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Dissolved Organic Matter Quality in a Shallow Aquifer of Bangladesh: Implications for Arsenic Mobility

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Abstract

In some high As groundwater systems, correlations are observed between dissolved organic matter (DOM) and As concentrations, but in other systems such relationships are not observed. The role of labile DOM as the main driver of microbial reductive dissolution does not explain the variation in these relationships. Other processes that may also influence arsenic mobility include complexation of As by dissolved humic substances, and competitive sorption and electron shuttling reactions mediated by humics. To evaluate such humic interactions, we characterized the optical properties of whole waters sampled from groundwater, spanning an age gradient in Araihaazar, Bangladesh. Further, we analyzed fulvic acids (FA) isolated from large volume samples for optical properties, C and N content and ¹³C-NMR spectroscopic distribution. Old groundwater (> 30 years old) contained primarily sediment-derived DOM and had significantly higher ($p < 0.001$) dissolved arsenic concentration than groundwater that was < 5 years old. Younger groundwater had DOM spectroscopic signatures similar to surface water DOM and characteristic of a sewage pollution influence. Associations between dissolved arsenic, iron, and FA concentration, and fluorescence properties of isolated FA suggest that aromatic, terrestrially-derived FAs promote arsenic-iron-FA complexation reactions that enhance arsenic mobility.

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Supporting Information

Detailed methods, supporting tables, and supporting figures are provided in the Supporting Information.

1. Introduction

In Bangladesh, it is estimated that ~45 million people are exposed to groundwater arsenic (As) concentrations that are above the World Health Organization guideline value of $10 \mu\text{g L}^{-1}$. Although it is generally agreed that microbially-mediated reductive dissolution of As-bearing Fe-minerals^{2,3} is the dominant mechanism for enrichment of As in reducing groundwater in the Bengal Basin and elsewhere, the role of dissolved organic matter (DOM) remains enigmatic in part due to a lack of characterization of DOM composition. Compared to the well-recognized role of labile DOM fueling microbial reduction, demonstrated in several field studies of Bengal groundwater^{4–7}, less is known about the role of humic DOM in arsenic-laden groundwater. Though humic DOM is considered to be recalcitrant to biodegradation⁸, it is involved in both biological and chemical reactions that may influence the mobility of arsenic⁹. For instance, microbial reduction of oxidized humic quinones has been shown to be an important step in the cascade that results in Fe reduction¹⁰. Quinone moieties in dissolved and solid-phase humic substances are able to shuttle electrons and accelerate microbial reduction of Fe^{10–13} and other terminal electron acceptors and are able to regenerate and continue to serve as electron shuttles¹⁴.

In addition to the biological role for humic DOM, evidence for humic DOM influencing As mobility through abiotic mechanisms such as competitive sorption and complexation has emerged in laboratory studies. It was demonstrated that competition with As for sorption sites on hematite¹⁵ and goethite¹⁶ minerals resulted in arsenic desorption. Humic DOM has also been shown to abiotically oxidize and reduce arsenic from soils and sediments^{16, 17}. Of particular interest is the formation of complexes between humic substances, Fe, and arsenic, which act to keep arsenic in solution under reducing conditions. Direct complexation was initially suggested between dissolved humics and arsenate^{18, 19} and arsenite^{20, 21}. More recently, Fe-bridging for ternary complex formation involving humic substances, Fe, either arsenate^{22, 23} or arsenite²⁴ has been invoked as the mechanism responsible. Indeed, X-ray absorption spectroscopy results demonstrate that ternary As—Fe-humic DOM complexes form by inner-sphere binding of As(V) to Fe(III)-humic DOM complexes²⁵.

Field observations also support that the quality of DOM, in particular the humic DOM fraction, exerts an influence on As mobility. First, significant positive correlations²⁶, significant negative correlations²⁷, or no correlations at all²⁸ have all been observed between groundwater As and DOC concentrations. This variation suggests that the role of DOM is not limited to that of an electron donor for microbial reactions, and is instead more complex due to the heterogeneous nature of DOM. Humic DOM, including dissolved humic and fulvic acids, participate in a potential suite of reactions, such as complexation, competition,

and electron shuttling that likely also influence arsenic mobility. Second, studies in the Bengal Basin have implicated the sediments as a likely source of DOM in reducing groundwater^{29–32}. Further, optical spectroscopic analyses of Bangladesh groundwater demonstrated that the DOM in groundwater with elevated As concentrations was largely derived from aromatic and lignaceous organic compounds already in the sediments³³. Taken together, the role of the humic DOM pool in reducing aquifers of the Bengal Basin merits further exploration.

