Available online at www.sciencedirect.com

SCIENCE DIRECT®

Int. J. Miner. Process. 75 (2005) 113–122

INTERNATIONAL JOURNAL OF MINERAL PROCESSING

www.elsevier.com/locate/ijminpro

Selective separation of pyrite from chalcopyrite and arsenopyrite by biomodulation using *Acidithiobacillus ferrooxidans*

M.N. Chandraprabha[a], K.A. Natarajan[a,*], P. Somasundaran[b]

[a]*Department of Metallurgy, Indian Institute of Science, Bangalore-560 012, India*
[b]*Langmuir Center for Colloids and Interfaces, Columbia University, New York, 10027, USA*

Received 16 March 2004; received in revised form 3 August 2004; accepted 3 August 2004

Abstract

This paper discusses the selective depression of pyrite from chalcopyrite and arsenopyrite by biomodulation using *Acidithiobacillus ferrooxidans* under natural conditions of pH. The effect of bacteria–mineral interaction on the surface charge of mineral and bacterial cell was studied by microelectrophoresis. Adhesion experiments were conducted to establish the relationship between cell adhesion to specific minerals and the electrokinetic behaviour of the minerals subsequent to interaction with cells. Effect of bacterial interaction on the xanthate-induced flotation of all the minerals was assessed. Adhesion of *A. ferrooxidans* on pyrite was rapid and tenacious and subsequent to interaction with cells, pyrite remained hydrophilic even in presence of xanthate collector. The collector, on the other hand, was able to render good flotability to chalcopyrite even after interaction with bacterial cells. Copper activated arsenopyrite was able to retain its hydrophobicity in presence of cells due to poor attachment kinetics of cells to the mineral surface. Thus, by suitably conditioning with the cells and collector, it was possible to effectively depress pyrite from chalcopyrite and arsenopyrite.

Keywords: *Acidithiobacillus ferrooxidans*; pyrite; chalcopyrite; arsenopyrite; biomodulation; selective flotation

1. Introduction

Separation of pyrite from other associated metal sulphides is desirable for the economical extraction of their valuable metals. Chalcopyrite and arsenopyrite are often associated with pyrite and selective depression of the pyrite from these would prove economically beneficial for further extraction. Separation of pyrite from chalcopyrite is conventionally achieved by selective depression of pyrite using depressants like cyanide under alkaline conditions. On the other hand, separation of pyrite from arsenopyrite has been a problem due to their similar flotation behaviour with sulphidic collectors. Conventional techniques for separation of pyrite from either chalcopyrite or arsenopyrite utilize depressants like cyanide or oxidizing agents like permanganates and magnesia–ammonia mixture for selective oxidation of arsen-

* Corresponding author. Tel.: +91 80 3600120; fax: +91 80 3600472.
E-mail address: kan@metalrg.iisc.ernet.in (K.A. Natarajan).

0301-7516/$ - see front matter © 2004 Elsevier B.V. All rights reserved.
doi:10.1016/j.minpro.2004.08.014

opyrite (Randol, 1992; Yen and Tajadod, 1999). Thus, there is still a need for less-expensive and environmentally benign reagents.

The utility of microorganisms in selective flotation or depression of sulphides and oxides has been previously reported (Yelloji Rao et al., 1992; Deo and Natarajan, 1998; Santhiya et al., 2000; Patra and Natarajan, 2003). However, no significant research has been reported on the separation of pyrite from associated ferrous sulphides. Recently, Sharma et al. (1999) have studied the effect of bacterial conditioning on the behaviour of pyrite and chalcopyrite. In our previous study (Chandraprabha et al., 2004), we were able to achieve selective separation of pyrite from a mixture of pyrite and chalcopyrite by collector interaction and biomodulation using *Acidithiobacillus ferrooxidans*.

In the present study, selective separation of pyrite from a mixture of chalcopyrite and arsenopyrite by biomodulation and collector interaction has been investigated in detail.

2. Materials and methods

2.1. Minerals

Arsenopyrite was obtained from Wards Scientific, USA, pyrite from Alminrock Indser Fabricks, India, and chalcopyrite from Gregory, Bottley and Lloyd, UK as pure handpicked mineral samples. The mineral samples were dry ground in a porcelain ball mill and dry sieved to obtain different size fractions. The −106+75-μm fraction was used for flotation studies. The −37-μm fraction was further ground in a Retsch mortar grinder. The particle size analysis of this sample was carried out using a Malvern Mastersizer 3000-model and the mean size was found to be ≈5 μm. This fraction was used for adsorption and electrokinetic studies. The minerals were stored in a desiccator under nitrogen atmosphere. The surface area was estimated by BET nitrogen specific surface area method and was found to be 1.26 m^2/g for pyrite, 1.61 m^2/g for chalcopyrite and 1.77 m^2/g for arsenopyrite respectively. Purity of the mineral samples were ascertained by mineralogical studies and X-ray diffraction using a JDX-8030 X-ray diffractometer system. The purity of the mineral samples was 99.9% for pyrite, 99.4% for chalcopyrite and 98.7% for arsenopyrite, respectively.

