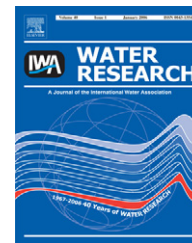


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Mathematical model for predicting microbial reduction and transport of arsenic in groundwater systems

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ARTICLE INFO

Article history:

Received 1 July 2006

Received in revised form

2 February 2007

Accepted 15 February 2007

Available online 30 March 2007

Keywords:

Arsenic

Microbial reduction

Precipitation

Reactive transport

Iron-reducing bacteria

Shewanella sp

ABSTRACT

A mathematical model was developed for describing the transport of arsenic, coupled with microbially-mediated biogeochemical processes. The biogeochemical characteristics of arsenic reactive transport processes were investigated in both batch and column tests, which showed that As(V) was reduced to As(III) by *Shewanella* sp., with the reduced arsenic species subsequently removed by precipitation. The breakthrough data obtained from the column experiments were used for the calibration of the arsenic reactive transport model. The reactive transport model, which only incorporated microbial reduction processes, showed a large discrepancy in predicting the observed As(III) concentration profiles, particularly later in the experiments. However, the model matched the experimental data much better with the inclusion of a term describing the precipitation process. Our results indicated that the precipitation reaction can be a major sink during microbially-mediated arsenic reactive transport. The proposed model provides a useful framework for predicting the transport of arsenic in saturated groundwater aquifers.

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1. Introduction

Arsenic from both anthropogenic and geologic sources is commonly found at trace levels within the subsurface, where it may exist in either the arsenopyrite (FeAsS), realgar (AsS) or orpiment (As₂S₃) forms (Newman et al., 1998). Arsenic is carcinogenic, and can also cause other human health effects, such as black disease and diabetes (Smedley and Kinniburgh, 2002). Understanding the mobility of arsenic in subsurface environments is important for evaluating its possible environmental and economic effects (Williams et al., 2003). The World Health Organization (WHO) suggests a maximum concentration limit (MCL) for arsenic in drinking water of 10 µg/L (WHO, 1993). This stringent standard has a significant impact on the management of arsenic contaminated sites,

since the technology for removing arsenic from groundwater may not be readily available at a local level.

Arsenic can be present in both organic and inorganic forms, and has four potential oxidation states (−3, 0, +3 and +5). In natural systems, arsenic mostly exists in inorganic forms, as oxyanions of arsenite, As(III), or as arsenate, As(V) (Adriano, 2001). In general, the inorganic forms of arsenic are more toxic than the organic forms. The inorganic forms of As(V) and As(III) have different sorption properties and toxicity levels (Anderson and Bruland, 1991; Bowell, 1994). As(III) is always more toxic than As(V) (Matschullat, 2000). Under oxidizing conditions, such as in well-mixed or oligotrophic surface waters, the predominant inorganic form of arsenic is arsenate (As(V) as H₂AsO₄[−] and HAsO₄^{2−}). On the other hand, under reducing conditions, such as ground water in an

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doi:10.1016/j.watres.2007.02.017

organic-rich environment, the predominant form is arsenite (As(III) as H_3AsO_3^0 and H_2AsO_3^-) (Oremland and Stolz, 2003). As(V) can be strongly adsorbed onto the surface of several common minerals, such as ferrihydrite; whereas, As(III) adsorbs less strongly to these minerals. Therefore, in natural systems, As(III) is more mobile than As(V) (Oremland and Stolz, 2003). The distribution of arsenic species [As(III) and As(V)] in natural water is highly dependent on the redox potential (Eh) and pH values (Smedley and Kinniburgh, 2002).

Since sorption affects the mobility of arsenic in ground water systems, several controlled laboratory studies have been conducted to understand the sorption of arsenic species onto various types of soil mineral. These studies have reported that As(III) primarily sorbs onto sulfide minerals and metal oxides/oxyhydroxides (Dzombak and Morel, 1990; Huerta-Diaz and Morse, 1992). Islam et al. (2004) suggested that arsenic adsorbed onto sediment surfaces could be mobilized into groundwater by anaerobic respiration of Fe(III) reducing bacteria. In another study, Newman et al. (1997) reported that the As(V)-reducing bacterium, *D. auripigmentum*, could simultaneously reduce As(V) and SO_4 , producing H_2S due to metabolic activity, which could precipitate the reduced As(III).

Previous studies on arsenic reactive transport have considered either the batch-scale equilibrium sorption or column-scale coupled adsorptive transport under abiotic conditions (Williams et al., 2003; Darland and Inskeep, 1997). None of these studies have considered the effects of the microbial reaction kinetics. Since microbial processes have a major influence on the fate of arsenic in aquifers (Newman et al., 1998), it is necessary to develop a comprehensive reactive transport model that can simultaneously describe microbially-mediated bio-chemical reactions as well as other advection–dispersion processes.

The objectives of this study were to develop and calibrate a reactive transport model that can describe the microbially-mediated reduction of As(V) to As(III) in saturated groundwater aquifers. The model was calibrated using both batch and column data. The biogeochemical kinetics was developed based on the observations from batch experiments, where a bacterial population was allowed to degrade organic matter by the consumption of arsenic as an electron acceptor. The kinetic reaction model was coded as a reaction module within the reactive transport model, RT3D (Clement, 1997; Clement et al. 1998). The results obtained from column experiments were then used to test the performance of the developed reactive transport model.

2. Model development

2.1. Conceptual model

Haque and Johannesson (2006) investigated the evolution of arsenic species along the path of groundwater flow in Florida, USA. Their study revealed that As(V) was the dominant species near the recharge area, and the concentration of As(III) progressively increased along the flow path, where the redox conditions were changed from oxic to anoxic. Further down-gradient, where the sulfide concentrations produced

due to microbially-mediated SO_4 reduction were considerably high, the reduced arsenic was removed from groundwater. The field data showed that the microbial oxidation of organic matter, reductive dissolution of Fe(III) oxides/oxyhydroxides, and SO_4 reduction and pyrite precipitation reactions influenced the evolution of arsenic and its speciation along the flow path. Newman et al. (1997, 1998) also indicated that microorganisms play critical roles in both the reduction and removal of As(V) from groundwater.

