

Distribution of sorbed phenanthrene and pyrene in different humic fractions of soils and importance of humin

B. Pan^a, B.S. Xing^b, W.X. Liu^a, S. Tao^{a,*}, X.M. Lin^a, X.M. Zhang^a, Y.X. Zhang^a,
Y. Xiao^a, H.C. Dai^a, H.S. Yuan^a

^a Laboratory for Earth Surface Processes, College of Environmental Sciences, Peking University, Beijing 100871, China

^b Department of Plant, Soil and Insect Sciences, University of Massachusetts, Stockbridge Hall, Amherst MA 01003-7245, USA

Received 19 June 2005; received in revised form 3 November 2005; accepted 8 November 2005

Humic fractionation after sorption experiment revealed that humin fraction is the main region for slow and nonlinear sorption of phenanthrene and pyrene.

Abstract

Contributions of fulvic–humic acids (FA/HA) and humin (HM) to sorption of phenanthrene (PHE) and pyrene (PYR) in a soil were differentiated using a humic separation procedure after multi-concentration sorption experiments. It was found that the amount of solutes in FA/HA did not change significantly after 48 h, while that in HM increased continuously and slowly up to the end of the experimental period (720 h), indicating that HM was the main region for slow sorption. Based on the fitting results using Freundlich equation, it was found that nonlinearity of both solutes was greater in HM than in FA/HA, consistent with the sorption characteristics of individually extracted HA and HM in a separate experiment. The observed nonlinearity of the solute distribution was confirmed by using three other soil samples with organic carbon contents ranging from 0.7 to 7.9%. Distribution dynamics of PHE and PYR among various fractions were also discussed.

© 2005 Elsevier Ltd. All rights reserved.

Keywords: Sorption; Distribution; Nonlinearity; Soil organic matter; PAHs

1. Introduction

Sorption is a major process determining the fate and bio-availability of hydrophobic organic chemicals (HOCs) in soils. It has been reported that soil organic matter (SOM) is the predominant sorbent for HOCs (Pignatello, 1998). Although exact sorption mechanisms by SOM are unclear, a great progress has been made towards the understanding of sorption of HOCs by SOM. One of major achievements is the emergence of two models describing the sorption property of SOM, namely dual mode model (DMM) (Xing and Pignatello, 1997) and distributed reactivity model (DRM) (Weber et al., 1992). Consistently, both models characterize SOM as

a multi-domain sorbent, generally linear and nonlinear, or rubbery and glassy sorption domains. The sorption of HOCs by mineral particles was commonly linear, and can be neglected if organic carbon content >0.1% (Schwarzenbach and Westall, 1981).

Researchers have presented a great deal of evidence to support these models from the aspects of generally observed non-ideal sorption isotherms and spectroscopic data (e.g., NMR). For example, time dependence of Freundlich parameters of sorption isotherms was recognized in several studies (Xing and Pignatello, 1996; Weber and Huang, 1996). The sorption capacity parameter, K_F , increased while nonlinearity factor, n , decreased with time, indicating that the sorption sites are not evenly distributed. The sites on surface region are less abundant and readily available to bulk solution compared to those in the interior region. Weber and Huang (1996) proposed a three-domain model (exposed inorganic surface, amorphous

* Corresponding author. Tel./fax: +86 10 62751938.

E-mail address: taos@urban.pku.edu.cn (S. Tao).

and condensed soil organic matter) based on the observation that K_F and n in water–soil systems varied functionally with logarithmic time.

Another line of evidence was obtained by conducting sorption experiments using separated SOM fractions, such as humic acid (HA) and humin (HM). A widely reported result is that isotherm nonlinearity and hysteresis increased in the order of $HA < soil < HM$ (Xing and Pignatello, 1997; Chiou et al., 2000; Gunasekara and Xing, 2003). Because the procedure of fraction separation involves dissolution of humic substances in alkaline solution and acid precipitation to form HA aggregates, one may argue that the separated fractions may not reflect the original sorption characters in the original soil. Therefore, to further explore sorption behavior of HOCs in soils, more direct information on how HOCs distribute among soil fractions is preferred.

