Mineralogy, Bacterial Diversity, and Bioavailable Trace element Loading in the Silt/Clay Fraction of Surficial Soils from Iraq and Kuwait

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Abstract:

In arid environments, winds, storms, and other physical disturbances can suspend surficial sands, silts, and clays and subsequently transport these materials over distances from meters to hundreds of kilometers. Exposure to burdens of airborne "dust" during mass transport events such as sand storms has been linked to human health effects including respiratory problems and disease. Bulk surface soil samples were collected from Iraq and Northern Kuwait in the summer of 2007. Samples were mechanically-sieved to determine the relative grain size distributions and the size distribution of the silt/clay fraction was further quantified using Laser Diffraction Particle Size Analysis (LDPSA). The silt/clay fraction was then characterized geochemically using Energy-Dispersive X-ray Fluorescence (ED-XRF) and clay minerals were identified using X-ray Diffraction (XRD). Physiologically-based fluid extractions were conducted on the same samples and analyzed by High Resolution Inductively-Coupled Mass Spectrometry (HR-ICP-MS) to investigate the potential bioavailability of trace metals associated with the silt-clay fraction. Results suggest grain size, clay content, and mineralogy influence bioavailable trace metal loading in the readily-suspendible silt/clay fraction of Iraq and Kuwait surficial soils but proximately to urban environments can result in significantly higher loadings. Comparing trace metal loadings to bacterial species diversity on the silt/clay fraction of these soils, as determined from length heterogeneity polymerase chain reaction (LH-PCR) on DNA extracts, suggests that high trace metal loading can lower bacterial species diversity, potentially indicating toxicity. Understanding this potential linkage is important to address questions related to human health in Iraq, Kuwait, and other arid regions.

Study Area and Methods:

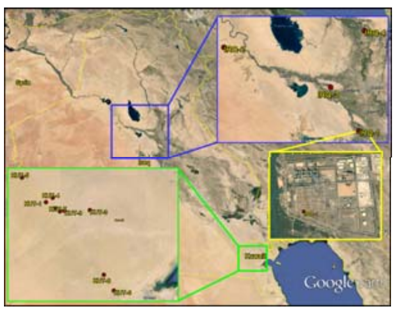
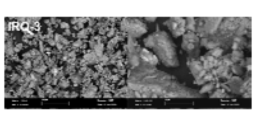


Figure 1. Study area showing locations in Iraq and Kuwait where bulk surface soil samples (upper 0-2 cm) were collected in the summer of 2007. Each surficial soil sample is a composite of 5 x ~ 200 ml samples of soil collected ~ 25 - 50 m apart for a total of ~ 1 L of soil. The area in blue shows sampling locations in Iraq (IRQ-1 through 4). The area in yellow highlights the urban environment around sampling location IRQ-1. The area in green shows sampling locations in Northern Kuwait (KUT-1 through 9).

Figure 1 shows locations where surface soil samples were collected in summer, 2007. After collection, bulk samples were mechanically-sieved to determine relative grain size distribution.

- Material < 75 µm in diameter was classified as the silt/clay fraction and then analyzed for grain size distribution using a Beckman-Coulter LS13 320 Series Laser Diffraction Particle Size Analyzer (LDPSA).
- Clay mineralogies in the silt/clay fraction were determined by X-Ray Diffraction (XRD).
- Major, minor, and trace elemental composition of the silt/clay fraction (< 75 µm) was determined using a Spectro XEPOS Energy-Dispersive X-Ray Fluorescence (ED-XRF) Spectrometer. Physiologically-based fluid extractions were then performed and extracts analyzed using a Thermo-Finnigan ELEMENT 2 High Resolution Inductively-Coupled Mass Spectrometry (HR-ICP-MS).
- Total DNA extraction was performed on ~ 0.5 g of the < 25 µm fraction of bulk surficial soil and analyzed by length heterogeneity polymerase chain reaction (LH-PCR).



