

Bioleaching of arsenic in a drinking water treatment process

Khondoker Mahbub Hassan, Kensuke Fukushi, Fumiyuki Nakajima and Kazuo Yamamoto

ABSTRACT

Arsenic is found to exist within the shallow zones of groundwater in many countries and a large number of drinking water treatment units have been installed to combat this arsenic poisoning. Microbial activity can greatly affect the mobilization of arsenic under anaerobic conditions when coupled with the oxidation of organic matter. In this study, the bioleaching of arsenic in response to organic matter inclusion in a drinking water treatment process has been elucidated. The decomposition of organic matter due to the microbial action has led to an anaerobic condition within the accumulated sludge in the treatment unit and hence caused the bioleaching of arsenic, above 130 $\mu\text{g/L}$, with the effluent water. Nevertheless, the biofilm structure, related to the oxygen penetration, had significant influence on the bioleaching of arsenic even in the circumferential aerobic condition of the biofilm. For a biofilm thickness around 400 μm , an anaerobic bottom part was noticed and hence a clear step-up concentration of arsenic was observed in the associated bulk liquid. This study suggests that organic matter would have a sufficiently important influence on the microbiological transformation of arsenic to warrant its consideration in designing safe remediation strategies in the context of arsenic removal processes for drinking water.

Key words | arsenic and iron treatment, biofilm, bioleaching, drinking water, organic matter

INTRODUCTION

Arsenic contamination of groundwater has emerged as a major public health problem in many countries like Argentina, Bangladesh, Cambodia, Canada, Chile, China, Germany, Hungary, India, Mexico, Mongolia, Myanmar, Nepal, Pakistan, Romania, Thailand, USA, Vietnam, etc., around the world (Mukherjee *et al.* 2006). At circumneutral pH values present in most environments, the predominant forms of inorganic arsenic are arsenate (As(V)) and arsenite (As(III)). Ahmann *et al.* (1997) found that As(III) is much less sorptive and more mobile than As(V). Considering the lethal impact of arsenic on human health, environmental authorities have taken a more stringent attitude towards the presence of arsenic in water. As the diagnosis and medication of arsenic-related diseases are difficult, the

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treatment of contaminated water as a preventive measure appears to be an effective alternative to combat arsenic poisoning. Use of granular ferric hydroxide (Driehaus *et al.* 1998), iron-impregnated activated carbon (Petruševski *et al.* 2002) and iron-manganese-amended activated alumina (Dhiman & Chaudhuri 2007) were found to be effective in removing the arsenic from groundwater. Based on the established biological iron oxidation from groundwater (Dimitrakos *et al.* 1997), arsenic removal by adsorption and co-precipitation onto the flocs of iron hydroxides and subsequent sand filtration has become a very popular technique. Moreover, there was an indication of As(III) oxidation by iron-oxidizing bacteria, leading to improved overall removal efficiency (Katsoyiannis & Zouboulis 2004).

Adopting this technique, different types of arsenic and iron removal units (AIRU) shown in Figure 1 were designed and installed in many regions. In a large-scale municipal water supply system in Bangladesh, having a treatment capacity around 3,000 m³/d (Ahmed 2001), this technique is termed an arsenic and iron removal plant (AIRP).

Microbial activity can greatly affect the mobilization of arsenic under anaerobic conditions by either an indirect or a direct mechanism (Zobrist *et al.* 2000). The former is the reductive dissolution of iron hydroxide minerals, leading to the release of associated arsenic into solution. The latter is the direct reduction of As(V) associated with a solid phase to the less adsorptive As(III). Dissimilatory arsenate-reducing bacterium (DARB), which achieve growth by the respiratory reduction of arsenate to arsenite, are able to mobilize arsenic via the reduction of arsenate contained in minerals or contaminated sediments. Oremland & Stolz (2003) reported that the reaction is energetically favorable when coupled with the oxidation of organic matter because the arsenate/arsenite oxidation/reduction potential is +135 mV. The bioleaching of arsenic is an oxidation–reduction process, where organic matter is oxidized to supply the required electrons and ferric iron or arsenate is reduced by acting as an electron acceptor. This process is

incorporated within a sequential terminal electron acceptor reaction framework, including aerobic, ferric-iron-reducing and arsenate-reducing processes (Lim *et al.* 2007). According to another study, the reduction of arsenate plays a relatively minor role in the solubilization of arsenic sorbed to iron hydroxides and arsenic release from contaminated soils and sediments proceeds considerably faster under conditions favoring dissimilatory reduction of ferric iron leading to the dissolution of sorbing phases (Langner & Inskeep 2000). Oremland & Stolz (2003) concluded that DARB isolated to date are physiologically diverse and capable of using arsenate as electron acceptors for growth and most of them are strict anaerobes. Because organic matter is not only potentially reactive toward arsenic but also ubiquitous in natural waters, typically found at concentrations between 1 and 50 mg/L as total organic carbon (TOC), its potential influence on arsenic mobility is great (Redman *et al.* 2002).