In our conceptual model, As(V) was assumed to be the dominant species present in natural aerobic systems. The influx of natural or anthropogenic organic substrates was assumed to promote microbial processes, which would result in the depletion of oxygen and a lowering of the redox potential. Under reducing condition, As(V)-reducing bacteria would respire adsorbed and/or dissolved As(V), with its conversion to As(III) (Oremland and Stolz, 2003; Haque and Johannesson, 2006) and subsequent removal by the precipitation process.

2.2. Mathematical model

2.2.1. Solute transport

Solute transport equations describe both the multi-dimensional fate and transport of aqueous- and solid-phase species in saturated porous media (Clement, 1997):

$$\frac{\partial C_k}{\partial t} = \frac{\partial}{\partial x_i} \left(D_{ij} \frac{\partial C_k}{\partial x_j} \right) - \frac{\partial}{\partial x_i} (v_i C_k) + \frac{q_s}{\phi} C_{sk} \pm r_k, \quad k = 1, 2, \dots, m, \quad (1)$$

$$\frac{\partial \tilde{C}_{im}}{\partial t} = \pm \tilde{r}_{im}, \quad im = 1, 2, \dots, (n - m), \quad (2)$$

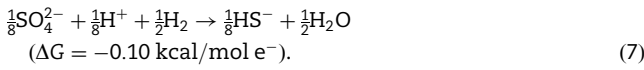
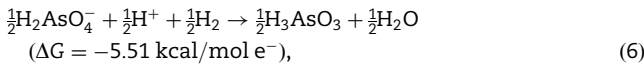
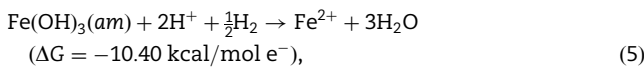
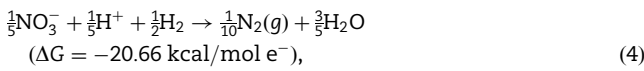
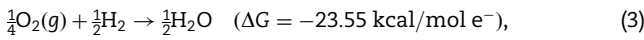
where n is the total number of species, m the total number of aqueous-phase or mobile species, $(n-m)$ the total number of solid-phase or immobile species, C_k the aqueous-phase concentration of the k th species (mg/L), \tilde{C}_{im} the solid-phase concentration of the im th species (mg/kg), D_{ij} the hydrodynamic dispersion coefficient (cm^2/h), v_i the pore water velocity (cm/h), q_s the volumetric flux of water per unit volume of the aquifer that represents the sources and sinks ($\text{cm}^3/\text{h}/\text{cm}^3$), ϕ the porosity, C_{sk} the concentration of source/sinks (mg/L), r_k represents all the aqueous-phase reaction rate terms (mg/L/h) that describe the mass of the species removed or produced per unit volume over time, and \tilde{r}_{im} represents all the solid-phase reaction rate terms (mg/kg/h).

The RT3D model requires the groundwater flow code, MODFLOW (A MODular three-dimensional finite-difference groundwater FLOW model), to generate a groundwater head distribution, and adopts the operator-split strategy, which allows the user to define various reaction models (Clement, 1997; Clement et al., 2000). A new kinetic model was developed, and then incorporated into the RT3D code in order to describe the fate and transport of arsenic in the subsurface environment.

2.2.2. Arsenic transformation and transport

As described in the conceptual model, the arsenic transformation reaction is modeled as an oxidation–reduction process, where a carbonaceous substrate is oxidized to supply

the required electrons and As(V) is reduced by acting as an electron acceptor. The As(V)-reduction process is incorporated within a sequential terminal electron acceptor reaction modeling framework, including aerobic, denitrifying, Fe(III)-reducing, As(V)-reducing and sulfate-reducing processes (note that the above processes are listed in the order in which they are expected to occur). Using H₂ (produced from the carbon source) as the ultimate electron donor (hereafter referred to as ED) and O₂, NO₃⁻, Fe(III), As(V), and SO₄²⁻ as electron acceptors (hereafter referred to as EA), the stoichiometry of different degradation processes can be described by the following set of biochemical reactions (Newman et al., 1998):



Several field studies have shown that first-order biodegradation kinetics can be used to approximate ED degradation processes via various pathways (Lu et al., 1999; Wiedemeier et al., 1996). Using this approach, a first-order decay term was used to describe the ED degradation process, a Monod term to account for the presence of an appropriate electron acceptor, and an inhibition term to simulate inhibition due to the presence of one of the electron acceptors earlier in the reaction sequence (i.e., an EA with higher Gibbs free energy). For example, in the presence of oxygen, nitrate or Fe(III), the reduction of As(V) is generally inhibited, because oxygen, nitrate, and ferric iron reducers derive more energy from the substrate than the As(V)-reducers. The kinetic models for the bio-chemical system can be written as follows:

$$r_{ED,O_2} = k_{ED,O_2} \cdot [ED] \cdot \left(\frac{[O_2]}{K_{EA,O_2} + [O_2]} \right), \quad (8)$$

$$r_{ED,NO_3^-} = k_{ED,NO_3^-} \cdot [ED] \cdot \left(\frac{[NO_3^-]}{K_{EA,NO_3^-} + [NO_3^-]} \right) \cdot \left(\frac{k_{i,O_2}}{k_{i,O_2} + [O_2]} \right), \quad (9)$$

$$r_{ED,Fe(III)} = k_{ED,Fe(III)} \cdot [ED] \cdot \left(\frac{[Fe(III)]}{K_{EA,Fe(III)} + [Fe(III)]} \right) \cdot \left(\frac{k_{i,O_2}}{k_{i,O_2} + [O_2]} \cdot \frac{k_{i,NO_3^-}}{k_{i,NO_3^-} + [NO_3^-]} \right) \quad (10)$$

$$r_{ED,As(V)} = k_{ED,As(V)} \cdot [ED] \cdot \left(\frac{[As(V)]}{K_{EA,As(V)} + [As(V)]} \right) \cdot \left(\frac{k_{i,O_2}}{k_{i,O_2} + [O_2]} \cdot \frac{k_{i,NO_3^-}}{k_{i,NO_3^-} + [NO_3^-]} \cdot \frac{k_{i,Fe(III)}}{k_{i,Fe^{3+}} + [Fe(III)]} \right), \quad (11)$$

