

Complexation and release isotherm of arsenic in arsenic-humic/fulvic equilibrium study

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Abstract. Soil organic fractions were extracted from 2 surface soils of West Bengal, one of which was arsenic-contaminated, and were fractionated into fulvic and humic acid fractions following standard procedures. These, and one humic acid sample synthesised in the laboratory, were characterised by pH-potentiometric titrations, viscometric measurements, visible spectrophotometry, and surface tension determinations. The results have been correlated with coiling–decoiling behaviour, as well as aliphatic/aromatic balance of these acids. The stability constant ($\log K$) of the complexes formed by these natural humic/fulvic and synthetic humic acids with arsenic in aqueous phase was evaluated by the ion-exchange method. These $\log K$ values tend to suggest that the organo-arsenic complexes were quite stable. Further, the $\log K$ values displayed an approximately inverse relationship with the viscosity-average molecular weight, excess time needed to attain a stable higher pH during the pH-potentiometric titration, and also the surface excess (in water) of the given humic/fulvic acid samples. These trends were explained in terms of the relative hydrophobic character of the acids, and their resultant tendencies to complex arsenic in the aqueous phase.

The release potential of arsenic from the arsenic–humic/fulvic complexes by soluble phosphate and nitrate salts was also examined in terms of the appropriate exchange isotherms. In general, phosphate demonstrated a greater degree of exchangeability with arsenic than did nitrate, at both the lower and higher concentrations.

Additional keywords: arsenic–humic/fulvic complex, isotherms, stability constant.

Introduction

Arsenic is a widely occurring toxic metalloid in natural ecosystems. Arsenic input to the environment, either through natural processes of weathering of arsenic-bearing rocks and/or use of arsenic-contaminated groundwater for irrigation, or through a host of anthropogenic activities (such as mining operations, smelting of base metal ores, combustion of coal, and application of arsenicals as agricultural pesticides), is well documented (Welch *et al.* 1988; Sanyal 1999; Sanyal and Nasar 2002). There has been rapidly growing global concern about arsenic contamination in drinking water (WHO 2001). Arsenic contamination of groundwater in the Gangetic alluvial zones of West Bengal, India (and in the contiguous districts of Bangladesh), has assumed the proportions of a drinking water-related health hazard for millions of people. As much as 38 865 km² of West Bengal is reported to be severely affected (Chowdhury *et al.* 2000). Two major hypotheses, both of geogenic origin, have been proposed to account for such widespread arsenic contamination in the groundwater in parts of West Bengal and Bangladesh confined within the delta bound by the

rivers Bhagirathi and Ganga-Padma. Of these 2 hypotheses, namely the arsenopyrite oxidation hypothesis (Mandal *et al.* 1996) and the ferric oxyhydroxide reduction hypothesis (Bhattacharya *et al.* 1997; Nickson *et al.* 1998), the latter is more consistent with the experimental observations reported for the aquifer sediments and water of the Bengal delta basin (Sanyal 1999; Sanyal and Nasar 2002). According to this hypothesis, an anoxic condition of the aquifer causes arsenic mobilisation from arsenic-bearing sediments into the groundwater aquifer. The maintenance of such anoxic condition is proposed to be facilitated by the widespread practice of wetland paddy cultivation in the affected belt.

The main focus of attention of this contamination, until recently, has been exclusively on arsenic contamination in groundwater-derived drinking water. However, since groundwater is also used extensively for crop irrigation in the arsenic belt of West Bengal, the possibility of a build-up of arsenic in agricultural soils and agronomic produce was anticipated. Elevated arsenic levels in groundwater-irrigated soils in West Bengal are now well documented (Sanyal and Nasar 2002). Arsenic uptake by crop plants

grown in soils contaminated with a high concentration of arsenic and irrigated with arsenic-laden groundwater, has also been reported (ICAR 2001; Abedin *et al.* 2002). These findings have thus demonstrated the pathways, other than drinking water, through which arsenic may enter the human food chain. This calls for immediate attention, since what remains essentially a point and fixed source of arsenic contamination as for drinking water (e.g. a tubewell discharging contaminated water) may well become a diffuse and uncertain source of contamination when arsenic finds its way into the food web, accompanied with possible biomagnifications up the food chain.

Soil acts as a major sink of arsenic inflow to agroecosystems (Livesey and Huang 1981; Carey *et al.* 1996; Ghosh *et al.* 2002; Mukhopadhyay 2002), thereby reducing the availability of the toxicant to the plant species and allowing the surface water bodies to remain largely free of arsenic.

It is in this context that a thorough understanding of soil–arsenic interactions is necessary to judge how well such a sink would be able to contain the toxin, and prevent it from entering the food chain via plant uptake and leaching to surface run-off and groundwater. The soil organic fraction including humic acid (HA) and fulvic acid (FA) is known to combine with metal ions, clays, pesticides, and several organics because of high specific surface area and chelating abilities (Sanyal 2001). Further, the humic substances also behave as good accumulators of toxic heavy metals, following the formation of metal–humate complexes (chelates) with different degrees of stability (Manunza *et al.* 1995; Datta *et al.* 2001; Sanyal 2001). In the present study, we examined the humic/fulvic substances (extracted from contaminated soils and/or prepared synthetically in the laboratory) in terms of the important physico-chemical properties (including surface properties) and recorded the resulting organo-arsenic complexation equilibria. The stability constants of such organo-arsenic complexes should indicate the ability of the native (or incorporated) soil organic fraction to bind arsenic within soil matrix.

