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Coal flotation using a biosurfactant from *Pseudomonas aeruginosa* as a frother

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Abstract−A biosurfactant-producing bacterial species (*Pseudomonas aeruginosa*) was grown in a mineral solution with gas oil as the source of carbon and energy. The biosurfactant was recovered from the solution by collecting the foam on the surface and drying. It had critical micelle concentration of 100 ppm. Froth characterization showed that the biosurfactant was superior to methyl isobutyl carbinol (MIBC) in terms of froth height and stability. The biosurfactant was examined in coal flotation as a frother. The combustible matter recovery of 72-79% with 10-15.5% ash content supporting 55-57.5% separation efficiency seemed promising enough to introduce the biosurfactant from *Pseudomonas aeruginosa* as a new frother.

Key words: Flotation, Coal, *Pseudomonas aeruginosa*, Rhamnolipid Biosurfactant, Frother

INTRODUCTION

Application of microorganisms in flotation has been under investigation in recent years. Microorganisms can act as surface modifiers, depressants, collectors, or dispersing agents. In bioflotation, microorganisms adhere to the surface of an ore and change its surface characteristics. The adhesion may be passive, and due to abiotic interaction of microbial cell wall and ore surface [1], or it may be accompanied by active oxidation/reduction of ore elements by bacteria [2].

Different microbial species have been used in bioflotation. Among others, the following species have been more frequently used: *Thiobacillus ferrooxidans* and *Acidithiobacillus ferrooxidans* as pyrite depressants in flotation of sulfide minerals [3-9], *E. coli* as a collector of quartz [1], *Staphylococcus carnosus* and *Bacillus firmus* as collectors of apatite [10], *Paenibacillus polymyxa* as surface modifier in iron ore flotation [11], *Mycobacterium phlei* as collector for hematite [12], *Bacillus subtilis* as dolimite depressant [13], and *Rhodococcus opacus* as collector of hematite in hematite-quartz flotation [14].

Microbial products such as biosurfactants, organic acids, and organic solvents can also be applied to mineral flotation. These biochemicals can be produced and recovered as concentrated forms in bioreactors in large amounts. They can replace conventional chemicals, and make the flotation more economical, and safer regarding environmental concerns. Although there is rather a large body of literature on the direct use of microorganisms, less attention has been paid to the use of microbial products in flotation. In this research we report the production of a biosurfactant in concentrated form from a bacterial species named *Pseudomonas aeruginosa*, the biosurfactant frothing characterization, and its application to coal flotation as a new frother.

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MATERIALS AND METHODS

1. Coal

A coal sample was obtained from Zarand coal washing plant. The coal sample was classified in two parts based on particle size: −300 μm (with the ash content of 33.81%), and +300 μm (with the ash content of 21.94%, the largest particles were about 500 μm in size). The samples were stored in nylon bags in a cool place.

2. Microorganism and Biosurfactant Production Procedure

A strain of *Pseudomonas aeruginosa* was obtained from the laboratory of biotechnology at Shahid Bahonar University of Kerman. The species had been previously reported to be able to produce a glycolipid type biosurfactant (rhamnolipid) [15]. A mineral medium with the following composition was used for the bacterial growth and biosurfactant production: KH_2PO_4, 3.4 g/L; K_2HPO_4, 4.3 g/L; $(NH_4)_2SO_4$, 4 g/L; $MgCl_2 \cdot 7H_2O$, 0.2 g/L; $CaCl_2 \cdot 2H_2O$, 0.04 g/L; $FeSO_4$, 0.03 g/L. The medium was supplemented with a trace element solution having the following composition: $MnCl_2$, 0.04 g/L; $NaMoO_4$, 0.08 g/L; $CuSO_4$, 0.006 g/L; H_3BO_3, 0.013 g/L; $ZnSO_4$, 0.06 g/L. Each liter of the final mineral medium for growth contained 25 mL of the trace element solution. All of the minerals were of analytical grade. To produce biosurfactant, 4 mL gas oil was added to one liter of the mineral medium to serve as the source of carbon and energy for the bacterial species (gas oil was used as an insoluble carbon source for the bacterium to stimulate biosurfactant production). The medium was aerated in a bioreactor at 30 °C for four days. During aeration, excess gas oil was added to the medium to compensate for gas oil evaporation. Intensive foaming was observed in the bioreactor, which was an indication for biosurfactant production. At the end of fermentation, the whole fermentation broth was centrifuged at 3,000 rpm for 15 minutes. The microbial pellet was discarded, and the layer of foam formed on the surface of the broth was separated by using a funnel. The foam was dried to obtain crude biosurfactant. Each liter of the fermentation medium gave about 3 grams crude biosurfactant. The material was stored at 4 °C for

later experiments.