This study elucidates the potential role that dissolved humic substances, namely fulvic acid (FA), which is the predominant humic fraction in the DOM pool of natural waters, play in As mobility through a systematic characterization of DOM chemistry. A characterization of groundwater dissolved fulvic acids has not previously been carried out in As-rich reducing groundwater environments, although extracts of sediments organic compounds have been analyzed and provide some insight into sedimentary DOM character. In several regions with elevated groundwater As concentration, Reza et al. (2010, 2011)^{34, 35} performed absorbance and fluorescence analyses of whole water samples and ¹H- NMR, ¹³C-NMR, and Fourier Transformed Infrared spectral analyses of organic compounds extracted from sediments using an acid-alkaline method. These studies demonstrated that the extracted sedimentary organic compounds from the Meghna and Brahmaputra floodplains had higher aromaticity and fluorescence intensity than those from the Ganga floodplain and could be involved in complexation reactions with arsenic. These studies using sedimentary organic compounds provide additional motivation for investigating the role of humic DOM. Noting, however, that DOM in groundwater of the Bengal Basin likely has diverse sources in addition to sedimentary organic matter, there is a need to evaluate the influence that both humic DOM, isolated directly from groundwater, as well as non-humic DOM may have on arsenic mobility.

Therefore, the goal of this study is to investigate the interactions between DOM fractions, arsenic, and iron to elucidate potential mechanisms for arsenic mobilization. Here we combine FA isolation, elemental analysis, ¹³C-NMR spectroscopy, UV-vis absorbance spectroscopy, and fluorescence spectroscopy to characterize dissolved humic substances in groundwater of varying ages and varying dissolved As concentrations that represent the range of hydrogeological and biogeochemical conditions typical of the Holocene shallow aquifer of Bangladesh. The association between fulvic acid content, iron, and arsenic was also investigated to evaluate if the presence of humic DOM could help explain variability in DOC and arsenic concentration correlations noted in previous field studies.

2. Methods

Groundwater and surface water samples (Tables S1 and S2) were from Araihaazar upazila, Bangladesh located approximately 30 km northeast of the capital city Dhaka. It lies within the floodplain of the Old Brahmaputra River, an abandoned river channel that has been filled through recent sedimentation³⁶ and reduced to a small stream (Figure S1). The study area was chosen because the redox condition of the shallow Holocene aquifer and in turn, groundwater As level and age, spans nearly the entire range found in Bangladesh^{37–41}. Groundwater samples were collected from monitoring wells installed at Site K described in

Radloff et al. (2008) and Radloff (2010)^{42, 43} and Site B described in Zheng et al. (2005)⁴⁴. The two sites are approximately 2 km apart and are in a primarily rural setting with numerous ponds and irrigated rice and vegetable fields.

To isolate FA, a total of 6 large volume water samples (~ 215 L) were collected from a small stream next to the field where monitoring wells were installed (Fig. S1), three wells from Site K and two wells from site B in October 2009. The three samples from Site K represented young groundwater with $^3\text{H}/^3\text{He}$ age⁴⁵ of < 5 years old (K12 and K10) and old groundwater with > 30 years according to $^3\text{H}/^3\text{He}$ dating (K8). Two samples were also collected from Site B (B-11 and B-14) for which the groundwater was known to be ~ 19 years old³⁹ to ensure that the age range of 5-30 years was represented. The wells were pumped for 10 to 30 minutes, until readings of temperature, conductivity, pH and Eh on multiprobes in a flow-through cell stabilized. Then, ~215 L of groundwater were collected into cubitainers and driven to the University of Dhaka, Bangladesh where they were then filtered, acidified, and pumped through columns packed with cleaned XAD-8 resin⁴⁶. After rinsing loaded columns with nanopure water, the FA was back-eluted with base and run through a cation exchange column. FAs were shipped on ice to the University of Colorado, where they were freeze-dried and analyzed for C and N content with an elemental analyzer and C functional group distribution with solid state ^{13}C -nuclear magnetic resonance (^{13}C -NMR) spectroscopy. The surface water sample was collected and processed in the same way. Prior to isolation, an aliquot of the large volume samples was reserved and analyzed for total dissolved As and Fe concentrations, DOC concentration, and UV-visible absorbance and fluorescence spectroscopy (see Supporting Information for details).