$$r_{ED,SO_4^{2-}} = k_{ED,SO_4^{2-}} \cdot [ED] \cdot \frac{[SO_4^{2-}]}{K_{EA,SO_4^{2-}} + [SO_4^{2-}]} \cdot \left(\frac{k_{i,O_2}}{k_{i,O_2} + [O_2]} \cdot \frac{k_{i,NO_3^-}}{k_{i,NO_3^-} + [NO_3^-]} \cdot \frac{k_{i,Fe(III)}}{k_{i,Fe(III)} + [Fe(III)]} \cdot \frac{k_{i,As(V)}}{k_{i,As(V)} + [As(V)]} \right), \quad (12)$$

where r_{ED,O_2} is the ED (i.e., natural organic matter or introduced carbon substrate) degradation rate using oxygen as the EA (mM/h), r_{ED,NO_3^-} the degradation rate using nitrate (mM/h), $r_{ED,Fe^{3+}}$ the degradation rate using Fe(III) (mM/h), $r_{ED,As(V)}$ the degradation rate using As(V) (mM/h), $r_{ED,SO_4^{2-}}$ the degradation rate using sulfate (mM/h), [O₂] the oxygen concentration (mg/L), k_{ED,O_2} the first-order degradation rate constant (h⁻¹) for the ED using oxygen as the EA, K_{EA,O_2} the half saturation constant for oxygen (mg/L), and k_{i,O_2} the oxygen inhibition constant (mg/L); similar nomenclature is used in subsequent reactions.

The transport equations for the EDs and various EAs in saturated porous media are written as follows:

$$R_{ED} \frac{\partial [ED]}{\partial t} = \frac{\partial}{\partial x_i} \left(D_{ij} \frac{\partial [ED]}{\partial x_j} \right) - \frac{\partial (v_i [ED])}{\partial x_i} + \frac{q_s}{\phi} [ED]_s - r_{ED,O_2} - r_{ED,NO_3^-} - r_{ED,Fe(III)} - r_{ED,As(V)} - r_{ED,SO_4^{2-}}, \quad (13)$$

$$R_{O_2} \frac{\partial [O_2]}{\partial t} = \frac{\partial}{\partial x_i} \left(D_{ij} \frac{\partial [O_2]}{\partial x_j} \right) - \frac{\partial (v_i [O_2])}{\partial x_i} + \frac{q_s}{\phi} [O_2]_s - Y_{O_2/ED} \cdot r_{ED,O_2} \quad (14)$$

$$R_{NO_3^-} \frac{\partial [NO_3^-]}{\partial t} = \frac{\partial}{\partial x_i} \left(D_{ij} \frac{\partial [NO_3^-]}{\partial x_j} \right) - \frac{\partial (v_i [NO_3^-])}{\partial x_i} + \frac{q_s}{\phi} [NO_3^-]_s - Y_{NO_3^-/ED} \cdot r_{ED,NO_3^-}, \quad (15)$$

$$R_{Fe(II)} \frac{\partial [Fe(II)]}{\partial t} = \frac{\partial}{\partial x_i} \left(D_{ij} \frac{\partial [Fe(II)]}{\partial x_j} \right) - \frac{\partial (v_i [Fe(II)])}{\partial x_i} + \frac{q_s}{\phi} [Fe(II)]_s + Y_{Fe(II)/ED} \cdot r_{ED,Fe(III)}, \quad (16)$$

$$R_{As(III)} \frac{\partial [As(III)]}{\partial t} = \frac{\partial}{\partial x_i} \left(D_{ij} \frac{\partial [As(III)]}{\partial x_j} \right) - \frac{\partial (v_i [As(III)])}{\partial x_i} + \frac{q_s}{\phi} [As(III)]_s + Y_{As(III)/ED} \cdot r_{ED,As(V)}, \quad (17)$$

$$R_{SO_4^{2-}} \frac{\partial [SO_4^{2-}]}{\partial t} = \frac{\partial}{\partial x_i} \left(D_{ij} \frac{\partial [SO_4^{2-}]}{\partial x_j} \right) - \frac{\partial (v_i [SO_4^{2-}])}{\partial x_i} + \frac{q_s}{\phi} [SO_4^{2-}]_s - Y_{SO_4^{2-}/ED} \cdot r_{ED,SO_4^{2-}}, \quad (18)$$

where R is used to represent the retardation coefficient for the various transported species, r represents the various biodegradation rate terms, as described in Eqs. (8)–(12), Y the stoichiometric yield coefficient (mg/mmol or mM/mM), and D_{ij} the dispersion tensor (cm²/h). Square brackets represent the concentrations of each species (mM for ED and mg/L for the others).

The above reactive transport model is a general framework for the modeling arsenic transport in a groundwater aquifer where a number of EAs exist. With the exception of the microbially-mediated arsenic reactive transport equation (i.e., Eq. (17)), the sequential reactive transport equations for oxygen, nitrate, Fe(II) and sulfate have been successfully used

to predict the reaction transport of a BTEX plume (Lu et al., 1999). Therefore, our focus was on testing the appropriateness of the arsenic reactive transport model developed in this study. Laboratory experiments were primarily designed to study the arsenic transport scenarios under the conditions where oxygen and As(V) were available as EAs. Consequently, the reactive transport equations associated with nitrate, Fe(III) and sulfate species were not considered when describing experimental data sets developed in this study.

3. Laboratory experiments

3.1. Flow and tracer tests

The schematic diagram of the experimental setup used for our column tests is shown in Fig. 1. As shown in this figure, the circular column consists of a feed jar, a feed pump (piston pump), an inlet, an outlet and five sampling ports. The length and diameter of the column were 100 and 10 cm, respectively. The column was fitted with a pair of piezometers to measure the overall head loss through the system. The column was packed with synthetic silica beads, with diameters ranging from 0.18 to 0.25 mm. As(V) and As(III) have distinct adsorption properties when interacting with natural soil minerals; this would considerably complicate the analyses of the microbial reduction of As(V) to As(III) in soil-water systems. Synthetic silica beads were used as the porous medium to avoid the complexities due to differential sorption.