Materials and methods

Characterisation of soils

Surface (0–0.15 m) soil samples were collected from an arsenic-contaminated area, namely Ghentugachhi (Chakdah Block, District Nadia, West Bengal), and an arsenic-free area, Gayeshpur (Haringhata Block, District Nadia, West Bengal). These soil samples were subjected to detailed characterisation with respect to the important physico-chemical properties (pH, EC, organic carbon, particle-size distribution, CEC, amorphous hydrous oxides of Fe, specific surface area, etc.) following the standard methods (Page 1982).

Humic acid samples were synthesised/extracted from soil by the following procedures:

- (i) Chemically synthesised HAs from α -amino acid (cysteine) and hydroquinone by means of oxidation with alkaline peroxodisulfate: Equal amounts (15 g each) of hydroquinone and cysteine were

dissolved separately in a 0.1 M NaOH solution. Hydroquinone was added to a warm solution of amino acid with constant stirring, followed by gradual addition of potassium peroxodisulfate (3 g). Thereafter, the solution was refluxed on a water bath for 90 min, using an air condenser. The refluxed materials were rendered alkaline (up to pH 9.0) with 0.1 N NaOH and were centrifuged to remove alkali-insoluble impurities. The centrifugate was acidified with 0.1 N HCl to pH 2.0 and kept overnight at room temperature for complete precipitation. The coagulated humic materials were separated from the soluble parts by centrifugation. The separated HA was redissolved in alkali followed by reprecipitation with acid and then separated from the soluble part by centrifugation. It was then dialysed against distilled water until free of chloride (Coulson *et al.* 1959).

- (ii) Natural HA and FA extracted by following the standard procedure from the 2 surface soils from Ghentugachhi and Gayeshpur: Soils were dried in air, ground, and passed through a 2-mm sieve. The soils were then shaken with 0.5 N Na₂CO₃ in a Winchester bottle and kept overnight. The supernatant was siphoned off and acidified to pH 2.0–3.4 with HCl to precipitate HA. The system was kept overnight to ensure complete precipitation. The processes of dissolution in alkali and reprecipitation of HA were repeated once more. The supernatant was siphoned off and the precipitate was dialysed until chloride-free. The supernatant was adjusted to pH 8.0 and treated with BaCl₂ to precipitate the FA as Ba-fulvate. The Ba-fulvate was dialysed until chloride-free, then passed through an H⁺ ion resin column to obtain FA (Kononova 1961). The arsenic-HA/FA complexation equilibria were examined by resorting to an ion-exchange method (Schnitzer and Skinner 1966).

The different HA/FA samples used in the present study were denoted as follows:

Humic/fulvic acid	Starting material
HA ₁	Humic acid extracted from Ghentugachhi Soil
HA ₂	Humic acid extracted from Gayeshpur Soil
HA ₃	Hydroquinone + cysteine
FA ₁	Fulvic acid extracted from Ghentugachhi Soil
FA ₂	Fulvic acid extracted from Gayeshpur Soil

Determination of contents of carboxyl and phenolic (OH) groups of humic and fulvic acids by the pH-potentiometric titration

Taking equal volumes of HA suspension in a set of bottles, different amounts of standard NaOH (0.01 N) solution were added. The suspensions were stirred well by a glass rod and the corresponding pH readings were measured with the help of the pH meter. The suspensions were then kept overnight to ensure equilibration and the next day the pH values of these suspensions were measured again. The carboxylic and phenolic (OH) group contents were recorded from the acidity equivalents of the first and the final inflexion points of the titration curves (Schnitzer and Khan 1972).

A fall in pH at the higher pH side of each alkali-treated HA sample was observed on standing for several hours after titration with alkali, and each pH was corrected back to the previous higher pH on fresh addition of the same alkali. Repeated fresh alkali additions at intervals of several hours were found to stabilise the (higher) pH (10.8) of the alkali-treated HA solutions finally. The total amount of fresh alkali needed to attain the stable higher pH as well as the time that elapsed after the initial completion of the titration were recorded for each HA sample.

Viscometric measurement

Prior to the viscosity measurement, all the carboxylic acid groups in each HA/FA sample were converted to the carboxylate form by the addition of

a requisite amount of alkali (NaOH) corresponding to the first inflexion point of the potentiometric titration curves.

The viscosities of the HA/FA suspensions were measured by the use of a Ubbelohde viscometer, having an average flow time of 399.0 s for 11 mL of distilled water at 31°C.

The temperature was kept constant at 31°C (within $\pm 0.1^\circ\text{C}$) by the use of a water thermostat provided with an electrical contact thermometer along with a solid-state relay unit. For each sample, viscosities were measured at 5 different concentrations. The mean flow time at each concentration was recorded. The densities of suspensions were also measured by the use of a specific gravity bottle. The specific viscosity (Eqn 1) at various concentrations for each HA sample was recorded and a plot of $\frac{\eta}{\eta_0} - 1$ v. \sqrt{c} constructed. The A and B coefficients of the Jones-Dole's equation (Chen and Schnitzer 1976) were obtained from the intercept and slope of the best-fit linear plot as above, according to the relationship:

$$\frac{\left(\frac{\eta}{\eta_0} - 1\right)}{\sqrt{c}} = \frac{\eta_{sp}}{\sqrt{c}} = A + B\sqrt{c} \quad (1)$$

where η and η_0 denote the respective viscosity coefficients at the same temperature of the HA suspension, having a percent concentration 'c', and water, while A and B are empirical constants sensitive to interactions between the disperse phase particles and those between the disperse phase and dispersion medium, respectively. The $B_{\text{expt.}}$ values of the humate anion were then obtained by subtracting the literature value of B_{Na^+} (Stokes and Mills 1965) from the $B_{\text{expt.}}$ value of sodium humate, i.e. $B_{\text{expt.}} = B_{\text{Na-HA}} - B_{\text{Na}^+}$ (Sanyal and Mandal 1983).