2.2. Microorganism and preparation of cell pellet

The bacterial culture used was a strain of *A. ferrooxidans* that was isolated from Hutti Gold Mines (HGML) and is referred to as TfH6. The purity was ascertained by the procedure outlined by Karavaiko (1988). The bacteria were cultured in sterile 9K medium developed by Silverman and Lundgren (1959). The bacterial count was monitored by direct counting under a Leitz phase contrast microscope (Labrolux K Wild MPS12) using a Petroff Hausser counter.

The grown culture was initially filtered through Whatman 42 filter paper to remove the precipitates. The filtrate was centrifuged at 10,000 rpm for 20 min in a Sorvall RC-5B refrigerated high-speed centrifuge at 5 °C to obtain the cell pellet. The pellet obtained was resuspended in pH 2 H_2SO_4 solution and then centrifuged as before to obtain metabolite-free cells.

2.3. Adsorption studies

The cell pellet from a culture of known cell concentration was suspended in 100 ml 10^{-3} M KCl solution at the desired pH in 250-ml standard Erlenmeyer flask. The mineral sample (1 g) was pulped to the cell suspension and the slurry obtained was agitated on a rotary shaker at 200 rpm for 30 min for equilibration. After equilibration, the slurry was vortex mixed for 1 min to remove loosely held cells, centrifuged at 2000 rpm for 5 min to settle the mineral particles and the cell number of the supernatant was recorded. For experiments on adhesion kinetics, the above procedure was repeated at regular intervals and the cell data with respect to time recorded.

2.4. Electrokinetic studies

The electrophoretic mobilities of the mineral samples before and after interaction with the bacterial cells were determined using a Malvern Zetasizer 3000 instrument. KCl solution (10^{-3} M)

was used as the base electrolyte in all the experiments. The mineral sample (1 g) was interacted with the desired cell concentration and pH for the required time. The slurry after interaction was centrifuged at 2000 rpm for 3 min to settle only the mineral particles. The mineral particles were resuspended in KCl solution, vortexed to remove loosely held cells and washed two to three times. The mineral sample so obtained was equilibrated in KCl solution that was preadjusted to the desired pH before taking the measurements.

Electrophoretic mobility of cells after interaction with the minerals was also recorded by equilibrating the interacted cells in 10^{-3} M KCl solution at different pH. In all the experiments, five readings were recorded and the results reported represent the average value. The standard deviation was <0.1.

2.5. Microflotation studies

The flotation of the mineral samples was carried out using a modified Hallimond tube (Fuerstenau et al., 1957) by passing nitrogen gas at the flow rate of 40 ml/min for 3 min. Prior to flotation the mineral (1 g) was conditioned with deionised double distilled water (200 ml) at the desired pH for 5 min followed by addition of collector (potassium isopropyl xanthate) and further conditioning for 10 min. The floated and tailing samples were collected separately, filtered, dried and weighed. For flotation studies after interaction with cells/collector, the mineral samples were interacted with the collector/bacterial cells at the desired pH. The supernatant was carefully removed and the interacted mineral was transferred to the Hallimond tube and floated as before.

2.6. Differential flotation studies

Differential flotation of pyrite–chalcopyrite–arsenopyrite mixture was carried out by interacting 1:1:1 mixture of the minerals either together or separately with cells and collector. This mixture was then transferred to the Hallimond tube and floated as described earlier. Both the floated and settled fractions were analyzed for their arsenic, copper and iron content using a Jobin Yvon inductively coupled plasma spectrophotometer (ICP).

All the experiments were carried out in triplicate and the results reported represent the average value. The maximum deviation was within ±5% of the average values.