To provide the geochemical context for the FA isolate chemistry data, a total of 24 groundwater samples were also collected from Site K, along with a total of 10 surface water samples representing the Old Brahmaputra River, local streams and ponds in Araihaazar, in March 2008 (Table S1). These “whole” water samples were analyzed for anions and cations, dissolved As and Fe concentrations and speciation, DOC concentration, and UV-visible absorbance and fluorescence spectroscopy (see Supporting Information for details).

For both FA isolates and whole water samples, key metrics from UV-vis absorbance and fluorescence spectroscopy were evaluated. Specific UV absorbance⁴⁷ was utilized to provide information about the aromaticity of whole waters, whereas for FAs the aromatic C content was measured directly with solid state ^{13}C -NMR. Three dimensional fluorescence excitation emission matrices (EEMs) were acquired and instrument specific corrections, inner-filter correction, Raman normalization, and blank subtraction were applied. EEMs were fit to the parallel factor analysis (PARAFAC) model of Cory and McKnight (2005)⁴⁸, and individual component loadings are presented in Raman units (RU) and as percentages of total fluorescence. The relative amount of amino acid-like fluorescence, taken as the sum of tyrosine-like and tryptophan-like PARAFAC components (C8 + C13 in Cory and McKnight, 2005), which are described as the more biologically labile components in the fluorescence literature, was tracked in all samples. The fluorescence source index (FI) was also calculated⁴⁸ to provide information about the DOM sources in whole waters and FAs. The FI has provided consistent and reliable information on the sources of DOM and humic substances in natural waters that were consistent with characterization using other

techniques. For example, Mladenov et al. (2008) isolated FAs from wetland surface water and found that spatial changes in the FA content, C:N ratios, C functional groups, and FI reflected an influx of plant-derived organic compounds during flooding⁴⁹.

A multivariate analysis of variance (MANOVA) was employed to test whether groundwater chemistry (As, Fe, sulfate, DOC, total dissolved nitrogen (TDN), SUVA, FI, and percent amino acid-like fluorescence) was significantly different among surface water (n = 10), groundwater samples in age group of < 5 years (n = 10), 5 – 30 years (n = 6), and > 30 years old (n = 8) (vegan package, R project). Additionally, an analysis of variance (ANOVA) was also used to determine significance in As concentration difference between data sets using a two-tailed t-test (R project).

3. Results and Discussion

Surface water DOM and FA isolate

Surface water samples, consisting of the old Brahmaputra river (OBR) channel, local streams near sites K and X, and ponds from sites K and Fe (Table S1) displayed the highest average DOC concentration than groundwater of any age (Table 1). Because whole water samples were obtained in March when the groundwater tables are substantially higher than the surface water tables⁴¹, the river and stream waters are primarily groundwater fed, with the water chemistry subject to “redox trapping” during groundwater discharge^{50, 51}. “Redox trapping” means that redox-sensitive elements such as As, Fe and Mn have a tendency to be removed from solution and “trapped” in sediments under oxidizing conditions, and, therefore, their concentrations are lower than when reducing conditions prevail. The low surface water concentrations of As, Fe, and Mn compared to those in groundwater and similar concentrations of Ca, which is not redox sensitive, in surface water and groundwater are consistent with this discharge process (Table 1). Additionally, because the surface water environment is oxic, sulfate concentrations in surface water are higher than in groundwater. The low RI of surface water DOM (Table 1) also reflects this oxic state and the presence of more oxidized quinone-like moieties⁵² than in groundwater. Because surface water DOC concentrations are two to three times higher than those in groundwater, it is likely that additional allochthonous inputs from the surrounding landscape and significant autochthonous biological inputs contribute to the high DOC concentrations. The FI value is the highest in surface water samples, indicating a microbial source, which would support autochthonous inputs such as bacteria and algae (Table 1). This is not surprising considering the sanitation conditions of rural Bangladesh, where human waste is frequently disposed of directly into surface water bodies⁵³. The EEMs of the whole surface water samples showed a peak in the region of amino acid-like fluorescence near excitation/emission of 275/350 nm, and provide further evidence of a microbial DOM source in this water type (Figure 1). However, there is also substantial humic fluorescence in the Peak A and C regions (Figure 1) of the whole water samples, which reflects contributions from soil and terrestrial plant sources to the DOM pool in surface water. Thus, the DOM of whole surface water is characterized by protein-like as well as humic DOM.