3. Surface Tension Measurement

The surface tension was measured by using the ring method. The measurement was conducted under ambient conditions with a Kruss tensiometer (Krüss, Optische-Mechanische Werkstatten, Germany) equipped with a platinum ring. The volume of the samples was 10 mL. Each measurement was replicated three times and an averaged value was reported.

4. Froth Characterization

The experiment for froth characterization was performed in a glass cylinder with the height of 40 cm and the inner diameter of 3 cm. An 85 mesh ceramic air sparger was at the bottom of the column. To determine frothability, the variation in froth height was measured as a function of air flow rate; and to determine froth stability, froth half life (the time after terminating aeration over which the equilibrium height of froth decreases to half) was measured. Froth characteristics of the biosurfactant were compared with those of methyl isobutyl carbinol (MIBC) as a conventional frother.

5. Coal Flotation Tests

The flotation experiments were done with the biosurfactant as a frother. The effects of particle size, biosurfactant concentration, collector (gas oil) concentration, and solid percentage on the percentage recovery of combustible matter, ash content, and separation efficiency were investigated. A one-half fraction of 2^4 factorial design (4 factors each at 2 levels) was used for the experiments. Table 1 shows the factors and their levels. Flotation tests were carried out in a Denver cell of 1 L capacity. Tap water (pH 7) was used throughout the tests. The impeller speed of the flotation chamber was set at 1,000 rpm for all tests. The aeration rate was 3.2 L min^{-1}. The temperature was 25±1 °C. Each sample was conditioned for 5 min. In all tests, the collector was added before biosurfactant. The conditioning period for the collector was 2 min, while it was 1 min for the biosurfactant. Flotation was continued for 6 min. Samples were collected during this time period (at 30, 60, 90, 150, 210, and 360 s) to have an insight into the kinetics of the flotation. The samples were filtered, dried, weighed, and analyzed for ash content, to determine the recovery percentage of combustible matter and separation efficiency. The software MINITAB 14.1 was used to analyze the flotation results. The data at each time were considered as a replicate (time served as a blocking factor) in statistical analysis of the effects of the factors on responses (percentage recovery of combustible matter, ash content, and separation efficiency).

Table 1. Factors and their levels used in the flotation experiments (187 g coal was employed in experiments with 14% solid content. For the experiments with 7% solid content, the figure was 93 g)

Run no.	Particle size (μm) (A)	Solid percentage (B)	Biosurfactant conc. (ppm) (C)	Collector conc. (kg/t) (D)
1	−300	14	90	2
2	−300	14	50	4
3	−300	7	90	4
4	−300	7	50	2
5	+300	14	50	2
6	+300	7	90	2
7	+300	7	50	4
8	+300	14	90	4

RESULTS AND DISCUSSIONS

1. Froth Characterization of the Biosurfactant

Different concentrations of the biosurfactant in distilled water were prepared, and the surface tension for each sample was measured. The same was done for MIBC. Fig. 1 shows the results. The results indicate the better ability of the biosurfactant to reduce surface tension of water compared to MIBC. Both of the surfactants have a critical micelle concentration of about 100 ppm.

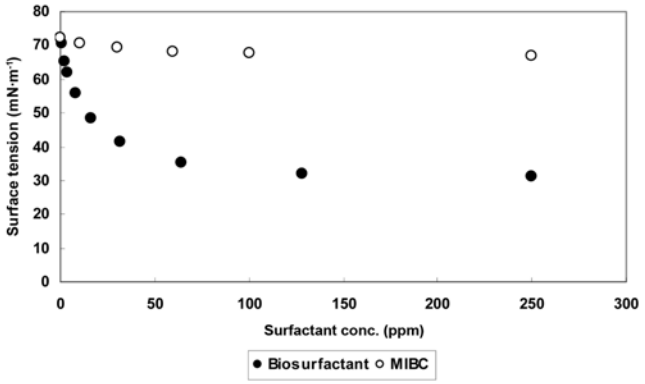

Fig. 1. Comparison between the biosurfactant and MIBC in reducing surface tension of distilled water.

