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Geochemical and microbial effects on the mobilization of arsenic in mine tailing soils

Keun-Young Lee · Kyoung-Woong Kim · Soon-Oh Kim

Abstract Arsenic (As) contamination has become a serious environmental problem in many countries. We have performed batch-type leaching experiments on mine tailing soils collected from three abandoned mine areas in South Korea with the objective of evaluating the effect of indigenous bacterial activity on As mobilization. The analysis of physicochemical properties and mineralogical compositions of the samples indicated that the secondary minerals or phases formed as a result of the oxidation or alteration of primary minerals were associated with the labile and bioleachable fractions of As. Compared to simulated abiotic processes using sterilization, the indigenous bacteria activated using a carbon source were able to enhance the dissolution of As under both aerobic and anaerobic conditions. The bacterial dissolution of iron (Fe) and manganese (Mn) was found to occur simultaneously with the dissolution of As, suggesting that the main bacterial mechanism was via the dissimilatory reduction of Fe(III), Mn(IV), and As(V). An anaerobic environment was more favorable for the prominent dissolution of As in the tailing soils. These results indicate that the mobilization of As can be enhanced in the oxygen-depleted part of the tailing dump, particularly with the infiltration of organic substrates. The difference in the degree of As lixiviation between the three tailing soils was found to be related to the bioavailability of As as well as the original biomass in the tailing soils.

Keywords Arsenic · Bioavailability · Metal reducing bacteria · Microbially mediated leaching · Tailing soil

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Introduction

Arsenic (As) contamination has recently been recognized as one of the most crucial environmental problems facing many countries, including the USA, Canada, Mexico, China, Bangladesh, Vietnam, and India. Most environmental problems associated with As are the result of mobilization under natural conditions. The significantly potentially high adverse effect of As on the ecosystem is well known. The tailings from abandoned mine areas as well as As contamination of agricultural soils and crops pose particularly serious management issues (Morin and Calas 2006; Wang and Mulligan 2006). Even though inorganic As in soils is in itself harmful, immobile As can be easily transformed into a mobile phase as

the result of changes in environmental conditions, thereby posing an additional potential environmental hazard. The contamination of ground and surface water by As from soils and sediments represent significant threats to human health (WHO 2004) and, consequently, it is essential to fully understand the transport characteristics of solid phase As (Zhang and Selim 2005). The effects of geological and biological weathering and changes in the redox potential on the fate of solid phase As have been well studied (Masscheleyn et al. 1991; Ribet et al. 1995; Oremland and Stolz 2003). However, these factors always coexist in the natural environment; therefore, the geochemical as well as microbial effects have to be simultaneously considered. Unfortunately, such multilateral studies are significantly lacking.

The typical primary mineral of As is arsenopyrite (FeAsS). The oxidation of arsenopyrite in abandoned mine areas may result in the formation of scorodite (FeAsO$_4 \cdot$ 2H$_2$O), with the release of As. Such oxidative environments are mainly acidic, with high concentrations of iron (Fe) and As(V) (Juillot et al. 1999; Harvey et al. 2006; Bluteau and Demopoulos 2007). Under oxidizing conditions, dissolved As may be retained in the soil due to its adsorption onto clays and metal oxides, particularly those of manganese (Mn), aluminum (Al), and Fe oxides. Within tailing deposits, the oxidation of sulfide minerals typically results in the formation of abundant secondary ferric (oxy)hydroxides or (oxy)hydroxysulfates on tailing surfaces. Arsenic and other metal elements may be coprecipitated or adsorbed onto these secondary minerals (Mok and Wai 1994; Ribet et al. 1995).

To date, biogeochemical investigations on the fate of As have revealed that microorganisms play a crucial role in the chemical speciation and cycling of As in nature (Newman et al. 1998). Microbial As(V)-reducing mechanisms can be classified into two types: detoxification and dissimilation. As(V), with its structural similarity to phosphate, readily enters microbial cells via phosphate-uptake proteins. Microorganisms have developed a detoxification strategy to protect themselves against the toxic effects of As, i.e., the capability to reduce As(V) to As(III) by means of an As resistance system (ars), which is functional in both aerobic and anaerobic environments (Ji and Silver 1995). Some anaerobic bacteria are able to gain energy by coupling As(V) reduction to the oxidation of H$_2$ or organic carbon. These dissimilatory As-reducing

bacteria use As(V) as a terminal electron acceptor during respiration (Ahmann et al. 1994). The bacterial reduction of As can enhance the mobility of As in groundwater, as As(III) is more mobile than As(V).

In addition to these direct mechanisms that are related to the oxidation state of As, several indirect processes also influence the mobilization of As. In general, As can be immobilized via its sorption onto Fe and Mn oxides. However, it has been reported that As seems to be remobilized by the dissolution of these oxides due to microbial reductions of Fe and Mn and that this process appears to be ubiquitous in anoxic environments (Lovley 1993; Nealson and Saffarini 1994; Lee et al. 2005). In aerobic environments, however, the oxidation of arsenic sulfide minerals by S- or Fe-oxidizing bacteria does not promote the sorption of As onto metal oxides, but, rather, promotes the release of As(III) into the aqueous phase (White et al. 1998). This type of mechanism is often applied as a remediation technology tool when treating soils and sediments contaminated by heavy metals—i.e., bioremediation—on the pretext of bioleaching (Seidel et al. 2004).

The focus of the study reported here was the elucidation of the effect of indigenous bacterial activity on the mobilization of As. To this end, we performed microbially mediated leaching experiments under both aerobic and anaerobic conditions. The major mechanisms controlling the mobilization of As in the subsurface environment is discussed and the potential hazard of solid phase As is highlighted. In addition, the physicochemical and mineralogical factors are related to the extent of microbially mediated lixiviation of As in the target tailing soils.