Nevertheless, biofilms consist of living cells, dead cells and cell debris in a matrix of extracellular polysaccharide (glycocalyx) attached to a surface. Biofilms are multiphase systems that consist of solids and of a liquid phase in the void space between the solids. Most biofilms are spatially heterogenous, characterized by complex assemblages of cell types and gradients of physical/chemical parameters

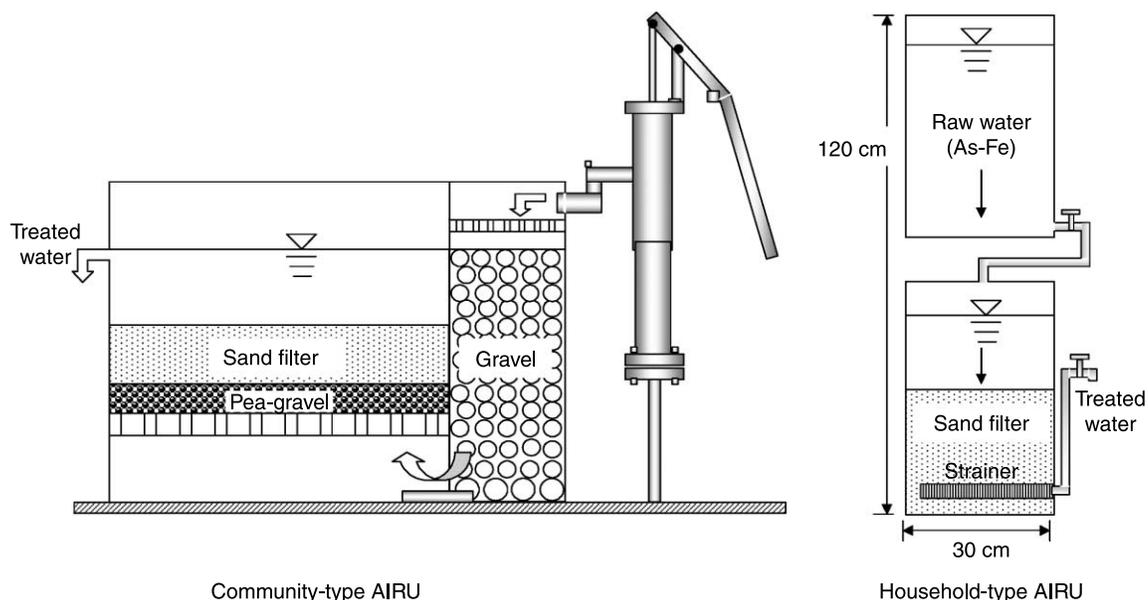


Figure 1 | Typical arsenic and iron removal units (AIRU) used in many regions.

(Rapporteur *et al.* 1989). As a result of these spatial gradients, the microbial species and density, the volume fraction of the water phase (porosity) and the tortuosity of the biofilms have to change as the depth of the biofilm increases. These spatial distributions of biotic and abiotic components in turn affect the mass transfer mechanisms and diffusivities in biofilms (Sun *et al.* 1989; Zhang *et al.* 1994). Biofilm structure is highly stratified. Density increases with depth, while porosity and the effective diffusivity for dissolved oxygen decrease with biofilm depth. Oxygen penetration, which is critical to the oxidation/reduction reactions of aerobic respiration, varies with the structure of the biofilm and the utilization rate of the cells (Bishop *et al.* 1995). In microelectrode studies for dissolved oxygen concentration across the depth of biofilm, fully anaerobic conditions were noticed at variable depths depending on the total thickness of the biofilm (Hibiya *et al.* 2004). The gradual development of biofilm on the settled sludge in the AIRU in the presence of organic matter may get thick enough to become anaerobic inside and hence could cause the bioleaching of arsenic with the effluent water.

This study revealed the bioleaching of arsenic from the accumulated sludge in the AIRU in response to organic matter inclusion in the treatment process. Two major aspects were focused on: (1) the microbial transformation of arsenic in anaerobic condition and (2) the influence of the biofilm structure, related to the oxygen penetration, on the bioleaching of arsenic.

METHODS

The bioleaching of arsenic in response to organic matter inclusion in the drinking water treatment process was investigated, using a simulated AIRU, in the laboratory. Contaminated groundwater was prepared artificially, using selected constituents, considering the purpose of this research work.