Water was injected from the bottom of the column, using a piston pump, at various constant flow rates, with the resultant gradient measured. The results of the flow experiments are shown in Fig. 2. Using these data, the estimated average hydraulic conductivity of the porous medium was found to be 150 cm/h. A tracer test was conducted by

continuously injecting bromide (Br^-) solution, with an initial concentration of 300 mg/L, into the column. The breakthrough data were modeled, using MT3D (Zheng, 1990), with estimated dispersivity value (α) found to be 0.05 cm. The porosity and dry bulk density were found to be 0.38 and 1.54 g/cm³, respectively. Details of the flow and transport parameters are summarized in Table 1.

3.2. Bacterial strain and culture medium

A facultative mixed culture, including Fe(III)-reducing bacteria, mainly *Shewanella* sp., was used to inoculate the column. The microbial culture was isolated from the inter-tidal flat sediments in the southwestern region of Korea. *Shewanella* sp. is a common microorganism that grows well either aerobically, in the presence of oxygen, or anaerobically, in the presence of metal species, such as Fe(III) and As(V). *Shewanella* sp. is capable of anaerobic growth, using lactate as the ED and As(V) as the EA (Zobrist et al., 2000).

The basal culture medium contained the following ingredients (g/L): 2.5 NaHCO₃, 0.08 CaCl₂·2H₂O, 1.0 NH₄Cl, 0.2 MgCl₂·6H₂O, 10 NaCl, 0.4 K₂HPO₄·3H₂O, 7.2 HEPES (hydroxyethylpiperazine-*N'*-2-ethanesulfonic acid), 0.5 yeast extract, and a 10 trace mineral and 1 vitamin solution. No exogenous electron carrier substance (i.e., anthraquinone disulfonate) and reducing agent (i.e., cysteine) were added to the medium. The trace mineral solution contained (g/L): 1500 Nitrilotriacetic acid, 200 FeCl₂·4H₂O, 100 MgCl₂·6H₂O, 20 sodium tungstate, 100 MnCl₂·4H₂O, 100 CoCl₂·6H₂O, 1000 CaCl₂·2H₂O, 50 ZnCl₂, 2 CuCl₂·2H₂O, 5 H₃BO₃, 10 sodium molybdate, 1000 NaCl, 17 Na₂SeO₃, 24 NiCl₂·6H₂O. The vitamin solution contained (g/L): 0.02 biotin, 0.02 folic acid, 0.1 B₆ (pyridoxine) HCl, 0.05 B₁ (thiamine) HCl, 0.05 B₂ (riboflavin), 0.05 nicotinic acid (niacin), 0.05 pantothenic acid,

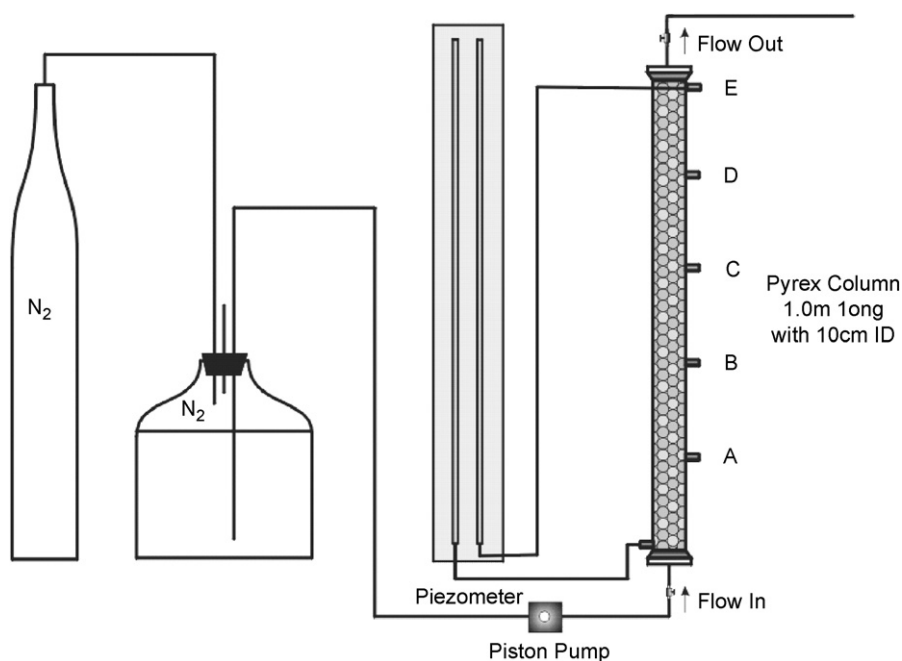


Fig. 1 – Schematic diagram of the experimental setup for the column tests.

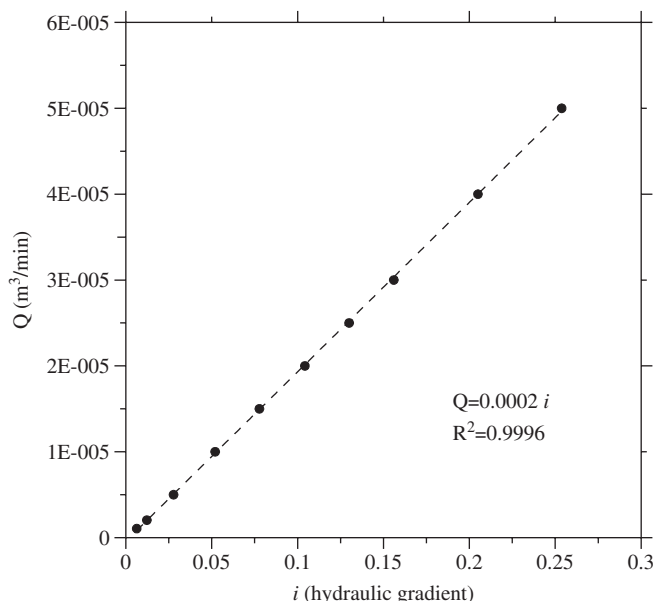


Fig. 2 – Determination of the hydraulic conductivity (K) using Darcy's law.