The $B_{\text{expt.}}$ values were approximated to the respective intrinsic viscosities, $[\eta]$ (Chen and Schnitzer 1976), which were used to compute the viscosity-average molecular weight (M) of the humic/fulvic substance by setting $K = 7.3 \times 10^{-4}$ and $\alpha = 0.65$ (Hansen and Schnitzer 1969; Relan *et al.* 1984) in the modified Staudinger's equation given below:

$$[\eta] = K(M)^\alpha \quad (2)$$

Surface tension measurement

Surface tension of aqua-humic acid at various concentrations was measured with the help of a du Nouy's tensiometer, maintaining the experimental temperature at $31^\circ \pm 0.1^\circ\text{C}$. The pH of each solution was maintained at 7.0. Surface tension values were measured at the air-water interfaces. The surface tension of water was taken as 71.09 dyne/cm at 31°C. The experiments were then repeated by taking the given HA/FA in a 0.1 M NaCl solution, while maintaining a pH of 7.0.

The surface excess, τ , was computed for both the aqua HAs and those in presence of 0.1 M NaCl by using the slope of γ v. c plots in the Gibbs adsorption equation:

$$\tau = -(1/RT) \times c \times (d\gamma/dc) \quad (3)$$

where T is absolute temperature, γ is surface tension of the solution, c is concentration in g/100 mL.

Visible spectrophotometric measurement

The optical densities at 465 nm (E_4) and 665 nm (E_6) for the dilute solutions of HA/FA (Na-form) were recorded using a Systronix visible spectrophotometer (Model 106) at pH 7.0 and 9.0.

Determination of stability constant of organo-arsenic complexes

The complexation equilibria between arsenic and the humic/fulvic substances were examined by following the standard method developed by Schnitzer and Skinner (1966) and the stability constants of the arsenic-humic/fulvic complexes formed recorded.

Aliquots of 0–10 mL of FA/HA solution were pipetted into 50-mL volumetric flasks and diluted to approximately 30 mL with distilled water. To each flask 5 mL of 1 N NaCl solution was added, followed by 6 mL of aqueous solution of $\text{Na}_2\text{HAsO}_4 \cdot 7\text{H}_2\text{O}$ containing 200 mg As/L. The pH was then adjusted to 5.0 by the addition of a few drops of 0.1 N NaOH or 0.1 N HCl solution as the case may be. Finally, the volume of each flask was made up to 50 mL with distilled water.

H^+ -saturated cation exchange resin (0.5 g), along with 50 mL of arsenic/HA or FA solution, was taken into a 250-mL conical flask with glass stopper and shaken for 1 h. Tests indicated that equilibrium was reached within this time. The exchange resin was then removed by filtration and the filtrate plus washings were taken to dryness on a hot water bath and then digested with 5:1 HNO_3 - H_2SO_4 on a hot plate (Schnitzer and Skinner 1966). The complete digestion took 1.5–2.0 h and the temperature attained during digestion was about 130°C. Each digest was diluted with distilled water to a volume of 100 mL. The concentrations of the arsenic in these aliquots were then measured by the atomic adsorption spectrophotometer coupled with the hydride generator unit (Model GBC-932B).

The logarithm of the stability constant of the arsenic/humate or fulvate complex ($\log K$) was determined using the following relationship:

$$\log[(\lambda_0/\lambda) - 1] = \log K + x \log[\text{HA}] \quad (4)$$

where λ_0 is the distribution constant in the absence of HA/FA, λ is the distribution constant in the presence of HA/FA, $\log K$ is stability constant of arsenic/HA or FA complex, x is number of mol HA/FA that combine with 1 mol arsenic, and [HA] is concentration of HA/FA in mol per L. λ_0 was determined from the following expression:

$$\lambda_0 = \frac{\alpha_0 V}{(100 - \alpha_0)g} \quad (5)$$

where α_0 is percent of total metal bound to exchange resin, $(100 - \alpha_0)$ is percent of total metal remaining in solution, V is volume of solution (100 mL), and g is weight of exchange resin (0.5 g).

Lambda (λ) was measured in the same manner as for λ_0 , but with differing HA/FA concentrations. The value of x can be obtained separately from the slope of the best-fit linear plot of $\log[(\lambda_0/\lambda) - 1]$ v. the different HA/FA concentrations, while the $\log K$ value is taken from the intercept (Eqn 4).

Exchangeability of phosphate and nitrate (replacing anions) with humic-arsenate complexes

The exchangeability of phosphate and nitrate with humic/fulvic-arsenate complexes was determined by placing equal portions of HA and FA suspensions in stoppered bottles. A measured amount of aqueous arsenic solution of known strength (25 mg/L) was added to each bottle. The bottles were shaken for 1 h and kept overnight for equilibration. The resulting arsenic-HA/FA complexes were dialysed against distilled water and used in the exchange studies.

Equal portions (2 mL) of arsenic-HA/FA complex were placed in centrifuge tubes, and to these were added increasing amounts of each electrolyte (25 mg P/L and 25 mg N/L as phosphate and nitrate, respectively). The total volume was adjusted to 25 mL in all the cases, maintaining a pH of 5.0. The tubes were shaken for 2 h and kept overnight for equilibration. The mixtures were then centrifuged at 5000 r.p.m. and the amount of arsenic released was determined by using an atomic absorption spectrophotometer (Model GBC-932B) (Pal and Chakravarti 1987).

