

Shahrood University of  
Technology

*Journal of Mining and Environment (JME)*

Journal homepage: [www.jme.shahroodut.ac.ir](http://www.jme.shahroodut.ac.ir)



Iranian Society of  
Mining Engineering  
(IRSME)

## Bioflotation and Bioleaching as an Alternative Method for Desulphurization and Ash Reduction in Tabas Coal

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### Article Info

Received 14 May 2023

Received in Revised form 9 July 2023

Accepted 7 October 2023

Published online 7 October 2023

DOI: [10.22044/jme.2023.13127.2387](https://doi.org/10.22044/jme.2023.13127.2387)

### Keywords

Tabas coal  
Coal desulfurization  
Biodesulfurization  
Bioflotation  
Ash removal  
Bioleaching

### Abstract

Tabas coal possesses favorable plastometric properties that make it suitable for use in metallurgical industries as coking coal. However, its high sulfur content, which stands at approximately 2%, poses a significant environmental pollution risk. Additionally, reducing ash content to below 10% is a critical objective of this study to prevent a decline in coal's thermal efficiency in the metallurgical industries. This research work investigates the removal of sulfur and ash from Tabas coal samples using the biological methods including bioflotation and bioleaching. Initially, a combination of mesophilic bacteria including *Acidithiobacillus ferrooxidans*, *Acidithiobacillus thiooxidans*, and *Leptosprillum ferrooxidans* were employed in the bioflotation method to detain pyrite sulfur in the Tabas coal samples. The highest reduction percentages of pyrite sulfur and ash were equal to 62% and 54.18%, respectively. In the next stage, bioleaching experiments were conducted, the effect of the test time, percentage of bacteria by volume, percentage of coal solids, and absence of bacteria on the amount of sulfur and ash removal was investigated. The test time emerged as the most critical factor. The best sulfur removal was achieved using bioleaching, with a maximum removal of 72.43%, observed for the PE coal sample. Bioflotation also achieved significant sulfur removal, with a maximum removal of 61% observed for the same sample. On the other hand, the best ash removal was achieved using bioflotation, with a maximum removal of 68.98% observed for the PE coal sample, and a maximum removal of 69.34% observed for the B4B2 coal sample using bioleaching. Finally, this research work conducted a comparison of biological methods to determine the amount of sulfur and ash reduction achieved. The results showed that both bioleaching and bioflotation were effective for coal desulfurization and ash removal, with bioleaching performing slightly better for sulfur removal and bioflotation performing slightly better for ash removal.

## 1. Introduction

As one of the most abundant fossil fuels on the planet, coal has played a significant role in powering industries and communities for centuries. However, the environmental impact of coal consumption cannot be ignored, especially when it comes to the emission of sulfur dioxide [1]. Sulfur exists in coal as four forms: inorganic sulfur, organic sulfur, sulfate, and elemental sulfur. Pyrite sulfur is the most abundant form of mineral sulfur found in coal [2, 3]. Organic sulfur is bound to the

structural components of coal by strong covalent bonds, making it difficult to separate from coal using traditional processing methods. Therefore, effective methods are necessary to remove organic sulfur from coal [4]. The second most significant impurity in coal is ash, which can cause thermal loss and damage in thermal furnaces. In industries, the amount of ash in coal should generally be below 10%. If it exceeds this amount, processing methods can be used to separate ash from coal [5].

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Tabas region in Iran has substantial coal reserves with low ash content, making Tabas coal ideal for use as coking coal in metallurgy. However, their high sulfur content prevents their profitable utilization. Therefore, extensive research is being conducted to find suitable desulfurization methods or combinations of methods to reduce the sulfur content of these coals.

Jorjani *et al.* (2004) investigated the application of a combination of microwave irradiation and peroxyacetic acid washing in a batch reactor to reduce sulfur content [6]. Another study investigated the application of various reagents including  $\text{Fe}_2(\text{SO}_4)_3$ ,  $\text{FeCl}_3$ ,  $\text{NaOH}$ ,  $\text{CH}_3\text{OH}$ ,  $\text{HNO}_3$ , and  $\text{H}_2\text{O}_2$  to remove sulfur and ash from Tabas coal in a batch reactor [7]. Alam *et al.* (2008) conducted a study on Tabas coal and achieved successful reduction of its ash and total sulfur content through froth flotation and leaching with nitric acid. Nitric acid was found to be more effective than  $\text{HCl}$  in the leaching process, and the Taguchi orthogonal experiment was used to optimize the experimental parameters [8]. However, chemical desulfurization of coal is not regarded as a cost-effective and environmentally friendly method [9]. Although chemical desulfurization techniques are commonly considered the most efficient way to eliminate both organic and inorganic sulfur, they are intricate, expensive, and necessitate elevated temperatures [10].

Alternatively, biological desulfurization is considered to be more environmentally friendly, with easy installation, low energy consumption, and high attractiveness for removing sulfur from coal [11]. Biological desulfurization methods such as bioflotation and bioleaching rely on the use of microorganisms and their extracellular polymeric substances (EPS) to remove sulfur from coal [11]. The bioflotation process uses microorganisms and bacteria instead of some of the chemicals used in the flotation process. Bioleaching, on the other hand, involves the use of microorganisms to break down the sulfur compounds in coal and convert them into soluble forms [12, 13]. Therefore, considering both environmental concerns and investment costs, the biological method was considered as the most appropriate method for removing sulfur coal when compared to other methods. Misra *et al.* (1996) investigated *Acidithiobacillus ferrooxidans* as a potential pyrite depressant for coal desulfurization, marking the first time this microorganism had been utilized for this purpose [14]. The study found that the floatability of pyrite using sodium isopropyl

xanthate (PIX) collector was significantly reduced from over 90% to less than 45% after bacterial treatment, achieved by increasing the pH from 1 to 7. Vijayalakshmi *et al.* (2002) conducted a study on the use of *P. polymyxa* to remove ash from coal samples [15]. In 2009, Amini *et al.* conducted a study on the impact of *Acidithiobacillus ferrooxidans* on pyrite depression during coal flotation, and compared the findings with those obtained using sodium cyanide [16]. A study conducted on biodesulfurization of Tabas coal in a pilot plant investigated the effect of particle size and pulp density on coal biodesulfurization [17]. Another study investigated biodesulfurization of high sulfur content coal concentrate from Tabas coal preparation plant. Golshani *et al.* (2013) used a mixed culture of mesophilic microorganisms, and evaluated the effects of pH, particle size, iron sulfate concentration, pulp density, and bioleaching time on sulfur reduction [18]. Etemadifar *et al.* (2017) used heterotrophic microorganisms in removal of sulfur from coal using growing and resting cells of *Rhodococcus erythropolis* strains. Xu *et al.* (2020) investigated the effects of various factors on the biodesulfurization of high sulfur coal from Shanxi using *Thiobacillus ferrooxidans*, *Escherichia coli*, and *Pseudomonas putida*. The results showed that *Pseudomonas putida* was the dominant strain, and achieved the highest sulfur removal [19].

In this study, the potential of biological methods (bioflotation and bioleaching) for removing sulfur and ash from Tabas coal samples was investigated. The study employs mesophilic bacteria including *Acidithiobacillus ferrooxidans*, *Acidithiobacillus thiooxidans*, and *Leptospirillum ferrooxidans* in the biodesulfurization of the Tabas coal samples. The use of mesophilic bacteria in the bioleaching and bioflotation of coal samples is an innovative approach to the removal of pyrite sulfur and ash. Mesophilic bacteria are active at moderate temperatures and acidic pH levels, making them ideal for this context. The bioleaching experiments were conducted to investigate the impact of various factors such as test time, percentage of bacteria by volume, percentage of coal solids, and the absence of bacteria on the removal of sulfur and ash. The study shows that biological methods can play a significant role in reducing the environmental impact of coal consumption and promoting sustainable approaches in the coal industry.