Preparation of artificial contaminated groundwater

The concentration of influent arsenate, having high adsorption capacity with the iron hydroxide solid phase, usually

determines the overall arsenic removal performance in the AIRU treatment process (Katsoyiannis & Zouboulis 2004). Thus, the contaminated groundwater was prepared artificially using arsenate (H_3AsO_4 standard solution, Merck) and ferrous sulfate ($\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$) in Milli-Q water and then the pH was adjusted to 7 by using sodium hydroxide (NaOH) reagent. In course of sludge accumulation within the filter bed of the laboratory-scale AIRU, arsenic and iron were spiked in the influent water to concentrations of 500 $\mu\text{g/L}$ and 5 mg/L, respectively, and operated for 10 bed volumes (volume of permeate/volume of filter bed) of effluent water. The selected 10 bed volumes, and hence a preliminary stage of the AIRU operational condition, was considered in this study to observe the potential bioleaching of arsenic in response to organic matter inclusion in the treatment process. In field AIRUs, the bioleaching of arsenic would be even severe due to the large amount of deposited sludge in the long-term operational condition of the treatment unit. In real situations, the organic contamination (as TOC) is mainly contributed by humic and fulvic acids originated through chemical and microbial decomposition of terrestrial and aquatic animals as well as plants. The humic and fulvic substances are not easily biodegradable and thus arsenate-reducing bacteria could hardly use them in the aquatic environment. However, several studies identified that the organic matter contamination in groundwater of the shallow reducing aquifer was frequently combined with high concentrations of nitrogen and phosphorus (Bhattacharya & Mukherjee 2001; Stollenwerk *et al.* 2007). The source of this contamination was suspected to be the wastewater from pit latrines as well as the grey water, which were highly biodegradable. Thus, in a laboratory study, the simulated wastewater organic matter stock solution was prepared, using tryptone (T), yeast extract (Y) and glucose (G) in pure water, following Standard Methods (American Water Works Association 2005) for plate count (G:Y:T = 1:2.5:5 wt/wt) excluding agar, which would give a preferable environment for the growth of bacteria. Laboratory reagent grade chemicals from Becton, Dickinson and Company (BD), USA were used in the above preparation. In another observation, an antibiotic, tetracycline hydrochloride 8 mg/L, was added to control the microbial activity (Berovic & Knezevic 1995) in the bioleaching process.

Simulation of the laboratory-scale AIRU

In order to minimize the hazardous and toxic waste generation during the research activity in the laboratory, a small cross-sectional area (diameter, $\phi = 25$ mm) was taken into consideration for the development of the laboratory-scale AIRU (Figure 2). A coarse-grained sand (effective size, $D_{10} = 0.4$ mm; uniformity coefficient, $U_C = 1.7$) filter bed having a depth of 300 mm was used for the reactor. The filtration rate was maintained at 0.6–0.7 m/h, which is the highest permissible filter loading rate for the slow sand filtration process (Montgomery Consulting Engineers Inc. 1985). The effluent was collected through a pipe connected at the bottom of the sand filter. The outlet of the effluent pipe was located at such a level that the filter bed would always be submerged under water, which is a common practice in the arsenic and iron removal units at the field level in Bangladesh and India (Ahmed 2001). A tracer study, spiking sodium chloride (NaCl) solution in the influent water, was carried out in the simulated AIRU in the laboratory. Normalized concentrations were then calculated as a fraction of the influent concentration. No short-circuiting was observed in this tracer study and a dispersed plug flow was evident.

In the real field situation, the AIRU is usually operated in both the “continuous flow” and the “intermittent flow” modes and occasionally it is kept inoperative for a few days.

The bioleaching of arsenic, in the presence of organic matter, from the accumulated sludge in the filter bed was observed under the inoperative condition of the AIRU. Subsequently the water samples were collected from the outlet of the effluent pipe for laboratory analyses. In an additional study to avoid atmospheric re-oxidation, in the bioleaching process, a flexible rubber cap was inserted within the AIRU at the top level of the supernatant water above the sand filter and allowing no headspace (Figure 2(c)). The rubber cap could move vertically along the cylindrical wall of the AIRU due to a suction effect while collecting the effluent water samples using a syringe.

Development of biofilm above the As–Fe sludge

In the AIRU treatment process, the As–Fe sludge is usually accumulated within the pore space of the granular sand media as well as the top surface of the sand filter bed. In response to organic matter inclusion in the treatment process, the biofilm would form in both locations of the As–Fe sludge. Due to the diverse spatial locations of the biofilm, the diffusivity of oxygen within the biofilm would not be identical and hence the bioleaching phenomena of arsenic would be different with different locations. Thus, in the laboratory study, to ensure a uniform environmental condition for the growth of the biofilm above a plain sludge surface, synthetic groundwater, contaminated with arsenic