3. Results and discussion

3.1. Electrokinetic studies

The effect of bacterial interaction on the surface charge of the minerals was accessed by electrokinetic measurements. The results obtained are portrayed in Fig. 1a. All the three minerals exhibited acidic isoelectric points, i.e., at pH 3.25 for pyrite, pH 2.4 for chalcopyrite and pH 2.5 for arsenopyrite, and beyond this pH the electronegative character of the minerals increased with increase in pH. Interaction with *A. ferrooxidans* cells for 30 min shifted the isoelectric point of pyrite to a pH value of 4. The electronegative character of the mineral was also reduced after interaction with the cells. This lowering of electrophoretic mobility can be due to the amino groups on the cell surface. The electrophoretic mobility of chalcopyrite after interaction with bacterial cells shifted to pH 3.3. The electronegative character of the mineral also reduced after interaction with cells. The shift in isoelectric point and also the reduction in electrophoretic mobility of chalcopyrite are lower when compared to that of pyrite. Similar interaction with bacterial cells did not have significant effect on the charge character of arsenopyrite. As observed from the figure, the isoelectric point for the interacted mineral was located at pH 2.8, as against pH 2.5 for the pure mineral. The reduction in electrophoretic mobility of arsenopyrite was also lower when compared to that of pyrite or chalcopyrite. This difference in the behaviour of the minerals consequent to interaction with bacterial cells may be due to the preferential adsorption of cells onto pyrite and chalcopyrite, compared to that on arsenopyrite.

The surface of bacterial cells also exhibits changes upon interaction with the minerals and the observed electrokinetic behaviour of the bacterial cells before and after interaction with the minerals in depicted in Fig. 1b. The isoelectric point of cells was located at pH 2.3. Interaction with minerals shifted the isoelectric point of cells to higher pH as a consequence of specific

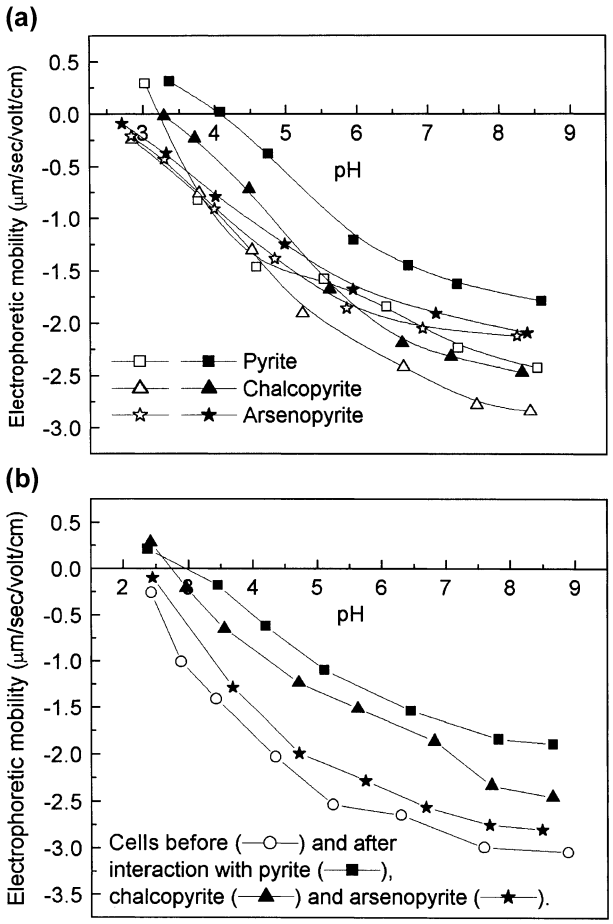

Fig. 1. Effect of cell–mineral interactions on the electrophoretic mobilities of (a) pyrite, chalcopyrite and arsenopyrite, and (b) bacterial cells with respect to pH. Open symbols represent electrophoretic mobilities before interaction and closed symbols after interaction.

adsorption of cells on minerals (Devasia et al., 1993). The shift in the isoelectric point was higher for pyrite-interacted cells (pH 3) when compared to chalcopyrite-interacted cells (pH 2.7). The bacterial cells did not exhibit similar shift in isoelectric point upon interaction with arsenopyrite. Owing to the formation of proteinaceous secretion on the bacterial cell surface due to interaction with minerals (Devasia et al., 1993), the electrophoretic mobility of the cells decreased upon interaction with minerals. This reduction in the electrophoretic mobility was higher for pyrite-interacted cells when compared to chalcopyrite-interacted or arsenopyrite interacted cells.

The observed difference in the electrokinetic behaviour of the minerals and cells consequent to their mutual interaction confirms that the cells exhibit selectivity towards the minerals.