## 2. Materials and Methods

### 2.1. Coal sample preparation

Coal samples from Parvadeh 4 and Eastern Parvadeh (PE) layers in Tabas coal mines were prepared for the bioflotation and bioleaching experiments. The experiments were conducted on the primary feed sample of the Eastern Parvadeh (PE), the primary feed sample of the 4-layer C<sub>1</sub> (B<sub>4</sub>C<sub>1</sub>), and the 4-layer B<sub>2</sub> (B<sub>4</sub>B<sub>2</sub>) sample, which were crushed to dimensions of +180 microns and -350 microns. To conduct the tests, the amount of sulfur (organic, pyrite, and sulfate) was analyzed

based on the ASTM-D3177 standard analysis, and the amount of ash was analyzed based on the ASTM-D3174 standard, as shown in Table 1. The amount of sulfated sulfur in each coal sample was negligible, and therefore not taken into account. SEM images and polarizing light microscope images were taken from the coal samples before conducting experiments to obtain a preliminary understanding of the forms of pyrite and coal macerals and to investigate their effect in reducing sulfur and ash from coal. The SEM images of coal samples are shown in Figure 1, and the polarized light microscope images are shown in Figure 2.

**Table 1. Analysis of sulfur and ash content of Tabas coal samples.**

Samples	Total sulfur (%)	Pyritic sulfur (%)	Organic sulfur (%)	Ash (%)
PE	1.85	1	0.85	11.8
B <sub>4</sub> C <sub>1</sub>	2.1	1.51	0.59	15.65
B <sub>4</sub> B <sub>2</sub>	2.07	1.17	0.9	25.9

The coal used for the bioleaching experiments was prepared from two layers of Parvadeh 4 and Eastern Parvadeh (PE) from Tabas coal mines for chemical and bioleaching tests. For the sample of Eastern Parvadeh (PE), experiments were conducted on the primary feed of the coal, which was crushed to dimensions of +180 microns and -350 microns, as well as on the flotation concentrate of the product with dimensions of +180 microns. The sample of the primary feed and its flotation concentrate are referred to as PE and PE-C, respectively. For the Parvadeh 4 coal sample, flotation tests were carried out separately for the C<sub>1</sub> and B<sub>2</sub> layers on the 0.5 mm granulation range of the primary feed. Additionally, heavy liquid tests were conducted on the granulation range of +0.5 mm of the primary feed for each of the layers C<sub>1</sub> and B<sub>2</sub>. The flotation concentrate of -0.5 mm and heavy liquid concentrate of +0.5 for each of the layers were then mixed and crushed into two fractions (+180 to -350 microns) using a rod mill, yielding two fractions known as B<sub>4</sub>C<sub>1</sub> and B<sub>4</sub>B<sub>2</sub>, respectively.

### 2.2. Microorganisms, culture media, and analytical methods

Bacterial cultures were obtained from the R&D Center of the Sarcheshme Copper Complex (Sarcheshme, Kerman Province, Iran). The mixed cultures consisted of mesophilic bacteria including *A. ferrooxidans*, *Leptospirillum ferrooxidans*, and *A. thiooxidans*.

The two mixed cultures were grown separately in 9K liquid medium, which contained (per liter) 3.0 g of (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>, 0.5 g of MgSO<sub>4</sub>·7H<sub>2</sub>O, 0.5 g of K<sub>2</sub>HPO<sub>4</sub>, 0.1 g of KCl, and 0.01 g of Ca(NO<sub>3</sub>)<sub>2</sub>·H<sub>2</sub>O. The cultures received Fe<sup>2+</sup> (9 g/L), added as ferrous sulfate and elemental sulfur (10 g/L) as energy sources. The initial pH was adjusted to 1.8 with H<sub>2</sub>SO<sub>4</sub>. The cultures consisting of 120 mL of medium and 30 ml of inoculum were maintained in 250 mL shake flasks at 150 rpm and 34 °C for mesophilic bacteria, and at 45 °C for moderately thermophilic prokaryotes.

To estimate the cell counts, samples were taken at intervals and measured using a Neubauer counting chamber (0.1 × 1/400 mm<sup>2</sup>) and a Zeiss optical microscope under 1000 × magnification. The pH and oxidation-reduction potential (ORP, Pt vs. Ag/AgCl) values were also measured at intervals using a Mettler Toledo pH/ORP meter.

The amount of sulfur (organic, pyrite, and sulfate) was analyzed based on the standard ASTM-D3177, and the amount of ash was analyzed using ASTM-D3174 [20, 21]. The Alborz-based Iran Mineral Processing Research Center (IMPRC) conducted all analyses for this work. It should be noted that the amount of sulfated sulfur in each of the coal samples was negligible, and was therefore not taken into account. Figure 1 describes the methodology used in this study to decrease sulfur and ash from Tabas coal using bioflotation and bioleaching techniques

### 2.3. Bioflotation experiments

To conduct standard flotation tests, a Denver D12 laboratory flotation cell was used. The mechanical stirrer operated at a fixed speed of 1000 rpm, while the aeration rate was set to 100 ml/min. Pulp with a solids content of 30% and pH 8.0 (natural pH value of coal slurry) were prepared for bioflotation tests, which was conditioned under constant agitation. During conditioning, the pH of the solution was adjusted to 8 and monitored throughout the process. A bacterial culture was added as biodepressant, followed by the addition of the diesel oil as collector and pine oil as frother. The bioflotation experiments involves the use of a combination of mesophilic bacteria (*Acidithiobacillus ferrooxidans*, *Acidithiobacillus thiooxidans*, and *Leptospirillum ferrooxidans*) to depress pyrite in coal. The bacteria were not adapted to the Tabas coal sample, but were grown and multiplied for 10 days until the bacteria count reached  $8 \times 10^7 \frac{\text{cell}}{\text{ml}}$ . After the trivalent iron deposit was formed, the solution containing bacteria was separated from the iron deposits using filter paper. The bacterial count decreased to  $2 \times 10^7 \frac{\text{cell}}{\text{ml}}$  after filtration. To prepare the coal suspension with bacteria, the solution containing bacteria in the amount of 10-15% (V/V) was added to the flotation cell containing coal. The preparation time of bacteria with coal suspension was 20 and 60 min under the optimal conditions used in the previous our experiments. The bioflotation test involved collecting the froth for 10 minutes, followed by filtering and washing the froth and tailings with water to eliminate any sulfate produced by bacterial treatment. The washed products were then dried, weighed, and subjected to ash and sulfur content analysis.

### 2.4. Bioleaching experiments

The main goal of the bioleaching experiment is to remove high amounts of pyrite sulfur. To achieve this, a combination of mesophilic bacteria (*Acidithiobacillus ferrooxidans*, *Acidithiobacillus thiooxidans*, and *Leptospirillum ferrooxidans*), which have the ability to oxidize pyrite, was used in a ratio of 1:1:1. In this experiment, K9 medium was chosen for the growth of bacteria, for the salts used to promote better bacterial growth. Additionally, 47.4 g/L of divalent iron or green alum and 10 g/L of sulfur were added to the

bacterial solution. Bacteria were grown in 250 mL Erlenmeyer flasks, with 100 mL of bacterial solution and K9 culture medium. It should be noted that 10% (v/v) mixed mesophilic bacteria and 90% culture medium were used for bacterial growth. Also during bacterial growth (14 days), the pH was adjusted to 1.7. The bacteria were grown at 34 °C in an incubator with a stirring speed of 150 rpm. Finally, bacterial growth was monitored by measuring ORP and pH daily. It should be mentioned that the bacteria culture was not adopted with the Tabas coal sample. After two weeks of bacterial growth and multiplication, when the bacterial count reached 108 cells/mL, bioleaching experiments were initiated. It should be noted that the bioleaching tests were conducted indirectly and in two stages.