Results:

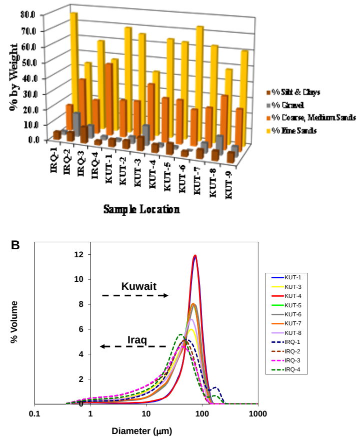


Figure 2. (A) Relative grain size distribution of Iraq and Kuwait bulk surface soil samples (~ 30 – 100 g) after mechanical sieving through #5, #40, and #200 ASTM mesh size sieves. **(B)** Size distribution of the silt/clay (< 75 µm) fraction of Iraq and Kuwait bulk surface soil samples from analysis using a Beckman-Coulter LS13 320 Series Laser Diffraction Particle Size Analyzer (LDPSA) with methods modified from *McCarthy (2008)*.

Table 1. Silt/Clay Size Analysis Summary

Sample	% < 10 µm	Mean Diameter (µm)	Specific Surface Area (µm^2)
IRQ1	10.5	37.3	1643
IRQ2	21.7	22.4	2862
IRQ3	22.1	21.8	2885
IRQ4	16.2	26.8	2290
KUT1	2.5	62.7	793
KUT3	13.9	31.2	2042
KUT4	3.3	60.4	874
KUT5	8.1	41.7	1438
KUT6	7.5	44.0	1343
KUT7	9.2	39.8	1584
KUT8	12.5	33.8	1893

Figure 2A shows surface soils from Iraq and Kuwait were generally 5-10% silt/clay. The silt/clay fraction of surface soils from Iraq had a lower mean diameter and a greater specific surface area than the silt/clay fraction of surface soils from Kuwait (**Fig. 2B; Table 1**).

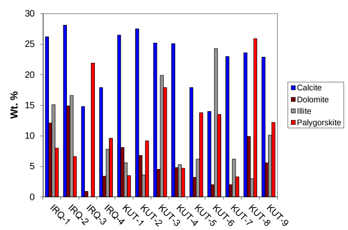


Figure 3. Relative % (by weight) of select clays in the silt/clay (< 75 µm) fraction of Iraq and Kuwait bulk surface soil samples as determined by XRD (CuKα radiation at 55kV and 120mA, 0.02 degree (2θ) stepsize, 10 sec/step, over angular range of 5-70°). Patterns were fitted with an initial seven-phase model developed from search/match using calculated powder patterns from the NIST/ICSD crystallographic database of minerals and refined using Jade 8 whole pattern fitting.

Figure 3 shows the relative % (by weight) of select clay mineral phases in the silt/clay fraction of surface soils from Iraq and Kuwait. Note that a significant percentage of each sample is palygorskite, a clay mineral common to arid regions of the Middle East.

Table 2. Major, minor, and trace elemental composition of in the silt/clay (< 75 µm) fraction of Iraq and Kuwait surface soil samples as determined using a Spectro XEPOS ED-XRF Spectrometer. ~2-5 g of material was analyzed under He purge using NIST 2709 San Joaquin Soil and CRM PACS-2 and MESS-3 reference standards.

		Mean	Min	Max
Mg (%)	Iraq	4.0	2.8	4.8
	Kuwait	4.7	4.2	5.4
Al (%)	Iraq	4.0	3.2	4.6
	Kuwait	4.0	3.6	4.5
Si (%)	Iraq	14.8	12.9	17.1
	Kuwait	17.0	16.0	18.5
K (%)	Iraq	1.0	0.7	1.2
	Kuwait	0.9	0.8	1.0
Ca (%)	Iraq	11.6	9.5	12.8
	Kuwait	12.2	10.6	13.0
Fe (%)	Iraq	3.4	2.7	4.2
	Kuwait	3.6	3.1	4.0
Sr (mg/kg)	Iraq	687.6	440.7	1033.7
	Kuwait	554.7	317.3	1051.5