3.2. Adsorption studies

The adhesion of *A. ferrooxidans* onto pyrite, chalcopyrite and arsenopyrite as a function of time is depicted in Fig. 2. Attachment kinetics with respect to pyrite and chalcopyrite was similar and was very fast with the equilibrium attained within 15 min. Presence of copper ions in the chalcopyrite lattice does not seem to affect the kinetics of adhesion of cells on its surface. But the cell density on the surface of chalcopyrite was lesser when compared to that on pyrite. Adsorption isotherm

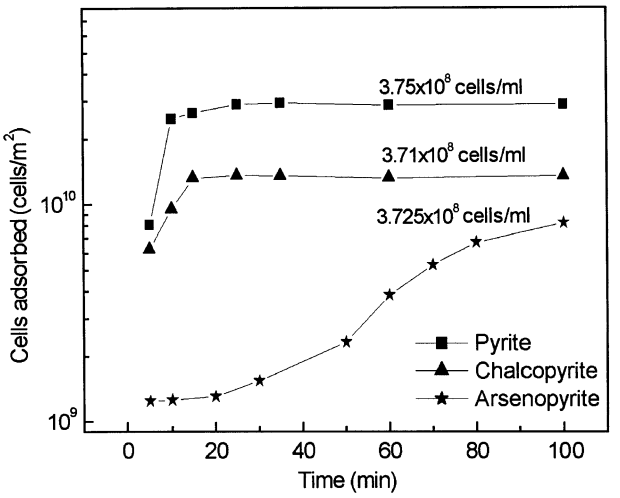

Fig. 2. Adsorption kinetics of *A. ferrooxidans* on pyrite, chalcopyrite and arsenopyrite.

studies revealed that the adsorption density of cells on the surface of pyrite was twice that on chalcopyrite (data not shown), confirming that the bacteria shows selectivity towards pyrite due to the presence of toxic copper ions in the lattice of chalcopyrite.

On the other hand, the kinetics with respect to arsenopyrite was very slow and even after 1 h incubation the decrease in the solution cell number was not comparable with that observed in presence of pyrite or chalcopyrite. The adsorption density on pyrite, chalcopyrite and arsenopyrite after 20 min incubation with 3.75×10^8 cells/ml of cells were 2.75×10^{10}, 1.36×10^{10} and 1.31×10^9 cells/m^2, respectively. Rapid oxidation of the active arsenopyrite mineral in acidic media results in the formation of an arsenic rich surface layer (Wang et al., 1992). At high pH values, arsenic in the form of insoluble arsenate is associated with the surface iron hydrooxide deposits. Presence of such toxic arsenic species on the surface of arsenopyrite hinders the adhesion kinetics and results in poor surface coverage of cells on it. Similar observation was reported by Fernandez et al. (1995). Adsorption isotherm studies revealed that the cell density on pyrite was almost four times more than that on arsenopyrite (data not shown). Attachment of iron-oxidizing *A. ferrooxidans* to pyrite is shown to be due to the specific binding protein apo-rusticyanin (Blake et al., 2001), which bound preferentially to pyrite relative to other iron-containing sulphides. This further explains the selectivity exhibited by the cells towards pyrite.

This observed difference in the adhesion behaviour of cells towards the minerals explains the changes observed in the electrokinetic behaviour of the minerals consequent to interaction with the cells. Poor adhesion of cells on the arsenopyrite surface resulted in insignificant changes in the isoelectric point of arsenopyrite consequent to interaction with cells as compared to pyrite and chalcopyrite.

3.3. Microflotation studies

Flotation behaviour of pyrite, chalcopyrite and arsenopyrite with 0.5 and 1 mM potassium isopropyl xanthate (PIPX) as collector is depicted in Fig. 3a. Flotability of pyrite and arsenopyrite was maximum in the acidic pH range of 4–6 and it reduced with increase in alkalinity. On the other hand, chalcopyrite exhibited good flotability both in the acidic and alkaline pH region. The recovery of pyrite, in the acidic pH range of 4–6, was approximately 85% and 98% with 0.5 and 1 mM PIPX collector, respectively. The recovery of arsenopyrite in the pH range 5–6, where the mineral exhibits maximum flotability, was approximately 62% and 80% with 0.5 and 1 mM PIPX collector, respectively. Recovery of chalcopyrite was nearly stable with pH and was found to be 95% with 0.5 mM and 98% with 1 mM PIPX collector.