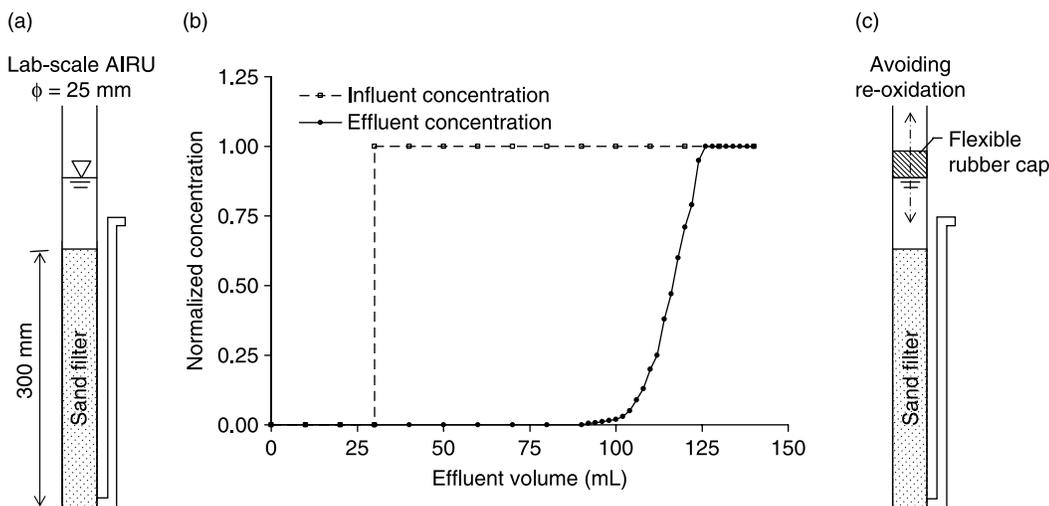


Figure 2 | Tracer study, spiking sodium chloride (NaCl) solution in the influent water, in laboratory-scale AIRU: (a) lab-scale AIRU, (b) experimental results of tracer study, where normalized concentrations are shown as a fraction of the influent concentration and (c) special experimental set-up of the lab-scale AIRU avoiding atmospheric re-oxidation.

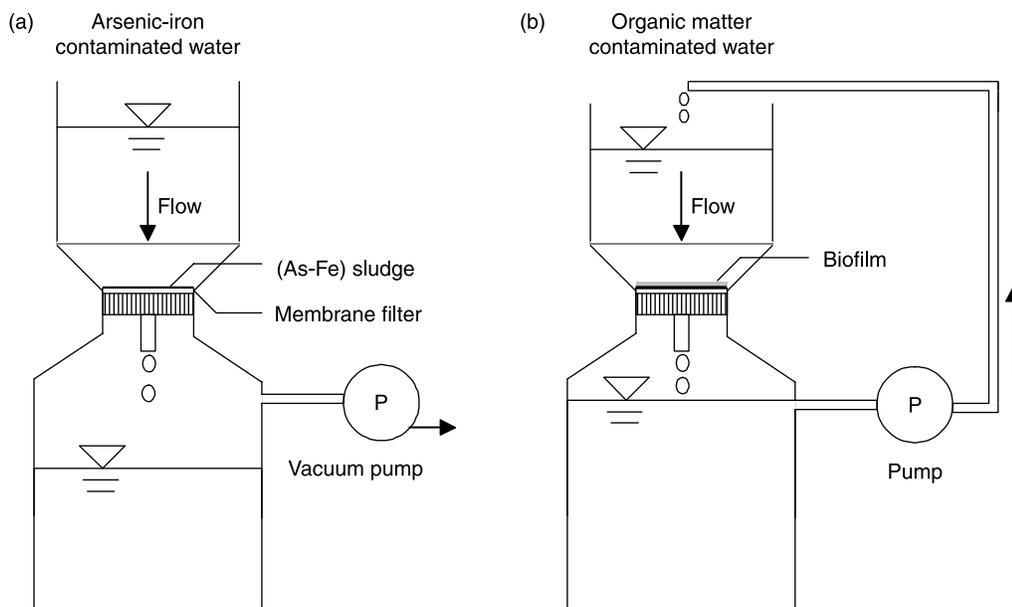


Figure 3 | Development of biofilm above the As-Fe sludge in membrane filter: (a) accumulation of As-Fe sludge in membrane filter and (b) formation of biofilm above the sludge surface.

and iron, was filtered through a $1.2\ \mu\text{m}$ pore sized nitrocellulose membrane (Figure 3). Due to aeration, the ferrous iron got oxidized to the insoluble ferric form and the aqueous arsenic was adsorbed on it. The formed As-Fe complex was then accumulated within the pore space and above the surface of the membrane and thus a plain sludge surface was obtained. The sludge membrane sheet was then shifted to a separate reactor, where simulated wastewater having organic matter concentration of $30\ \text{mg/L}$ as TOC was continuously re-circulated to facilitate the atmospheric aeration as well as the flow of water to develop the biofilm above the As-Fe sludge.