A more direct observation may be conducted by alkaline extraction after the loading of organic chemicals and this procedure has been frequently used in bound residue studies (Northcott and Jones, 2000 and references therein) and sequestration investigation (Guthrie et al., 1999). The separation procedure was the same as that employed for sorption experiments using separated humic fractions. These studies addressed the mass balance of organic pollutants in fulvic acid (FA), HA and HM (Kohl and Rice, 1998; Nam and Kim, 2002), the relationship of bioactivity and distribution of organic pollutants in SOM fractions (Guthrie and Pfaender, 1998; Macleod and Semple, 2003), and the sequestration mechanism of HOCs in HA and HM (Guthrie et al., 1999). Chung and Alexander (1999) observed the concentration dependence of sequestration and bioavailability of organic pollutants in soil.

This work was designed to take a detailed look at the sorption behavior of HOCs in soil humic fractions employing the conventional humic separation method after multi-concentration sorption experiments. Two polycyclic aromatic hydrocarbons, phenanthrene (PHE) and pyrene (PYR), were chosen as model compounds. The concentration dependence of distribution of PHE and PYR in fulvic and humic acids (FA/HA) and HM fractions was investigated. The distribution characteristics were described by Freundlich parameters as in sorption experiment. Although the separation procedure may somewhat alter the existing distribution of solutes in soil humic fractions, this approach was expected to provide additional direct data on the contribution of different humic fractions to the sorption, and to present a new angle to assess multi-domain sorption concept. This line of research would be a step forward towards a better

understanding of the behavior of organic chemicals inside SOM, thus provide important information to understand HOCs sequestration in soils and sediments.

2. Experimental section

2.1. Sorbent preparation

Four soils with various organic carbon contents were collected from northern suburb of Beijing, China. The soils were gently mixed and ground using wood rods and sieved through a 2 mm sieve. The soils were kept moist and in dark at room temperature. For sorption experiments, the soils were wet weighed and the dry weights were calculated by subtracting moisture contents, which were calculated based on the weight differences before and after heating the soils overnight at 105 °C. The moist soil samples were used because it takes too long for sorption to reach equilibrium due to slow wetting (Todorku et al., 2003). The total organic carbon (TOC) of the four soils, measured on a TOC analyzer (Shimadzu 5000A), spanned an order of magnitude from 0.7 to 7.9% and more than 55% of the organic carbon was in HM fraction (Table 1). The background concentrations of the solutes were 20–40 and 10–100 times lower than the lowest concentration of PHE and PYR added in the soil for the sorption experiment, respectively. The desorption of PHE and PYR in 0.01 M CaCl₂ and 200 mg/L HgCl₂ solution was not detectable from the original soil samples, thus the background PHE and PYR in the soil samples was ignored for the sorption calculation.

The humic fractions of soil B were also separately extracted and purified for comparison. Briefly, a base solution of 0.1 M NaOH and 0.1 M Na₄P₂O₇ was mixed with the soil (50:1, v:w) to extract FA/HA. After 12 h of equilibration, the mixture was centrifuged at 1000g for 10 min and the supernatant was decanted. The extraction procedure was repeated three times until a light yellow extract was reached. The extractions were gathered and the HA was precipitated with HCl. The insoluble solid residue was referred to as HM. The HA and HM were washed using distilled water until a negative test of chloride using AgNO₃, freeze-dried and crushed into <500 μm particles.

2.2. Chemicals and solutes

PHE and PYR (purity > 98%) were from Acros Co. and were dissolved separately in methanol as stock solution. All the other chemicals and solvents used were better than

Table 1
Soil properties

	Moisture content (%)	pH	TOC ^a (%)	FA/HA ^a (%)	HM ^a (%)	PHE ^b (μg/g)	PYR ^b (μg/g)	Clay (%)	Silt (%)	Sand (%)
Soil A	9	7.84	0.70	0.31	0.39	0.034	0.003	2.5	30.1	67.3
Soil B	22	7.61	2.89	1.16	1.73	0.018	0.024	2.8	34.5	62.7
Soil C	28	6.4	4.53	1.94	2.59	0.056	0.030	2.1	48.0	49.9
Soil D	27	7.09	7.90	3.36	4.54	0.071	0.055	2.6	34.6	62.8

^a Expressed as carbon content in whole soil.