Results and discussion

Some important physicochemical properties as well as the Taxonomic Classification of the experimental soils are shown in Table 1. The soils were neutral to slightly alkaline in

Table 1. Important physicochemical properties of soils

Sand	Silt (%)	Clay	Texture	pH ^A	EC ^A (dS/m)	Organic C (g/kg)	SSA ^B (m ² /g)	CEC ^C	Exchang. cations ^C		Amorphous Fe (%)	As ^D (mg/kg)	P ^D (mg/kg)	
								Na	K	Ca + Mg				
2.1	40.1	57.8	Silty clay	7.89	0.40	3.9	134	29.2	0.08	0.87	11.03	0.52	1.21	68.5
<i>Ghentugachhi, Typic Haplustepts^E</i>														
13.6	64.8	21.6	Silty clay	7.83	0.28	5.3	74	22.0	0.03	0.33	5.98	0.25	0.34	43.6
<i>Gayeshpur, Typic Haplustepts^E</i>														

^A(1 : 2.5). ^BSpecific surface area. ^C(cmol (+)/kg). ^DOlsen extractable. ^ESoil Taxonomy.

Table 2. Functional group (i.e. content of -COOH and phenolic -OH groups) analysis (cmol/kg) of humic/fulvic acid samples from pH-potentiometric titration curves

Humic acid: HA₁, extracted from Ghentugachhi soil; HA₂, extracted from Gayeshpur soil; HA₃, synthesised from hydroquinone + cysteine.

Fulvic acid: FA₁, extracted from Ghentugachhi soil; FA₂, extracted from Gayeshpur soil

HA/FA sample	Amount of carboxyl (COOH) group	Amount of phenolic (OH) group	Total acidity
HA ₁	18.0	16.0	34.0
HA ₂	84.0	26.0	110
HA ₃	15.0	14.0	29.0
FA ₁	194	41.0	235
FA ₂	67.0	25.0	92.0

nature with varying organic carbon and clay contents. The oxalate-extractable (amorphous) Fe content was higher in the Ghentugachhi soil than the Gayeshpur soil, as were the Olsen-extractable (0.5 M NaHCO₃, pH 8.5) arsenic and phosphorus contents, specific surface area and CEC.

The present HA/FA samples had higher carboxylic acidity than the phenolic (OH) group contents (Table 2). The FA sample extracted from the Ghentugachhi soil (FA₁) had the highest total acidity with the remaining samples arranged sequentially as follows: FA₁ > HA₂ > FA₂ > HA₁ > HA₃. The range of total acidity of the given humic/fulvic substances agreed, in general, with those reported by Ghosh and Schnitzer (1980), Saha and Sanyal (1988), Tomar *et al.* (1992a), and Datta *et al.* (2001).

Table 3 records the experimental B values (B_{expt.}) of the HA/FA samples, which are related to the modification of micro-viscosity of water in the neighbourhood of the disperse phase particles, compared with the bulk viscosity. As stated earlier, the slope of the best-fit linear plot of η_{sp}/\sqrt{c} v. \sqrt{c} by the least square method (Fig. 1) provides the B_{expt.} value. The data for the present HA/FA samples suggest reinforcement of 'local' water structure leading to an increase in viscosity of HA/FA solutions (relative to water) and a positive viscosity B coefficient. On the basis of such viscometric studies, Mandal and Sanyal (1984) proposed a means of arriving at the folded shape by way of comparing the experimental B coefficient (B_{expt.}) of the Jones-Dole's equation with the additive value, B_{theor.}, computed by summing up the B contributions due to the presence of various hydrophilic groups in humic acid molecules.

The HAs (HA₁ and HA₂) extracted from soils showed a relatively higher B_{expt.} value than did the HA synthesised from hydroquinone and cysteine (HA₃). The FA (FA₁) extracted from the Ghentugachhi soil showed lower B_{expt.} than that of Gayeshpur soil (FA₂). The B_{expt.} values of HA/FA are expected to increase with an increase in carbon number in the acid molecules, provided that the solutions/suspensions are dilute and thus the disperse phase or the solute-solute interactions negligible (i.e. the A coefficient of Eqn 1 is negligibly small) (Sanyal 2001). At the same time, the coiled character of the HA/FA molecules may also be responsible for the changes of the B_{expt.} values (Sanyal 1984). The B_{expt.} values of the HAs (HA₁ and HA₂) and to some extent of FA (FA₂) extracted from the soil samples were higher (Table 3)

Table 3. Evaluation of intrinsic viscosity ($[\eta]$) and molecular weight (M) of humic/fulvic acid samples from viscometric analysis

HA/FA sample	B _{expt.} of Na humate/fulvate anion (per cent ⁻¹)	B _{expt.} of humate/fulvate anion (per cent ⁻¹) [B _{expt.} = B - B _{Na}]	$[\eta]$ (100 mL/g)	Molecular weight (M) of HA/FA
HA ₁	0.7320	0.7316	0.7316	41 254
HA ₂	0.3380	0.3376	0.3376	12 557
HA ₃	0.1528	0.1524	0.1524	3704
FA ₁	0.0910	0.0906	0.0906	1661
FA ₂	0.5540	0.5536	0.5536	26 869