		Mean	Min	Max
V (mg/kg)	Iraq	151.8	80.0	278.0 (IRQ-1)
	Kuwait	104.3	87.6	125.9
Cr (mg/kg)	Iraq	464.1	219.4	620.7
	Kuwait	2272.5	789.7	4383.2 (KUT-1)
Mn (mg/kg)	Iraq	565.5	445.1	720.7
	Kuwait	674.2	552.4	1014.0
Ni (mg/kg)	Iraq	129.3	93.7	196.8 (IRQ-1)
	Kuwait	110.4	97.5	126.7
Cu (mg/kg)	Iraq	29.6	25.9	34.6
	Kuwait	24.3	21.4	35.5
Zn (mg/kg)	Iraq	163.6	56.8	365.5 (IRQ-1)
	Kuwait	55.2	47.9	60.6
As (mg/kg)	Iraq	6.5	6.0	6.9
	Kuwait	5.4	2.7	12.2 (KUT-3)
Pb (mg/kg)	Iraq	19.5	15.6	23.7
	Kuwait	12.7	9.5	19.4

Table 2 shows the relative major, minor, and trace elemental composition of the silt/clay fraction of surface soils from Iraq and Kuwait. Significantly high trace elemental concentrations are highlighted with the location. **Table 3** shows the yields of physiologically-based fluid extractions on the silt/clay fraction of surface soils samples. Note the high yield of vanadium (and nickel) in sample IRQ-1 in both simulated GF and PSF.

	% Extracted in GF				
Sample	As	Cu	Ni	Pb	V
IRQ-1	18.1	11.1	7.0	48.8	26.0
IRQ-2	26.6	10.5	3.5	31.5	8.0
IRQ-3	25.5	13.2	4.3	65.1	6.1
KUT-1	56.7	8.0	1.4	27.9	1.7
KUT-2	21.4	12.0	1.5	53.3	2.0
KUT-3	27.1	8.8	2.2	20.0	8.8
KUT-4	100.7	5.6	1.3	19.6	2.1
KUT-5	12.7	7.5	1.5	23.2	2.3
KUT-6	42.8	12.5	3.7	20.9	3.7
KUT-7	28.1	12.5	2.7	67.2	4.0
KUT-8	65.2	8.6	2.9	15.0	4.7
KUT-9	22.5	7.3	1.7	22.0	2.8

<u>Gastric Fluid (GF)</u>
0.4M Glycine; pH 1.5 (HCl)
37°C; 1 hour
(*Schaider et al., 2007; U.S. EPA, 2004*)

<u>Phagolysosomal Simulant Fluid (PSF)</u>
20 mM Potassium Hydrogen Phthalate
110 mM NaCl
6.0 mM Glycine
1.0 mM Na_2HPO_4
0.5 mM Na_2SO_4
0.20 mM $CaCl_2$
50 ppm Benzalkonium Chloride*
pH 4.5; 37°C; 5 days

	% Extracted in PSF				
Sample	As	Cu	Ni	Pb	V
IRQ-1	16.2	2.5	2.9	<MDL	9.8
IRQ-2	10.6	3.8	0.6	<MDL	2.3
IRQ-3	18.7	4.0	0.8	0.7	1.4
KUT-1	11.1	10.2	0.4	<MDL	0.7
KUT-2	12.4	9.7	0.4	0.7	0.8
KUT-3	26.5	2.4	0.4	<MDL	3.9
KUT-4	9.6	7.5	0.3	<MDL	0.9
KUT-5	7.8	11.6	0.5	<MDL	0.9
KUT-6	12.7	10.9	1.2	<MDL	1.1
KUT-7	22.2	5.6	0.5	<MDL	1.2
KUT-8	37.2	7.1	0.7	<MDL	1.8
KUT-9	17.3	6.5	0.4	<MDL	0.9

Table 3. Physiologically-based fluid extraction yields for the silt/clay fraction (< 75 µm) of Iraq and Kuwait bulk surface soil samples. Extractions were performed using methods modified from *Schaider et al. (2007)* and analyzed using a Thermo-Finnigan ELEMENT 2 HR-ICP-MS.