Materials and methods

Sampling and site description

Arsenic-contaminated tailing soil samples were collected from three abandoned mine areas in South Korea: Myungbong (MB), Songcheon (SC), and Nakdong (ND). These sites are known to be seriously contaminated by As and heavy metals. The MB mine is located in the southwestern region of the Korean Peninsula, whereas the SC and the ND mines are

located in the mid-eastern region. Mining at these sites began in the early 1900s. The MB and the SC mines were productive gold (Au) mines until 1970 and 1943, respectively, while the ND mine was actively operated for the mining of As, bismuth (Bi), and Fe until 1989. Since their closure, these three mines have been abandoned without any proper management, resulting in detrimental impacts on the surrounding environments.

Tailing soil samples, produced by the weathering of tailings dump, were carefully collected from each mine within a depth of 30 cm from the surface of the tailing-dumping area. A composite sampling method was applied: a representative sample was prepared by mixing at least five sub-samples taken within a 10 m^2 area, with at least three representative samples obtained from each mine. Some of the samples were air-dried and sieved through a no. 10 sieve (diameter 2 mm) for physicochemical characterization, while other samples were kept without the drying step for the microbially mediated leaching experiments. To maintain the original environment for the survival of indigenous bacteria, we stored the moist tailing soil samples in polyethylene vinyl containers at room temperature.

Physicochemical characterization of tailing soil samples

The sieved tailing soils were used for all physicochemical analyses. Soil pH, organic matter content, and particle size distribution were investigated as the principal properties of each sample. To determine the total concentrations of As and heavy metals [Al, cadmium (Cd), copper (Cu), lead (Pb), zinc (Zn), cobalt (Co), chromium (Cr), nickel (Ni), Mn and Fe], each sample (0.5 g) was digested with aqua regia (4 ml; concentrated HNO_3:HCl, 1:3), and heated at 70°C on a shaker for 1 h (Ure 1995). Distilled water (6 ml) was then added to the solution, with the final samples filtered through 0.45-μm polytetrafluoroethylene membrane filters.

Sequential extraction techniques were used (Wenzel et al. 2001) to examine the chemical speciation of each As fraction in the tailing soils. The mode of As occurrence was classified as non-specifically sorbed phases (step 1), specifically sorbed phases (step 2), amorphous and poorly crystalline hydrous oxides of Fe and Al phases (step 3), well-crystallized hydrous oxides of Fe and Al phases (step 4), and residual phases (step 5). The sequential extraction of other heavy metals was originally suggested by Tessier et al. (1979), where each step of the method can be described as the exchangeable fraction (step 1), carbonate and specifically adsorbed fraction (step 2), Fe–Mn (hydro)oxides (step 3), organic matter and sulfide (step 4), and the residual fraction (step 5). The specific procedures for each sequential extraction are shown in Tables 1 and 2.

All reagents used were of American Chemical Society (ACS) grade, and all analytical procedures carried out on the samples were repeated in duplicate. The analytical data were assessed during all procedures using a quality control system, including duplicates, blanks, and standard reference samples, to check the accuracy and precision. All duplicate measurements demonstrated relative standard deviations (%RSD) of <5%. Reagent blanks for all the extractants were analyzed in parallel with the samples; there were negligible As levels in all cases.

The As concentrations in all samples were determined using atomic absorption spectrometry-graphite furnace (AAS-GF; PE model 5100PC; Perkin–Elmer, Foster City, CA), with atomic absorption spectrometry-flame (AAS-Flame; Perkin–Elmer) used for all the other heavy metals.

Table 1 The sequential extraction procedure for arsenic (modified from Wenzel et al. 2001)

Step (target phase)	Extractant	Step time
I. Non-specifically sorbed phases	0.05 M $(NH_4)_2SO_4$, 20°C, continuous agitation	4 h
II. Specifically sorbed phases	0.05 M $(NH_4)H_2PO_4$, 20°C, continuous agitation	16 h
III. Amorphous and poorly crystalline hydrous oxides of Fe and Al phases	0.2 M NH_4-oxalate buffer, pH 3.25, 20°C, continuous agitation in the dark	4 h
IV. Well-crystallized hydrous oxides of Fe and Al phases	0.2 M NH_4-oxalate buffer, 0.1 M ascorbic acid, pH 3.25, 96 ± 3°C, continuous agitation in the light	30 min
V. Residual phases	HCl and HNO_3 (3:1), 70°C, continuous agitation	1 h

Table 2 The sequential extraction procedure for iron and manganese (modified from Tessier et al. 1979)

Step (target phase)	Extractant	Step time
I. Exchangeable fraction	0.5 M MgCl$_2$, pH 7, 25°C, continuous agitation,	2 h
II. Carbonate and specifically adsorbed fraction	1 M NaOAc, pH 5 (adjusted with HOAc), 25°C	6 h
III. Fe–Mn (hydro)oxides	0.04 M hydroxyl-ammonium hydrochloride (NH$_2$OH HCl) in 25% HOAc, 95°C, occasional agitation	7 h
IV. Organic matter and sulfide	0.02 M HNO$_3$ and 30% H$_2$O$_2$ (adjusted to pH 2), 85°C, 2 h, occasional agitation	6 h
	30% H$_2$O$_2$ (adjusted to pH 2) added, 85°C, 3 h, occasional agitation	
	After cooling, 3.2 M ammonium acetate (NH$_4$OAc) in 20% (v/v) HNO$_3$ added	
V. Residual fraction	HCl and HNO$_3$ (3:1), 70°C, continuous agitation	1 h

Mineralogical analyses

The main mineral constituents of the samples were measured using X-ray diffractometry (XRD) (Siemens model D5005 Diffractometer; Cu-Kα 40 kV and 35 mA). The 2θ range and the scanning speed were 3°–90° and 0.02°/s, respectively. The original tailing samples were embedded in epoxy resin and then polished for mineralogical studies using reflection microscopy, scanning electron microscopy–energy dispersive spectrometry (SEM–EDS; model JSM-6380LV; JEOL, Japan) and electron probe micro-analysis (EPMA; model JXA-8100; JEOL). The SEM–EDS and back-scattered electron (BSE) images with the EPMA maps were used to characterize the elemental composition of individual grains within each tailing sample.