^b Exhaustedly extracted on accelerated solvent extractor and analyzed on gas chromatography–mass spectrometer.

analytical grade (Beijing Chemical Reagent Co.). Hexane was distilled in glassware for purification and the fluorescence spectrum of purified hexane showed stable and low baseline in the wavelength ranges of PHE and PYR.

2.3. Sorption experiment

Three separate sets of experiments were conducted in this study: (1) sorption dynamics of PHE or PYR on soil B (TOC, 2.89%) were determined at six concentrations, from 10 to 380 ng/mL for PHE and from 2 to 20 ng/mL for PYR. The sampling times were 1, 12, 24, 48, 72, 168, and 720 h; (2) sorption of PHE or PYR by extracted HA and HM from soil B was evaluated after fractionation of HA and HM as done in the literature (Gunasekara and Xing, 2003; Xing, 2001); (3) sorption experiments of other three soils (samples A, C, and D with TOC of 0.7–7.9%) were performed in the same concentration range as for the first set of the experiments at time 48 h, by which the apparent equilibrium was approximately reached.

The experimental procedure was adopted from Xing (2001). Briefly, the stock solution was diluted sequentially to a series of concentrations distributed evenly on a log scale in 0.01 M CaCl₂ and 200 mg/L HgCl₂ solution. HgCl₂ was used as biocide because it was reported as one of the best soil sterilization methods to minimize soil property change (Wolf et al., 1989). The methanol:solute ratio was the same in all the sorption systems, and the volume ratio of methanol was below 0.1% to avoid co-solvent effect (Weber and Huang, 1996). The sorption experiments were conducted in 10 mL centrifuge tubes equipped with ground glass stoppers. Soil sample of 0.02–0.1 g (wet weight) was mixed with 10 mL PHE or PYR solution in a tube that was sealed with a glass stopper immediately. The stopper was tightly bound with the tube externally by parafilm, which was not in contact with solution. The headspace was kept minimal to reduce solute vapor loss. The tube was kept in dark and rotated vertically on a rotator (30 rpm, Glas-Col lab rotator, RD9924CE) for a desired contact time, and centrifuged at 1000g for 10 min. The supernatant fluid was poured out carefully to prevent the loss of soil particles, and subjected to solute analysis. The water resided in the soil after centrifugation and decanting was calculated by the weight difference, and the solute in the residual water was taken into account during the fractionation experiment below. The experiments for PHE and PYR were conducted separately. All experiments including the procedure blanks were run in duplicate, and control samples of PHE or PYR were run in duplicate with samples for the time points longer than 2 days for calibration. The highest variation was below 10%.

2.4. Base extraction of FA/HA after sorption

All the soils in the first set of sorption experiment were subjected to SOM fractionation after solid–water separation. As in the humic substance separation procedure, the soils were extracted three times by the base solution (0.1 M NaOH and 0.1 M Na₂P₂O₇). All the extracts were mixed in a 25-mL colorimetric cylinders. This process of fractionation is a traditional

way to extract humic substances. However, the solutes in the base extracts may not be fully attributed to those only bound with FA/HA, since the HM-bound solutes may desorb in the process of extraction. Thus, the alkaline extracts may include the FA/HA bound solutes and any solute desorbed from HM during the extraction, but the residue solutes in HM would actually represent the highly resistant solute fraction bound in the soil. The FA/HA was not further fractionated because the acid precipitation may cause redistribution of solutes in FA and HA.

2.5. Measurement of the solutes

Since the brown extracts in base solution could not be analyzed by fluorescence photometry directly, all the aqueous samples were extracted by purified hexane in half volume of the samples. The water–hexane was mixed vigorously, and two drops of ethanol were added to diminish emulsification. The recoveries of the hexane extraction exceeded 98% for both PHE and PYR according to the results of a preliminary experiment.