Discussion:

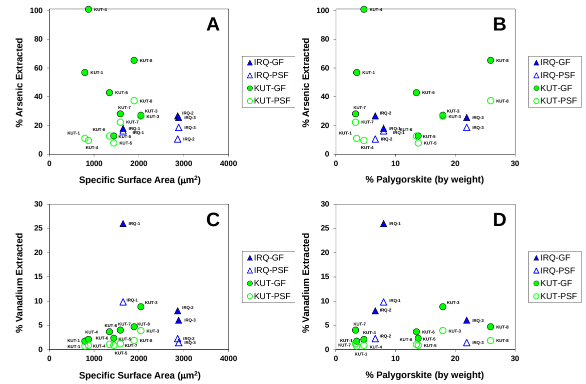


Figure 4. Physiologically based fluid extractions yields for As plotted vs. **(A)** specific surface area and **(B)** palygorskite content and for V vs. **(C)** specific surface area and **(D)** palygorskite content of the silt/clay (< 75 µm) fraction of Iraq and Kuwait bulk surface soil samples.

Figure 4 shows physiologically-based extraction yields for As and V plotted against specific surface area and palygorskite content. There is no clear relationship between extractable As or V and specific surface area or palygorskite content but the sample from IRQ-1 stands out as a clear outlier for extractable vanadium. IRQ-1 is clearly from an urbanized environment (**Fig. 1**). Vanadium may indicate contamination from oil and/or oil refinement (Al-Jebouri et al., 2014).

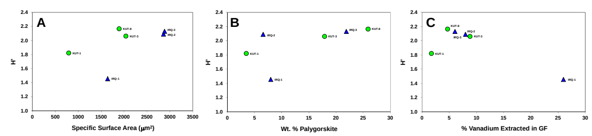


Figure 5. Shannon-Weiner Diversity Index [H' = -Σ p_iLn(p_i)] values (< 25 µm fraction) plotted vs. **(A)** specific surface area **(B)** palygorskite content, and **(C)** % V extracted in simulated gastric fluid for the silt/clay (< 75 µm) fraction of Iraq and Kuwait surface soil samples. Total DNA extraction was performed using a BP Medical FastDNA SPIN kit. Following extraction, total DNA concentrations were quantified using the Quant-iT Pico Green dsDNA Assay (Invitrogen) and analyzed by LH-PCR using primers designed to amplify variable regions 1 and 2 of the 16S rDNA gene from prokaryotic organisms. Amplified PCR fragments were separated by capillary electrophoresis (ABI 310 Genetic Analyzer) and analyzed with Genetic Analysis Collection Software (v 2.0) and GeneMapper (v 4.0).

Figure 5 shows the Shannon-Weiner Diversity Index for silt/clay (< 75 mm) fraction of Iraq and Kuwait surface soil samples plotted against specific surface area, palygorskite content, and % V extracted in simulated GF. There appears to be a relationship between bacterial species diversity, specific surface area, and palygorskite content, however, bacterial species diversity is clearly lower in the sample with high extractable V (IRQ-1).

Conclusions:

•Physiologically based fluid extractions suggest a significant proportion of the trace element load in the readily-suspendible silt/clay fraction of Iraq and Kuwait surface soils may be bioavailable, especially in soils near urban environments.

•Trace metal bioavailability is a function of anthropogenic loading with specific surface area, speciation, and clay content and mineralogy also likely factors.

•High bioavailable trace metal loading in the readily-suspendible silt/clay fraction of Iraq and Kuwait surface soils may lower bacterial species diversity indicating potential toxicity.

Acknowledgements: U.S. Naval Research Laboratory (Code 6114 and 6900); Kathleen McCarthy, Harvard University School of Public Health; and Britt Argow, Wellesley College. This research was supported in part by the Office of Naval Research (Code 342).