Table 1 – The hydraulic and transport properties of the porous medium measured in the column

Parameter	Value
Column length, L	100.0 cm
Column diameter, D	10.0 cm
Mean grain diameter, d	0.21 mm
Porosity, ϕ	0.38
Bulk density, ρ_b	1.54 g/cm ³
Hydraulic conductivity, K	150.0 cm/h
Dispersivity, α	0.05 cm
Average linear velocity, v	2.01 cm/h

0.001 B12 (cyanobalamine) crystalline, 0.05 PABA (P-amino-benzoic acid) and 0.05 lipoic acid (thioctic). The basal culture medium for facilitation of microbial growth was adjusted to pH 7.5–8.0, using 1 M NaOH, and boiled on a hot plate under a stream of N₂ gas to create an anoxic environment. The boiling method reduced the dissolved oxygen (DO) to below 1.0 mg/L; further chemical analysis of the boiled basal medium showed that the boiling method also reduced the As(V) to As(III).

3.3. Microbial batch and column tests

Both batch and column tests were completed to assess the microbial reduction and reactive transport of arsenic in groundwater systems. Water samples were withdrawn from sampling ports of the experimental column or from serum bottles of the batch experiments using a 5 mL plastic syringe fitted with a 23G 1 1/2 needle. The samples were filtered (0.2 μ m), and the redox potential (Eh), DO, pH, total As, As(III), lactate and acetate were analyzed. The samples were filtered to help remove most of the colloidal material and micro-organisms, which can affect the dissolved As(III/V) ratio. The

Eh, DO and pH values were determined at room temperature, in an anoxic environment, using Orion model 9678BN platinum redox electrodes, 083010F DO probe and 9107BN pH electrode, respectively. The electrode was cleaned routinely without disassembling. To remove precipitate that forms on the outside wall or tip of the electrode, it was rinsed with distilled water. We checked the electrode routinely with 900011 solution (Thermo Electron Corporation, Beverly, MA, USA) to find an evidence of malfunction (Bates, 1973). The measurements of total arsenic and As(III) were performed using hydride generation atomic absorption spectrometry (Perkin Elmer-5100). Operating parameters of analytes were set as recommended by the manufacturer and the percent differences on checking standards were ranged from 0.38% to 4.4% (average = 2.02%). The separation of As(III) from the total arsenic was performed using a solid-phase extraction cartridge (Supelco, 3 mL LC-SAX). The solid-phase extraction cartridge separates the As(V) from samples by allowing As(III) to pass through the cartridge, with the As(V) being retained (Le et al., 2000). The measurements of lactate and acetate were performed using high-performance liquid chromatography (HPLC). Scanning electron microscopy, with energy dispersive X-ray (SEM-EDX) analysis, was used to examine the morphology and chemistry of the precipitates.

Biologically active batch tests were first conducted to evaluate the arsenic biotransformation parameters. These batch tests were initiated with the addition of 10 mM of lactate, 0.01 mM of As(V) and 5 mL of cell suspension to 400 mL of basal medium under a headspace of N₂ gas. After the initiation process, the initial concentrations of As(V) and As(III) were 492.7 and 117.2 μ g/L, respectively. Batch tests were performed in duplicates, under strict anaerobic conditions, at 25 °C, without shaking. Batch control tests, without bacteria, were also conducted to quantify the abiotic losses in the system. In the bio-active reactors, *Shewanella* sp. reduced the dissolved As(V) to As(III) within 28 h, with the concurrent

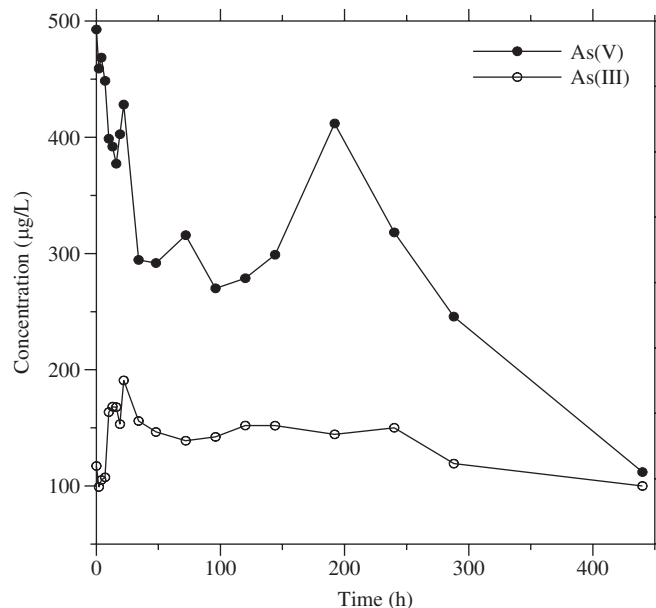


Fig. 3 – Variations in the As(V) and As(III) concentrations in the batch tests incubated with *Shewanella* sp. Data shown in the figure are the average of two replicate batch experiments.

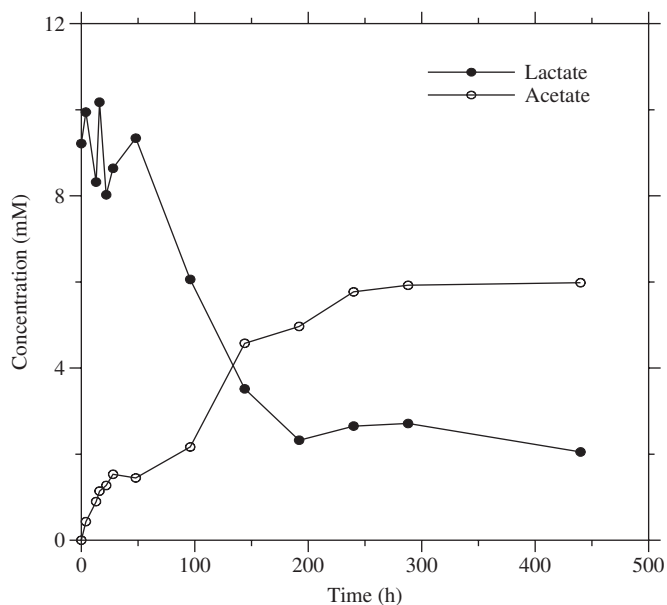


Fig. 4 – Lactate and acetate concentrations over time in the batch tests incubated with *Shewanella* sp. Data shown in the figure are the average of two replicate batch experiments.

