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Challenges from North to South
Des défis du Nord au Sud

On the effect of sophorolipids on different fractions of mine tailing and the speciation of arsenic

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ABSTRACT

Mine tailings produced after extracting gold from its ore, contain high levels of arsenic and heavy metals. As a result, mine tailings are one of the main environmental contamination sources. The objective of the present study was to investigate the effects of sophorolipids on the speciation and mobilization of arsenic, as well, on the morphology and composition of the mine tailings. The result from the sequential extraction of untreated and treated sample showed that in a continuous setting, a solution of 1% sophorolipids at pH 5 was able to change the morphology of the sample and mobilize arsenic associated with all fractions of the mine tailing sample. By using the soil washing in the continuous setup the total removal of 99% of iron and 98.8 % of arsenic was achieved. The results drawn from these investigations will be used to develop an efficient, environmentally friendly, and economically feasible system for the treatment of mine tailings.

RÉSUMÉ

Les résidus miniers produits après l’extraction de l’or de son minerai, contiennent de hauts niveaux d’arsenic et de métaux lourds. En conséquence, les résidus miniers sont l’une des sources principales de pollution de l’environnement. L’objectif de la présente étude était d’étudier les effets des sophorolipides sur la spéciation et la mobilisation de l’arsenic, ainsi que sur la morphologie et la composition des résidus miniers. Le résultat de l’extraction séquentielle des échantillons traités et non-traités a montré que dans un test continu, une solution contenant 1% de sophorolipides à un pH de 5 peut changer la morphologie de l’échantillon et mobiliser l’arsenic associé à toutes les fractions de résidus miniers de l’échantillon. En utilisant le lavage dans la configuration continue, 99% du fer et 98,8% de l’arsenic ont été enlevés. Les résultats tirés de cette investigation seront donc utilisés pour développer un système efficace, respectueux de l’environnement et économiquement viable pour le traitement des résidus miniers.

1 INTRODUCTION

Arsenic is a toxic metalloid and the 20th most abundant element in the earth’s crust (Mandal and Suzuki, 2002). Among the common valence states of Arsenic in the earth’s crust, arsenite [As (III)] and arsenate [As (V)] are the most abundant forms of arsenic. Exposure to a high level of this metalloid, or even long term exposure to lower levels of arsenic, is linked to many short term and long term health problems. The maximum level of inorganic arsenic recommended for the dry components of soil is 32 mg/kg in a residential area, and 640 mg/ kg in a commercial area (Environment Agency, 2009).

Arsenic is found in gold mineral deposits in the form of arsenopyrite. In the giant mine gold ores contain 2 to 3 percent arsenopyrite (FeAsS). To extract gold, the ores proceed through step by step processes of crushing and grinding, flotation, roasting, and cyanidation (Mudroch et al., 1989). These processes generate mine tailings that contain high levels of arsenic, cyanide and heavy metals such as chromium, manganese and zinc. According to Mudroch et al. (1989), the high level of zinc in these tailing is the result of using zinc for precipitating gold. It has been estimated that the mine tailings from the past mining activities at Giant Mines, Yellowknife contain a quarter-million tonnes of arsenic, which are deposited into nearby Great Slave Lake. These mine tailings are one of the main environmental contamination sources. Reports have indicated that heavy metals and arsenic from these mine tailings are transported to the local ground or surface waters, in addition to accidental release of mine tailings due to malfunctioning chambers, or seeping of the contaminated water from storage areas (Constantine & Price, 1983, Keeling & Sandlos, 2012). The drastic increase of arsenic in the sediment of The Great Slave Lake during past century coincides with the opening of gold mines in the area. One of the main sources of pollution of these sediments is baker creek, which receives effluent from the mine tailing in the giant mine, and carry the pollution to The Great Slave Lake (Wayland, 2004).

Over the past few decades, significant advances have been made on the treatment of contaminated soil and sediment. Some of these methods have been shown to be successful in removing heavy metals from soil, but most are not environmentally friendly. In recent years, many researchers studied the effectiveness of using environmentally friendly additives for metal extraction procedures (Mulligan et al., 2001). One of the promising methods that has grabbed the attention of the researchers in the recent years is using biosurfactants to enhance the process of removing the contaminants from the soil and sediment (Mulligan, 2005). Biosurfactants have shown success for removing hydrocarbons from soil, but the studies on the ability of biosurfactants to remove metals and metalloids from soil is still in the research phase.