Microbially mediated leaching experiments

All of the obtained tailing soil samples were used in batch-type leaching experiments. The leaching solution was prepared by mixing 6.6 mM of glucose and 3.3 mM of lactate as the carbon sources. Then, 20 g of sterilized (autoclaved at 121°C) and non-sterilized samples were separately agitated in two bottles containing 200 ml of leaching solution and the bottles incubated on a rotating shaker (180 rpm) at 30°C for 30 days under both aerobic and anaerobic conditions. Anaerobic experimental conditions were achieved by performing all procedures in an artificially made anaerobic chamber under a 90% N$_2$, 5% H$_2$, and 5% CO$_2$ atmosphere. An aliquot of the leaching solution was collected during the experiments, and the

pH, oxidation–reduction (redox) potential (Eh), and dissolved oxygen (DO) were measured. The As(total), As(III), Fe(total), Fe(II) and Mn(total) concentrations in the samples were also analyzed. The biomass during the early stage of the leaching experiments (after 2 and 4 days) was analyzed from the optical density (OD) at 600 nm using a UV spectrophotometer (model UV-1201PC; Shimadzu, Japan). To eliminate any disturbance in the optical densities due to the soil particles and colloidal suspensions in the samples, the optical densities of non-sterilized and sterilized samples were simultaneously measured, and the difference between those two values was regarded as the OD originating from biomass.

The samples were allowed to pass through a silica-based anion–exchange cartridge (LC-SAX SPE tube; Supelco, Bellefonte, PA) to determine As(V) and As(III) speciation (Le et al. 2000). The As concentrations of the original and pretreated samples, indicating the As(total) and As(III), respectively, were determined using AAS–GF, as mentioned previously. The Fe(II) in the samples was determined using a colorimetric procedure employing ferrozine (Stookey 1970). The total Fe and Mn concentrations were analyzed using AAS-Flame.

Results and discussion

Geochemical characteristics of tailing soils

The initial pH range of the soils was between 5 and 6. The organic matter content of the ND tailing soil was higher than those of the MB and the SC tailing soils

Table 3 Physicochemical properties and total concentrations of target elements and the mineral constituents found in the tailing soil samples

Physicochemical properties	MB	SC	ND	Natural contents[a]
pH	5.09	5.7	5.58	
Organic matter content (%)[b]	<0.005	0.013	2.635	
Composition (%)[c]				
Sand	68.5	55.5	88.5	
Silt	28.43	41.74	10.24	
Clay	3.07	2.76	1.26	
Texture	Sandy loam	Sandy loam	Sand	
Target elements (mg/kg)				
Al	3,892	1,484.6	31,980	72,000 (700 to <10,000)[d]
Cd	1.60	1.04	4.34	0.35 (0.01–2)
Cu	15.18	23.28	164.70	25 (<1–700)
Pb	157.66	104.36	73.96	19 (<10–700)
Zn	120.4	486	235.4	60 (<5–2,900)
Co	4.78	2.76	27.44	9.1 (<3–70)
Cr	6.74	4.77	46.94	54 (1–2,000)
Ni	12.83	12.33	31.50	19 (<5–700)
Mn	252.6	243.6	945.6	550 (<2–7,000)
Fe	14,770	21,740	105,100	26,000 (100 to >1,000,000)
As	1,480	3,380	4,780	7.2 (0.1–97)
Mineral constituents			Calcite	
	Calcite	Calcite	Illite	
	Kaolinite	Kaolinite	Jarosite	
	Muscovite	Muscovite	Kaolinite	
	Quartz	Quartz	Montmorillonite	
	Scorodite	Scorodite	Muscovite	
			Quartz	
			Scorodite	

MB, Myungbong; SC, Songcheon; ND, Nakdong

[a] Natural contents in soils from U.S. Geological Survey Professional Paper 1270 (1984)

[b] Organic matter content (%) was calculated from measured loss-on-ignition(LOI%) (Ball 1964; FitzPatrick 1983)

[c] Sand, 50–2,000 μm; silt, 5–50 μm; clay, <5 μm

[d] Median (range)

(Table 3). Based on the soil texture, as suggested by the USDA (United States Department of Agriculture), the MB and SC tailing soils were classified as sandy loam and that of the ND tailing soil as sand.

Chemical analyses revealed that the tailing soils were seriously contaminated with several heavy metals and As, especially compared with the range of heavy metals content in the natural background (Table 3). The tailing samples from the three sites showed similar elemental compositions, but somewhat different concentrations. The concentrations of almost all of the target elements, with the exceptions of Pb and Zn, were higher in the samples from the ND mine than in those from the MB and SC mines. In the case of As content, the total concentrations were in the order: ND (4780 mg/kg) > SC (3380 mg/kg) > MB (1480 mg/kg).