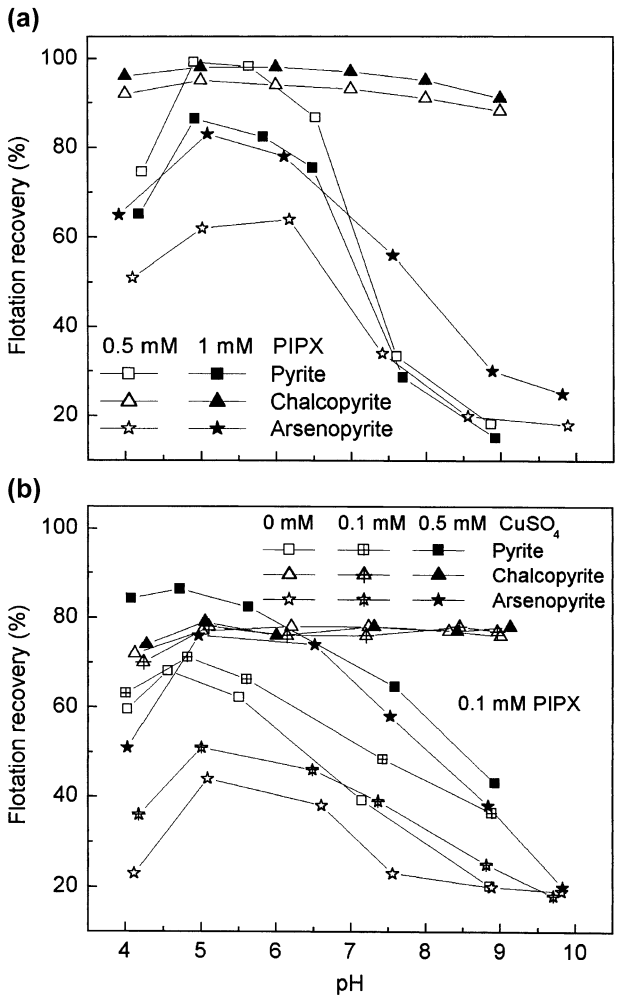

Fig. 3. (a) Flotation of pyrite, chalcopyrite and arsenopyrite at different collector (PIPX) concentration. (b) Effect of copper sulphate activator on xanthate-induced flotation of pyrite, chalcopyrite and arsenopyrite.

3.3.1. Effect of conditioning with copper sulphate

Arsenopyrite exhibited poor flotability with PIPX as collector and hence activation by copper ions becomes necessary to obtain better recovery. The effect of copper activation on the flotation recovery of all the minerals was tested and the results are depicted in Fig. 3b. In the acidic pH range of 5–6, the maximum recovery of arsenopyrite with 0.1 mM PIPX increased from 43% to 51% approximately when conditioned with 0.1 mM of copper sulphate solution. This was further enhanced to 76% when the concentration of copper sulphate was increased to 0.5 mM. Similar conditioning of pyrite mineral increased its maximum flotability from 67% to 71% with 0.1 mM copper sulphate, which further increased to 86% with 0.5 mM copper sulphate. Conditioning with copper ions had no effect on the flotability of chalcopyrite.

Recovery of arsenopyrite showed significant increase when conditioned with the activator. The activation effect was more prominent in the alkaline region and we observe that the activation due to copper ions renders good flotability to the mineral up to pH 8. Pyrite recovery was also improved but to a lesser extent when compared to that of arsenopyrite. As against this, chalcopyrite recovery was not affected much due to activator conditioning.

3.3.2. Effect of conditioning with cells

The effect of bacterial interaction on the flotation recovery of the minerals was studied. The results obtained are shown in Fig. 4 represented by curves with closed symbols. Upon conditioning with the bacterial cells for 5 min prior to collector conditioning, the recovery of pyrite declined to less than 25% even in presence of 0.5 mM PIPX collector. This confirms rapid and tenacious attachment of the bacterial cells on the pyrite surface, which hinders the adsorption of xanthate species on the surface, thereby reducing its flotability significantly.

Under similar conditions, chalcopyrite exhibited 65% flotability with 0.5 mM PIPX at all pH. It was earlier observed that the adsorption kinetics of cells on pyrite and chalcopyrite surfaces was similar, but the cell density on chalcopyrite surface was lower when compared to that on pyrite. The nature of the alkylxanthate species formed on the surfaces of pyrite and chalcopyrite are also shown to differ. While the dialkyl dixanthogen species formed on the pyrite surface is physisorbed on its surface, the alkylxanthate species are strongly chemisorbed at the copper sites as copper(I) alkylxanthate on the surface of chalcopyrite (Mats and Persson, 1994). Since the copper sites on the chalcopyrite surface remain unoccupied by cells, the xanthate collector is able to render sufficient flotability even in presence of cells. Thus, the minerals exhibited different flotability consequent to interaction with the cells and collector.

Recovery of arsenopyrite was not significantly affected due to conditioning with cells as observed for pyrite and chalcopyrite. In the pH range 5–6, the recovery with 0.5 mM PIPX, consequent to interaction with cells, was 50.5% approximately as against 62.2% obtained without interaction. As observed earlier adhesion kinetics of cells was slow on arsenopyrite when compared to other minerals resulting in poor surface coverage after 5 min interaction with cells. Effect of xanthate ions on the surface was thereby not significantly affected. However, since the recovery of arsenopyrite is poor activation by copper ions becomes necessity.