Approximately 1 mL of the hexane was used for fluorescence analysis (Hitachi, F-2500) after the water–hexane interface was clear. The parameters of fluorescence photometry were set as follows: slit 2.5 nm, voltage 700 V, emission wavelength 300–450 nm, excitation wavelength 293 nm for PHE and 272 nm for PYR. The peak heights at 365 and 385 nm were used to quantify PHE and PYR, respectively.

2.6. Recovery of sorption experiment

The overall recovery test of sorption experiment demands to know the quantity of solutes both in aqueous and in solid (Fig. 1). A preliminary study showed that the extraction efficiency of PHE and PYR from base solution of HA by hexane was higher than 95% (Fig. 2). Because HA could be easily dissolved by alkaline solution, the sorbed amount of PHE or PYR could be quantified by hexane extraction of the dissolved HA after sorption experiment (Fig. 1). A recovery test of sorption experiment using isolated HA was conducted based on this observation. As presented in Fig. 2, the recoveries of the sorption experiment of PHE and PYR for isolated HA in this study were both higher than 93%. Furthermore, no degradation was observed as indicated by similar chromatographic patterns of GC–MS over the equilibration time. Therefore, during the fractionation of the soils after sorption experiment, the change of solutes was attributed to the irreversible binding with HM fraction. The following mass balance was employed for data manipulation:

The initial input of PHE or PYR

$$= \text{PHE or PYR in water} + \text{PHE or PYR in FA/HA} \\ + \text{PHE or PYR in HM}$$

2.7. Data analysis

The average of the duplicates was used for data analysis. In order to compare the distribution of solutes in SOM fractions

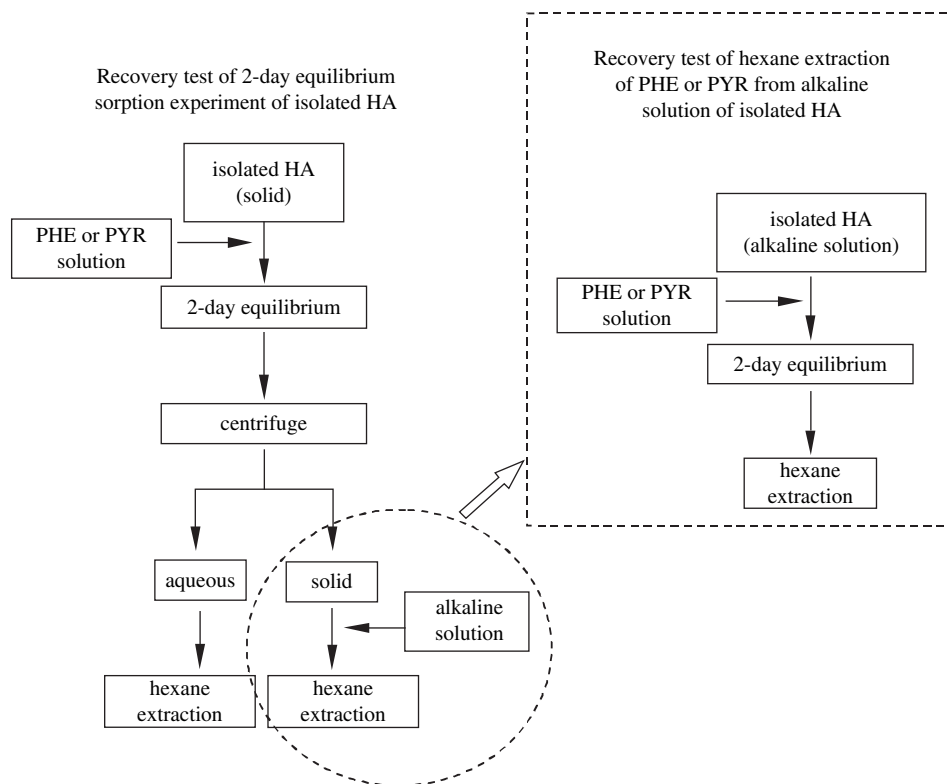


Fig. 1. Flow chart of recovery tests of sorption experiment.