The analytical results obtained from the five-step sequential extraction indicated that the modes of As occurrence in each tailing were entirely different, as
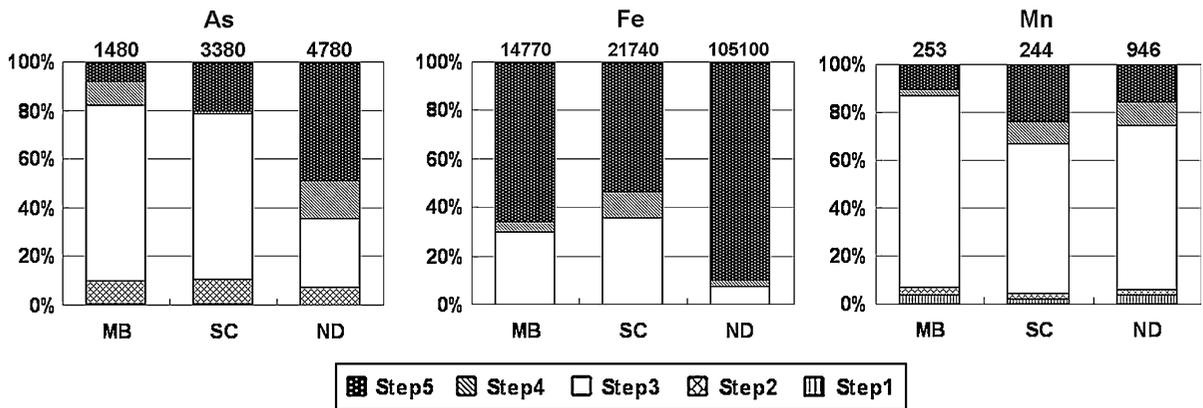
Fig. 1 Total concentrations and chemical speciations of As, Fe, and Mn in the Myungbong (*MB*), Songcheon (*SC*), and Nakdong (*ND*) tailing soil samples. Step 1, non-specifically sorbed; step 2, specifically sorbed; step 3, amorphous/Fe–Mn (hydro)oxides; step 4. well-crystalline/organic matter and sulfide; step 5, residual phases. The values at the *top* of each column show the total concentrations of each element (mg/kg)

shown in Fig. 1. The As contents in the MB and SC tailing soils were mainly partitioned in the labile fraction (82.7 and 78.6%, respectively) with respect to the As extracted via steps 1–3. In particular, most of As was found to be associated with amorphous and poorly crystalline hydrous Fe oxides. Conversely, the fraction of labile As was relatively lower (35.8%) in the ND tailing soil. Given these above results, it is essential to determine the levels of Fe and Mn in the tailing soils, as As is likely to be strongly associated with the oxides of these two metals. The results from the sequential extraction of Fe and Mn from each tailing soil indicated that the main fraction of Fe was residual, whereas those of Mn were oxides or hydroxides (Fig. 1). The level of the (hydro)oxides portion of Fe in the ND tailing sample was relatively lower (7.7%) than those in the MB and SC tailing soils (30.6 and 35.8%, respectively). The differences in the fractionation of Fe, Mn, and As can be attributed to a variety of geochemical factors, such as the composition of the main minerals and the physicochemical conditions of surrounding environments, as well as the type and extent of the controlling geochemical processes (Savage et al. 2000; Paktunc et al. 2004). The results of the sequential extraction revealed the proportions of Fe, Mn, and As in the labile fractions to be relatively higher in the MB and SC tailing samples than in the ND tailing sample. Accordingly, it can be speculated that the MB and SC tailings had been more affected by weathering.

Mineralogical composition and results of SEM–EDS and EPMA

The main mineral constituents observed from the XRD analyses are shown in Table 3. The mineral compositions of the MB and SC tailings were found to be very similar, with scorodite ($FeAsO_4 \cdot 2H_2O$) identified as the predominant secondary mineral that included Fe and As in both tailings. Even though the tailings were likely to contain primary sulfide minerals, including Fe and As, such as pyrite or arsenopyrite, no corresponding XRD results were identified due to their low levels.

The BSEs and corresponding EDS spectra for each tailing sample are presented in Fig. 2. In addition, the X-ray maps created using the EPMA shows the distribution of O, Fe, S and As (Fig. 3). The brighter grains likely correspond to the primary sulfide minerals and the lighter surrounding rims to the secondary minerals formed due to the oxidation or alteration of the primary sulfides. In the case of the MB tailing, pyrite (FeS_2) was observed to be the dominant primary sulfide mineral, with the surrounding rim oxidized to a form of Fe oxide. The oxidation of pyrite was definitely confirmed in the EPMA mapping. The brighter grains were enriched with S and depleted of O, but these two elements exhibited opposite behaviors in the darker area, thereby indicating that the formation of secondary oxides in the surrounding area was due to the alteration of primary sulfides. Although no As was detected in the MB
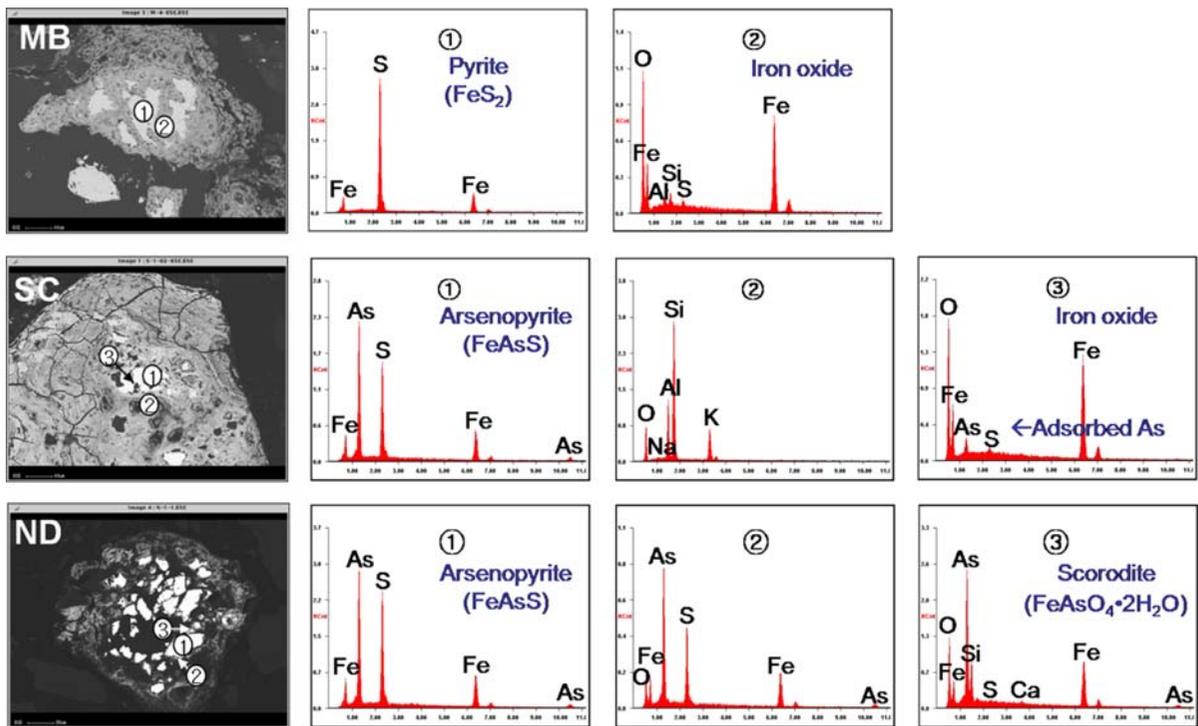
Fig. 2 Back-scattered electron (*BSE*) images (*left*) and the energy dispersive spectroscopy (*EDS*) spectra of tailing soil samples (*1*, *2*, *3*) collected at Myungbong (*MB*, *top row*), Songcheon (*SC*, *middle row*), and Nakdong (*ND*, *bottom row*)