To conduct the experiments, 250 mL Erlenmeyer flasks were used, with only 100 mL of the solution placed in the flask and the rest of the space left empty for ventilation. The experiments were conducted at a temperature of 34 °C, stirring speed of 150 rpm, bacterial inoculation rate of 10-20% (v/v), coal solid percentage of 5-10% (w/v), and initial pH of 1.7 for 10-20 days, with the aim of reducing sulfur and ash content in the coal samples. For each coal sample, a test without the presence of bacteria (sterile environment) was also performed under the same conditions to determine the effect of bacterial presence on sulfur and ash removal. To perform the experiment in a sterile environment, only K9 culture medium was used along with coal samples and 3% (w/w) thymol. Thymol has antibacterial properties and prevents unwanted bacterial growth in the environment. During the tests, pH and ORP were checked in the coal samples every 3-4 days. Additionally, the weight of the solutions was monitored during the experiment, so that, in case of solution evaporation, an alternative amount of distilled water adjusted to pH = 1.7 with 5 M sulfuric acid was added to the solution. By doing this, the reduction of the sample size was avoided. It should be noted that after the completion of the test and filtration of the coal samples, the remaining solid was washed with 10% (v/v) hydrochloric acid to remove any remaining iron on the coal, which could unintentionally increase the coal ash content. Finally, the samples were dried in an oven for 5-6 hours at 60-70 °C degrees Celsius, and then sent for sulfur and ash analysis.

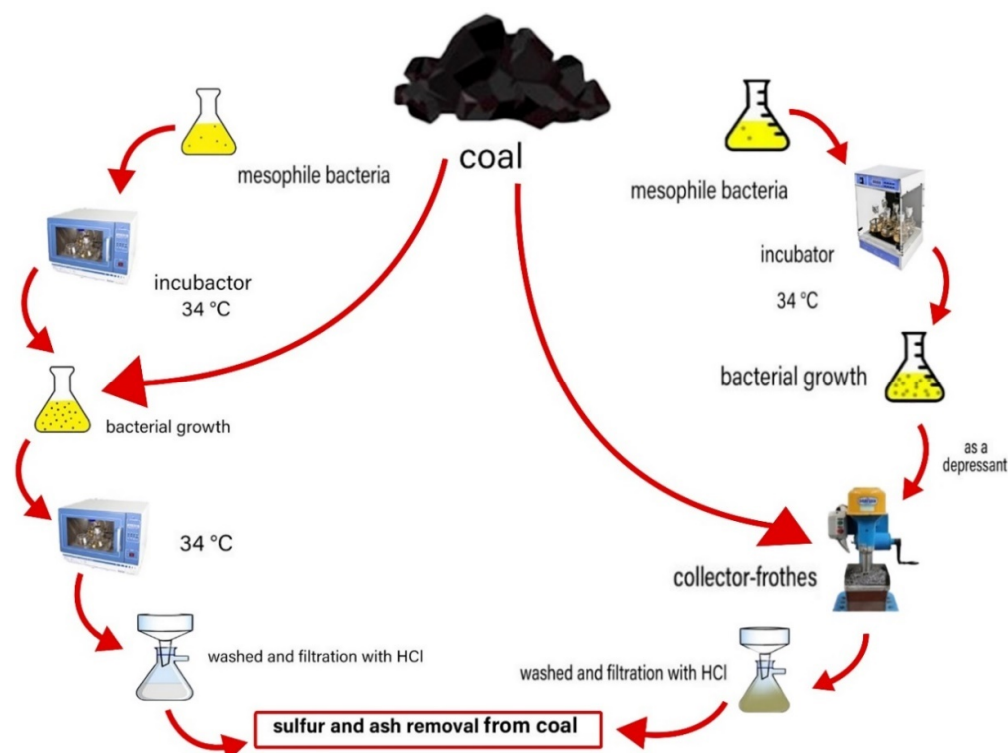


Figure 1. Methodology used in this study to decrease sulfur and ash from Tabas coal using bioflotation and bioleaching techniques.

### 3. Results and Discussion

#### 3.1. Coal sample characterization

Based on the SEM images shown in Figures 2, a significant amount of pyrite mineral can be seen in the sample of the primary feed of Eastern Parvadeh (PE), which has accumulated and formed masses in some areas. Pyrite mineral is accompanied by calcite and dolomite minerals in some parts, and mixed with clay mineral in the sample. In addition, there are some minor minerals such as ankerite, pyrrhotite, and galena. Regarding the sample B<sub>4</sub>C<sub>1</sub>, a significant amount of free pyrite is observed, which is mixed with clay minerals in some parts. The inclusion of pyrite and calcite can also be seen in a few parts of the sample, and some iron oxide, mainly in free form, was identified. The sample contains a lot of clay minerals and some calcite, as well as minor minerals such as barite, smithsonite, and galena. For the sample of (B<sub>4</sub>B<sub>2</sub>), a significant amount of pyrite mineral is observed, which has accumulated and formed masses in some areas. Pyrite mineral is also mixed with clay minerals in some parts, and the involvement of pyrite and calcite, as well as pyrite and quartz, is observed in the sample. There are some clay minerals and some calcite in the sample, as well as a small amount of apatite mixed with quartz.

Based on the polarizing light microscope images shown in Figure 3, it is evident that the primary feed sample of Eastern Parvadeh (PE) coal contains Vitrinite and Fusinite macerals. Vitrinite is the pre-dominant maceral in the sample, comprising around 70-75% of it, while Fusinite constitutes an important but smaller proportion of around 8-10%. Pyrite, a waste mineral, is present in two forms within the sample. The first type is organic origin pyrite, less than 0.2 mm in size, and occurs in framboidal forms, accounting for around 2% of the sample. The second type is mineral origin pyrite, with an abundance of approximately 1-2%, and exhibits a replacement texture. In Layer B<sub>2</sub> of sample 4 (B<sub>4</sub>B<sub>2</sub>), pyrite with organic origin refers to pyrite that has formed through the biological processes such as the decay of organic matter in sediments or the activities of certain microorganisms. In these cases, the pyrite forms as microscopic crystals within the organic matter or as aggregates of small crystals known as framboids. Pyrite with mineral origin refers to pyrite that has formed through inorganic processes. This type of pyrite is usually formed during hydrothermal mineralization, which occurs when hot fluids rich in sulfur and metals flow through rocks and deposit minerals in open spaces or fractures. Pyrite with mineral origin is commonly

found in hydrothermal veins and can also occur in sedimentary rocks, where it may form through diagenesis, which is the process by which sediments are transformed into solid rock [22]. Vitrinite is again the most abundant macerals, comprising around 70% of the sample. Fusinite presents an amount of around 10%. Pyrite with

mineral origin accounts for around 2-3% of the sample. In layer C<sub>1</sub> of sample 4 (B<sub>4</sub>C<sub>1</sub>), Vitrinite again constitutes the majority of the sample, with an amount of around 75%. Fusinite makes up around 8% of the sample, and pyrite exhibits the same characteristics as that found in the primary feed of Eastern Parvadeh (PE) coal.