Wang and Mulligan (2009b)'s research demonstrated the effectiveness of rhamnolipids in removing arsenic and heavy metals from mine tailings.

The objectives of the present study were to determine the effect of sophorolipid biosurfactants on mine tailings, and its efficiency for removing arsenic from mine tailings. Furthermore, the effect of the sophorolipids and rhamnolipids on the speciation of metalloids and metals was investigated and discussed.

2 MATERIAL AND METHODS

2.1 Analytical apparatus used in this research

Inductively coupled plasma mass spectrometry (ICP-MS), Agilent 7700 (Tokyo, Japan) equipped with a plasma frequency-matching RF generator and an octopole reaction collision cell and with detection limit of 0.1 ppb for arsenic, LC-ICP-MS Agilent 7700 (Tokyo, Japan), Laser Scattering Particle Size Distribution Analyser (Horiba Partica, LA950V2- Kyoto Japan), SEM/ EDS-SEM: JEOL 35-cf equipped with EDAX system for elemental analysis (Hitachi S-3400N), Philips X'Pert Pro Multipurpose X-ray Diffractometer (XRD), Aokton Electronic pH meter, and Denver Analytical Balance (Denver Instrument, SI 234; NY, USA).

2.2 Experimental setup

The setup of the continuous experiment was composed of two Plexiglas columns (length: 15 cm and inner diameter: 1.5 cm), a column holder, tubing, peristaltic pump (Cole Parmer Instrument Company, Montreal Canada), and volumetric and Erlenmeyer flasks for preparing, storing the inlet and outlet streams.

2.3 Materials

2.3.1 Chemicals

Nitric acid (HNO_3), calcium chloride ($CaCl_2$), hydrochloric acid (HCl), sodium hydroxide (NaOH) were purchased from Fisher Scientific Co. (Canada). Arsenic trioxide (As_2O_3), sodium metaarsenate and dimethylarsinic acid (DMAA) were purchased from Sigma, USA. Monomethylarsinic acid (MMAA) was purchased from Welck, USA. The reagents were of analytical grade.

2.3.2 Sophorolipids

Sophorolipids (SL) used in this experiment were Ecover 41% sophorolipids (SL18), donated by Ecover Co., Belgium. The sophorolipids were a mixture of 30% acidic sophorolipids and 70% lactonic sophorolipids. They were produced by yeast *Candida bombicola*, while growing on a mixture of rapeseed oil and glucose (Develter and Lauryssen, 2010).

2.3.3 Rhamnolipids

The rhamnolipids (Rha) used in this research, JBR425, are generally a mixture of mono-rhamnolipids and di-rhamnolipids. It was purchased from JENEIL Biosurfactant Co. (Saukville, WI, USA).

2.3.4 Mine tailings sample

Mine tailings sample was collected from the south tailing pond of Giant Mine, Yellowknife, Northwest Territories, Canada. The samples were collected from different depths of the pond, from the surface to a depth of four feet. In the laboratory, the samples were kept in an air tight container to preserve the mine tailings' physiochemical qualities.

2.4 Preliminary measurement of element concentration

By using a Niton *X-ray fluorescence* (*XRF*) analyzer, elements in the specimen and their approximate concentrations were detected. This preliminary measurement shows the elements present in the media, which have a concentration higher than the XRF detection limit (for example > 0.1 $\mu g.kg^{-1}$ for arsenic).

2.5 Measurement of element concentration with ICP-MS analyzer

To analyse and quantify the target elements within a sample with ICP-MS, the elements should be in a soluble form. Therefore, samples went through acid digestion according to EPA Method 3050, then were filtered (with a 0.2 µm filter), diluted and acidified further by using nitric acid (1%) and hydrochloric acid (0.5%) to be analyzed by ICP-MS. The ICP-MS was able to measure the trace metals in the sub parts per trillion ranges. It broke down samples to ions in high temperature argon plasma, and then analyzed based on their mass-to-charge ratio. The process was completed in three modes; without gas, with helium, and with hydrogen. Afterward, by using the correction equation (equation 1), the interference of argon and chlorine is corrected and a more realistic concentration arsenic is obtained.

$$75\text{As} = 75\text{As} - [3.127 \text{ x } (77\text{Se} - (0.322 \text{x } 78\text{Se}))]$$

EQUATION 1

Which can be interpreted as:
75As signal = total signal in mass 75 – (3.127 x (signal in mass 77 – (0.322 x signal in mass 78)))

2.6 Particle size distribution

Particle size distribution of untreated and treated mine tailing samples were measured by using a Laser Scattering Particle Size Distribution Analyzer (Horriba, LA-950). This analyzer is able to accurately determine particle size and determine the mean diameter of particles.