Although we have only one fulvic acid isolate of a local stream water (K-SW) sample, the characteristics of this surface water FA isolate are illustrative. This FA isolate had >40% C

content, which was greater than the C content of all groundwater FA isolates (Table 2). Further, the combination of high SUVA, reflecting terrestrial DOM contributions, and high FI and amino acid-like fluorescence, reflecting autochthonous microbial contributions, would also suggest that, like the whole water sample, this fulvic acid was derived from a combination of microbial constituents as well as soil and vascular plant sources in the area surrounding the river.

Groundwater DOM and FA isolates

The extensive spatial survey of groundwater from different wells and depths showed that there were significant differences in DOC concentration as well as in the quality of DOM among groundwater of different ages. Differences in DOC concentration, SUVA, FI, and %AA-like fluorescence of small volume samples were significant between young, < 5 year old, groundwater and the two groups of older groundwater (Table 1). All groundwater samples have comparable RI values consistent with reducing conditions indicated by elevated levels of Fe and Mn concentrations in groundwater compared to surface water (Table 1). Further, the results showing significantly ($p < 0.00001$) higher concentrations of As, Fe, Ca, and TDN, mostly as ammonia (Table S2) and lower sulfate concentrations in older groundwater than in young groundwater (Table 1) are consistent with progressively greater reducing conditions as groundwater ages and subsequent occurrence of mineral weathering (calcite dissolution) and redox reactions^{39, 44}. Additionally, significant differences, determined with MANOVA ($R^2 = 0.38$; $p < 0.0001$), were also observed for DOC, SUVA, FI, and percent amino acid-like fluorescence between groups of groundwater. The lower FI, lower AA-like fluorescence, and higher SUVA in the two groups of older groundwater samples suggest inputs of “terrestrial” sources, such as lignaceous and aromatic C compounds, common in peat or other dispersed sedimentary organic matter. One explanation for the more terrestrial DOM signatures in the older groundwater is that DOM is mobilized from sediments as a consequence of microbial Fe reduction and Fe mineral dissolution. This scenario would be consistent with other studies showing that sedimentary organic matter contributes substantially to the groundwater DOM pool^{54, 55}. DOM mobilization from sediments is supported by the results of sediment incubations from our earlier study conducted also at Araihasar, which showed an increase in DOC and Fe concentrations and DOM terrestrial fluorescence signatures after incubation with native groundwater³³.

Similarly, the higher fulvic acid content of large volume samples B11, B14 and K8, representative of older groundwater (> 5 years old), than in the younger K10 and K12 groundwater (Table 2) indicates that the older groundwater has higher amounts of dissolved humic substances, most likely originating from organic material in the sediments. Also, the FAs isolated from large volume samples of older groundwater had higher aromatic C concentration, lower FI values, and lower amino acid-like fluorescence than FA isolates of young groundwater (Table 3), consistent with the trends observed in the small volume samples that point to a terrestrial, sedimentary DOM source. The lower amino acid-like fluorescence was also evident in EEM spectra of the large volume samples of old groundwater (Figure 1), which were instead dominated by fulvic acid-like fluorescence.

By contrast, several lines of evidence suggest that DOM in younger groundwater (< 5 years old) is influenced by microbial sources. The average FI value of small volume samples of young groundwater is similar to surface water, and the average value of %AA-like fluorescence is the highest amongst all water types (Table 1), likely reflecting the influence of sewage-polluted surface water on young groundwater. This influence was identified at the same study site⁵⁶ in young groundwater, which has been shown to contain high counts of the fecal indicator bacterium *E. Coli*⁵⁷. The lowest average SUVA value in young groundwater suggests that the young groundwater is the least influenced by terrestrially-derived DOM among all water types (Table 1). Similarly the large volume samples of young groundwater from sites K10 and K12 had low fulvic acid content (Table 2), and the FA isolated from those sites had higher FI and amino acid-like fluorescence and lower %C and molar concentrations of aromatic C (Table 3) than older groundwater.