Analytical techniques

Laboratory analysis for total arsenic and iron concentrations in sample water was carried out by inductively coupled plasma mass spectrometry (ICP-MS, HP 4500, Yokogawa). The analytical detection limits for arsenic and iron were $0.3\ \mu\text{g/L}$ and $0.1\ \mu\text{g/L}$, respectively. For the isobaric interference of ArCl^+ in the arsenic analysis, the EPA recommended corrections (EPA Method 200.8) were applied. Arsenic species were quantified using high-performance liquid chromatography (HPLC) coupled with

ICP-MS (Ohno *et al.* 2007). An anion exchange column (Gelpack: GL-IC-A15, Hitachi Chemical Co. Ltd., Tokyo, Japan) and an eluting phosphate buffer ($5\ \text{mM}\ \text{KH}_2\text{PO}_4$) at pH 8.5 were used in this determination. The instrumental parameters were as follows: injection volume, $50\ \mu\text{L}$; flow rate, $1.5\ \text{mL/min}$; column temperature, 40°C . The value of pH was measured using a pH meter (HACH Co., USA). In the tracer study using NaCl, the electrical conductivity was measured with a conductivity meter (Twin Cond B-173, Horiba Ltd., Kyoto, Japan). The TOC concentrations in the simulated wastewater and in other water samples were determined by using a TOC analyzer (TOC- V_{CSH} , Shimadzu), employing high-temperature combustion catalyzed oxidation, which allowed a detection limit of $0.4\ \text{mg/L}$. Dissolved oxygen (DO) concentrations within the accumulated sludge in the sand filter bed as well as in the biofilm were observed using a microelectrode with a $10\ \mu\text{m}$ tip diameter and a micromanipulator having a vertical resolution of $10\ \mu\text{m}$ (OX10, MM33-2, Unisense). The developed biofilm over the settled sludge was taken out from the reactor and kept in the freezer at -80°C , so that it would remain in its original position while taking the horizontal sections using a microtome. The depth of slicing was fixed at $50\ \mu\text{m}$ for each layer starting from

the top to the bottom of the biofilm. For collecting samples of each layer, three different pieces of biofilm were used and they were kept in a plastic vial and the weight of the specific layer after collection was determined by a sensitive digital balance. The sliced biofilms were digested in a closed microwave system (Multiwave 3000, Perkin–Elmer) in 10% HNO₃ solution and in a temperature range of 165 ± 5°C for 20 min following Standard Methods (American Water Works Association 2005). After digestion, the water samples were adjusted to 1% HNO₃ concentration and filtered through a 0.45 µm PTFE filter for the analysis of arsenic and iron using ICP-MS.

RESULTS AND DISCUSSION

Microbial transformation of arsenic in laboratory-scale AIRU

The bioleaching of arsenic, in the presence of organic matter, from the accumulated sludge in the filter bed was monitored in the laboratory-scale AIRU under the inoperative condition. The initial decreasing pattern of the effluent arsenic concentration (Figure 4(a)) might be due to less stable iron–arsenic complexes, which were driven out at the first stage. In addition to this, biological iron oxidation (Dimitrakos *et al.* 1997) in aerobic conditions would contribute to the greater retention of iron–arsenic sludge. However, afterwards, significantly high concentrations of arsenic in the AIRU effluent water were observed in the case of simulated wastewater inclusion in feed water. In course of the biodegradation of organic matter (Figure 4(c)), aerobic oxidation was expected to precede the other reactions because oxygen reducers would derive more energy (i.e. higher Gibbs free energy) from the substrate than the iron and arsenic reductions. Thus, the depletion of dissolved oxygen due to microbial action led to an anaerobic-reducing environment within the accumulated sludge in the filter bed and hence caused the bioleaching of arsenic by anaerobic bacteria. The ratio of As (III)/As (V) concentrations in the effluent water was around 1.10 (data not shown), indicating the microbial reduction and dissolution of the ferric hydroxide sorbing phase as well as the dissimilatory arsenate reduction.