oxidation of lactate to acetate (Figs. 3 and 4). In the bio-active experiments, the lactate concentrations decreased from 10 to 3 mM (Fig. 4) and the As(V) from ~0.5 to 0.28 mg/L (Fig. 3), due to microbial respiration, within 200h. In addition to lactate oxidation as the ED for As(V)-reduction, *Shewanella* sp. may use lactate for the reduction of dissolved oxygen in the medium. The Eh also declined from +71.3 to -207.3 mV. The bacterium, *Shewanella* sp. used in this experiment, can begin to reach its death phase within 50h (Lee et al., 2007). In addition to *Shewanella* sp., other bacteria may use lactate for respiration during the experiments, as the microcosm used in

this study contained mainly *Shewanella* sp., but other species also. Although the decrease in the lactate concentration stopped within 200h, the As(III) concentration continued to increase, which may be attributable to changes in the thermodynamic conditions (reducing condition, low Eh) generated due to the microbial respiration process. Several specific details of the underlying biological As(V)-reduction processes remain unanswered, but from an electrochemical viewpoint, the bacteria alter the local Eh and pH conditions, which in turn, shift the thermodynamic conditions, which can facilitate the reduction of arsenic.

The control, without *Shewanella* sp., showed no appreciable changes in the lactate, As(V) and Eh values during the course of the experiments. On the other hand, in the systems inoculated with bacteria, the concentration of As(III) decreased after 28 h (Fig. 3), with a significant amount of black precipitates formed after 13 days. The continuous increase in the acetate concentration indicated that precipitation was associated with the microbial oxidation of lactate, coupled with the microbial reduction of As(V). SEM-EDX analysis of the formed precipitates showed the bacteria had formed ~1 µm-sized ball shaped precipitates, consisting mainly of iron, calcium and sulfur (data not shown). The chemistry of the precipitate indicated that the reduced arsenic may be precipitated as arsenic sulfide. Aliquots of the precipitates were dissolved with 4.5 mL of 1 M HCl to extract the As, and diluted with 4.5 mL of deionized water. The total arsenic,

As(III) and As(V) contents of the precipitates were 38.5, 18.1 and 20.4 mg/kg, respectively. The As(III) in the precipitates may be photolytically oxidized during the dissolution with HCl for the quantification of the arsenic in the precipitates. From the batch test results, the first-order degradation rate of lactate, and the first-order removal (by precipitation) rate of As(III) were determined, the results of which are summarized in Table 2. The batch test also indicated that a hydraulic retention time of approximately two days would be required to allow the bacteria to reduce the As(V) to As(III).

As a second phase, biologically active, column-scale transport experiments were completed to investigate the transformation of arsenic and its subsequent reactive transport. For inoculation of the column, about 10 mL of lactate solution (0.5 M) and 6 mL of cell suspension were injected into a sterilized column, via each port; the column was left to stand for 24 h to facilitate microbial adaptation to the column surroundings and remove any dissolved oxygen from the column. The influent solution was prepared by the addition of 10.0 mM of lactate and 0.01 mM of As(V) to the basal medium, where As(V) was reduced to As(III) due to the anoxic conditions of the basal medium which had a DO of only 0.63 mg/L. The initial concentrations of As(V) and As(III) in the influent solution were 271.44 and 450.0 µg/L, respectively. The prepared solution was injected through the column at a rate of 1.0 mL/min. The laboratory temperature was maintained at 25 °C to facilitate microbial growth. Variation of the As(III) and As(V) concentrations over time, at a point 60 cm from the inlet (i.e., sampling port C in Fig. 1), are shown in Fig. 5. These data showed that the Eh values continuously declined, finally reaching –266.8 mV after 34.5 h; this indicated that microbial respiration actively occurred in the column test. Later, the concentrations of As(III) and As(V) became lower than those at the early stage of the test, due to precipitation of the arsenic species, which was consistent with the results obtained from the microbial batch tests. The breakthrough

Table 2 – Summary of the reaction parameters for validation of the developed model

Parameter	Value
Substrate reaction terms by electron acceptor of oxygen	
k_{ED,O_2}	0.035 h ⁻¹
k_{EA,O_2}	0.05 mg/L
$Y_{O_2/ED}$	0.096 mg/mmol
$Y_{Ac/ED,O_2}$	1.0 mM/mM
Substrate reaction terms by electron acceptor of As(V)	
$k_{ED,As(V)}$	0.007 h ^{-1a}
k_{i,O_2}	0.1 mg/L
$Y_{As(V)/ED}$	0.149 mg/mM
$Y_{Ac/ED,As(V)}$	1.0 mM/mM
Precipitation of As(III)	
$K_{As(III)}$	0.005 h ^{-1a}
First-order decay of As(V) for both reduction and precipitation	
$K_{As(v)}$	0.005 h ^{-1a}

^a Values obtained from the laboratory experiments .

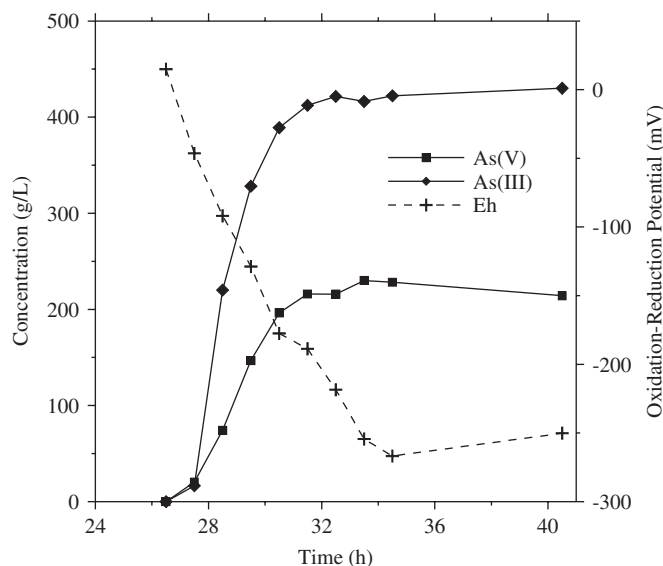


Fig. 5 – Variations of the As(V) and As(III) concentrations at sampling port C of the column over time.

concentration values for As(V) and As(III) were used to calibrate the arsenic reactive transport model.