Non-humic DOM

In addition to humic DOM in Araihaazar groundwater, non-humic DOM is present in the groundwater and influences the optical spectroscopic properties measured in this study. For the same five large volume samples we characterized the fulvic acids, described above, as well as whole water, which includes fulvic acids as well as carbohydrates, low molecular weight organic acids, and other non-humic DOM. The FI and % amino acid-like fluorescence of whole water samples prior to FA isolation (Table S1) were higher than for the FA isolates (Figure 2). The non-humic fraction that is also present in the whole water samples, therefore, reflects contributions to the DOM pool from microbial and other non-humic DOM sources. FI values of whole waters are known to be higher when compared to FA isolates, in part, because of the presence of other fluorescent microbial compounds that would act to raise the FI⁵⁸. These microbial sources may include microbial exudates or microbial products from lysed microbial cells, which may derive from sewage-influenced surface water drawn laterally⁵⁹ or to depth or from the sediments themselves. Other microbial consortia, including bacterial taxa capable of Fe, As, and humics reduction, which are key members of the community in Araihaazar sediments⁶⁰, may also contribute to the amino acid-like fluorescence, which is known to represent biodegradable DOM⁶¹, observed in the whole water DOM.

Humic DOM and As mobility

Having illustrated the differences between two pools of groundwater DOM, humic and non-humic, we now attempt to shed light on the variable relationship, either positive, negative or a lack thereof, between groundwater As and DOC concentrations observed in several field studies^{26–28}. There was no significant correlation between whole water DOC concentration and As in this set of shallow groundwater samples from Bangladesh (Figure S2) and only a weak correlation for the 5 large volume samples (Figure 2). Instead, it was with the humic DOM component expressed as FA concentration of groundwater that we observed significant linear relationships with As and Fe (Figure 2). We acknowledge that the sample size is small; however FA isolation is a highly representative technique⁴⁶. More importantly, the quality of DOM change supports the transition of DOM and FA quality from non-humic, microbially-derived material to humic, terrestrially-derived material as the groundwater ages.

We interpret the correlation between the chemical characteristics (aromaticity, FI, % amino acid-like fluorescence) of FAs and both As and Fe (Figure 2) as indicating the involvement of humic DOM in reactions that maintain As in solution. Prior laboratory and field studies suggest a likely mechanism through formation of As—Fe-DOM complexes^{22–24}. Although humic DOM, As, and Fe may all be present as free molecules as a consequence of reductive dissolution, the stoichiometric relationship showing proportionately greater humic DOM content in water with greater dissolved As or Fe concentrations (Figure 2) implies potential complex formation among these species. Multiple laboratory studies have now shown that As is able to form ternary complexes with Fe and DOM²⁵. Using Pahokee Peat Humic Acid as their DOM source, Sharma et al. (2010) demonstrated that at neutral pH As(V) formed colloids and complexes with Fe-DOM, but not with DOM in the absence of Fe. Formation of complexes between As and DOM in the presence of metals but not in their absence was also demonstrated for DOM extract from compost⁶². Fulvic acids readily complex Fe at neutral pH, and As can bind strongly to such complexes via bridging mechanisms²⁴ to stay in solution. Therefore, it is not unexpected that humic DOM correlates well with As in the presence of Fe in the neutral pH groundwater of our study. Using a molecular weight of 470 Da for groundwater FA from a similar environment⁶³, we calculated that molar Fe:As:FA ratios were about 47:1.3:1 (see slopes of linear regressions in Figure 2). The average molar ratios of Fe to As for large volume samples from groundwater > 5 years old ranged from 40 to 68 in our study, which is slightly higher than the 20 to 55 range reported in laboratory studies of As—Fe-DOM complexes under controlled conditions^{23, 24}.