2.7 Moisture and organic content

The moisture content of the sample was calculated according to the ASTM method D2216. The organic content of the sample was measured according to EPA

Method 160.4. It was calculated by measuring the weight loss of the sample after calcination. For this, the weight of the total solid in the samples was measured after drying in an oven at 103-104^{o}C. Then, the samples were put in a muffle furnace for 50 minutes to be ignited at a temperature of 550^{o} C. After ignition, the weight of solid material combustible at 550^{o}C was calculated by subtracting the weight of the sample after calcination from the weight of the dried sample before going through ignition. The calculated weight loss was equal to the volatile residue of the sample and a rough approximation of the amount of organic matter present in the sample.

2.8 pH of the sample

The pH of the mine tailing samples was measured according to the method described by Schofield & Taylor (1955). It was done by measuring the pH of a mixture of 4 grams of the sample in 10 ml of 0.01 M calcium chloride (1g per 2.5 ml solution) using an electronic pH meter.

2.9 Column experiments

To evaluate the effectiveness of sophorolipids versus deionized water in mobilizing the arsenic from mine tailings, 50g of dried mine tailing sample was passed through a 100 µm sieve. Then it was compacted into a Plexiglas column (length: 15 cm and inner diameter: 1.5 cm). DI water, 1%, and 0.5% SL were passed through the column at the flow rate of 10 ml/min to minimize the pressure build-up and channeling through the sample. This flow rate was found to be suitable for this set-up, and it was chosen after repeated attempts and testing with a few different flow rates. After each 5-pore-volume, a sample was collected from the effluent, filtered, diluted and acidified to be analyzed with ICP-MS to determine arsenic concentration. Each run consisted of washing the sample with 60-pore-volume of solution. All experiments were conducted at room temperature and were repeated three times.

2.10 Selective sequential extraction

For the sequential extraction of the samples (both untreated and treated samples), the sequential extraction procedure described by Yong *et al.* (1993) was applied. Dried untreated mine tailing samples and treated mine tailings sample (washed by biosurfactants and dried) were put in six separate 50 mL centrifuge tubes (1 g of sample in each tube). In each step of extraction, the specimens in the tubes and the control tubes were subjected to different solutions, from deionized water to aqua regia. At the end of each step, the supernatant was collected and analysed to determine the percentage of target elements that were washed with each solution, or, to put it simply, the concentration and the percentage of elements associated with each portion of the medium.

3 RESULTS AND DISCUSSION

3.1 Main physiochemical properties of the mine tailings sample

The main physiochemical properties of the untreated mine tailings sample were as follows:

The moisture content of the sample was 19.8% and the pH of the sample shows to be alkaline (7.61).

The weight of volatile residue of the sample is a rough approximation of the amount of organic matter present in the sample and it was calculated to be 0.82 ± 0.07%. This falls in the range of organic matter in mine tailings (0 to 5.8%) reported by previous researchers (Pepper et al., 2012; Wu et al., 2006).

3.2 Result of preliminary measurement of element concentration with XRF

Results of the preliminary measurement of elements in the mine tailing sample has shown that Si. Al, Ca, As, Fe, K, Ti, Mn, Sc, Sb, Zn, Ni, Cu, Cr, Pb, Sn, Hg, V, Zr, Rb and Cd are present in detectable amounts. This preliminary measurement has also shown that the concentration of heavy metals/metalloids in the sample is a few times higher than the standard limits.

3.3 The results of measurement of elements' concentration with ICP-MS analyzer

The results from the ICP-MS can be found in Table 1. As it can be observed that high concentrations of arsenic and other heavy metals are present in the sample. The concentration of these elements are much higher than Canadian maximum standard limit for these elements in soil in industrial areas (CCME, 1997).Therefore the result obtained from this study shows the urgency of a cleanup plan for these tailings.

Table **1**: Results from ICP-MS analysis after digestion of mine tailing sample.