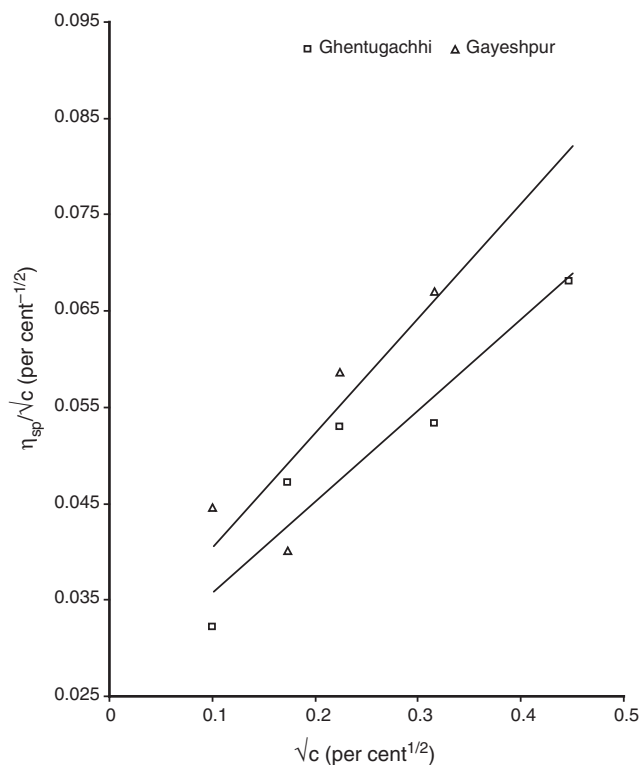


Fig. 1. Representative plots of η_{sp}/\sqrt{c} v. \sqrt{c} for fulvic acids extracted from Ghentugachhi and Gayeshpur soils.

than that of the synthetically prepared acid (HA₃). This may be related to the higher degree of stable coiling, and hence higher molecular weight of the former than the latter. Intrinsic viscosities, $[\eta]$, calculated from the $B_{\text{expt.}}$ values (Chen and Schnitzer 1976) were used for computing the viscosity-average molecular weight (M) by resorting to the modified Staudinger's equation (Eqn 2). A higher $B_{\text{expt.}}$ value leads to a higher molecular weight. The naturally occurring HAs and FAs generally showed the higher molecular weight than did the synthetic acid (Table 3).

Table 4 records the surface tension values (γ) of HA/FA samples that exhibit lower γ values than that of pure water at the same temperature. The decrease in surface tension with increasing concentration of HA/FA suspension is demonstrated in the representative plots of γ v. c (Fig. 2). The latter signifies the predominantly hydrophobic character of the present HA/FA samples which thus tend to minimise their contact surface area with water (Sanyal 2001). This is also in agreement with the positive surface excess (τ ; Eqn 3) of these HA/FA samples (Shaw 1970), more so for the HAs possessing higher τ values (Table 4). Further, the natural HAs (HA₁ and HA₂) exhibited much larger τ value than the synthetic HA (HA₃). This is again supported by the higher molecular weights of the former (Table 3).

A rise in τ for nearly all the HA and FA samples was observed in the aqueous solution of 0.1 M NaCl (Table 4),

which may be due to the higher degree of 'exclusion' of the predominantly hydrophobic HA/FAs in the presence of the strongly hydrated electrolyte (Sanyal 2001).

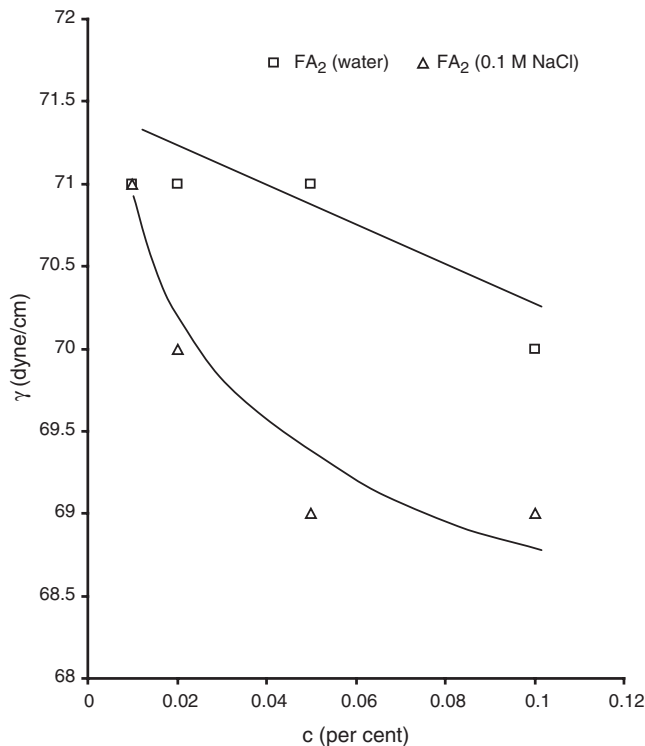
The (E_4/E_6) ratio of the given HA samples provides an index of aliphatic–aromatic balance, which, in turn, depends on the extent of coiling of the acid molecules. The aromatic moiety, being the most hydrophobic part, tends to hide itself in the innermost core of the coil, leaving the aliphatic moiety forming the outer periphery of the coil. This results in a greater absorbance by the aliphatic part (i.e. absorbance at 465 nm) than the aromatic part (i.e. absorbance at 665 nm) due to a particular conformational arrangement of the aliphatic and aromatic moiety (Kar and Sanyal 1990; Datta *et al.* 2001). The (E_4/E_6) values of the HA/FA samples (Table 5) being greater than unity (at both the pH 7.0 and 9.0) support the above contention. Further, between the HA and the corresponding FA fractions of the 2 experimental soils, the FA fraction generally exhibits a higher (E_4/E_6) value, in agreement with the findings of Tomar *et al.* (1992b), Rivero *et al.* (1998), and Sanyal (2001).