tailing, sequential extraction had already confirmed the existence of the labile form of As as sorbed and/or amorphous phases in these secondary oxides. Arsenopyrite (FeAsS) was observed as the primary sulfide in the SC tailing, with very similar oxidation or alteration to that found with the MB tailings. The EDS spectra suggest that the secondary minerals were Fe oxides, onto which the As appeared to be adsorbed. In addition, the EPMA mapping dramatically highlighted the difference in the elemental compositions between the primary sulfides and secondary oxides. As shown in Fig. 3, S and O exhibited the opposite behaviors between arsenopyrite (brighter area) and the Fe oxide grains (darker area). Arsenopyrite was also observed as the predominant primary mineral in the ND tailings, with scorodite formed as the secondary mineral as a result of oxidation. Compared to the distribution of As in the SC tailings, however, a significant abundance of As was detected in the ND tailing. This may be due to the partitioning of As in the non-mineral phases (e.g., in adsorbed forms) in the SC tailings, but in the mineral phases (e.g., scorodite) in the ND tailings.

These minerals or altered phases may relate to the behaviors of each element, especially Fe and As, in the microbially mediated environment. The leaching of Fe oxides via microbial dissimilatory reduction under anaerobic conditions can cause the release of As. In addition, secondary minerals, such as scorodite, can be dissolved through reductive dissolution, allowing Fe(II), As(III), or As(V) ions to be leached out (Papassiopi et al. 2003; Verplanck et al. 2008).

Microbially mediated leaching of As

The microbial effects on As leaching were investigated under both aerobic and anaerobic conditions. A significantly greater amount of As was leached from the non-sterile samples than from the sterile samples (control test) (Figs. 4, 5). In addition, the characteristics of As leaching were significantly different depending on the conditions and properties of the tailing samples. The order of dissolved As between the different tailing soils was SC > MB > ND. This sequence can be attributed to several factors, such as the bioavailable As fraction, the major mechanisms

affecting the reactions, and the biomass related to those mechanisms. The maximum As concentration in the leaching solution of the SC tailing soil, which had both a high concentration of As and a high labile fraction, occurred under anaerobic conditions. Although the ND tailing showed a relatively high total As concentration, the maximum concentration of As in the leaching solution during the experiments barely reached 10 mg/l (Fig. 4c). This low level seemed to be caused by the lower proportion of bioavailable As, as supported by the sequential extraction and mineralogical observations. The amount of biomass after 2 and 4 days also supports the difference in the degree of leaching in each experiment. Despite being activated by sufficient carbon sources, a significantly lower bacterial growth was observed in ND tailings (Fig. 6), probably due to the high levels of toxic heavy metals in the tailings. Several heavy metals and trace elements are required for the growth of microorganisms and are utilized during their growth processes. However, the excessive enrichment of trace elements could suppress the survival of microorganisms (Beveridge et al. 1997).

A much higher amount of dissolved As was obtained under anaerobic conditions than under aerobic ones, indicating an anaerobic environment was the more favored one for the prominent dissolution of As. Similar results in terms of increased levels of Fe, Mn, and As under anaerobic conditions were obtained in all experiments (Fig. 4). The bacterial dissimilatory reduction of Fe and Mn can be considered to be a major mechanism of anaerobic dissolution (Lovley 1991, 1993; Ahmann et al. 1997). After concentrations of those elements had increased in the leaching solutions, no decline was observed throughout the experiments. The Eh during the experiments did not attain a sufficient level for sulfate reduction (Eh < −200 mV) and, therefore, sulfide formation and metal precipitation did not occur. Based on the relative $-\Delta G$ of the redox couples, microbial respiratory preferences should sequentially deplete as follows: $O_2 \leftrightarrow H_2O$, $NO_3^- \leftrightarrow N_2$, $Mn^{4+} \leftrightarrow Mn^{2+}$, $NO_3^- \leftrightarrow NH_4^+$, $Fe^{3+} \leftrightarrow Fe^{2+}$, $SO_4^{2-} \leftrightarrow HS^-$, and $CO_2 \leftrightarrow CH_4$ (Scott and Morgan 1990). The microbial reduction of As(V) is also feasible because the free energy related to the transformation of As(V) to As(III) falls between the values needed for the reduction of Fe^{3+} and SO_4^{2-} (Sadiq 1990). However, we do not postulate that the microbial reduction of

As(V) is the most dominant mechanism for the leaching of As under anaerobic conditions for two reasons: first, in general, the amounts of microbially available Fe(III) and Mn(IV) in the tailing soils were relatively larger than that of As(V); second, Fe(III) and Mn(IV) are well known to show a relatively larger redox potential than As(V). For these reasons, indigenous bacteria preferentially utilize Fe(III) and Mn(IV) as electron acceptors under anaerobic conditions. Although most of the As originally existing in the tailing soils were in the As(V) form (determined by XANES, data not shown), As(III) was the main component measured in the leaching solution (more than 80%), indicating that the dissimilatory reduction of As(V) by indigenous bacteria occurred in the experiments conducted under anaerobic conditions.

