite particles as a function of pH. The *C. xerosis* cells had an isoelectric point at pH 2.0 while the isoelectric point of fluorite was near pH 10.0. Negatively charged organisms should readily adhere to the positively charged fluorite at neutral pH values. At pH 7.0, which gave best flocculation results, the *C. xerosis* cells have a zeta potential of -30 mV and fluorite particles of about + 60 mV.

DISCUSSION

The experimental data indicate that *C. xerosis* cells adhere to fluorite particles and promote flocculation of fine fluorite suspensions over a range of pH values. The quality of the flocs obtained are comparable to those produced with long chain polymers. The flocs are very “tight” and dense and contain little water. The flocculation occurs rapidly, within 1min., showing that these bacteria are more

efficient than the *Arthrobacter* species used by Deshpande (5) for clearing manganese mining wastewaters, which required a minimum of 30 min settling time.

The mechanism of flocculation has been discussed elsewhere and primarily occurs by cell bridging (7,14). This mechanism is specially efficient in systems where the microorganism is smaller or of

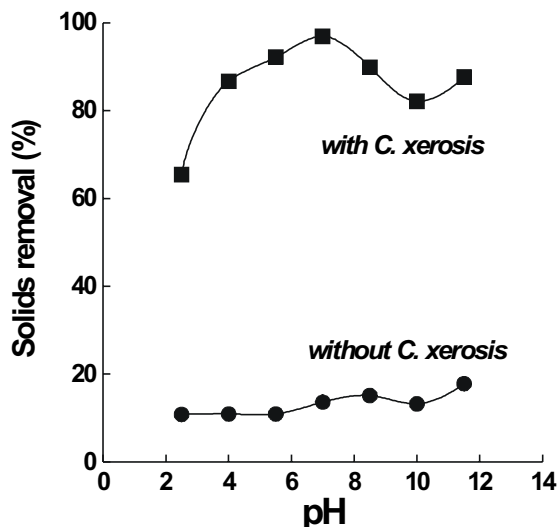


Figure 5. Solids removal efficiency of fluorite suspensions as a function of pH in the presence or absence of 40 mg/l of *Corynebacterium xerosis* cells (1 min settling time).

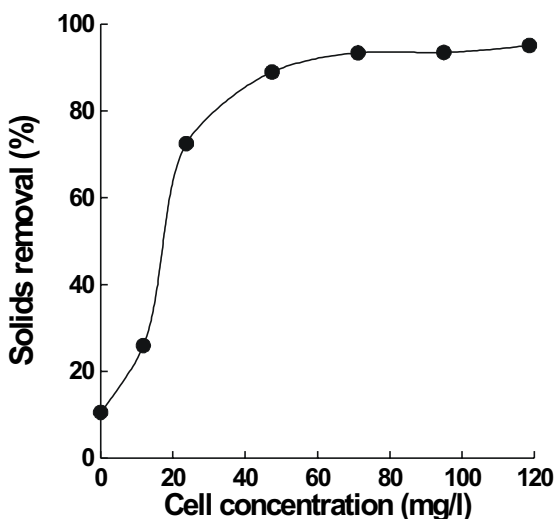


Figure 4. Solids removal efficiency of fluorite suspensions as a function of *Corynebacterium xerosis* cells (pH 7.0 +/- 0.2, 1 min settling time).

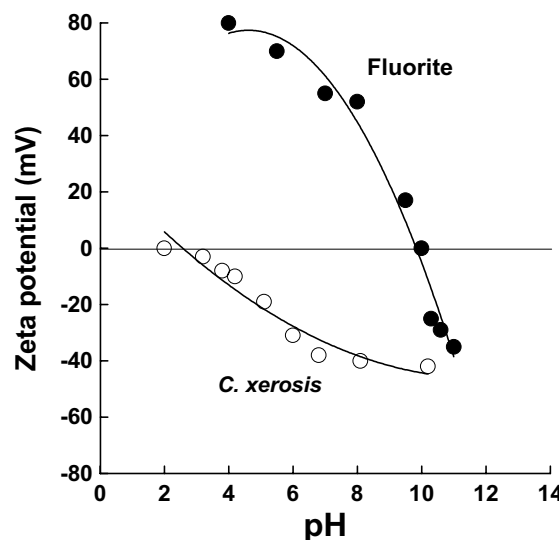


Figure 6. Zeta potential of *Corynebacterium xerosis* cells and fluorite particles as a function of pH. Ionic strength maintained by addition of 1×10^{-3} M NaNO_3 .