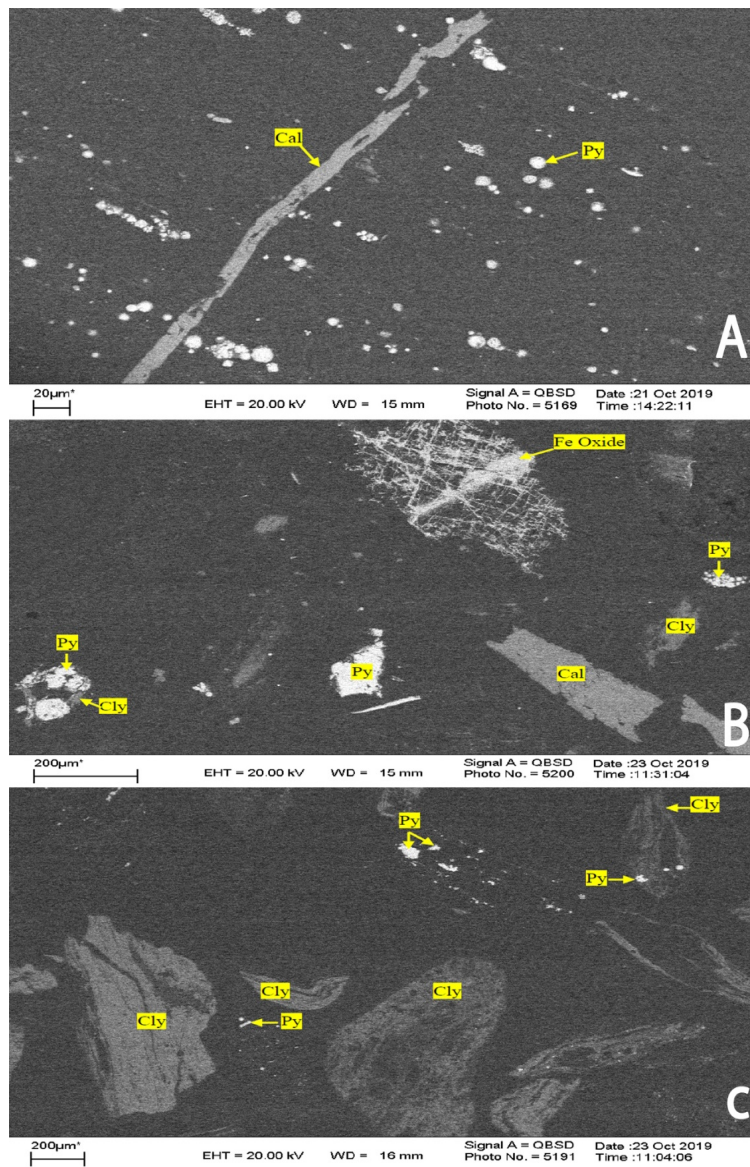


Figure 2. Scanning electron microscope (SEM) images of Tabas coal samples: A) PE, B) B<sub>4</sub>C<sub>1</sub>, C) B<sub>4</sub>B<sub>2</sub>, Py: Pyrite, Cal: Calcite, Fe-oxide: Iron oxide minerals, Cly: Clay Minerals.

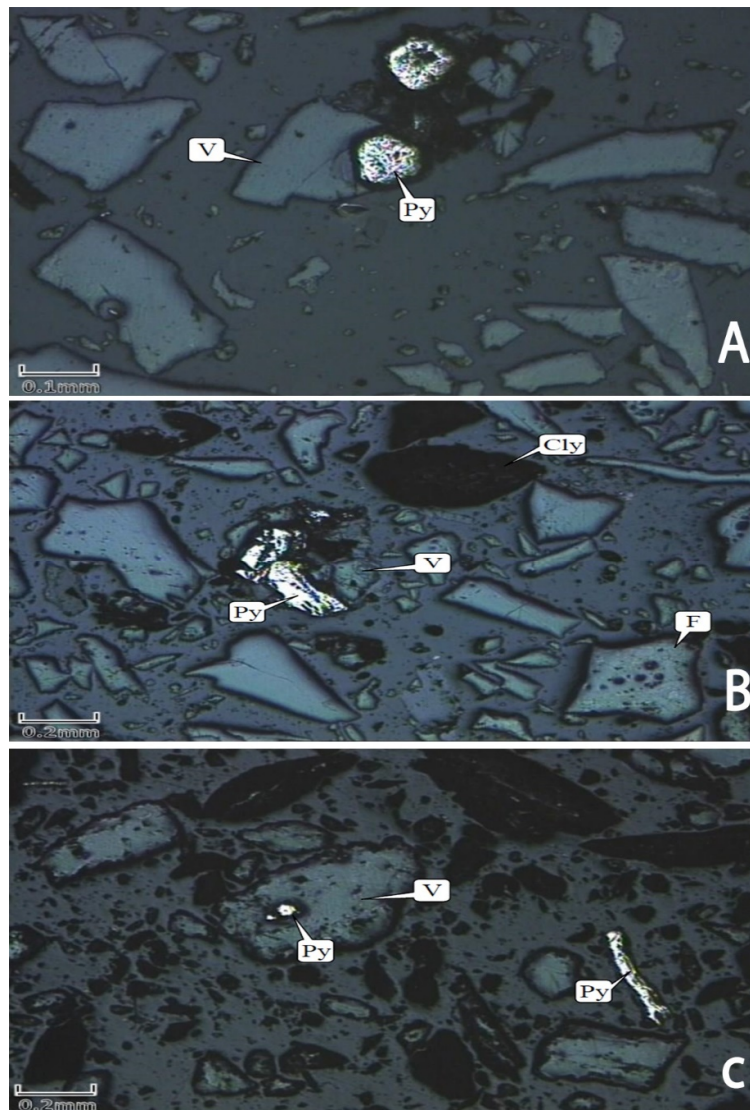


Figure 3. Polarized light microscope images of Tabas coal samples: A) PE, B) B<sub>4</sub>C<sub>1</sub>, C) B<sub>4</sub>B<sub>2</sub>. Py: Pyrite, Cly: Clay Minerals, F: Fusinite, V: Vitrinite.

### 3.2. Bioflotation

The flotation experiments with dextrin, a combination of mesophilic acidophilic bacteria (*Acidithiobacillus ferrooxidans*, *Acidithiobacillus thiooxidans*, and *Leptosprillum ferrooxidans*) was used for pyrite depression. This was done to compare the effect of organic and biological inhibitors. Previous studies have shown that using *Acidithiobacillus ferrooxidans* bacteria prevents the flotation of certain minerals such as pyrite [23]. In this study, all three coal samples were brought

into contact with each other during the bacteria preparation time of 20 and 60 min at pH = 3 with a volume percentage of bacteria of 10% and other optimized conditions in flotation experiments. After the preparation time of bacteria with coal, the pH reached = 7, and the collector was added at a rate of 400 grams per ton, followed by pine oil at a rate of 150 grams per ton. The effect of preparation time and volume percentage of bacteria on the reduction of pyrite sulfur and ash of primary coal feed samples was determined and presented in Table 2.

**Table 2. Effect of preparation time and volume percentage of bacteria on sulfur and ash reduction of primary feed samples using bioflotation.**

Samples	Time	Bacterial inoculum % (v/v)	Total sulfur (%)	Total sulfur removal (%)	Pyritic sulfur removal (%)	Ash removal (%)
PE	20	10	1.34	27.57	61	51.02
PE	60	10	1.33	28.11	61	52.17
PE	20	15	1.29	30.27	62	54.18
B <sub>4</sub> B <sub>2</sub>	20	10	2.05	0.97	0.85	66.14
B <sub>4</sub> B <sub>2</sub>	60	10	2.01	2.90	3.42	68.26
B <sub>4</sub> B <sub>2</sub>	20	15	1.96	5.31	8.55	69.34
B <sub>4</sub> C <sub>1</sub>	20	10	1.9	9.52	10.59	51.73
B <sub>4</sub> C <sub>1</sub>	60	10	1.88	10.48	12.58	56.04
B <sub>4</sub> C <sub>1</sub>	20	15	1.83	12.86	13.91	59.23

The results showed that increasing the preparation time from 20 to 60 min did not cause any specific change in the reduction of pyrite sulfur. Therefore, the preparation time of 20 min was chosen to continue the experiments. In the case of Eastern Parvadeh (PE) sample, increasing the volume percentage of bacteria from 10% to 15% resulted in the highest reduction of pyrite sulfur and ash, which were 62% and 54.18%, respectively. Similarly, an increase in the volume percentage of bacteria from 10% to 15% in 20 min resulted in an increase in the removal of sulfur, pyrite, and ash in all three samples of Tabas coal. The results also showed that increasing the preparation time from 20 to 60 min led to a decrease in pyrite sulfur in the flotation concentrate of the initial feed samples of layer B<sub>2</sub> of sample 4 (B<sub>4</sub> B<sub>2</sub>) and layer C<sub>1</sub> of sample 4 (B<sub>4</sub> C<sub>1</sub>). This decrease can be attributed to the additional time allowed for mesophilic bacteria to stick to the pyrite surface and change its surface from hydrophilic to hydrophobic. As the volume percentage of bacteria increased, more mesophilic bacteria could attach to the pyrite surface, increasing the probability of pyrite retention in the flotation cell.