The amount of fresh excess alkali (Table 6) needed by HA/FA to attain a stable, higher pH (when HA/FA molecules were unfolded more under thermodynamic compulsion; Sanyal 1984, 2001) evidently depends on the amount of the remainder acidic groups that were not accessible to alkali. The 'steric hindrance', associated with a particular conformation of these molecules due to their coiled nature, governs the phenomenon. Again, the time required to attain a stable higher pH depends on the ease of decoiling, which is inversely related to the intramolecular H-bonding. Formation of such bonds is often interrupted by the R group of the amino acid unit (in synthetic humic acid), which governs the 'flexibility' of the molecules. The naturally occurring humic samples, namely HA₁ and HA₂, required a longer time to attain the stable higher pH than did the HA₃ sample, despite consuming less fresh alkali. This may have resulted from a highly coiled, polycondensed structure (obtained through polymerisation, engaging the functional groups, e.g. –COOH and phenolic –OH groups) of these naturally occurring humic acids, as well as their much higher molecular weight. Such coil formation does not necessarily require the participation of the carboxylic and phenolic (OH) groups in intramolecular hydrogen bond formation, and hence not much 'excess acidity' gets locked up inside the coils. The same may hold for FA₁ and FA₂ as well. The presence of the small R group of the synthetically prepared acid (HA₃) than that in the natural HAs (HA₁ and HA₂) may have been responsible for the comparatively shorter time to attain the stable higher pH by the former for reasons explained earlier. According to the flexibility of the HA/FA coils, based on the time required (i.e. less time corresponds to higher degree of flexibility of the humic/fulvic molecule) to attain the stable higher pH (Table 6), the given

Table 4. Surface tension and surface excess of humic/fulvic acid samples at different concentrations of humic/fulvic acids

Surface tension of water at 31°C = 71.09 dyne/cm

HA/FA sample	Conc. (%) (c)	Surface tension (dyne/cm) in:		Surface excess (mol/cm ²) in:	
		Water (γ)	Presence of 0.1 M NaCl solution (γ')	Water ($\tau \times 10^{11}$)	Presence of 0.1 M NaCl solution ($\tau' \times 10^{11}$)
HA ₁	0.6	67.0			
	0.5	67.0	67.0		
	0.4	68.0	67.0	8.33	10.1
	0.3	69.0	68.0		
	0.1	71.0	69.9		
HA ₂	0.6	67.0			
	0.5	67.0	67.0		
	0.4	68.0	68.0	7.93	5.92
	0.3	69.0	68.0		
	0.1	70.0	69.0		
HA ₃	0.05	71.9	71.4		
	0.07	70.5	69.9	2.43	4.91
	0.10	69.8	69.3		
	0.12	68.1	68.8		
FA ₁	0.20	68.0			
	0.10	69.0	68.0		
	0.05	69.0	68.0	0.048	0.052
	0.02	69.0	68.0		
	0.01	69.0	69.0		
FA ₂	0.20	70.0			
	0.10	70.0	69.0		
	0.05	71.0	69.0	0.150	0.083
	0.02	71.0	70.0		
	0.01	71.0	71.0		

**Fig. 2.** Representative plots of surface tension (γ) v. concentration (c) for fulvic acid (FA₂) fraction extracted from Gayeshpur soil.

HA/FA samples can be arranged sequentially as follows: HA₃ = FA₁ > FA₂ > HA₂ > HA₁.

Table 7 records the values of x (number of mol HA/FA that combine with 1 mol arsenic) as well as $\log K$ (where K is the stability constant of arsenic-HA/FA complexes). At pH 5.0, the $\log K$ values of arsenic-humate/fulvate complexes varied from 2.63 to 8.35, the highest $\log K$ and x values being obtained with FA₁ (i.e. FA extracted from Ghentugachhi soil). The $\log K$ values were generally comparable to those reported earlier for heavy metal-humic complexes (Schnitzer and Skinner 1966; Matsuda and Ito 1970; Stevenson 1976; Stevenson 1977), but were larger than the values observed for stability constants of organometallic complexes formed by root exudates containing low molecular weight organic acids (Mench *et al.* 1988). It has been proposed that higher molecular weight HAs generally bind heavy metals more strongly than do the lower molecular weight acids (Young *et al.* 1982). The $\log K$ values for the present arsenic-HA/FA complexes fell in the following order: FA₁ > HA₃ > HA₂ > FA₂ > HA₁. These $\log K$ values exhibited negative relationships, especially with molecular weight ($r = -0.979$), excess time to attain the stable higher pH ($r = -0.711$) of HA/FA component, and also surface excess in water ($r = -0.303$), and positive relationships with total acidity ($r = 0.460$) and (E₄/E₆) at pH 7.0 ($r = 0.414$). As stated earlier, hydrophobicity of

Table 5. Spectrophotometric (E_4 and E_6) analysis of humic/fulvic acid samples at two different pH values

HA/FA sample	E_4/E_6 at pH:	
	7.0	9.0
HA ₁	5.54	6.82
HA ₂	13.0	10.3
HA ₃	2.79	2.67
FA ₁	14.6	16.2
FA ₂	6.96	11.6

Table 6. Amount of fresh alkali and the period of time required to attain a stable higher pH (10.8) of humic/fulvic acid