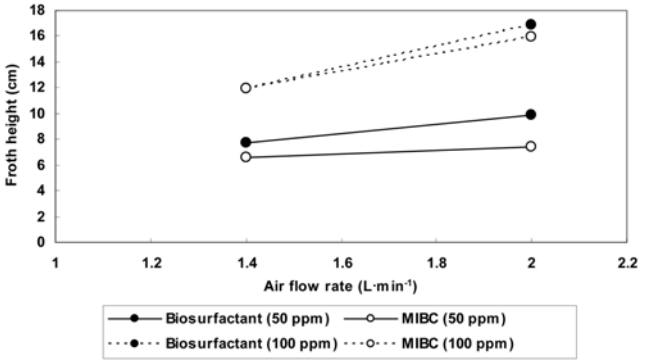

Fig. 2. Froth height as a function of air flow rate.

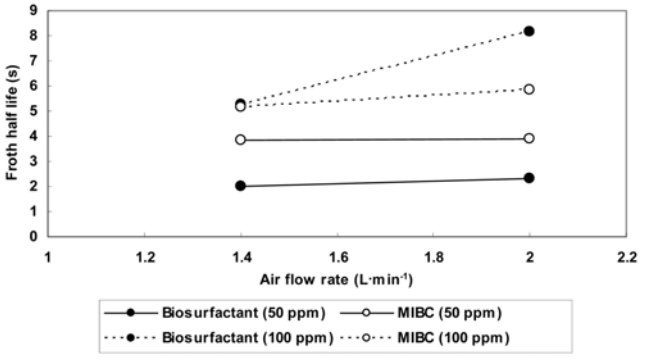

Fig. 3. Froth half life as a function of air flow rate.

Frothers, which are usually non-ionic heteropolar molecules, are added to create a reasonably stable froth. The polar end of a frother molecule forms hydrogen bonds with water. The non-polar end concentrates at the air/water interface. This phenomenon alters the surface tension of water. Changing the surface tension of water is a measure of the surface activity of surfactants. In general, an increase in surface activity results in increased frothability and froth stability [16]. Surface tension measurements showed that at equal concentrations, the biosurfactant had higher surface activity. So the biosurfactant may be expected to show better frothability compared to MIBC.

Since the ability to reduce the surface tension of water is not the only parameter for the evaluation of the frothability of a surfactant, the height and stability of the froths produced by the biosurfactant were compared with those of MIBC. For this purpose the height of froth was measured as a function of aeration rate at two different surfactant concentrations. Fig. 2 shows the results. A statistical paired t-test showed a significant difference between froth heights pro-