In this study, mixed culture mesophilic bacteria were used to oxidize and modify the surface of pyrite sulfur in the Tabas coal samples during a series of bioleaching and bioflotation experiments. The aim was to investigate whether bacterial treatment could enhance pyrite depression before flotation, and the results showed that pyritic sulfur removal increased. Possible mechanisms for the depression of pyrite flotability in this system could be due to bacterial action, and adsorption of bacterial cells or their extracellular products, which are hydrophilic on pyrite surface. Previous research works indicated that bacterial culture had the main role in the depression of pyrite flotability [17, 18].

The exact mechanism for biomodification of pyrite flotability is still uncertain, but the adsorption of the biomass (bacterial cells and extracellular compounds) is the most likely mechanism. The previous studies suggest that the bacteria selectively attach to the pyrite surface, and their extracellular products may adsorb selectively on the pyrite particles. The pyrite surfaces may be covered with a layer of organic materials, making the pyrite more hydrophilic [11, 19].

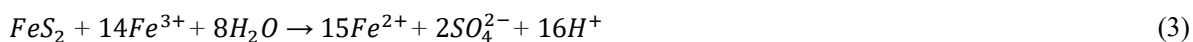
Interestingly, the state of surface oxidation of pyrite appears to have little effect on the biodepression ability of the bacterial culture, possibly because the mesophilic acidophilic bacteria (*Acidithiobacillus ferrooxidans*, *Acidithiobacillus thiooxidans*, and *Leptosprillum ferrooxidans*) can catalyze the oxidation of all the iron and sulfur species present on the pyrite surface. Future research may reveal the nature of the bonding mechanism between the bacteria and the pyrite surface.

### 3.3. Bioleaching

Equations 1 to 3 show that the concentration of ferric sulfate and pyrite oxidation is directly related to the increase of ferrous sulfate oxidation in an environment with the presence of iron-oxidizing bacteria. Therefore, the reduction rate of pyrite sulfur is crucial in conducting these tests. Biological oxidation is based on the ability of bacteria to oxidize sulfur compounds in coal and convert them into compounds that are soluble in water. Acidophilic iron and sulfur-oxidizing microorganisms can oxidize inorganic sulfur compounds and pyrite. Changes in acidity, total iron, and trivalent iron can be used to evaluate the desulfurization of coal during the biological process [3, 24].

The desulfurization of coal during the biological process can be evaluated by changes in acidity, total iron, and trivalent iron.





The results of Tables 3-5 showed that increasing the time and bacterial volume from 10% to 20% led to a higher removal of sulfur and ash in all three samples. The highest reduction of sulfur and ash was achieved during 20 days of bioleaching with a 20% volume of mesophilic bacteria. The study also found that organic sulfur was significantly removed from the coal sample along with pyrite sulfur. However, the biological process only affects pyrite sulfur and does not impact organic sulfur. The removal of organic sulfur up to 60% can be attributed to the acidic media with a pH of 1.7 within 10 to 20 days of bioleaching. The best bioleaching result to remove sulfur and coal ash was obtained during 20 days with 20% bacteria. In contrast, the absence of microorganisms significantly reduced the rate of sulfur and ash removal. However, about 50% of pyrite sulfur and 32.43% of the total sulfur in the PE sample were still removed, possibly due to the presence of coal in acidic conditions.

The result presented in Figure 4 showed a decrease in pH from the first day, which continued

until the 11th day, indicating proper bacterial activity in the solution. During this time, the bacteria broke the bond between sulfur and iron in the pyrite in coal, converting insoluble sulfides to water-soluble sulfate compounds. The divalent iron released from pyrite was converted to trivalent iron by losing electrons and remaining in the solution, forming sulfated compounds such as sulfuric acid and trivalent iron, which lowered the pH. The ORP, or iron(III) to soluble iron (II) ratio, from the fourth-day growth suggested excellent bacterial activity in the solution. The bacterium converts iron(II) to iron(III) by accepting electrons from iron (II) separated from the pyrite bond, eventually increasing the ORP.

The study also investigated the effect of pulp density on the removal of sulfur and ash from Tabas coal samples; the results were presented in Table 4. At a pulp density of 10%, only about 4-5% of sulfur was reduced compared to a pulp density of 5%, but in all cases, the total sulfur was below 1%. The results suggest that this type of coal is very suitable for use in the steel industry.

**Table 3. Effect of bacterial inoculum and time on the removal of sulfur and ash from Tabas coal samples with a pulp density of 5% bioleaching.**

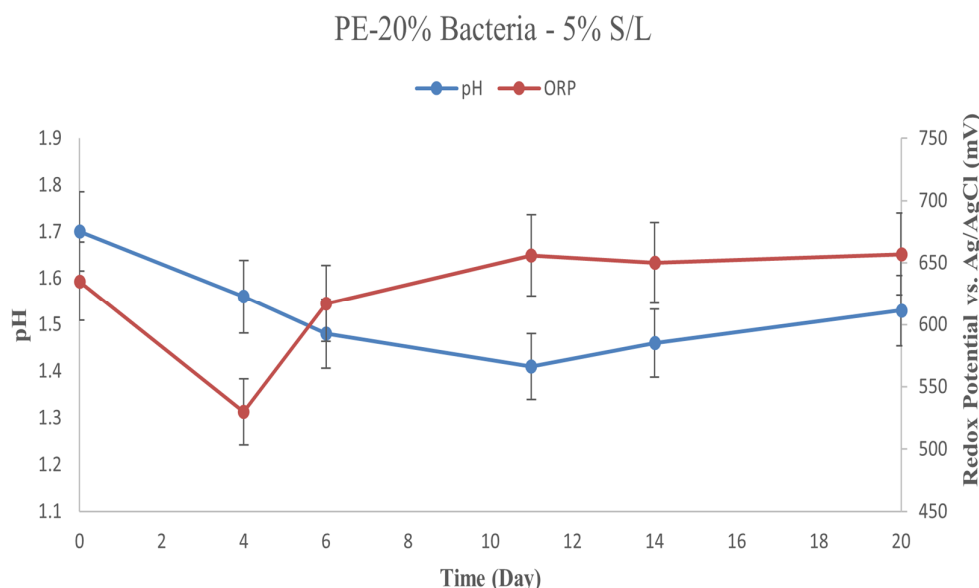
Samples	Bacterial inoculum % (v/v)	Time (day)	Total sulfur (%)	Total sulfur removal (%)	Pyritic sulfur removal (%)	Ash removal (%)
PE	10	10	0.8	56.76	79	63.12
PE	10	20	0.63	67.03	90	67.09
PE	20	10	0.66	64.32	84	65.18
PE	20	20	0.51	72.43	92	68.98
$B_4C_1$	10	10	0.84	55.78	61.14	44.46
$B_4C_1$	10	20	0.73	61.58	67.78	49.31
$B_4C_1$	20	10	0.78	58.95	64.44	46.71
$B_4C_1$	20	20	0.62	67.36	74.44	52.12
$B_4B_2$	10	10	1.12	51.72	77.88	31.12
$B_4B_2$	10	20	0.91	60.77	89.38	35.71
$B_4B_2$	20	10	0.98	57.76	84.07	32.33
$B_4B_2$	20	20	0.81	65.09	91.15	37.12

**Table 4. Removal of sulfur and ash from Tabas coal samples in the absence of microorganisms in 5% pulp density and 20 days.**

Samples	Bacterial inoculum % (v/v)	Total sulfur (%)	Total sulfur removal (%)	Pyritic sulfur removal (%)	Ash removal (%)
PE	0	1.25	32.43	50	36.18
B <sub>4</sub> C <sub>1</sub>	0	1.54	18.95	23.71	23.18
B <sub>4</sub> B <sub>2</sub>	0	1.93	16.81	23.89	19.17