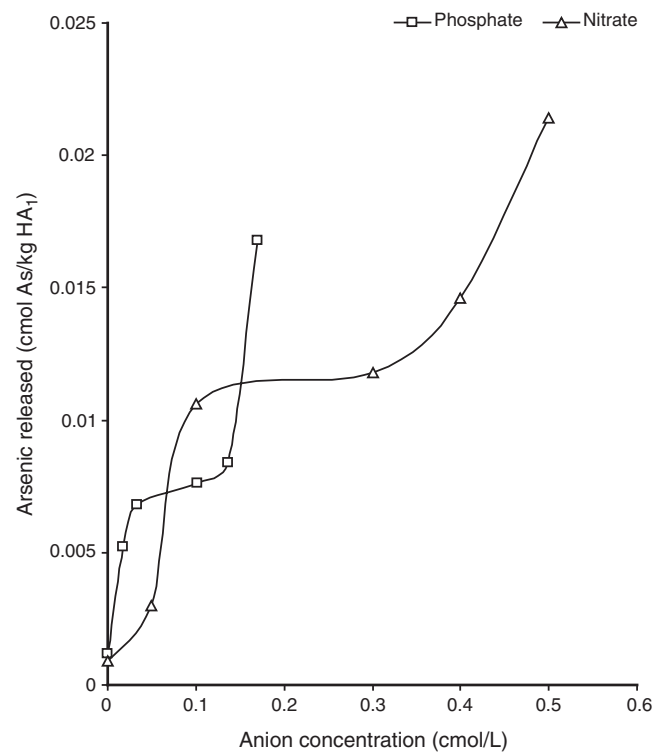
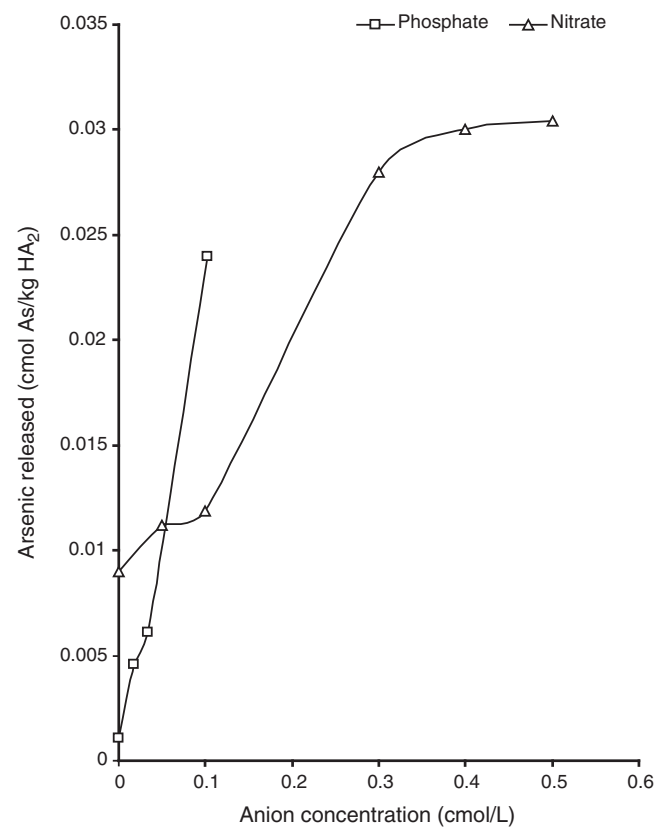
HA/FA sample	Amount of fresh alkali required (mol/kg)	Time required (h)
HA ₁	0.600	92
HA ₂	0.730	90
HA ₃	10.1	72
FA ₁	4.10	72
FA ₂	5.60	87

Table 7. Stability constant ($\log K$) and mole ratio (x) of HA/FA–arsenic complexes at pH 5.0

HA/FA sample	x	$\log K$
HA ₁	0.505	2.63
HA ₂	1.530	7.91
HA ₃	1.530	8.32
FA ₁	1.830	8.35
FA ₂	0.802	4.31

the humic/fulvic substances is expected to increase with molecular weight, and consequently lead to an increase in surface excess in water and also the excess time needed for attaining the stable higher pH during the pH-potentiometric titration of the HA/FA samples. This would cause a likely fall in the stability of the complex formed with aqueous arsenic by the given HA/FA substances, and hence the negative relationship between $\log K$ on one hand, and the viscosity–average molecular weight, excess time, and surface excess in water of the present HA/FA samples on the other. The rather weak positive correlation of $\log K$ with total acidity of the given HA/FA tends to suggest that both the phenolic and the carboxylic groups, rather than one or the other, may have been involved in the resulting complex formation process. This is supported by the findings of Khalili (1990) and Stevenson (1991) as reviewed recently by Sanyal (2001).

Figures 3, 4, 5, 6 illustrate the release isotherms of arsenic from the humic (Figs 3 and 4) and fulvic (Figs 5 and 6) complexes due to treatment of the latter with phosphate and nitrate salts. The release isotherm for both the phosphate and

**Fig. 3.** Release of arsenic from HA₁–As complex by aqueous phosphate and nitrate anions.**Fig. 4.** Release of arsenic from HA₂–As complex by aqueous phosphate and nitrate.