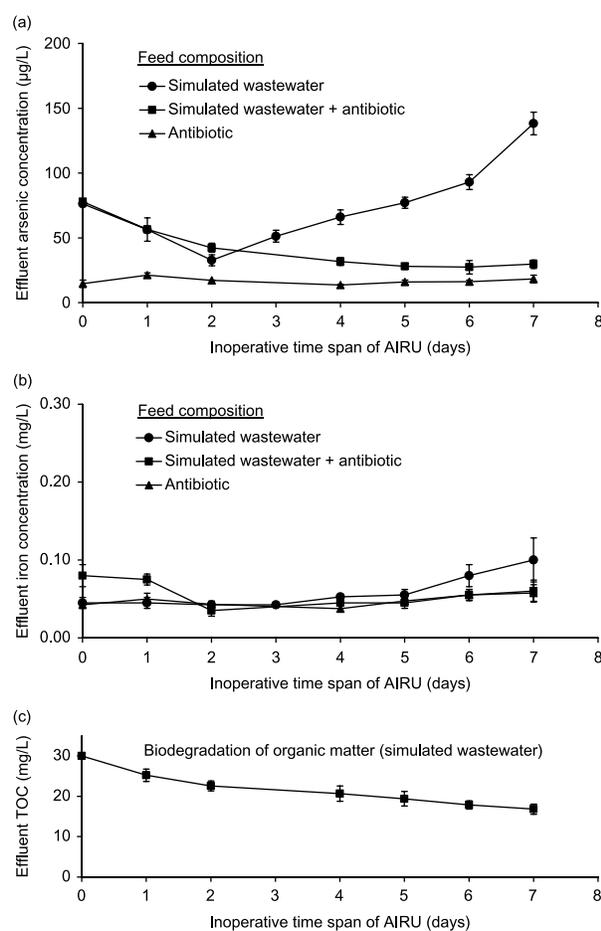


Figure 4 | Effect of organic matter, using simulated wastewater of TOC concentration 30 mg/L, on the bioleaching of arsenic and iron from the accumulated sludge in the lab-scale AIRU under the inoperative condition: (a) bioleaching of arsenic, (b) bioleaching of iron and (c) biodegradation of organic matter. Symbols represent the average of triplicate samples and error bars show the range of standard deviation.

In contrast, the effluent iron concentrations in the bioleaching experiment were found insignificant and the concentrations never exceeded 0.15 mg/L (Figure 4(b)).

Thus, it may be considered that the arsenic–iron sludge, retained mostly in the top portion of the sand filter bed, caused the bioleaching of arsenate, arsenite and ferrous iron under anaerobic conditions. While collecting the effluents, the aqueous ferrous iron, reduced in the bioleaching process, re-oxidized to insoluble ferric form due to the intrusion of supernatant aerobic water into the sand filter bed and was trapped by the mechanical straining mechanism, whereas a significant portion of arsenate and arsenite escaped due to inadequate iron hydroxide sorption sites in the bottom part of the sand filter bed, and they

eventually appeared with the effluent water. In this process, re-adsorption of arsenate onto the re-oxidized ferric hydroxides would also occur to some extent.

In order to verify the above speculation, “re-oxidation of the reduced ferrous iron to insoluble ferric form due to the intrusion of supernatant aerobic water into the sand filter bed”, a special experimental set-up in the AIRU was produced (Figure 2(c)) to avoiding atmospheric aeration. In this special study, in comparison to the previous ordinary experimental set-up, significantly higher concentrations of both arsenic and iron were observed in the effluent water using similar organic matter concentrations in the AIRU feed water (Figure 5). Possibly in this case the supernatant water above the sand filter bed remained anoxic due to the microbial activity in the biodegradation of organic matter. Consequently, high concentrations of arsenic and iron appeared with the effluent water. The effect of the antibiotic itself in the leaching process of arsenic and iron was also studied and it was found that the effluent arsenic concentrations were always less than 20 µg/L (Figure 4(a)). In the case of the controlled microbial activity, using antibiotic in addition to the simulated wastewater, the effluent arsenic concentrations were found to be slightly higher in comparison to the antibiotic itself. The sorption equilibrium, established in the arsenic–iron sludge, might be interrupted due to the competing anions in the organic matter (Redman *et al.* 2002) and consequently the desorbed arsenic would appear with the effluent water.

With the intention to make sure that the bioleaching of arsenic from the accumulated sludge in the filter bed

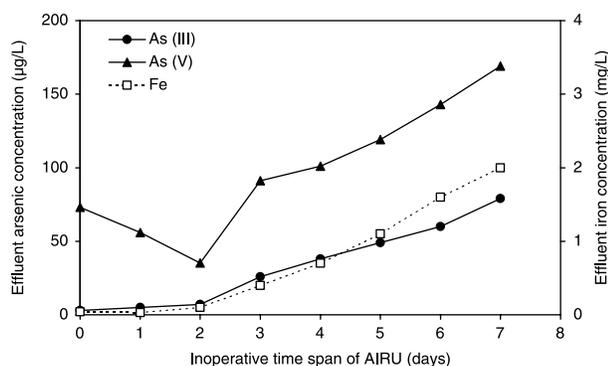


Figure 5 | Effect of organic matter, using simulated wastewater of TOC concentration 30 mg/L, on the bioleaching of arsenic and iron from the accumulated sludge in the lab-scale AIRU under the inoperative condition in a special experimental set-up, avoiding atmospheric re-oxidation.