**Table 5. Effect of pulp density on the removal of sulfur and ash from Tabas coal samples in 20 days with 20% bacterial inoculum.**

Samples	Pulp density (%)	Total sulfur (%)	Total sulfur removal (%)	Pyritic sulfur removal (%)	Ash removal (%)
PE	5	0.51	72.43	92	68.98
PE	10	0.6	67.57	89	67.56
B <sub>4</sub> C <sub>1</sub>	5	0.62	67.36	74.44	52.12
B <sub>4</sub> C <sub>1</sub>	10	0.71	62.63	65.55	50.11
B <sub>4</sub> B <sub>2</sub>	5	0.81	65.09	91.15	37.12
B <sub>4</sub> B <sub>2</sub>	10	0.89	61.64	87.61	35.91



**Figure 4. Evaluation of pH and ORP during 20 days bioleaching with 20% bacterial inoculum% (v/v) on the PE sample.**

In the bioflotation experiments, the use of mesophilic acidophilic bacteria was found to enhance the depression of pyrite flotability in coal samples. The results suggest that the adsorption of bacterial cells or their extracellular products, which are hydrophilic on the pyrite surface, could be the possible mechanism for this effect. Moreover, increasing the volume percentage of bacteria from 10% to 15% resulted in an increase in the removal of pyrite sulfur and ash in all the three Tabas coal samples. This could be attributed to the fact that more mesophilic bacteria could attach to the pyrite surface, increasing the probability of pyrite retention in the flotation cell. The decrease in pyrite

sulfur in the flotation concentrate of the initial coal feed samples due to the additional time allowed for mesophilic bacteria to stick to the pyrite surface and change its surface from hydrophilic to hydrophobic.

The bioleaching experiments showed that the biological process based on the ability of bacteria to oxidize sulfur compounds in coal can significantly remove pyrite sulfur and ash. Increasing the time and bacterial volume from 10% to 20% led to a higher removal of sulfur and ash in all the three Tabas coal samples. The highest reduction of sulfur and ash was achieved during 20 days of bioleaching with a 20% volume of

mesophilic bacteria. The acidic media with a pH of 1.7 within 10 to 20 days of bioleaching could be responsible for the removal of organic sulfur up to 60%. The ORP suggested excellent bacterial activity in the solution, and the bacterium converts iron(II) to iron(III) by accepting electrons from iron(II) separated from the pyrite bond, eventually increasing the ORP. The results suggest that the absence of microorganisms significantly reduced the rate of sulfur and ash removal, indicating the crucial role of bacteria in the bioleaching process.

Overall, the results suggest that the use of mesophilic acidophilic bacteria in bioflotation and bioleaching experiments can effectively enhance the removal of pyrite sulfur and ash from coal samples. The possible mechanisms for the depression of pyrite flotability in the bioflotation system could be due to bacterial action and the adsorption of bacterial cells or their extracellular products, which are hydrophilic on the pyrite surface. The bioleaching process based on the ability of bacteria to oxidize sulfur compounds in coal can significantly remove pyrite sulfur and ash, and the acidic media with a pH of 1.7 within 10 to 20 days of bioleaching could be responsible for the removal of organic sulfur. The ORP suggested excellent bacterial activity in the solution, and the absence of microorganisms significantly reduced the rate of sulfur and ash removal, indicating the crucial role of bacteria in the bioleaching process.

#### **3.4. Bioleaching and bioflotation for coal desulfurization and ash removal**

In this study, mixed culture mesophilic bacteria were used to modify the surface of pyrite sulfur in the Tabas coal samples during a series of bioleaching and bioflotation experiments. The aim was to investigate whether bacterial treatment could enhance pyrite depression to achieve desulfurization and ash removal of coal. The results showed that both bioleaching and bioflotation were effective for coal desulfurization and ash removal presented in Figure 5, with bioleaching performing slightly better for sulfur removal and bioflotation performing slightly better for ash removal. The best sulfur removal was

achieved using bioleaching, with a maximum removal of 72.43% observed for the PE coal sample. Bioflotation also achieved significant sulfur removal, with a maximum removal of 61% observed for the same sample. On the other hand, the best ash removal was achieved using bioflotation, with a maximum removal of 68.98% observed for the PE coal sample, and a maximum removal of 69.34% observed for the B<sub>4</sub>B<sub>2</sub> coal sample using bioleaching. The results are consistent with previous studies that have shown that bacterial action and adsorption of bacterial cells or their extracellular products on pyrite surface which are the possible mechanisms for the depression of pyrite flotability during bioflotation [25, 26]. The exact mechanism for biomodification of pyrite flotability is still uncertain, but the adsorption of the biomass (bacterial cells and extracellular compounds) is the most likely mechanism [4, 27, 28]. The bioleaching process is based on the ability of bacteria to oxidize sulfur compounds in coal and convert them into compounds that are soluble in water. Acidophilic iron and sulfur-oxidizing microorganisms can oxidize inorganic sulfur compounds and pyrite, resulting in the reduction of sulfur content in coal [4, 29]. Changes in acidity, total iron, and trivalent iron can be used to evaluate the desulfurization of coal during the biological process. The study found that organic sulfur was also significantly removed from the coal sample along with pyrite sulfur during bioleaching, but the biological process only affects pyrite sulfur and does not impact organic sulfur. The removal of organic sulfur up to 60% can be attributed to the acidic media with a pH of 1.7 within 10 to 20 days of bioleaching. The best bioleaching result to remove sulfur and coal ash was obtained during 20 days with 20% bacteria. The results suggest that a combination of both methods may be most effective for coal desulfurization and ash removal, with the choice of method depending on the specific coal type and desired outcome. Further research is needed to fully understand the mechanisms underlying each method and to optimize their use for coal desulfurization and ash removal.