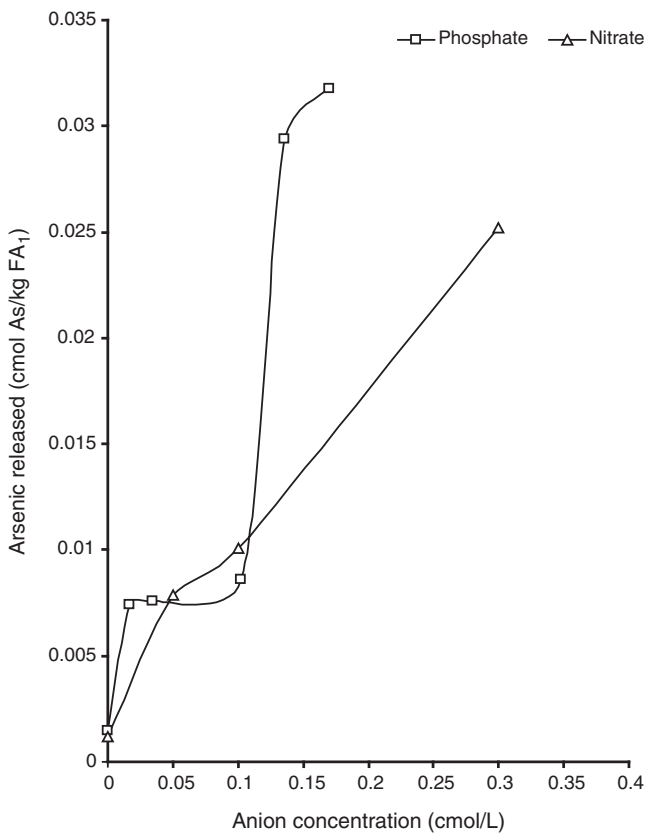


Fig. 5. Release of arsenic from FA₁-As complex by aqueous phosphate and nitrate.

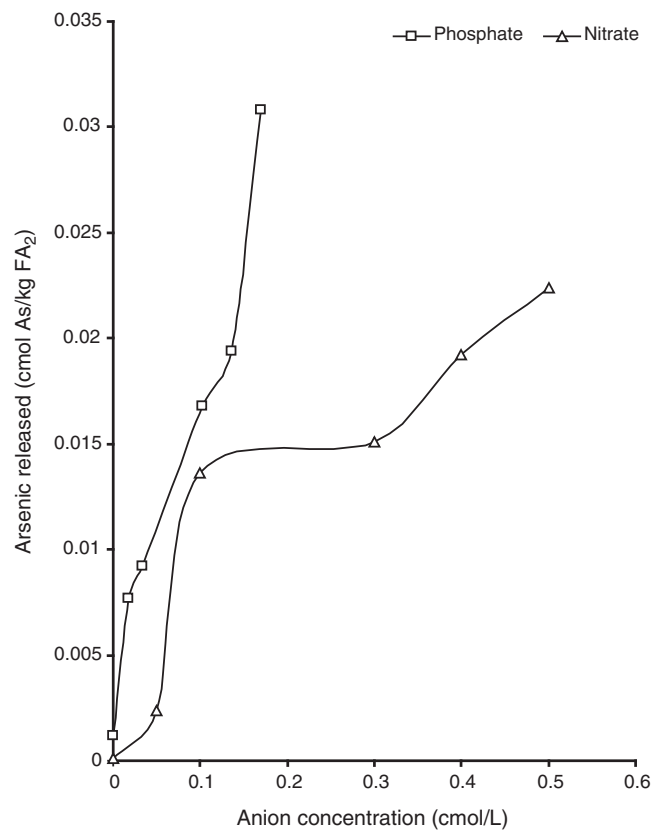


Fig. 6. Release of arsenic from FA₂-As complex by aqueous phosphate and nitrate.

nitrate ions suggested that the exchange process occurred in 2 or 3 stages (Figs 3 and 4). In the initial stage, the extent of exchange at equilibrium was comparatively high, followed by a limiting equilibrium exchange, for instance, for nitrate. These figures suggested that phosphate, in general, demonstrated a greater tendency to displace arsenic than did nitrate at both the lower and the higher concentrations. The release isotherm of fulvic-arsenic complex (Figs 5 and 6) also exhibited similar trends to the humic-arsenic complexes. Such basically 2-phase exchange of arsenic from its organic complexes by phosphate and nitrate may possibly reflect the initial exchange from relatively loosely held sites (e.g. outer-sphere complexation sites), followed by that from relatively strongly bonded organo-arsenic sites, possibly the inner-sphere complexation sites (Sanyal 2001). Further, the relative replacing power of phosphate and nitrate may also be related to the relatively high ability of phosphate to replace arsenic from the common sorption sites in soils, clays, and soil organic matrix by way of offering stronger competition for these sites through the inner-sphere complex formation, i.e. through specific sorption (Hingston *et al.* 1971; Manning and Goldberg 1996). On the other hand, the greater electronegativity, smaller size, and coordination of the nitrate ion implies its lesser affinity to sorption (compared

with phosphate on a given site) at identical concentrations to phosphate.

Conclusions

The findings from the studies on arsenic-humic/fulvic complexation equilibria are expected to elucidate the stability of these complexes, demonstrating their dependence on the nature and properties of the humic polymer, which, in turn, would affect the retention/release of such toxic metal in soil matrix. The release isotherms of arsenic from its humic and fulvic complexes on treatment with phosphate and nitrate salts demonstrated a greater degree of exchangeability of arsenic by phosphate than by nitrate from such organo-arsenic complexes, although the extent of exchange was small in both the cases. The findings are expected to have some bearing on the ability of the native soil organic fraction to sorb arsenic, thereby moderating its toxicity in the soil/crop system, and via the latter, its entry in the food chain. However, the tropical soils were poor in soil organic matter and it may thus be practical to adopt appropriate management practices (such as recycling of crop residues, incorporation of organic manures, etc.) to improve the soil organic matter stock and hence arsenic retention in the arsenic-affected soils. We are currently investigating some of these aspects.

Acknowledgments

The financial assistance of the Indian Council of Agricultural Research, New Delhi, India during the entire course of the study is gratefully acknowledged.

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Manuscript received 19 July 2003, accepted 20 May 2004