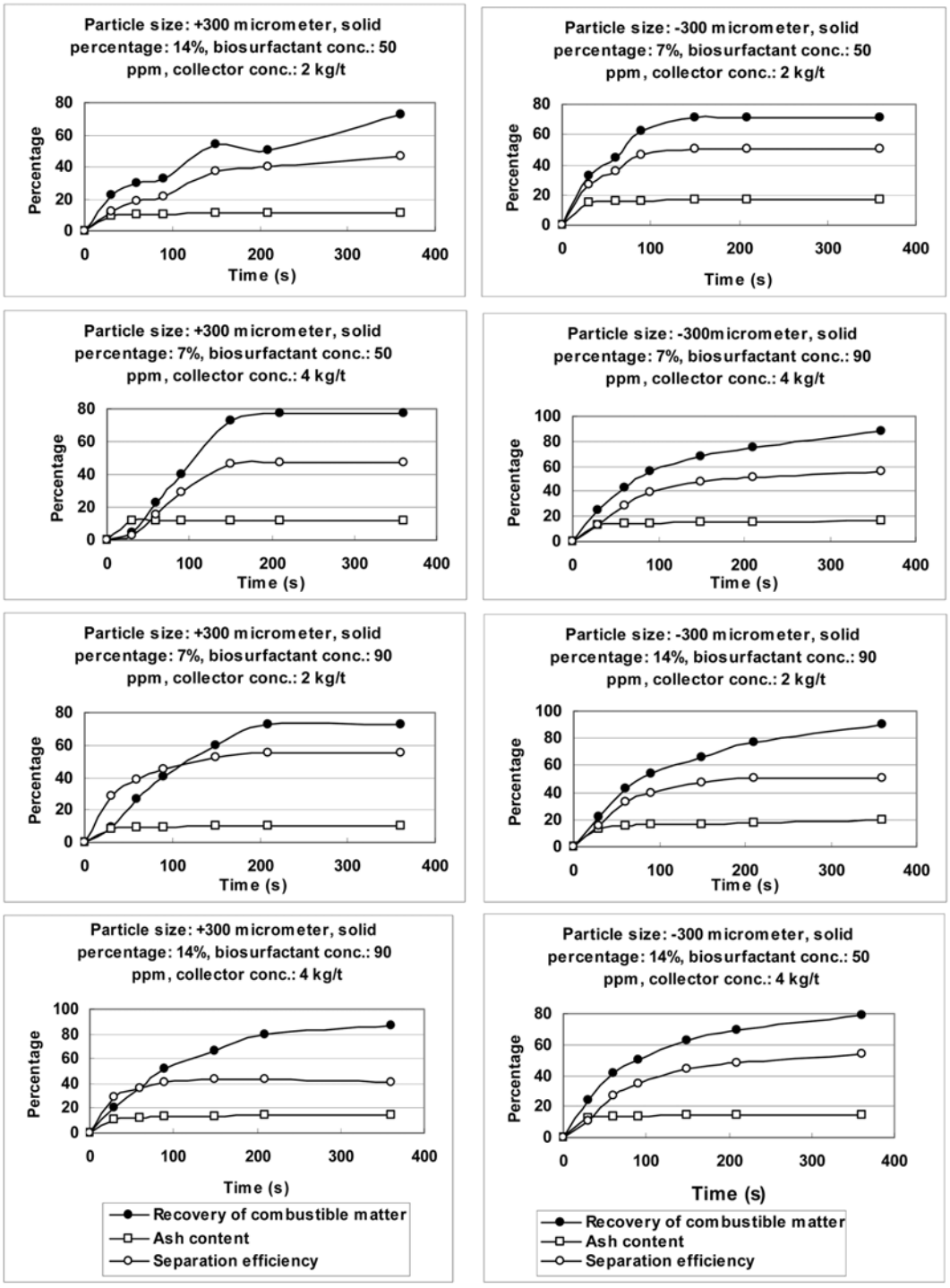

Fig. 4. Results of flotation under different combination of factor levels as a function of time.

duced by the surfactants [17]. The biosurfactant produced higher froth heights compared to MIBC. The froth stability was also measured for both surfactants. Froth half life was considered as the measure of froth stability. Fig. 3 shows the results. The results indicate that at the lower concentration MIBC produces more stable froth, while at higher concentration the reverse is true. It seems that at higher concentration the interfacial adsorption density of biosurfactant is more than MIBC.

Overall, the froth characterization tests showed that the biosurfactant can compete with MIBC which is a common surfactant in flotation processes. *Pseudomonas aeruginosa* produces a biosurfactant named rhamnoloipid. The rhamnolipid molecules consist of one or two molecules of rhamnose, which is a methyl pentose sugar, linked to one or two molecules of β-hydroxy-decanoeic acid. Various carbon sources can be used by the microorganism to synthesize the biosurfactant. This biosurfactant can reduce the surface tension of water as low as 25 mN/m (Fig. 1 shows that the crude biosurfactant which was produced here could reduce surface tension of water to about 31 mN/m) [18]. Since this material has a biological origin, it is most likely to be biodegradable. Having good frothability and biodegradability makes this biosurfactant a promising frother in flotation.