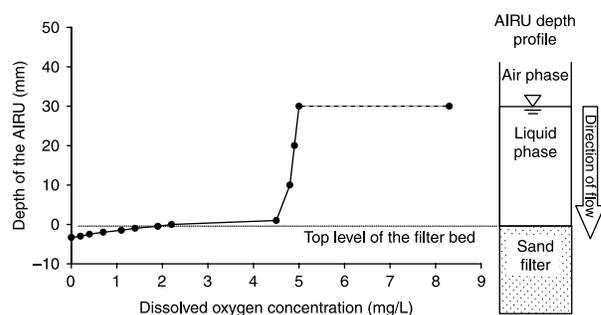


Figure 6 | Depletion of dissolved oxygen concentrations in the lab-scale AIRU in the bioleaching observation time on the seventh day due to the biodegradation of simulated wastewater organic matter of TOC concentration 30 mg/L.

occurred in the anaerobic conditions, the dissolved oxygen (DO) concentration profile in the lab-scale AIRU was observed in the ordinary experimental set-up using simulated wastewater in the feed water. The theoretical value of the air-saturated DO concentration was 8.3 mg/L (Figure 6). In the course of the biodegradation of organic matter, aerobic oxidation resulted in the depletion of DO concentration to 5–4.5 mg/L in the liquid phase. Within the sand filter bed, located at the bottom of the liquid phase, although the deoxygenation rate was similar the reoxygenation rate was very slow through the oxygen diffusion. Moreover, the accumulation of arsenic–iron sludge within the pore space of the water-saturated sand filter bed resulted in a limited diffusivity of oxygen and hence shifted to an anaerobic condition within 3.3 mm depth of the filter bed. Thus, it was evident that the bioleaching of arsenic and iron from the accumulated sludge in the sand filter bed was executed through the activity of anaerobic bacteria.

Biofilm structure related to the bioleaching of arsenic

The spatial distributions of biotic and abiotic components in biofilms affect the mass transfer mechanisms and diffusivities. Oxygen penetration was found to vary with the structure of the biofilm. Oxygen penetration depth was determined from the obtainable dissolved oxygen (DO) concentration profile across the depth of the biofilm. A completely anaerobic condition was noticed at the bottom part of the biofilm, at a depth greater than 200 µm (Figure 7). In the anaerobic condition, at the bottom part of the biofilm which was connected to the As–Fe sludge, microbial activity might lead to the bioleaching of arsenic

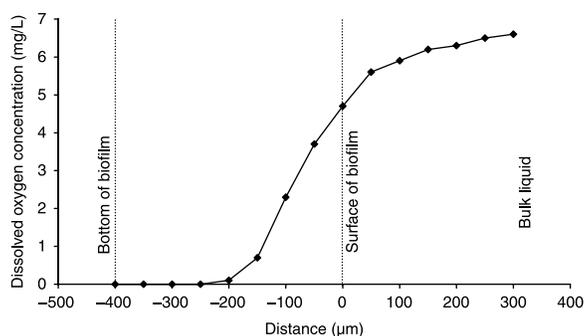


Figure 7 | Dissolved oxygen (DO) concentration profile across the depth of biofilm above the settled As–Fe sludge in presence of organic matter in the bulk liquid.

in the associated bulk liquid phase. However, the thin biofilm without an anaerobic zone was effective for the bio-oxidation process of organic matter from the standpoint of obtaining a larger aerobic zone throughout the entire biofilm.

In the course of biofilm development over the As–Fe sludge in the presence of organic matter, the bioleaching of arsenic and iron were monitored in the associated bulk liquid phase (Figure 8). Starting from the initiation of the experiment until the formation of the biofilm to a depth of 170 μm , the As(V) concentration was found around 20 $\mu\text{g/L}$. The sorption equilibrium, established in the As–Fe sludge, might be interrupted due to the organic matter inclusion in the bulk liquid and hence the desorbed arsenic would appear in the bulk liquid phase. When the biofilm thickness reached a depth of around 200 μm , a clear step-up

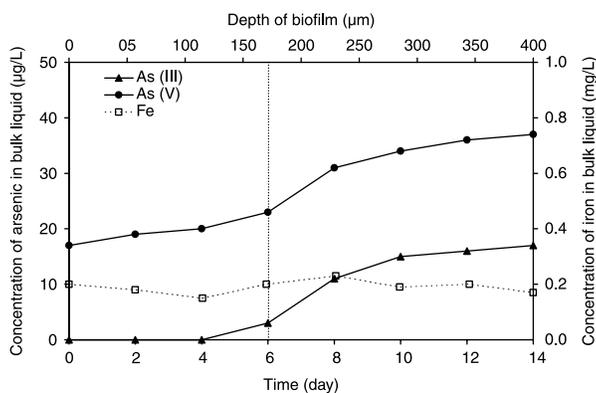


Figure 8 | Effect of biofilm structure, related to oxygen penetration, on the bioleaching of arsenic and iron from the underlying As–Fe sludge in the presence of organic matter in the bulk liquid. Dotted line represents the variation of iron concentration (secondary Y axis) in the bulk liquid with the gradual development of biofilm (secondary X axis). The time of observation was counted from initiation of this biofilm study.