Element	Concentration (mg/kg)	Element	Concentration (mg/kg)
Al	98.41×10^3	Zn	0.56×10^3
As	2.57×10^3	Mo	27.55
Ca	1.43×10^3	Sb	0.22×10^3
Cr	0.22×10^3	Li	0.74×10^3
Mn	4.6×10^3	B	0.76×10^3
Fe	212.6×10^3	Na	5.36×10^3
Ni	0.396×10^3	Mg	12.5×10^3
Cu	0.65×10^3	K	65.8×10^3

3.4 Particle size distribution of the sample

The results of the analysis of the mine tailing samples, using a Laser Scattering Particle Size Distribution Analyzer showed that in untreated sample indicates that the size of the grains in the mine tailing sample and their homogeneity place them in the same category as silt (4 to 62 µm) (Wentworth, 1922). The coefficient of uniformity

(Cu) of the untreated sample was: 2.38 [$CU=D_{60}/D_{10}$; CU= 9.29/3.91= 2.38], that represent a uniformity of the untreated mine tailing sample. The Cu of the sample which was treated with a solution of sophorolipids 1% in continuous experiments was: 2.1 [Cu=8.816 /4.19 =2.10], that shows an increase in the uniformity of the sample.

Decrease in the grain size results in higher total surface area and it is expected to adsorb the higher quantity of trace elements on the surface of grains. Although, sometimes when the size of the grains are so small, they are cementing together and form aggregates, so the surface area decreases (Horowitz and Elrick, 1987). The presence of organic matter or clay, as well, results in cementing the grains together, decreasing the effective surface area. Lower effective surface area results in a lower adsorption rate. Table 2 shows a comparison between the result of particle size analysis of the untreated and treated samples. As it can be seen in the Table 2 the distribution of the grains in untreated sample is skewed. The result shows that the majority of the grains are in the range of fine silt. There is small percentage of clay and for untreated sample, the skewness is toward the larger grains. As it can be seen in the Table 2, after treatment with sophorolipids 1% in continuous experiments the homogeneity of the sample increased.

Table 2. Result of particle size analysis of the untreated and treated samples.

Parameters (μm)	Untreated sample	Treated with	
		DI water	1%SL
Median Size	8.14	7.81	7.60
Mean Size	13.42	10.68	8.17
Std.Dev.	10.42	10.55	3.86
Geo.Mean Size	8.37	8.00	7.26
Geo.Std.Dev.	1.21	2.05	1.66
Mode Size	8.23	8.22	8.23

3.5 Effect of sophorolipid concentration on mobilization of arsenic

The first step of the continuous experiments consisted of washing the mine tailings with sophorolipid solutions with different concentrations (1%, 0.5% and 0.1%) at pH 7. As it can be observed in Figure 1, the highest rate of arsenic removal was obtained by introducing the highest concentration of sophorolipids being used (1%) and the lowest removal of arsenic was in the presence of lowest concentrations of sophorolipids (0.1%). Therefore, it can be assumed that there is a positive correlation between the rate of arsenic removal and the concentration of sophorolipids being used. There is a need for more investigations to determine the maximum concentration at which this statement applies. Higher concentrations of sophorolipids cause the particles in mine tailings to form floating clusters / colloids and clog the outlet filter.

The results show that sophorolipids with concentration 1% reached their peak removal at 20 pore volume washes but 0.5% and 0.1% sophorolipids reached their maximum removal on 35 and 45 pore volumes, respectively. Altogether, although sophorolipids with the concentration of 1% seem promising for arsenic removal, comparison of the sophorolipids being used and total removal, it can be concluded that 0.5% sophorolipids is the better choice.

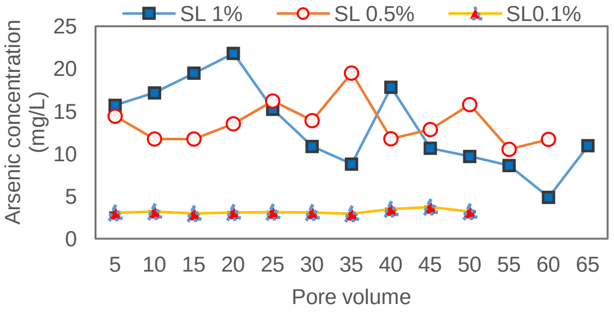

Figure 1: Removal of arsenic from mine tailing by using SL with different concentration in continuous setting.