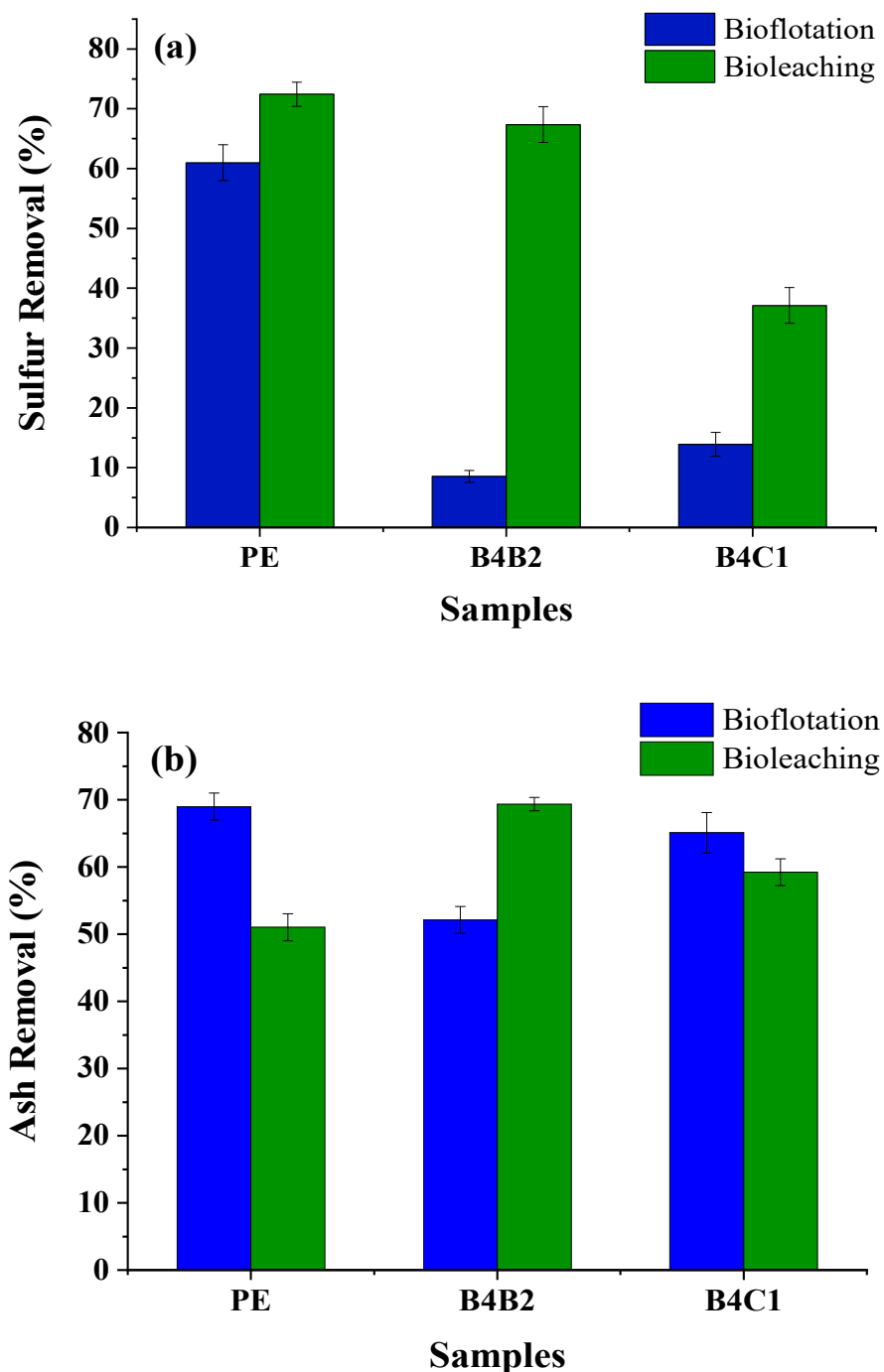


Figure 5. (a) Sulfur and (b) ash removal from Tabas coal samples using bioleaching and bioflotation methods.

#### 4. Conclusions

The objective of this study was to explore the potential of biological methods, specifically bioflotation and bioleaching, for removing sulfur and ash from the Tabas coal samples. A combination of mesophilic bacteria including *Acidithiobacillus ferrooxidans*, *Acidithiobacillus thiooxidans*, and *Leptosprillum ferrooxidans* was

used in the bioflotation method to remove pyrite sulfur in the Tabas coal samples. The study found that the use of this combination of bacteria in bioflotation was highly effective, achieving reduction percentages of up to 62% for pyrite sulfur and 54.18% for ash. In the subsequent stage of the study, bioleaching experiments were conducted to investigate the impact of various factors on the removal of sulfur and ash. The

findings revealed that test time was the most critical factor, with the best outcomes observed when the test conditions involved 20% bacteria by volume and a duration of 20 days. Under these circumstances, the Eastern Parvadeh (PE) sample recorded substantial reductions of ash, pyrite sulfur, and total sulfur. Upon comparing all of the methods employed in the study, the results indicated that bioleaching was the most effective method for achieving high levels of sulfur and ash reduction. In summary, this study demonstrated that biological methods such as bioflotation and bioleaching can effectively remove sulfur and ash from the Tabas coal samples. The results showed that both bioleaching and bioflotation were effective for coal desulfurization and ash removal, with bioleaching performing slightly better for sulfur removal and bioflotation performing slightly better for ash removal. The results of the laboratory experiments suggest that the use of mesophilic acidophilic bacteria in bioflotation and bioleaching processes can significantly enhance the removal of pyrite sulfur and ash from coal samples. However, it is important to consider the scalability of these experimental conditions for application to an industrial production line. One factor to consider is the cost-effectiveness of the proposed changes. The bacterial volume and bioleaching time used in the laboratory experiments may not be feasible or cost-effective for an industrial-scale production line. Therefore, further research would be needed to determine the optimal bacterial volume and bioleaching time that would provide the maximum benefits in terms of pyrite sulfur and ash removal while minimizing costs. Another factor to consider is the potential impact of the proposed changes on the overall production process and product quality. The increased bacterial volume and longer bioleaching time may have an impact on the processing time and product characteristics such as coal quality and combustion efficiency. Therefore, it is important to carefully evaluate the potential benefits and drawbacks of the proposed changes before implementing them on an industrial production line. While the experimental results are promising, further research and analysis would be needed to determine the feasibility and potential impact of applying the proposed changes to an industrial-scale production line. The conclusion section of the study could discuss these considerations and suggest directions for future research to explore the potential for industrial-scale application. The study could also emphasize the importance of carefully evaluating the potential benefits and drawbacks of any proposed changes

before implementing them on an industrial production line to ensure the optimal use of resources and the maintenance of product quality.

### Acknowledgments

The authors acknowledge the staff of the Iran Mineral Processing Research Center (IMPRC) for technical support.

### References

- [1]. Alam, H. G., Moghaddam, A. Z., & Omidkhan, M. R. (2008). The influence of process parameters on desulfurization of Mezino coal by HNO<sub>3</sub> / HCl leaching. *Fuel Processing Technology*, 0, 1–7.
- [2]. Amini, E., Hosseini, T. R., Oliazadeh, M., & Kolahdoozan, M. (2009). Application of acidithiobacillus ferrooxidans in coal flotation. *International Journal of Coal Preparation and Utilization*, 29(6), 279–288.
- [3]. *ASTM D3176-09 Standards, Standard Practice for Ultimate Analysis of Coal and Coke*. (2013). American Society for Testing and Materials.
- [4]. Aytar, P., Kay, C. M., Mutlu, M. B., & Çabuk, A. (2013). Coal desulfurization with acidithiobacillus ferrivorans, from Balya acidic mine drainage. *Energy and Fuels*, 27(6), 3090–3098.
- [5]. Çabuk, A., Koca, S., & Koca, H. (2014). Isolation and characterization of native microorganism from Turkish lignite and usability at fungal desulfurization, 116, 634–641.
- [6]. Dai, S., Jiang, Y., Ward, C. R., Gu, L., Seredin, V. V., Liu, H., Zhou, D., Wang, X., Sun, Y., Zou, J., & Ren, D. (2012). Mineralogical and geochemical compositions of the coal in the Guanbanwusu Mine, Inner Mongolia, China: Further evidence for the existence of an Al (Ga and REE) ore deposit in the Jungar Coalfield. *International Journal of Coal Geology*, 98, 10–40.
- [7]. Duz, M. Z., Saydut, A., Erdogan, S., & Hamamci, C. (2009). Removal of sulfur and ash from coal using molten caustic leaching, a case study from Hazro fields, Turkey, 27(6), 391–400.
- [8]. Eghbali, F., & Ehsani, M. (2010). Biodesulfurization of Tabas Coal in Pilot Plant Scale. *Iranian Journal of Chemistry and Chemical Engineering*, 29, 75–78.
- [9]. Ehsani, M. (2006). Desulfurization of Tabas Coals Using Chemical Reagents. *Iranian Journal of Chemistry and Chemical Engineering*, 25.
- [10]. El-Midany, A. A., & Abdel-Khalek, M. A. (2014). Reducing sulfur and ash from coal using *Bacillus subtilis* and *Paenibacillus polymyxa*. *Fuel*, 115, 589–595.
- [11]. Etemadzadeh, S. S., Emtiazi, G., & Etemadifar, Z.