In relation to aerobic microbially mediated leaching with the carbon source, the dissolution of As with Fe and/or Mn increased during these experiments (Fig. 5), suggesting the As adsorbed onto or coprecipitated with Fe and Mn (hydro)oxides was dissolved due to bacterial dissolution. There are a diverse number of feasible mechanisms. First, As detoxifying bacteria are ubiquitous in soil environments, and the transformation of As(V) to As(III) by these bacteria can enhance the mobility of As (Macur et al. 2001). This may be a major mechanism accounting for why mainly As was observed in the leaching solutions, as found at SC in this study (Fig. 5b). Second, S-oxidizing bacteria can promote the release of As (Bayard et al. 2006). The sulfate concentrations in the leaching solutions from the non-sterile experiments were slightly higher than those from the sterile experiments (data not shown), but no correlation was found between the dissolved As and sulfate concentrations, thereby excluding the oxidation of S as a result of the main As leaching mechanism. Thirdly, the biological production of carbon dioxide or organic acids can be sufficient to lower the local pH and indirectly accelerate the dissolution of the minerals (Ullman et al. 1996; Grantham et al. 1997; Liermann et al. 2000). The solution pH was found to be lower than the initial neutral range (7–8), extending into a somewhat acidic range (nearly 5–6) upon the dissolution of the target elements. Fourthly, although the reductions of Fe(III) and Mn(IV) are not thermodynamically favorable under aerobic conditions, local anoxia beneath individual cells is likely to further change an anaerobic
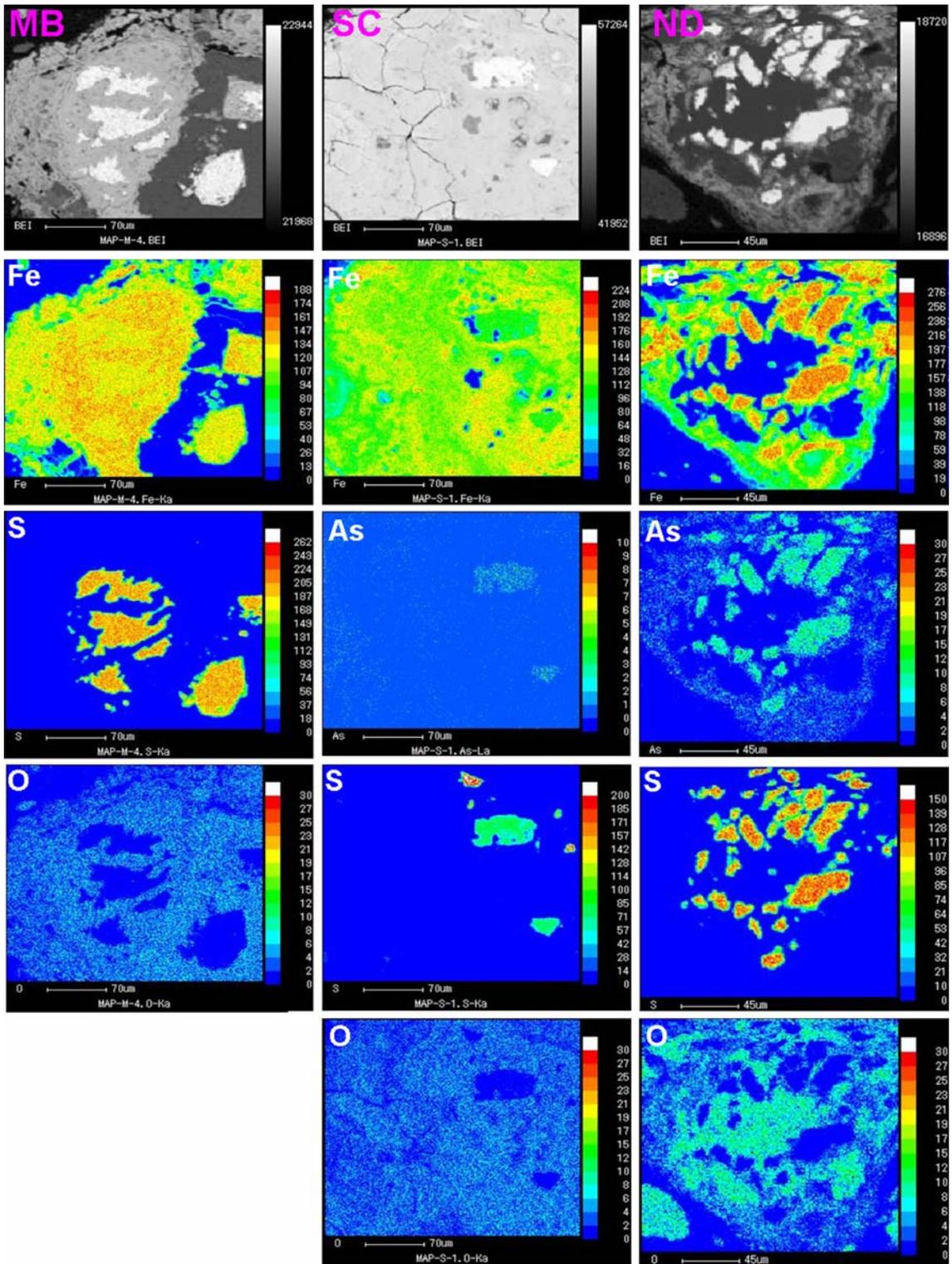
Fig. 3 Electron probe micro-analysis (EPMA) X-ray maps showing the distributions of Fe, As, S, and O in each sample collected at Myungbong (*MB*, *left*), Songcheon (*SC*, *middle*), and Nakdong (*ND*, *right*)
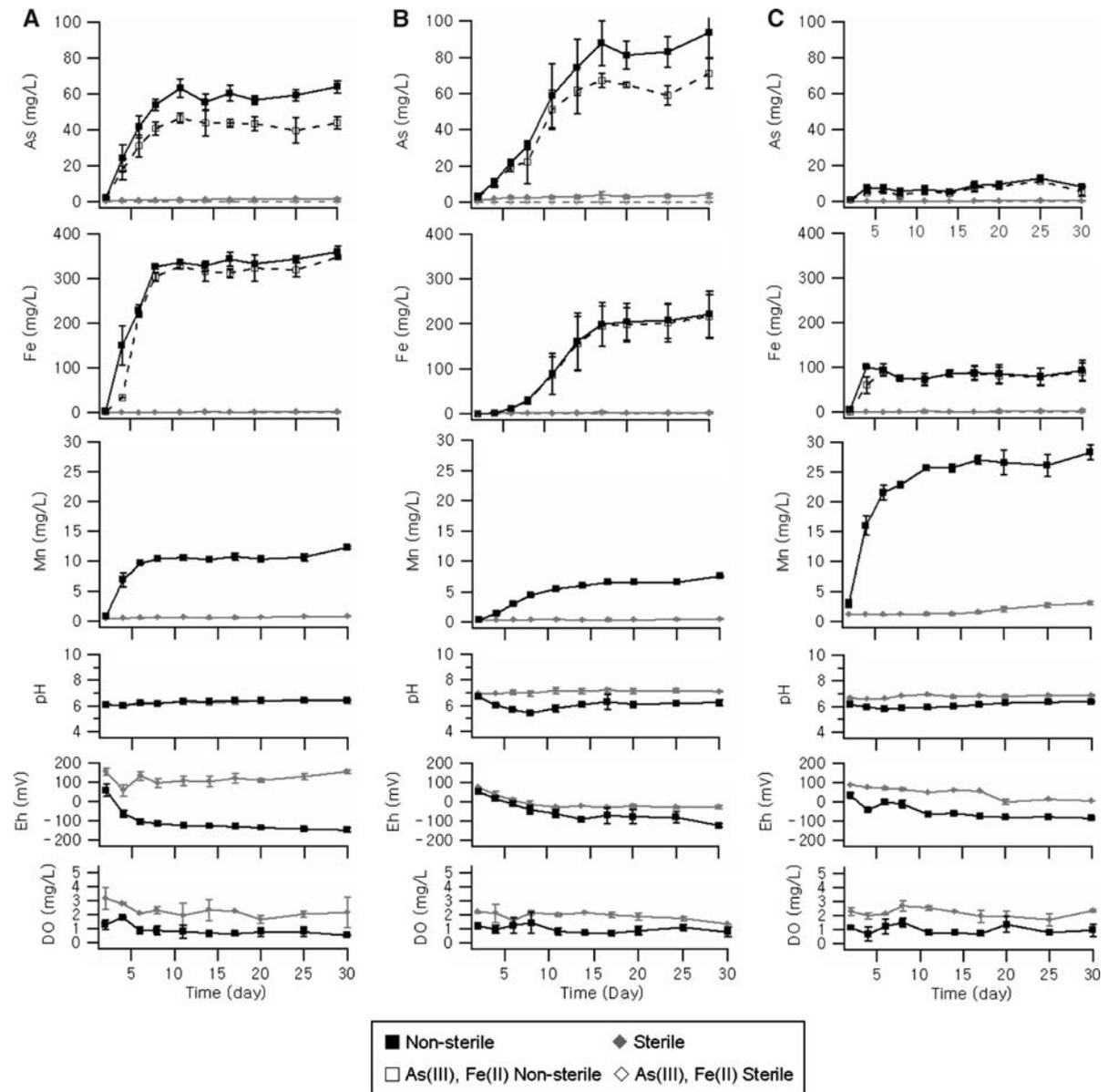
Fig. 4 Variations in the measured parameters and concentrations of As(total) (*solid line*), As(III) (*dotted line*), Fe(total) (*solid line*), Fe(II) (*dotted line*), and Mn(total) in the leaching solutions from the anaerobic experiments. **a** Myungbong, **b** Songcheon, **c** Nakdong. The *error bars* indicate the standard deviations of triplicate experiments. The *square* and *diamond* symbols correspond to the microbially mediated leaching and control experiments, respectively. *DO* Dissolved oxygen, *Eh* oxidation–reduction (redox) potential