Colloidal organic molecules also appear to have undergone associations with As and Fe. Prior to isolation of the FAs in this study, we recovered orange-colored organic matter on the filters after filtering the large volume water samples that were acidified to pH 2 to prevent Fe precipitation. This orange-colored retentate (material retained on filters) for three samples, K12, K8, and B11, was re-dissolved in 0.1 N HCl and its FA fraction was also isolated⁴⁶. The FA content of the filter retentates was substantially higher (FA of 18%, 28%, and 37% for samples K12, K8 and B11, respectively) than in the corresponding large volume groundwater samples (Table 2), suggesting that organic matter in the colloidal form was more humic than DOM in groundwater. The FAs of filter retentates also displayed the most terrestrially-derived signatures with the much lower mean FI values (FI values of 1.24, 1.23, and 1.29 for samples K12, K8 and B11, respectively) than dissolved FA isolates of groundwater same three sites (Table 3; mean FI = 1.54).

The evidence for Fe- and organic- colloids on As mobility is only emerging. Ultra-filtration of As and DOM rich groundwater from Hetao Plain, Inner Mongolia has demonstrated stronger association between As and smaller organic colloids than with larger Fe colloids⁶⁴. Guo et al. (2009) compared 43 paired unfiltered and 0.45- μ m filtered samples and found that larger amounts of As were trapped with small size organic colloids (<0.45 μ m). Additionally, SEM images, EDS analysis and synchrotron XRF analyses in their study confirmed the association of As with natural organic matter (NOM) with molecular weights of 5–10 kDa.

Implications for groundwater As enrichment

Deltaic regions with elevated arsenic concentrations in reducing groundwater tend to be rich in sedimentary organic compounds derived from the deposition of plant and animal biomass detrital compounds over time. In Bengal Deltaic sediments, Meharg et al. (2006) postulated that arsenic and organic matter were co-deposited during the Holocene⁶⁵, especially in productive coastal wetland ecosystems that developed at the time. Similar co-deposition of As and organic matter may be true for other deltaic environments with elevated arsenic in groundwater, such as the Red River Delta, the Mississippi River Delta, and the Pearl River Delta. Therefore the relationships between arsenic, Fe, and DOM that may promote arsenic mobility in reducing groundwater environments deserve further exploration. With respect to the linear relationships between As, DOM, and Fe observed in our study, it is possible that As—DOM-Fe complexes or colloids were sequestered in the sediments simultaneously during deposition, thousands of years ago. Co-deposition of organic matter and arsenic with Fe-containing minerals, as proposed by Meharg et al. (2006), and later ternary complex formation after reductive dissolution is a plausible scenario to explain why groundwater with elevated arsenic contains more terrestrially-derived, humic DOM than groundwater with low arsenic concentrations. Regardless of whether the association with arsenic is via dissolved complex or colloidal complex, our results suggest that the humic fraction of DOM, in particular, plays a key role in arsenic mobilization. Differentiating between the humic, chemically reactive, from the non-humic, biologically-reactive, fraction of DOM in reducing groundwater of deltaic aquifers is a first step to better elucidating that role.

In addition to its importance in maintaining arsenic in solution in reducing aquifers, the chemical reactivity of humic DOM influences treatment and remediation strategies used for dealing with high As concentrations in drinking water. For example, the presence of colloidal DOM-Fe-As associations may hamper filtration efforts⁶⁶, and competition with humic or fulvic acids inhibits several treatment processes, such as As sorption on Fe minerals⁶⁷, As(V) adsorption during coagulation with ferric chloride⁶⁸, and As sorption on Fe nanoparticles⁶⁹.

Despite the important role of organic matter in arsenic cycling and the clear value of elucidating its chemical structure to understand and predict biogeochemical reactions, much still remains to be understood regarding the quality of DOM found in groundwater of different age and arsenic concentrations. This study provides new information on the role of DOM fractions as being both biologically and chemically reactive, with the humic DOM acting to maintain arsenic in solution in reducing groundwater. In particular, we have shown that DOM in older groundwater (> 5 years old) is characterized by higher FA content, more terrestrial, sedimentary sources, and less microbial spectroscopic signatures than DOM in younger groundwater or surface water. The highly significant relationships we observed between As and Fe concentrations and FA content, the FI, and amino acid-like fluorescence of FA isolates further suggest that humic DOM-Fe-As interactions may have occurred long ago during the precipitation of arsenic-containing Fe minerals or recently as complexation reactions after free As was mobilized from sediments. The presence of humic DOM in groundwater and its variable source in dispersed sedimentary material may, therefore, may be another factor contributing to the large spatial variability in arsenic concentrations over

short distances. In addition to improving our understanding of the heterogeneity of elevated groundwater arsenic across the landscape, accounting for the quality of DOM in groundwater of different ages is important to the development remediation and treatment strategies.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Highlights