4. Calibration of arsenic reactive transport model

The general reactive transport equations presented in the previous section were appropriately revised to model our simple system. The following version of the mathematical model for arsenic reactive transport was used to simulate our column experiments:

$$R_{\text{As(III)}} \frac{\partial[\text{As(III)}]}{\partial t} = \frac{\partial}{\partial x_i} \left(D_{ij} \frac{\partial[\text{As(III)}]}{\partial x_j} \right) - \frac{\partial(v_i[\text{As(III)}])}{\partial x_i} + \frac{q_s}{\varphi} [\text{As(III)}]_s + Y_{\text{As(III)/ED}} \cdot r_{\text{ED,As(V)}}, \quad (19)$$

$$R_{\text{As(V)}} \frac{\partial[\text{As(V)}]}{\partial t} = \frac{\partial}{\partial x_i} \left(D_{ij} \frac{\partial[\text{As(V)}]}{\partial x_j} \right) - \frac{\partial(v_i[\text{As(V)}])}{\partial x_i} + \frac{q_s}{\varphi} [\text{As(V)}]_s - Y_{\text{As(V)/ED}} \cdot r_{\text{ED,As(V)}}. \quad (20)$$

The transport equations for the ED (i.e., lactate in this experiment), using oxygen and As(V) as the EAs, can be reduced from Eq. (13) to the following form:

$$R_{\text{ED}} \frac{\partial[\text{ED}]}{\partial t} = \frac{\partial}{\partial x_i} \left(D_{ij} \frac{\partial[\text{ED}]}{\partial x_j} \right) - \frac{\partial(v_i[\text{ED}])}{\partial x_i} + \frac{q_s}{\varphi} [\text{ED}]_s - r_{\text{ED,O}_2} - r_{\text{ED,As(V)}}. \quad (21)$$

The transport equation for the product (i.e., acetate in this experiment) due to oxidation of the electron donor can be expressed as

$$R_{\text{Ac}} \frac{\partial[\text{Ac}]}{\partial t} = \frac{\partial}{\partial x_i} \left(D_{ij} \frac{\partial[\text{Ac}]}{\partial x_j} \right) - \frac{\partial(v_i[\text{Ac}])}{\partial x_i} + \frac{q_s}{\varphi} [\text{Ac}]_s + Y_{\text{Ac/ED,O}_2} r_{\text{ED,O}_2} + Y_{\text{Ac/ED,As(V)}} r_{\text{ED,As(V)}} \quad (22)$$

where [Ac] is the acetate concentration (mM), $Y_{\text{Ac/ED,O}_2}$ the stoichiometric yield coefficient (mM/mM) for the acetate produced due to oxidation of the electron donor, with the reduction of oxygen; similar nomenclature was applied to $Y_{\text{Ac/ED,As(V)}}$.

The one-dimensional model domain was a 102 cm long and 2 cm wide section, discretized using 2 cm grid cells to simulate the experimental setup. Constant head boundaries were applied along the left and right ends of the model domain, with steady-state flow simulations then performed. The constant heads of the left and right boundaries were set at 10.0 and 9.5 cm, respectively. The hydraulic and transport properties used in the simulation are presented in Table 1, with the reaction parameters summarized in Table 2. The concentration values of the substrate (i.e., lactate), As(V) and As(III) on the left boundary were fixed at 10 mM, 0.27, and 0.45 mg/L, respectively.

In the first set of simulations, only the advection and dispersion processes were considered, with the microbial reduction terms excluded, i.e., last terms in Eqs. (19) and (20) were ignored. The breakthrough curves for As(V) and As(III) predicted by the model were compared against those observed in the column tests (Fig. 6a), which clearly showed the requirement for an additional term to account for these

discrepancies. The second simulation was completed after including the microbial reduction process (i.e., Eqs. (19) and (20) were solved). This second simulation did not match the observed As(III) breakthrough data either (see Fig. 6b), as the model did not consider removal due to arsenic precipitation, which was observed in both the batch and column tests.

Therefore, the model was revised to include a precipitation reaction term to better describe the reactive transport system for arsenic. A first-order term was used to describe the losses due to precipitation. In the batch tests, both As(III) and As(V) were found in the precipitates. The first-order removal rate of As(III) due to precipitation can be estimated by fitting the As(III) curves after the peak concentration in Fig. 3. Both the precipitation and reduction of As(V) contributed to the decrease in the aqueous phase As(V) concentration, and a first-order removal term would be effective for describing both the microbial reduction and precipitation of As(V). The effective first-order removal rate constant for As(V) was estimated from the data presented in Fig. 3. The reactive transport model equations for As(III) and As(V) were modified to accommodate the effective rate constant in the following manner:

$$R_{\text{As(III)}} \frac{\partial[\text{As(III)}]}{\partial t} = \frac{\partial}{\partial x_i} \left(D_{ij} \frac{\partial[\text{As(III)}]}{\partial x_j} \right) - \frac{\partial(v_i[\text{As(III)}])}{\partial x_i} + \frac{q_s}{\varphi} [\text{As(III)}]_s + Y_{\text{As(III)/ED}} \cdot r_{\text{ED,As(V)}} - k_{\text{As(III)}} \cdot [\text{As(III)}], \quad (23)$$

$$R_{\text{As(V)}} \frac{\partial[\text{As(V)}]}{\partial t} = \frac{\partial}{\partial x_i} \left(D_{ij} \frac{\partial[\text{As(V)}]}{\partial x_j} \right) - \frac{\partial(v_i[\text{As(V)}])}{\partial x_i} + \frac{q_s}{\varphi} [\text{As(V)}]_s - k_{\text{As(V)}} \cdot [\text{As(V)}], \quad (24)$$

where $k_{\text{As(III)}}$ is the first-order removal rate constant (h^{-1}) for As(III) due to precipitation and $k_{\text{As(V)}}$ is the first-order removal rate constant (h^{-1}) for As(V) removal due to microbial reduction and precipitation (Table 2).