The effect of conditioning with copper sulphate prior to flotation with collector was studied under the above conditions and the results obtained are shown in Fig. 4 represented by curves with open symbols. Recovery of pyrite increased to 40% in the acidic pH region when conditioned with the activator. The copper ions occupying the vacant sites on the pyrite surface fix the xanthate firmly and induce some hydrophobicity. Suppression of pyrite was therefore not efficient in presence of copper ions in the system. Presence of copper ions had only marginal effect on the flotability of chalcopyrite. The copper ions, on the other hand, showed marked activation of the arsenopyrite surface even in presence of cells. When

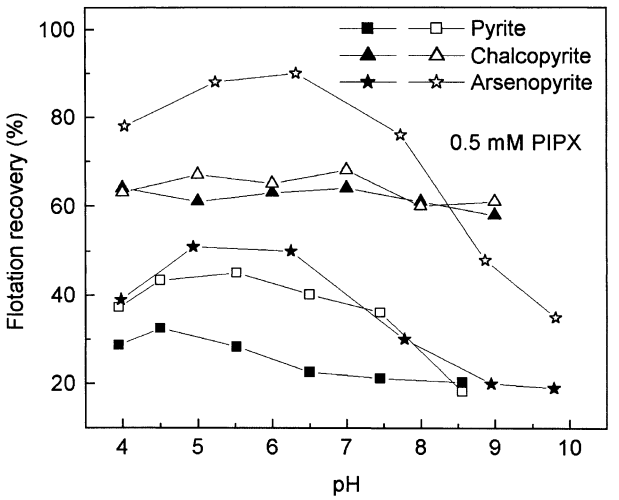

Fig. 4. Effect of cells on xanthate-induced flotation behaviour of pyrite, chalcopyrite and arsenopyrite in the absence (closed symbols) and presence (open symbols) of $CuSO_4$ activator.

conditioned with 0.5 mM copper sulphate prior to collector conditioning, the recovery obtained at pH 5.5 was 88.4% approximately as against 50.5% obtained without activator. Cu(II) can selectively adsorb onto the arsenic sites to form stable arsenides (Abeidu and Almahdy, 1980) namely Cu_3As and Cu_3As_2, which can fix xanthate firmly. Poor adhesion of bacterial cells and marked activation of the surface by the copper ions resulted in good flotability of arsenopyrite even in presence of cells.

3.4. Differential flotation of pyrite–chalcopyrite–arsenopyrite system

Microflotation studies suggest that biomodulation with *A. ferrooxidans* cells shows possibility of selective depression of pyrite from chalcopyrite and arsenopyrite. Results of the differential flotation studies of a 1:1:1 synthetic mixture of pyrite, chalcopyrite and arsenopyrite is tabulated in Table 1. Two conditions were tested. In the first case the minerals were interacted with the cells and/or reagents separately, and the interacted minerals were mixed together and floated. In the second case, the minerals were interacted together with the cells and/or collector and floated together.

When the minerals were conditioned separately and floated together, consequent to interaction with 4×10^8 cells/ml of bacterial cells and further conditioning with 0.5 mM collector, the recovery of pyrite, chalcopyrite and arsenopyrite in the floated fraction at pH 6.5 was about 25.1%, 63.2% and 52.3%, respectively. The recovery at the same pH when the minerals were conditioned together was 20.3%, 61.6% and 63.1% for pyrite, chalcopyrite and arsenopyrite, respectively. The recovery of pyrite, which was 25.1% when present alone, decreased to 20.3% when the minerals were conditioned together. The recovery of chalcopyrite also decreased, but to a lesser extent when compared to pyrite. Recovery of arsenopyrite, on the other hand, increased from 52.3% to 63.1%.The recovery of arsenopyrite at pH 4.5 enhanced from 56.3% to 69.8%, while the recovery of pyrite decreased from 24.4% to 19.3%. Studies by Mielczarski and Mielczarski (2003) on adsorption of xanthate on pyrite–chalcopyrite and pyrite–galena mixture revealed that galvanic contact inhibits xanthate adsorption on pyrite while the adsorption on chalcopyrite or galena significantly increased. This explains the observed increase in the recovery of arsenopyrite and the increased depression of pyrite.

Table 1
Differential flotation of pyrite–chalcopyrite–arsenopyrite (1:1:1) mixture after conditioning with cells followed by collector

Flotation pH	Experimental condition	Weight % in floated fraction		
		Pyrite	Chalcopyrite	Arsenopyrite
4.5	individually	24.4	65.4	56.3
	together	19.3	60.2	69.8
6.5	individually	25.1	63.2	52.3
	together	20.3	61.6	63.1

Conditions: PIPX: 0.5 mM (5 min). Cells (pH 4.5): 4×10^8 cells/ml (5 min).