Data shows that the concentration of the arsenic in the samples obtained from the effluent, after every pore volume, increases until it reaches a maximum removal, and then there is a decrease in arsenic removal, until the removal rate reaches a constant value. In the following experiments, the maximum value of the pore volume was 65. The obtained data shows that, not only higher concentrations resulted in higher removal of arsenic, but also, the solution with higher concentrations of sophorolipids reached its peak of removal in less pore volumes of washing.

3.6 Effect of pH on arsenic mobilization in the column experiment

A comparison between the data obtained from column experiment, when using 0.5% sophorolipids as the washing solution, at pH 2.5 versus pH 8 shows that in lower pH there is a higher arsenic removal (Figure 2). The total removal of arsenic after 60 pore volume wash at the pH 2.5 was 172.8 mg/kg, and at pH 8 it was 59.8 mg/kg, or 2.9 times less than what was removed at pH 2.5.

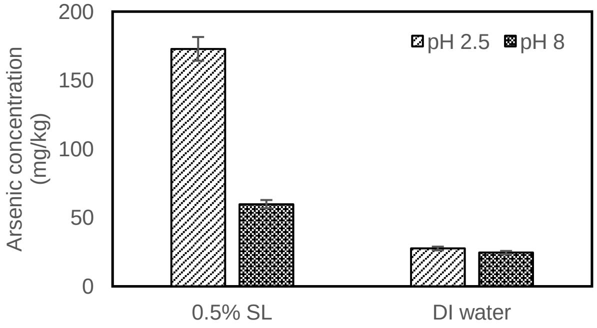

Figure 2: Total arsenic removed from the mine tailings after 65 pore volumes in the column experiments, using 0.5% SL and DI water at pH 2.5 and 8 as the washing solution.

Comparing the removal of arsenic at pH 2.5 in column experiment (Figure 2) shows that the total removal of arsenic between 5 to 60 pore volume washings, by using 0.5% sophorolipids was 6.8 times greater than the removal by deionized water (27.5 mg/kg).

Using 0.5% sophorolipids and deionized water at pH 8 shows that the sophorolipids solution was much more effective than deionized water with the same pH (Figure 3). By using deionized water as the washing solution, the total arsenic removal during 5 to 65 pore volume washing was 21.5 mg/kg. On the other hand, the total arsenic removal from 5 to 60 pore volume washing by using 0.5% sophorolipids was 59.8 mg/kg, 2.8 times greater than arsenic removed by deionized water.

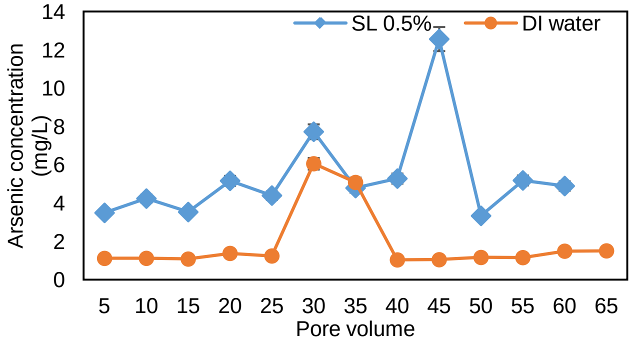

Figure 3: Removal of arsenic by DI water and 0.5 % SL at pH 8 in column experiments.

3.7 Iron mobilization

As it can be seen in the following graph (Figure 4), at the lower pH the removal rate of iron was much higher and the total iron removal from mine tailing sample at pH 2.5 was 3651.5 mg/kg and at pH 8 the total removal was 1401.4 mg/kg, or 2.6 times lower than removal at pH 2.5.

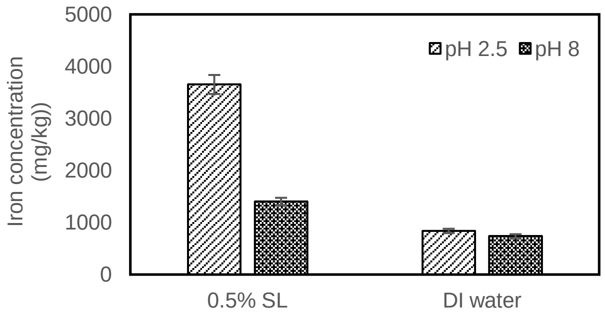

Figure 4: Total iron released from the mine tailings in the column tests after 60 pore wash by using 0.5% SL and DI water at pH 2.5 and 8.