pattern of the As(V) concentration was noticed in the bulk liquid. At this time, the bottom part of the biofilm was shifted to an anaerobic condition (Figure 7) which promoted the reductive dissolution of iron hydroxide minerals due to microbial activity (Zobrist *et al.* 2000), leading to the release of associated arsenic into solution. However, the iron concentration in the bulk liquid phase was not worth mentioning and found to be less than 0.2 mg/L throughout the study. The As(III) concentration in the bulk liquid phase was first detected in such a condition of the developing biofilm, at a thickness of 170 μm , that the dissolved oxygen concentration was almost depleted, being less than 1 mg/L, in the bottom part. Due to this enormous deficiency in dissolved oxygen concentration, it seemed that the activation of the dissimilatory As(V)-reducing bacterium was initiated. Subsequently, the As(III) concentration was found to be boosted up in the complete anoxic condition at the lower level of the biofilm. The As(III) concentration was found to be far less than the As(V) in the liquid phase, which was analogous to another study (Langner & Inskeep 2000).

From the stoichiometric point of view, the arsenate reduction did not occur corresponding to the decreasing concentration of TOC in the bulk liquid, because the aerobic condition was prevalent in the bulk liquid as well as in the top part of the biofilm while the anaerobic condition was identified only at the bottom portion of the biofilm. Although the concentration of As(V) decreased with anaerobic reduction, simultaneously reduction of the iron hydroxide solid phase from the underlying As–Fe sludge led to the release of As(V) in the bulk and anaerobic biofilm (Figures 8 and 9). However, the arsenate and iron

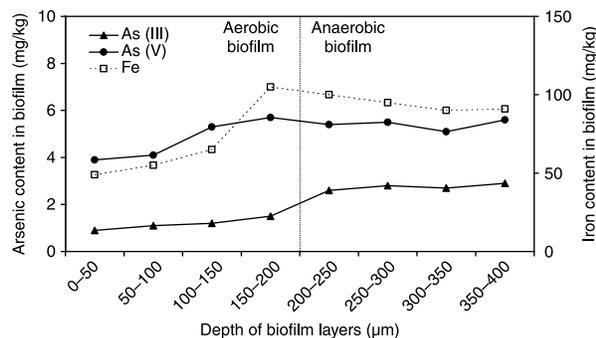


Figure 9 | Arsenic and iron contents in the sliced biofilm layers from the underlying As–Fe sludge in the presence of organic matter in the bulk liquid.

reduction, in the anaerobic condition, had a minor role in the overall TOC concentration in the effluent water. We did not have any experimental check for this explanation.

The arsenic and iron contents were analyzed in the sliced biofilm layers at different depths. The aerobic and anaerobic zones of the observed biofilm were found to be prevailing around the depths of 0–200 μm and 200–400 μm , respectively (Figure 9). The aerobic portion of the biofilm was found to have less accumulation of arsenic and iron content in comparison to the anaerobic part. The interface between the aerobic and anaerobic parts of the biofilm was found to have the maximum amount of arsenic and iron content. In the anaerobic condition, prevailing at the bottom part of the biofilm, microbial activity caused the reductive dissolution of iron hydroxide minerals, leading to the release of associated arsenic into solution, along with dissimilatory As(V) reduction. On reaching the aerobic portion of the biofilm at the top part, the aqueous ferrous iron re-oxidized to the insoluble ferric form and got wrapped with the biofilm. Re-adsorption of As(V) with this metal hydroxide solid phase may also occur to some extent. Possibly, this is the reason why the arsenic and iron contents were found to be higher in the anaerobic part of the biofilm and maximum at the anaerobic/aerobic interface.

CONCLUSIONS

The microbiological importance of organic matter interactions for arsenic mobility is large. This study revealed several consistent phenomena regarding the consequence of organic matter inclusion in feed water on the bioleaching of arsenic in the drinking water treatment process. There were two major aspects: (1) the microbial transformation of arsenic in anaerobic conditions and (2) the influence of the biofilm structure, related to the oxygen penetration, on the bioleaching of arsenic. In summary, the following conclusions can be drawn:

- In the inoperative condition of the AIRU, the biodegradation of organic matter led to an anaerobic condition within the accumulate sludge in the filter bed, which was evident in the microelectrode examination of the dissolved oxygen concentration.
- A high concentration of bioleached arsenic was noticed in the anaerobic condition of the sludge in the filter bed. However, the iron concentration was not worth mentioning in the bioleaching process.
- The thin aerobic biofilm over the As–Fe sludge did not show any bioleaching of arsenic in the associated liquid phase;
- An anaerobic bottom part, at depths greater than 200 μm , was noticed in the thick biofilm and in this case the bioleaching of arsenic was observed in the bulk liquid.
- In the sliced biofilm layers, the arsenic and iron contents were found to be higher in the anaerobic part in comparison to the aerobic portion of the biofilm and especially a maximum at the anaerobic/aerobic interface.

Nevertheless, these basic observations suggest that the microbial transformation of arsenic in response to organic matter inclusion in the treatment process would have a sufficiently important influence to affirm its consideration in designing safe remediation techniques for drinking water.

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