environment (Grantham et al. 1997; Roso et al. 2003). The consumption of oxygen by aerobic microorganisms during the early stage of the experiments depleted the oxygen levels; dissimilatory reducing bacteria can play a major role in the leaching of Fe, Mn and As.

In addition, the decrease in the Fe, Mn and As concentrations in the solution after a few days indicate that dissolved ions can be reprecipitated or readsorbed with declining bacterial activity. In particular, dissolved Fe(II) is easily adsorbed onto the surface of metal oxides, which induces goethite
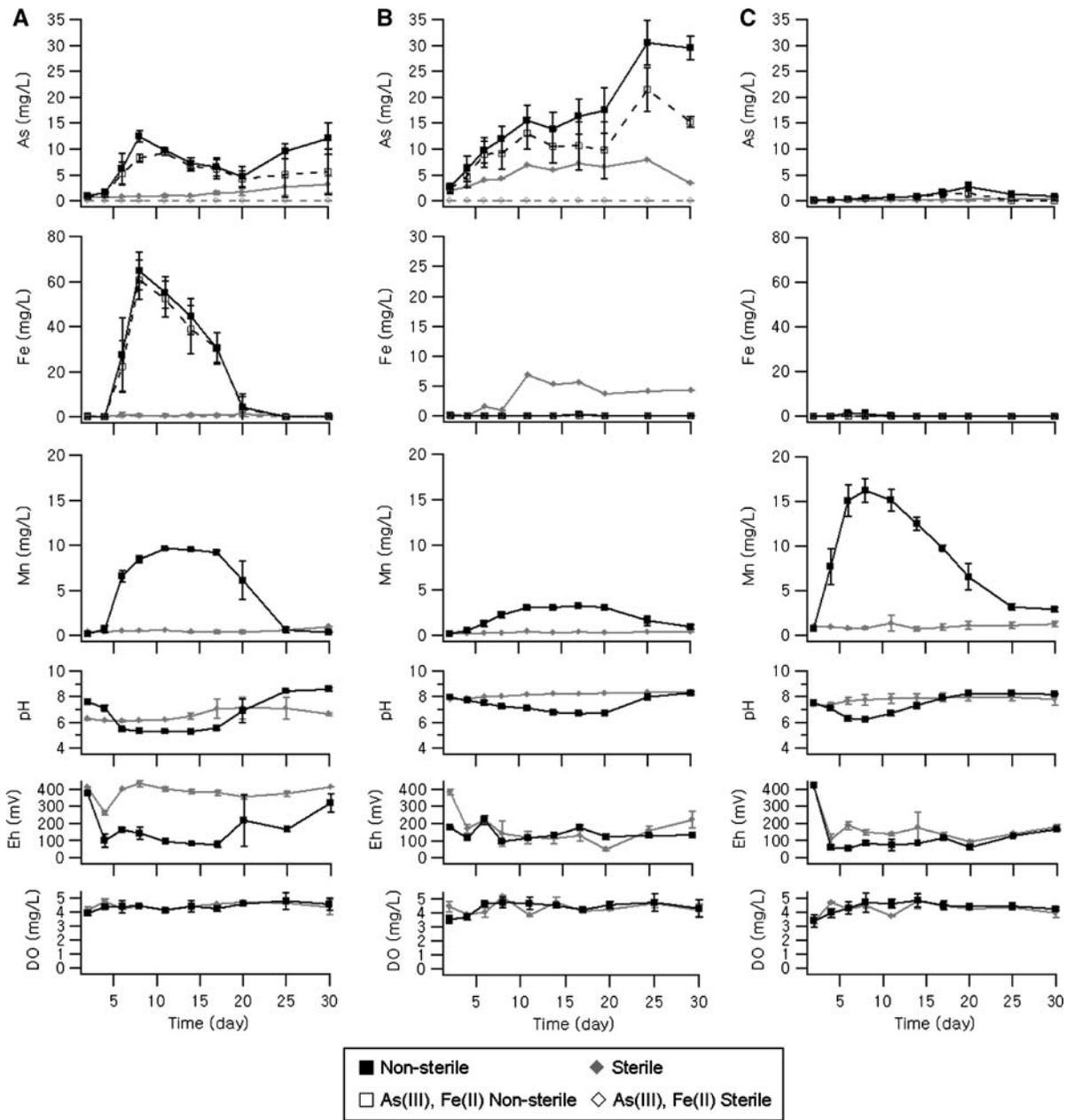
Fig. 5 Variations in the measured parameters and concentrations of As(total) (*solid line*), As(III) (*dotted line*), Fe(total) (*solid line*), Fe(II) (*dotted line*), and Mn(total) in the leaching solutions from the aerobic experiments. **a** Myungbong, **b** Songcheon, **c** Nakdong. The *error bars* indicate the standard deviations of triplicate experiments. The *square* and *diamond* symbols correspond to the microbially mediated leaching and control experiments, respectively. *DO* Dissolved oxygen, *Eh* oxidation–reduction (redox) potential

nucleation via the ferrihydrite-bearing systems (Cooper et al. 2006) as well by As. In the case of the ND tailings, which contained a relatively high concentration of labile Mn (Fig. 1), a large amount of dissolution was observed under both aerobic and

anaerobic conditions compared to that with Fe; this can be explained by the fact that the microbial reduction of Mn has priority over that of Fe, as previously mentioned (Scott and Morgan 1990). This finding leads us to conclude that the mobility of As in
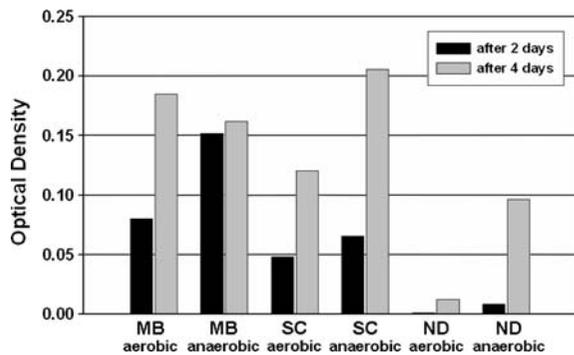
Fig. 6 Biomass measured during each experiment after 2 and 4 days

the tailing soils was significantly influenced by the fate of the dominant metals, such as Fe and Mn.

Conclusions

Compared to abiotic processes, the indigenous bacteria in the tailing soils promoted the degree of As dissolution in relation to the supply of carbon source under the experimental aerobic and anaerobic conditions tested in our study. The anaerobic environment was found to favor the prominent dissolution of As. These results demonstrate that the mobilization of As can be significantly increased from the oxygen-depleted portion of the tailing dump when microbially available organic substrates infiltrate. We postulate that the microbially enhanced lixiviation of As causes detrimental impacts in the local groundwater. The finding that the bacterial dissolution of Fe and Mn appeared to occur with the dissolution of As suggests the main mechanism involves the dissimilatory reductions of Fe(III), Mn(IV), and As(V) due to the presence of indigenous bacteria. Although the total As concentration was lower in the MB and SC tailing soils than in the ND tailing soil, the amount of As dissolved in the former two tailing soils was significantly larger than that in the ND tailing soil. The difference in the bioavailable portions of As between the tailings seems to be the result of the type and amount of secondary minerals or phases formed due to either oxidation or alteration of the primary minerals. Accordingly, the extent of microbially mediated leaching is speculated to be primarily influenced by the geochemical and mineralogical

features of each tailing soil. In addition, it is speculated that the biomass within the tailing soil is an important factor in terms of the dissolution of As. Our results therefore suggest that such a difference in the biogeochemistry of these tailing soils can influence differences in the environmental risk and extent of As leaching.

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