A comparison between the release of iron by using 0.5% sophorolipids and deionized water at pH 8 shows that the sophorolipid solution was much more effective than deionized water with the same pH (8). Using 0.5% sophorolipids resulted in 2.8 times more than the amount of iron washed by deionized water. The same comparison, but in pH 2.5 shows that in the acidic environment also, the total removal of iron by using sophorolipids was 4.9 times higher than by deionized water.

A comparison between the removals of iron by using washing solutions of deionized water at pH 2.5 and 8 shows that although the removal in some points overlap, but overall, the removal rate of iron from the mine tailing sample at lower pH was higher. The total removal after 60 pore volumes wash at pH 2.5 (834 mg/kg) was 1.14 times higher than that at pH 8 (734.37mg/kg) (Table 4).

3.8 Arsenic and iron distribution in fractions of untreated mine tailing

Table 3 shows the concentration of iron and arsenic in each fraction of the untreated mine tailing sample. As it can be seen, residuals contain the highest percentage of arsenic, and the organic fraction of the mine tailings contain the lowest percentage of arsenic. Arsenic distribution in mine tailing is as follow: organic fraction< water soluble fraction< exchangeable fraction < oxide/ hydroxide potion < carbonate fraction < residual fraction.

Iron concentration was differently distributed between the different fractions of the mine tailings. The iron concentration from the lowest to highest was associated with water soluble, organic fraction, exchangeable, oxide/ hydroxide, carbonate, and the residual fraction.

Some of the environmentally friendly methods of soil washing and flushing introduced by previous researchers (Wang and Mulligan, 2009a and b), such as using rhamnolipids, are able to remove the arsenic associated with the water soluble and exchangeable fractions of mine tailings. For the removal of arsenic from the other fractions of the sample, using conventional methods such as using strong acids or bases was suggested. In the following parts of the experiment, the effects of sophorolipids on the different fractions of mine tailing samples were studied.

Table 3: Concentration of arsenic and iron (mg/kg) extracted during SSE process from untreated mine tailing using 0.1% sophorolipids.

	Fe	As
Water soluble	38.7	12.9
Exchangeable	220.9	13.9
Carbonate	157.1	16.8
Oxide/hydroxide	14862.0	385.0
Organic	883.5	41.4
Residual	135284.0	2082.8
Total	151446.3	2552.8

3.9 Arsenic and iron distribution in fractions of the treated mine tailing sample

In this part of the experiments, the treated mine tailings sample, which was washed with a solution of 1% sophorolipids in a column experiment, was air dried. It was then subjected to the process of sequential extraction. The result from the sequential extraction of the treated mine tailings sample is shown in Table 4.

Table 4: Iron and arsenic extracted (mg/kg) from different fraction of treated mine tailing sample.

	Fe	As
Water soluble	52.9	14.1
Exchangeable	65.8	154.7
Carbonate	224.9	23.0
Oxide/hydroxide	1227.6	42.6
Organic	111.9	54.1
Residual	3771.4	136.5
Total	5454.6	425.1

As it can be seen in Table 4, the distribution of arsenic and iron in different fractions of the treated sample appears very similar to the untreated sample. Results from the sequential extraction shows that sophorolipids had an impact on all of the fractions of the mine tailings sample.

Wang and Mulligan (2009a) reported that rhamnolipids were able to remove arsenic from water soluble and exchangeable part of soil and sediments. The result from sequential extraction of sample which was treated with sophorolipids shows that sophorolipids are a superior choice than rhamnolipids for mobilizing metals and metalloids, as they are able to mobilize the arsenic from all fractions of the media.

3.10 Speciation of arsenic in untreated and treated mine tailing samples

The results from the analysis show that, among the extractable arsenic species from mine tailing samples, using deionized water with pH 6 as the washing solution, As (V) was the only species of arsenic detected. Washing with deionized water at pH 11 resulted in a small amount of As (III), but again the majority of the extracted arsenic was arsenic (V). The reason for this occurrence is that the majority or all of the arsenic associated with the water soluble fractions of mine tailings are As (V). As the mine tailing specimens that were used have been stored in tailing ponds for over 10 years, it was expected that arsenic and other metals/ metalloids were oxidized. Then again, by increasing the pH of the deionized water, the organic fraction of the mine tailings were subjected to being partially washed, therefore releasing the arsenic associated with the affected parts. Therefore the presence of As (III) in the solution with pH 11 shows that some of the arsenic is associated with the organic fraction of mine tailing are in the lower oxidation states, As (III).