1. Measurements of DOC concentration alone are not sufficient to explain As mobilization and DOM quality must be considered.
2. Old groundwater (> 30 years old) contains high As concentrations and DOM with humic and lignaceous chemical character.
3. Very young groundwater (< 5 years old) contains higher protein-like fluorescence than older groundwater and is low in dissolved arsenic.
4. Relationships between dissolved As, Fe, and fulvic acid suggest that complexation with humic DOM maintains As in solution.
5. Reductive dissolution is hypothesized to desorb As—Fe-DOM complexes and mobilize arsenic.

Figure 1.

EEMs of surface water (K-SW) and Site K and Site B groundwater whole waters collected for fulvic acid isolation (samples described in Table 2). Fluorescence intensities (z-axis) are in Raman units (RU). Regions of amino acid-like fluorescence (proxy for labile DOM) and humic-like fluorescence are marked.

Figure 2. Scatterplots of total dissolved As and Fe vs. a) DOC concentration, b) fluorescence index (FI), and c) amino acid-like (AA-like) fluorescence of large volume whole water groundwater samples at Site B and Site K, d) fulvic acid concentration of large volume samples, and e) fluorescence index and f) amino acid-like fluorescence of fulvic acids isolated from the large volume samples at Site B and Site K (sample labels shown in panel a). Regression lines shown only for significant relationships ($p < 0.05$).

Table 1

Chemistry of whole water samples collected from Araihaazar, Bangladesh in March 2008. Mean and standard deviations of age, depth, solute concentrations^a, and DOM optical properties^b for surface water and young (< 5 yr), old (5 – 30 yr), and very old (> 30 yr) groundwater collected from site K are shown.

	Surface water n = 10 ^c	< 5 year old groundwater n = 10	5 – 30 year old groundwater n = 6	> 30 year old groundwater n = 8	T-test results ^d
Age (years)	-	1.9± 1.5	10.5± 5.0	34.7± 1.6	-
Depth (m)	-	8.9± 2.2	11.1± 3.6	10.5± 3.2	G
As _{total} (µg L ⁻¹)	24± 14	37± 29	158± 107	281± 112	B – F
% As(V)	66 ± 30	16± 33	1± 1	4± 4	G
Fe _{total} (mg L ⁻¹)	0.11± 0.12	6.7± 5.3	10.3± 5.5	10.8± 2.0	E
SO ₄ (mg L ⁻¹)	19± 15	15± 23	4.4± 6.4	0.05± 0.07	C, E
Ca (mg L ⁻¹)	38± 4.0	39± 25	41± 3.0	57± 17	C, E, F
Mn (µg L ⁻¹)	149± 117	1885± 1054	2145± 1036	1252± 970	A – C
TDN (mg L ⁻¹)	1.37± 1.11	0.62± 0.60	1.20± 0.62	1.76± 0.58	A, E
DOC (mg L ⁻¹)	6.30± 2.98	2.78± 0.50	3.51± 0.38	3.65± 0.69	A – E
SUVA (L mg ⁻¹ m ⁻¹)	1.97± 1.22	1.18± 0.64	1.89± 0.45	2.06± 0.48	D, E
FI	1.69± 0.24	1.47± 0.10	1.38± 0.04	1.40± 0.03	A – E
RI	0.35± 0.07	0.50± 0.04	0.51± 0.02	0.48± 0.02	A, B, C, F
% AA	0.084± 0.034	0.10± 0.07	0.05± 0.01	0.04± 0.01	B, C, E

^a n = sample size; σ = mean; δ = standard deviation; dash = not measured or performed.

^b FI and RI are dimensionless; AA = amino acid-like fluorescence.

^c Sample size is 6 for As, % As(V), Fe, SO₄, Ca, and Mn values (Table S1).