Table 2
Effect of copper sulphate activation on the differential flotation of pyrite–chalcopyrite–arsenopyrite (1:1:1) mixture after conditioning with cells followed by collector

Flotation pH	Experimental condition	Weight % in floated fraction		
		Pyrite	Chalcopyrite	Arsenopyrite
4.5	individually	43.2	65.4	85.1
	together	18.6	60.5	81.3
6.5	individually	44.3	64.2	88.7
	together	23.1	62.4	84.2

Conditions: cells (pH 4.5): 4×10^8 cells/ml (5 min). $CuSO_4$: 0.5 mM (5 min), PIPX: 0.5 mM (5 min).

To further improve the separation, effect of copper sulphate conditioning was studied. The results obtained with 0.5 mM copper sulphate as activator are summarized in Table 2. The conditions maintained were similar to earlier experiment (Table 1). The recovery of pyrite, chalcopyrite and arsenopyrite at pH 6.5 was 23.1%, 61.4% and 84.2%, respectively. The results obtained show that the activation of pyrite by copper sulphate observed earlier (Fig. 4) was suppressed in the presence of more active arsenopyrite mineral. Copper ions would selectively migrate to the surface of arsenopyrite, which has a lower rest potential than pyrite/chalcopyrite. Preferential adsorption of xanthate on the active copper-activated arsenopyrite surface further reduces the flotability of pyrite. The flotability of chalcopyrite was only marginally affected by copper ions. Thus, although good flotation of arsenopyrite was obtained by copper activation, there was no marked improvement in the flotability of chalcopyrite.

In our earlier studies (Chandraprabha et al., 2004) on pyrite–chalcopyrite system, we demonstrated that good separation of pyrite and chalcopyrite was achieved by initially conditioning the minerals with the collector followed by interaction with bacterial cells. The cells were shown to depress collector interacted pyrite effectively while having no effect on chalcopyrite flotability. Similar protocol was implemented here to improve the recovery of chalcopyrite. The effect of such conditioning on the flotability of the three minerals was studied. The results obtained with 1:1:1 mixture of pyrite–chalcopyrite–arsenopyrite conditioned simultaneously are summarized in Table 3. The recoveries of the three minerals at pH 6.5 were 25.8% pyrite, 86.6% chalcopyrite and 55.4% arsenopyrite, respectively. When conditioned with copper sulphate prior to collector conditioning, the recoveries obtained were 21.2% pyrite, 84.3% chalcopyrite and 80.2% arsenopyrite, respectively. Similar separation was obtained at pH 4.5. Thus, in presence of the activator, effective separation of pyrite from chalcopyrite and arsenopyrite was achieved.

The beneficial aspects of biomodulation in promoting differential flotation of complex sulfides are evident from the results of this work. Compared to the use of organic collectors along with chemical-based activators and depressants, biomodulation can be expected to be beneficial both from economical as well as environmental view points. Only starvation amounts of any chemical flotation reagents would suffice (if at all) in biomodulation, thus effecting significant savings in the costs of flotation chemicals. The microorganisms used in the biomodulation process are indigenously present in the ore deposits and thus can be cultured easily without any additional costs. Further, the use of such environment-friendly and energy-efficient bacteria would lead to effective replacement of toxic and costly cyanide reagents.

Table 3
Differential flotation of pyrite–chalcopyrite–arsenopyrite (1:1:1) mixture after conditioning PIPX collector with followed by conditioning with cells

Flotation pH	Experimental condition	Weight % in floated fraction		
		Pyrite	Chalcopyrite	Arsenopyrite
4.5	without $CuSO_4$	22.7	83.4	58.1
	with $CuSO_4$	19.2	86.3	82.1
6.5	without $CuSO_4$	20.8	86.6	55.4
	with $CuSO_4$	21.2	84.3	80.2

Conditions: $CuSO_4$: 0.5 mM (5 min), PIPX: 0.5 mM (5 min). Cells (pH 4.5): 4×10^8 cells/ml (5 min).

4. Conclusions

The following major conclusions can be drawn from the present studies

1. Surface chemical studies revealed that the change in the surface character of the minerals consequent to interaction with *A. ferrooxidans* cells was most significant for pyrite followed by chalcopyrite. The effect on arsenopyrite was negligible in comparison to the other two minerals.
2. The observed electrokinetic behaviour of the minerals consequent to interaction with bacterial cells was confirmed by the adhesion studies, which revealed that the bacterial cells attached preferentially to pyrite surface compared to chalcopyrite and arsenopyrite surfaces. The kinetics of cell attachment and the surface coverage was very poor for arsenopyrite.
3. The selective preference of *A. ferrooxidans* for pyrite surface resulted in its depression even in presence of xanthate collector. The collector, on the other hand, was able to render good flotability to chalcopyrite surface even in presence of cells. Poor attachment kinetics and surface coverage of bacterial cells and preferential activation of the more active arsenopyrite surface by the copper sulphate activator resulted in good floatability of the mineral in presence of cells.
4. Conditioning of activator and collector interacted minerals with the bacterial cells yielded best separation of pyrite from chalcopyrite and arsenopyrite.