Further investigations on the effect of biosurfactants on the oxidation state of arsenic showed that the arsenic extracted by using rhamnolipids was a mixture of As (V) and As (III), although, the majority of the arsenic extracted with the rhamnolipids were As (V). The slight increase in the concentration of arsenic extracted at pH 11 can also be the effect of increasing pH on the organic fractions of the mine tailings.

The result of analyzing the extracted arsenic from the mine tailing samples by using a solution of 1% sophorolipids showed that the majority of the extracted arsenic was in the form of As (III) (the more mobile state of arsenic). Therefore, it can be assumed that sophorolipids are able to affect not only the water soluble fraction of mine tailing, but also other fractions of mine tailings. On the other hand, a solution of 0.1% sophorolipids doesn't affect the other fractions of the tailings. Table 4 displays the result of analyzing the extractable arsenic species from the batch experiments by using the LC-ICP-MS. It shows that the changes in the concentration of specific arsenic oxidation state, mobilized from the samples, depends on the biosurfactant being used and the biosurfactant concentration.

Table 5: Extractable arsenic species in the mine tailing by using sophorolipid and rhamnolipids and measured by HPLC- ICP-MS.

Biosurf.	**%**	**pH**	**Total As (mg/kg)**	**As(III) + DMA* (mg/kg)**	**As(V) (mg/kg)**
SL	0.1	6	4.87	0	4.8
SL	1	6	23.51	18.6	4.91
Rha.	0.1	6	4.78	0.16	4.62
Rha.	1	6	19.73	0.26	19.47
Rha.	1	11	35.56	1.74	33.82
control	NA	6	2.27	0	2.27
control	NA	11	36.32	0.25	36.07

*DMA: Dimethylarsonic acid

Furthermore, batch experiments were conducted to determine the effect of biosurfactants on the speciation of arsenic, and to determine if biosurfactants are able to reduce the oxidation state of arsenic. For this part, mixtures of 2 mg/L sodium arsenate and sophorolipids (0%, 0.1%, 1% and 5%) were prepared, and were placed in eight separate tubes (two tubes for each concentration). Tubes were put on a shaker, shaking at 60rpm for 4.5 weeks and at room temperature. After 24 hours, and after 7 days, 14 days, 30 days, samples were taken from each tube and prepared for analysis with LC-ICP-MS. The result of analysis showed that there were no changes in the oxidation states of arsenic. The only species of arsenic found in the samples was As (V). Therefore, the release of arsenic (III) from the mine tailing samples in the presence of biosurfactants is due to the presence of As (III) in the non-water soluble fractions of mine tailings, and the effectiveness of the biosurfactants in releasing arsenic from non-water soluble fractions of the specimen.

3.11 Backscattered electron imaging

The analysis of the back scattered electrons from the samples displays the composition image of the samples. The following images display the back scattered images of untreated samples and samples that have been treated by using different solutions: 1% sophorolipids, 0.5% sophorolipids, and deionized water.

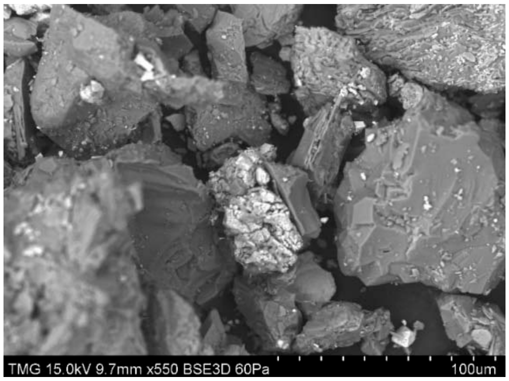

Figure 10: An SEM (backscattered image) of an untreated mine tailing sample.

The back scattered image of the untreated mine tailing sample in Figure 10 illustrates the compositional heterogeneity of the sample. There are crystals with metallic sheen that seem to be pyrite, while the white amorphous matter in the center of the image resembles a calcium compound. As it was mentioned before, during the analysis of the untreated mine tailing sample, the high metal content of the untreated samples limited the ability to increase the resolution. By increasing the magnification or voltage, the grains became charged, resulting in blurred images. For obtaining a more accurate understanding of the composition of the grains, the sample was analysed by the SEM-EDS.

In the backscattered image of a sample which was treated with a solution of 1% sophorolipids with 1000x magnification (Figure 11), the grains seemed more separated based on their composition, and very small crystals with metallic sheen were present. The composition of these crystals and the larger masses were analysed with the SEM-EDS to determine elemental composition of them.