- (2016). Heterotrophic Bioleaching of Sulfur, Iron, and Silicon Impurities from Coal by *Fusarium oxysporum* FE and *Exophiala spinifera* FM with Growing and Resting Cells. *Current Microbiology*, 72(6), 707–715.
- [12]. Golshani, T., Jorjani, E., Chehreh, C. S., Shafaei, S. Z., & Heidari, N. Y. (2013). International Journal of Mining Science and Technology Modeling and process optimization for microbial desulfurization of coal by using a two-level full factorial design. *International Journal of Mining Science and Technology*, 23(2), 261–265.
- [13]. He, H., Hong, F.-F., Tao, X.-X., Li, L., Ma, C.-Y., & Zhao, Y.-D. (2012). Biodesulfurization of coal with *Acidithiobacillus caldus* and analysis of the interfacial interaction between cells and pyrite. *Fuel Processing Technology*, 101, 73–77.
- [14]. Jorjani, E., Chelgani, S. C., & Mesroghli, S. (2008). Application of artificial neural networks to predict chemical desulfurization of Tabas coal, 87, 2727–2734.
- [15]. Jorjani, E., Rezai, B., Vossoughi, M., & Osanloo, M. (2004). Desulfurization of Tabas coal with microwave irradiation / peroxyacetic acid washing at 25 , 55 and 85 8 C, 83, 943–949.
- [16]. Kiani, M. H., Ahmadi, A., & Zilouei, H. (2014). Biological removal of sulphur and ash from fine-grained high pyritic sulphur coals using a mixed culture of mesophilic microorganisms. *Fuel*, 131, 89–95.
- [17]. Liu, T., Hou, J.-H., & Peng, Y.-L. (2017). Biodesulfurization from the high sulfur coal with a newly isolated native bacterium, *Aspergillus* sp. DP06. *Environmental Progress & Sustainable Energy*, 36(2), 595–599.
- [18]. Malik, A., Dastidar, M. G., & Roychoudhury, P. K. (2001). Biodesulphurization of coal: Effect of pulse feeding and leachate recycle. *Enzyme and Microbial Technology*, 28(1), 49–56.
- [19]. Milan, A. D., Ahmadi, A., & Hosseini, S. M. R. (2017). Biodesulfurization of a Coarse-Grained High Sulfur Coal in a Full-Scale Packed-Bed Bioreactor. *Solid State Phenomena*, 262, 207–210.
- [20]. Mishra, S., Pradhan, N., Panda, S., & Akcil, A. (2016). Biodegradation of dibenzothiophene and its application in the production of clean coal. *Fuel Processing Technology*, 152, 325–342.
- [21]. Misra, M., Bukka, K., & Chen, S. (1996). The effect of growth medium of *Thiobacillus ferrooxidans* on pyrite flotation. *Minerals Engineering*, 9(2), 157–168.
- [22]. San Martín, F., Kracht, W., Vargas, T., & Rudolph, M. (2020). Mechanisms of pyrite biodepression with *Acidithiobacillus ferrooxidans* in seawater flotation. *Minerals Engineering*, 145(October 2019), 106067.
- [23]. Shahbazi, M., Abdollahi, H., Shafaei, S. Z., Pourkarimi, Z., Jannesar Malakooti, S., Rahimi, M., & Ebrahimi, E. (2022). Desulfurization of Tabas coal using chemical (Meyers, Molten caustic leaching) and biological (bioleaching) methods. *International Journal of Mining and Geo-Engineering*.
- [24]. Subramanian, S., Santhiya, D., & Natarajan, K. A. (2003). Surface modification studies on sulphide minerals using bioreagents. *International Journal of Mineral Processing*, 72(1–4), 175–188.
- [25]. Vijayalakshmi, S. P., & Raichur, A. M. (2002). Bioflocculation of high-ash Indian coals using *Paenibacillus polymyxa*. *International Journal of Mineral Processing*, 67(1–4), 199–210.
- [26]. Xia, W., & Xie, G. (2017). A technological review of developments in chemical-related desulfurization of coal in the past decade. *International Journal of Mineral Processing*.
- [27]. Xu, J., Liu, X., Song, C., Du, Z., Wang, F., Chen, X., & Zhou, A. (2019). Environmental Effects Biodesulfurization of high sulfur coal from Shanxi : Optimization of the desulfurization parameters of three kinds of bacteria Biodesulfurization of high sulfur coal from Shanxi : Optimization of the desulfurization parameters of thr. *Energy Sources, Part A: Recovery, Utilization, and Environmental Effects*, 00(00), 1–19.
- [28]. Xu, J., Liu, X., Song, C., Du, Z., Wang, F., Luo, J., Chen, X., & Zhou, A. (2020). Biodesulfurization of high sulfur coal from Shanxi: Optimization of the desulfurization parameters of three kinds of bacteria. *Energy Sources, Part A: Recovery, Utilization, and Environmental Effects*, 42(18), 2297–2315.
- [29]. Yue, L., Jiao, Y., Wu, L., Rong, H., Fayek, M., & Xie, H. (2020). Evolution and origins of pyrite in sandstone-type uranium deposits, northern Ordos Basin, north-central China, based on micromorphological and compositional analysis. *Ore Geology Reviews*, 118, 103334.

## بیوفلوتاسیون و بیولیچینگ به عنوان روشی جایگزین برای گوگردزدایی و کاهش خاکستر از زغالسنگ طبس

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ارسال ۲۰۲۳/۰۵/۱۴، پذیرش ۲۰۲۳/۱۰/۰۷

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### چکیده:

زغالسنگ طبس دارای خواص پلاستومتری مطلوبی است که آن را برای استفاده در صنایع متالورژی به عنوان زغال کک سازی مناسب می‌کند. با این حال، محتوای بالای گوگرد آن، که تقریباً ۲ درصد است، خطر آلودگی زیست محیطی قابل توجهی را به همراه دارد. علاوه بر این، کاهش محتوای خاکستر به زیر ۱۰ درصد یک هدف مهم این مطالعه برای جلوگیری از کاهش بازدهی حرارتی زغالسنگ در صنایع متالورژی است. این کار تحقیقاتی به بررسی حذف گوگرد و خاکستر از نمونه‌های زغالسنگ طبس با استفاده از روش‌های بیولوژیکی شامل بیوفلوتاسیون و بیولیچینگ می‌پردازد. در ابتدا، ترکیبی از باکتری‌های مزوفیل شامل *Leptosprillum ferrooxidans* و *Acidithiobacillus thiooxidans*، *Acidithiobacillus ferrooxidans* در روش بیوفلوتاسیون برای حفظ گوگرد پیریتی از نمونه‌های زغالسنگ طبس استفاده شد. بیشترین درصد کاهش گوگرد پیریتی و خاکستر به ترتیب برابر با ۶۲ درصد و ۵۴/۱۸ درصد بود. در مرحله بعد، آزمایش‌های بیولیچینگ انجام شد، تأثیر زمان آزمایش، درصد باکتری بر حسب حجم، درصد جامد زغالسنگ و عدم وجود باکتری بر میزان حذف گوگرد و خاکستر بررسی شد. زمان به عنوان مهم‌ترین عامل ظاهر شد. بهترین حذف گوگرد با استفاده از بیولیچینگ، با حداکثر حذف ۷۲/۴۳ درصد، برای نمونه زغالسنگ PE به دست آمد. بیوفلوتاسیون نیز توانست میزان قابل توجهی از گوگرد را حذف کند، با حداکثر حذف ۶۱ درصد برای نمونه زغالسنگ PE مشاهده شد. از سوی دیگر، بهترین حذف خاکستر با استفاده از بیوفلوتاسیون به دست آمد، با حداکثر حذف ۶۸/۹۸ درصد برای نمونه زغالسنگ PE، و حداکثر حذف خاکستر با استفاده از بیولیچینگ ۶۹/۳۴ درصد برای نمونه زغالسنگ B4B2 مشاهده شد. در نهایت، در این کار تحقیقاتی مقایسه‌ای بین روش‌های بیولوژیکی برای تعیین میزان کاهش گوگرد و خاکستر انجام شد. نتایج نشان داد که هر دو روش بیولیچینگ و بیوفلوتاسیون برای گوگرد زدایی زغالسنگ و حذف خاکستر مؤثر بودند، روش بیولیچینگ برای حذف گوگرد و بیوفلوتاسیون برای حذف خاکستر بهتر عمل کرد.

**کلمات کلیدی:** زغالسنگ طبس، گوگردزدایی زغالسنگ، گوگردزدایی زیستی، بیوفلوتاسیون، حذف خاکستر، بیولیچینگ.