Acknowledgement

The authors gratefully acknowledge the financial support from the Department of Science and Technology (DST), India and National Science Foundation

(NSF), USA provided under the DST-NSF (INT-01-17627) collaborative research program.

References

Abeidu, A.M., Almahdy, A.M., 1980. Magnesia mixture as a regulator in the separation of pyrite from chalcopyrite and arsenopyrite. Int. J. Miner. Process. 6, 285–302.

Blake II, R.C., Sasaki, K., Ohmura, N., 2001. Does aporusticyanin mediate the adhesion of *Thiobacillus ferrooxidans* to pyrite? Hydrometallurgy 59, 357–372.

Chandraprabha, M.N., Natarajan, K.A., Modak, J.M., 2004. Selective separation of pyrite from chalcopyrite by biomodulation. Colloids Surf B Biointerfaces 37 (3-4), 93–100.

Deo, N., Natarajan, K.A., 1998. Studies on interaction of *Paenibacillus polymyxa* with iron ore minerals in relation to beneficiation. Int. J. Miner. Process. 55, 41–60.

Devasia, P., Natarajan, K.A., Satyanarayana, D.N., Ramananda Rao, G., 1993. Surface chemistry of *Thiobacillus ferrooxidans* relevant to adhesion on mineral surfaces. Appl. Environ. Microbiol. 59, 4051–4055.

Fernandez, M.G.M., Mustin, C., Donato, P., Barres, O., Marion, P., Berthelin, J., 1995. Occurrences at mineral–bacteria interface during oxidation of arsenopyrite by *Thiobacillus ferrooxidans*. Biotechnol. Bioeng. 46, 13–21.

Fuerstenau, D.W., Metzger, P.H., Seele, G.D., 1957. Modified Hallimond tube for flotation testing. Eng. Min. J. 158, 93–95.

Karavaiko, G.I., 1988. Methods of isolation, evaluation and studying of microorganisms. In: Karavaiko, G.I., Rossi, G., Agate, A.D., Groudev, S.N., Avakyan, Z.A. (Eds.), Biotechnology of Metals—Manual. Center for International Projects GKNT, Moscow, pp. 47–79.

Mats, V., Persson, I., 1994. Interactions between sulphide minerals and alkylxanthates. Colloids Surf., A Physicochem. Eng. Asp. 83, 207–217.

Mielczarski, E., Mielczarski, J.A., 2003. Influence of galvanic effect on adsorption of xanthate on pyrite, galena and chalcopyrite. In: Lorenzen, L., Bradshaw, D.J. (Eds.), Proceedings of XXII International Mineral Processing Congress. Cape Town, South Africa, pp. 866–873.

Patra, P., Natarajan, K.A., 2003. Microbially-induced flocculation and flotation for pyrite separation from oxide gangue minerals. Min. Eng. 16, 773–965.

Randol, 1992. Permanganate depression of FeAsS. In Innovations in gold and silver recovery, Phase IV 5. Randol International, pp. 2502. Chap. 17.

Santhiya, D., Subramanian, S., Natarajan, K.A., 2000. Surface chemical studies on galena and sphalerite in the presence of *Thiobacillus thiooxidans* with reference to mineral beneficiation. Min. Eng. 13, 747–763.

Sharma, P.K., Das, A., Hanumantha Rao, K., Forssberg, K.S.E., 1999. *Thiobacillus ferrooxidans* interaction with sulfide minerals and selective chalcopyrite flotation from pyrite. In: Parekh, B.K., Miller, J.D. (Eds.), Advances in Flotation Technology. Society for Mining Metallurgy and Exploration, ISBN: 0873351843, pp. 147–165.

Silverman, M.P., Lundgren, D.G., 1959. Studies on chemoautotrophic iron bacterium *Thiobacillus ferrooxidans*. J. Bacteriol. 77, 642–646.

Yelloji Rao, M.K., Natarajan, K.A., Somasundaran, P., 1992. Effect of biotreatment with *Thiobacillus ferrooxidans* on the floatability of sphalerite and galena. Miner. Metall. Process. 9, 95–100.

Yen, W.T., Tajadod, J., 1999. Selective depression of enargite and chalcopyrite. Proceedings of XXV International Mineral Processing Congress, Rome, Italy, pp. 49–55.

Wang, X.H., Ahlberg, E., Forssberg, K.S.E., 1992. Electrochemical study of surface oxidation and collectorless flotation of arsenopyrite. J. Appl. Electrochem. 22, 1095–1103.