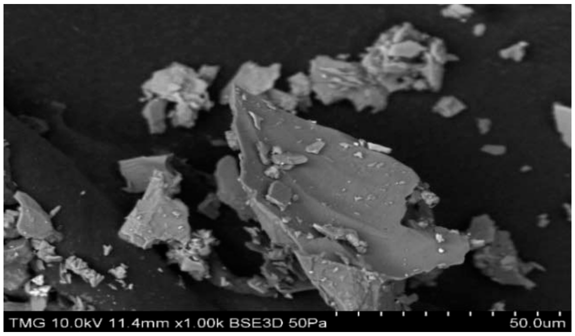

Figure 11: A backscattered image of a treated sample with 1% sophorolipids.

Figure 12 displays a backscattered (BSE) 1000x magnified image of a sample treated with 0.5% sophorolipid solution. A comparison between the images of the samples treated with 0.5% and 1% sophorolipid solutions, shows that in the samples which were treated with 0.5% SL, a greater variety of grain sizes and grain compositions were observed. On the other hand, Figure 13 illustrates the backscattered image of specimen which was treated with deionized water. In the BSE image of the sample which was washed with deionized water, a variety grain types and grain sizes are observed. Overall, the sample seems more heterogeneous than the samples which were washed with sophorolipid solutions.

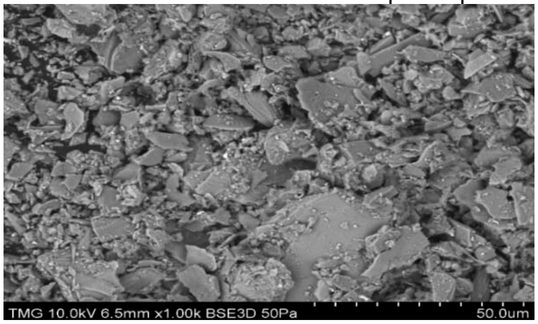

Figure 12: An SEM (backscattered image) of a treated sample with 0.5% sophorolipids

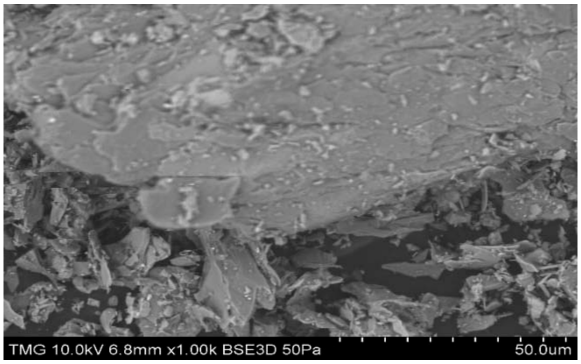

Figure 13: A backscattered image of a sample treated with deionized water.

By comparing the data from the samples (Figures 11, 12 and 13), it can be said that the surface of the grains varies depending on the type of the solution that has been used for the treatment. Comparing between the morphology and composition of the samples being treated with 0.5% or 1% sophorolipid solutions, with an untreated mine tailing sample show that different concentrations of sophorolipids make a noticeable difference on the morphology and composition of the sample. The higher concentration of sophorolipids affected the sample morphology more than the lower concentrations did. Furthermore, the SEM-EDS analysis shows a drastic increase in the concentration of metal/metalloids in the grains.

4 CONCLUSIONS

SEM analyses of the samples before and after being treated with sophorolipids of different concentrations, show that the morphology, homogeneity and composition of the samples were significantly affected by different concentrations of sophorolipids. A comparison between the results from the sequential extraction of untreated and treated samples show that sophorolipids were able to affect all fractions of the mine tailings.

The results from the speciation of arsenic showed that that the only water extractable species of arsenic was arsenate [As (V)]. However, speciation of the arsenic in the effluent from the batch experiments revealed that washing the mine tailings with a 1% sophorolipid solution resulted in the release of both arsenate [As (V)] and arsenite [As (III)] from the mine tailing specimen.

The high production rate, thus lower production price, and its efficiency in variety of pH, make sophorolipids a perfect candidate for being used in many applications, particularly for the bioremediation of soil and sediment.

The results drawn from these investigations will be used to develop an efficient, environmentally friendly, and economically feasible system for the treatment of contaminated mine tailings.

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