a similar size to the particles (6). In the present case, the median size of the particles was 12.5 μm and only 2% of the fluorite has a particle size smaller than 1 μm (approximate size of *C. xerosis* cells).

In many cases, the adherence of a microorganism to a mineral surface can be predicted by a calculation based on the Derjaguin-Landau-Verwey-Overbeek (DLVO) theory (6,18,19). According to this theory, the total long range interaction ($> 1\text{nm}$) between surfaces is a summation of van der Waals and coulombic interactions (1,4). This theory explains the optimum flocculation behavior at neutral pH values in the fluorite - *C. xerosis* system. At pH 7.0 the fluorite has a high positive charge (about + 60 mV) and the bacteria a high negative charge (about - 30 mV).

However, a specific adsorption interaction should not be neglected. Bacteria having similar electrokinetic behavior were not able to promote the same degree of flocculation of fluorite (8). It has been shown that some Gram-negative bacteria have specific adsorption mechanisms to solid surfaces, associated with the surface lipopolysaccharides (3), and it is possible that a similar type of interaction may occur with this Gram-positive organism. Gram negative bacteria tested by us had no effect on settling (results not shown). However, the members of the *Acintomycetaceae* have previously been shown to promote flocculation of fine particles (5) and one possibility is that this is due to the distinctive nature of their cell wall material (11). In addition, the *Corynebacterium* group can produce short chains of cells because of incomplete separation after cell division (2) and this leads to a pseudo-polymeric structure, promoting cell bridging.

The concentration of *C. xerosis* required to flocculate fine fluorite suspensions is lower than that of other microorganisms reported in the literature. For instance, flocculation of hematite suspensions with the yeast *Candida parapsilosis* requires about 50 kg/t of bioflocculant (12), while flocculation of fluorite with *C. xerosis* is possible with only 4 kg/t. However, the dosage required is still higher than that used in flocculation with long synthetic polymers. A long chain polyacrylamide (average molecular weight 1×10^6) flocculates some suspensions at concentrations below 0.1 kg/t.

Microorganisms are potential low cost flocculants for solid-liquid operations in mineral processing. Further studies are necessary to increase their competitiveness with common flocculants used

in the mining industry. These will include research to identify more bacterial strains able to adhere to mineral surfaces, investigate the mechanisms involved, develop procedures to isolate the biochemical fraction responsible for flocculation, and to optimize the production of the bioflocculants on large scale.

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RESUMO

Floculação de partículas finas de fluorita com a *Corynebacterium xerosis*

O tratamento de partículas finas dispersas em líquidos é comum em diversas indústrias e especialmente importante no processamento de minérios. A eficiência de operações de sedimentação pode ser substancialmente aumentada com a floculação. O objetivo deste trabalho foi estudar a floculação de partículas de fluorita com a bactéria *Corynebacterium xerosis*. A metodologia empregada para avaliar a floculação incluiu experimentos de floculação, microeletroforese e microscopia ótica. Os resultados demonstraram que as células da *Corynebacterium xerosis* se aderem na superfície da fluorita, promovendo a agregação das partículas. Flocos de alta qualidade podem ser obtidos rapidamente em pH 7,0 a uma concentração de microrganismos de 40 mg/l, dosagem esta consideravelmente menor do que aquelas previamente relatadas na literatura. Os resultados são discutidos em termos das propriedades superficiais do mineral e do microrganismo.

Palavras-chave: adesão, *Corynebacterium xerosis*, floculação, fluorita.

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