3. Coal Flotation using the Biosurfactant

The application of the biosurfactant from *Pseudomonas aeruginosa* to coal flotation was examined as a case study. Table 1 shows the experimental design for the flotation. Fig. 4 shows the results of the flotation experiments. The results indicate successful application of the biosurfactant to coal flotation. Fig. 5 shows the standardized effects [17] of the independent factors (particle size, solid percentage, the biosurfactant concentration, and the collector concentration) on the percentage recovery of combustible matter. The figure indicates a significant positive effect of the biosurfactant concentration on the percentage recovery of combustible matter. Fig. 6 shows the standardized effects of the independent factors on separation efficiency of flotation. The figure indicates that the increase in particle size and solid content has negative impacts on separation efficiency, while the increase in biosurfactant concentration has a positive effect, and the effect of the collector concentration is not significant. Similarly, the standardized effects of the independent factors on the ash content of the product can be observed in Fig. 7. In summary, under the condition tested, the best results were 72-79% combustible matter recovery with 10-15.5% ash content supporting 55-57.5% separation efficiency. Using these preliminary results and an optimization program, it is possible to obtain better flotation results.

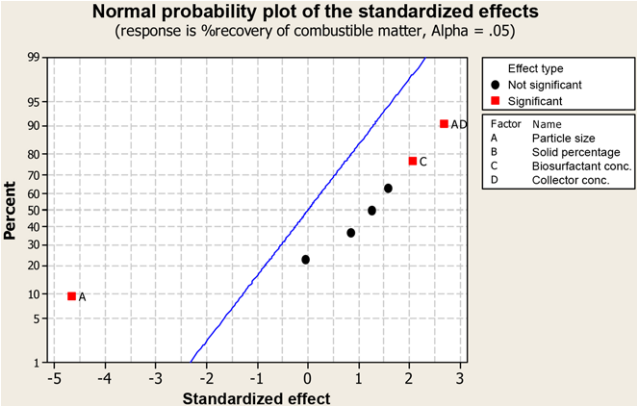

Fig. 5. Standardized effects of the factors on the recovery percentage of combustible matter.

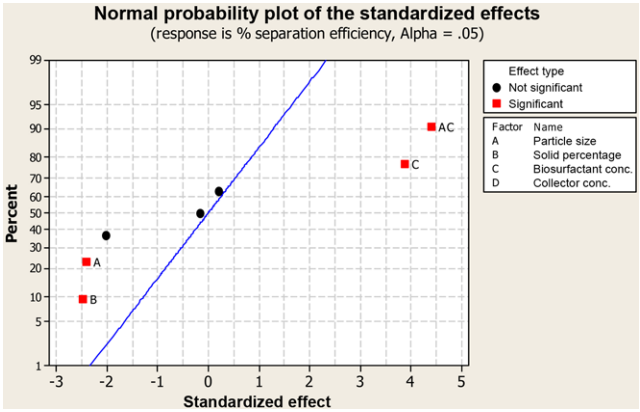

Fig. 6. Standardized effects of the factors on the separation efficiency of coal flotation.

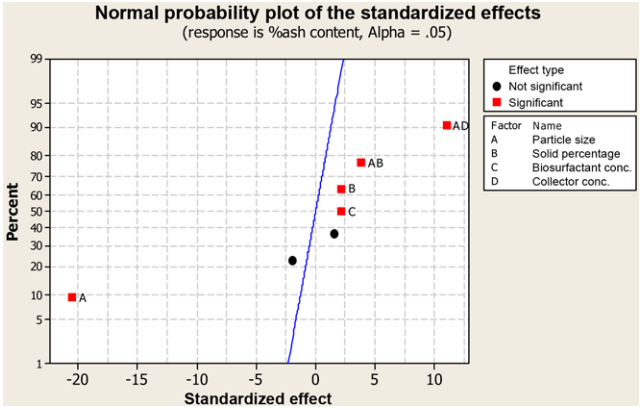

Fig. 7. Standardized effects of the factors on the ash content of the product.

CONCLUSION

Pseudomonas aeruginosa, which is a bacterial species, can be grown in a mineral medium with gas oil as the sole source of carbon and energy. The biosurfactant produced by the bacterium can be separated from the fermentation medium by foam separation and drying. The biosurfactant shows good frothability in terms of reducing surface tension of water, froth height, and froth stability. The case study on coal flotation showed that the biosurfactant could be successfully applied to coal flotation. To our knowledge this is the first report on the characterization and application of the biosurfactant from *Pseudomonas aeruginosa* as a frother in flotation.

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