^d T-test results show significant difference between: A) surface water and < 5 year old groundwater, B) surface water and 5 – 30 year old groundwater, C) surface water and > 30 year old groundwater, D) < 5 year old and 5 – 30 year old groundwater, E) < 5 year old groundwater and > 30 year old groundwater, and F) 5 – 30 year old and > 30 year old groundwater; G represents no statistical difference in any sample sets.

Characteristics of 6 large volume samples, including site description, solute chemistry (total dissolved arsenic (As_T), % arsenic as $As(III)$), total dissolved Fe (Fe_T), dissolved organic carbon (DOC), and fulvic acid (FA) content and concentration), and DOM optical spectroscopic characteristics (specific UV absorbance (SUVA), fluorescence index (FI), and amino acid-like (AA-like) fluorescence) collected from Site K and Site B groundwater and Site K surface water (K-SW) in October 2009.

Table 2

Sample	Site description			Solute chemistry					Optical spectroscopic characteristics				
	Latitude (degrees)	Longitude (degrees)	Depth (m)	Age ^a (yrs)	As _T (μg L ⁻¹)	%As (III)	Fe _T (mg L ⁻¹)	DOC ^b (mg C L ⁻¹)	FA content (% of DOC)	FA conc. (mg FA L ⁻¹)	SUVA ^c (L mg ⁻¹ m ⁻¹)	FI	AA-like fluorescence (% of fluorescence)
K-SW	23.7916	90.6101	0	N.A.	23.3	40	0.01	4.80	42.7	4.63	2.66	1.65	5.7
K12.1	23.7948	90.6284	7.50	<5	2.0	90	0.5	0.59	2.80	0.04	1.60	1.47	24
K10.2	23.7941	90.6281	11.0	<5	69	100	3.5	0.92	8.30	0.26	1.72	1.37	8.0
K8.3	23.7932	90.6280	14.8	>30	363	94	11.0	0.93	25.0	1.28	2.24	1.37	4.0
B11	23.780	90.640	11.4	5 - 30	213	95	9.6	2.08	13.3	0.54	2.52	1.46	3.3
B14	23.780	90.640	14.0	5 - 30	340	95	10.2	2.51	34.4	1.70	2.73	1.46	4.1

^aWater age category based on $3H/3He$ dating reported in Stute et al (2007)³⁹ and Radloff (2010)⁴⁵.

^bDOC concentrations are higher in large volume samples collected in Oct 2009 post monsoon than in small volume samples collected in Mar 2008 during dry season (see Table S1).

^cHigh SUVA values (> 5.0) may indicate additional Fe absorbance in the UV-range (Weishaar et al., 2003)⁴⁷.

Elemental content, ¹³C-NMR distribution of C functional groups of fulvic acid isolates, and optical spectroscopic characteristics of fulvic acids isolated from Site K and Site B groundwater and Site K surface water (K-SW).

Table 3

Sample	Elemental content			¹³ C-NMR distribution (%) and C functional group distribution ^a							Optical spectroscopic characteristics			
	C (%)	N (%)	C:N ratio	Aliphatic (60-80)	Hetero-aliphatic (90-60)	Acetal (110-90)	Aromatic (165-110)	Carboxyl (190-165)	Carbonyl (220-190)	Ar:Al ratio	SUVA (L mg ⁻¹ m ⁻¹)	FI	AA-like fluorescence (%)	
K-SW	44.3	2.9	15.28	48	15.62	0.01	19.43	13.61	3.27	0.40	4.36	1.69	11.0	
K12.1	45.2	1.41	32.1	46.1	8.57	1.92	24.7	16.1	2.58	0.54	3.86	1.57	4.9	
K10.2	28.8	1.30	22.2	63.2	11.4	0.12	14.1	10.6	0.59	0.22	5.16	1.46	4.1	
K8.3	15.4	0.77	19.9	47.1	5.76	3.34	30.2	11.9	1.75	0.64	4.71	1.49	3.7	
B11	51.3	1.57	32.7	53.1	9.30	0.56	19.6	16.0	1.47	0.37	3.96	1.57	3.5	
B14	50.6	1.50	33.7	45.3	10.4	2.98	25.8	14.1	1.50	0.57	4.15	1.57	3.3	

^a Parentheses list range of wavelengths for each functional group (nm).