Predictions using the modified reduction-precipitation reactive transport model matched the experimental data well (Fig. 6c). Since arsenic is subject to both microbial reduction and subsequent removal due to precipitation, the arsenic reactive transport model should have terms for considering both microbial As(V)-reduction and precipitation. The use of an effective first-order removal model, which incorporated both As(V)-reduction and precipitation, was found to be an effective approach for predicting the transport of As(V). Haque and Johannesson (2006) observed similar reactions in the field, and indicated the importance of understanding the evolution of arsenic species due to microbial reduction, with their subsequent precipitation along the path of a groundwater flow. Therefore, this reactive transport model, which has the capability to model both the microbial reduction and precipitation processes, can be useful tools for simulating the reactive transport of arsenic on a field scale.

5. Conclusions

A reactive transport model for describing the microbially-mediated transformation of arsenic species and their

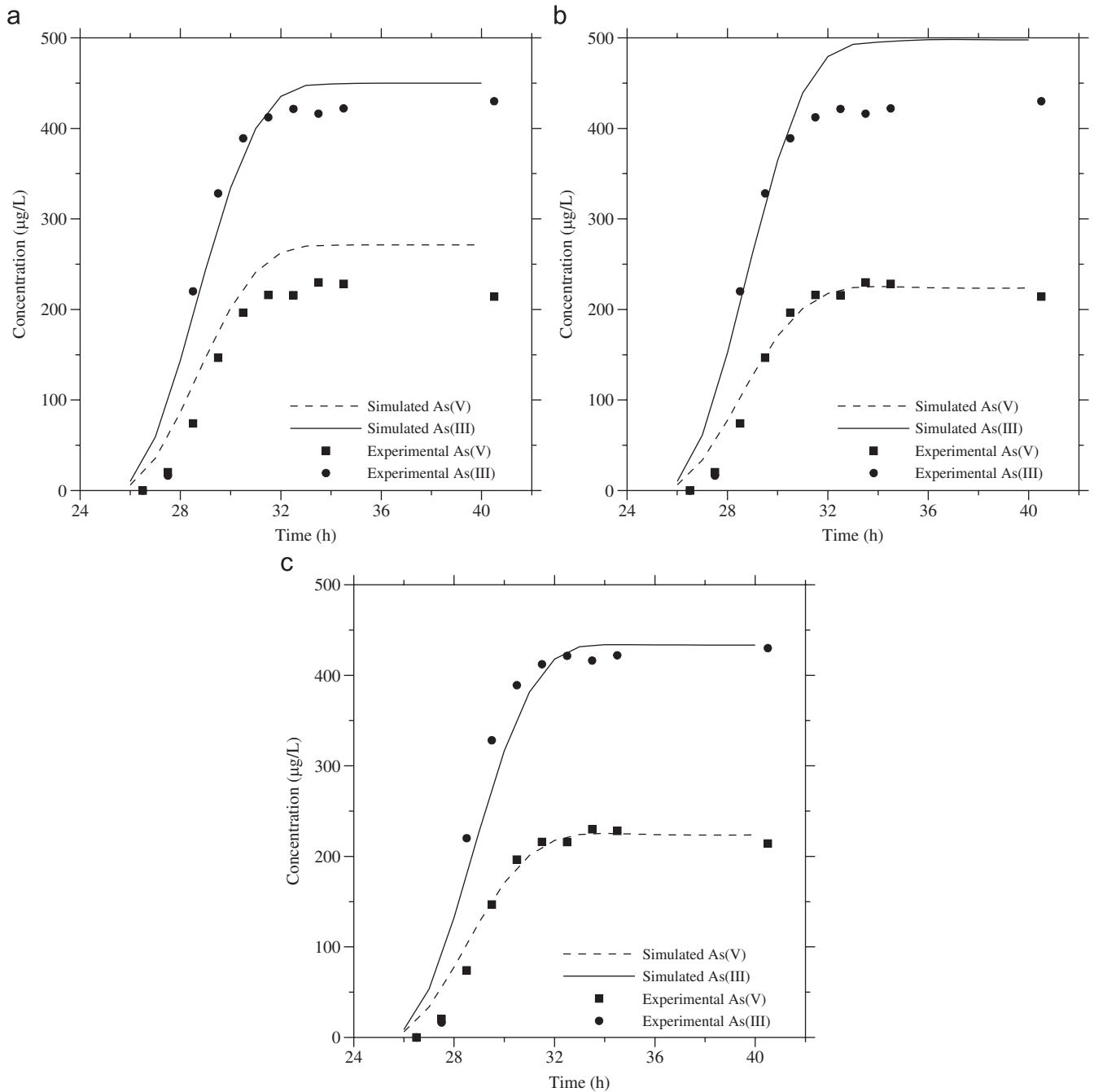


Fig. 6 – Comparison of the experimental breakthrough curves for arsenic with those obtained from the transport models: considering (a) advection and dispersion; (b) advection, dispersion and microbial As(V)-reduction; and (c) advection, dispersion, microbial As(V)-reduction and precipitation.

subsequent transport was developed and tested using both batch and column experiments.

The experimental data indicated an initial sharp drop in the Eh values, with the rapid reduction of As(V) to As(III) in this region. Furthermore, the rapid increase in acetate level indicated effective microbial activity in this region. Later, the aqueous concentrations of As(III) and As(V) were decreased due to precipitation. This precipitation phenomenon was also observed in the column tests. The batch and column tests showed microbial reduction of As(V) to As(III),

with the subsequent removal of arsenic species due to precipitation.

Several types of arsenic reactive transport model formulations were used to simulate the observed column results. The reactive transport model employing only microbial As(V)-reduction kinetics was not able to reproduce the observed breakthrough concentrations of As(III) and As(V). Therefore, a precipitation term was introduced to the microbial As(V)-reduction kinetic model; after which, the simulation results of the modified model well matched those found

experimentally. A similar reaction pathway, where arsenic was removed due to reduction followed by precipitation, was recently observed at a field site. The proposed arsenic reactive transport model can serve as a useful tool for predicting the fate and transport of arsenic species in groundwater systems.

Acknowledgments

This study is supported by the Korea Research Foundation Grant, funded by the Korean Government (MOEHRD) (R02-2004-000-10176-0), and by the BK 21 project of the Korean Government.

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