at various initial concentrations, the relative content in a fraction was calculated as follows:

$$\text{Relative content in a specific fraction} = \frac{\text{(PHE or PYR in the fraction)}}{\text{(initial addition of PHE or PYR)}}$$

The relative content is nondimensional and the sum of relative content in water, FA/HA and HM would be 1 according to the mass balance assumption. As mentioned earlier, the

distribution of solutes in different fractions could be characterized using isotherm parameters. Freundlich equation was used to fit the distribution data (Carmo et al., 2000):

$$C_s = K_F(C_e/S_{scl})^n$$

where C_s is the solid concentration ($\mu\text{g/g}$), C_e is aqueous concentration ($\mu\text{g/mL}$) and S_{scl} is the supercooled liquid-state solubility ($\mu\text{g/mL}$). The K_F has the same unit as C_s , $\mu\text{g/g}$, and independent of nonlinearity factor, n , thus the K_F s are comparable between isotherms. The equation was applied in water–soil, water–FA/HA, and water–HM systems. Since the concentrations of FA/HA and HM were expressed as the organic carbon content, C_s was expressed as the solute concentration in organic carbon ($\mu\text{g}/(\text{g of OC})$) in all the systems examined, thus the equation could be written as:

$$C_{soc} = K_{Foc}(C_e/S_{scl})^n$$

where C_{soc} is solute concentration per unit of organic carbon ($\mu\text{g}/(\text{g of OC})$) and K_{Foc} is organic carbon normalized Freundlich coefficient with the same unit as C_{soc} . The solute contents in water and FA/HA were measured directly, while the content in HM was calculated based on the mass balance (see the recovery test above). The fitting was processed using SigmaPlot 2001 (SPSS Inc.) and statistical analysis was performed using SPSS 11.0 (SPSS Inc.).

Organic carbon normalized sorption coefficient ($K_{oc} = K_{Foc}C^{n-1}C_{scl}^{-n}$), was calculated at two different solute

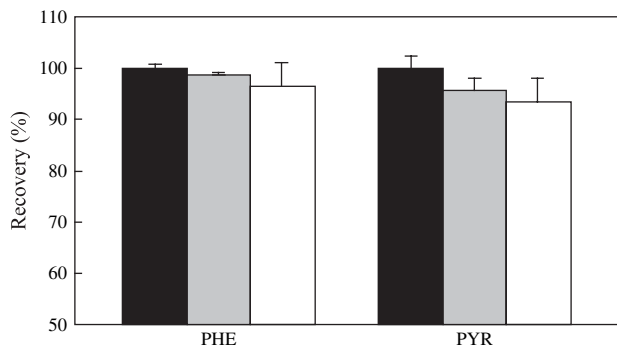


Fig. 2. Recovery test of PHE and PYR: black bars (■) are the recovery of extraction by hexane from water; gray bars (■) are the recovery of extraction by hexane from alkaline HA solution; and open bars (□) are the overall recovery of sorption experiment of isolated HA after 2-day equilibration (i.e., the second set of sorption experiments for HA particles).

Table 2
Physicochemical properties of solutes^a

Compound	$\log K_{ow}$	$\log K_{oc}$	T_m^b (°C)	ΔH^c (kcal/mol)	S^d ($\mu\text{g/ml}$)	S_{sc}^e ($\mu\text{g/ml}$)
Phenanthrene	4.57	4.36	101	4.450	1.290	5.973
Pyrene	5.18	4.92	150	5.541	0.134	2.166

^a K_{ow} s and K_{oc} s were from Karickhoff et al. (1979), and the other data were from Mackay et al. (1997).

^b Melting temperature.

^c Heating of fusion.

^d Solubility in water at 25 °C.

^e Supercooled liquid-state solubility.

concentrations in order to compare with that in the literatures (such as in Mackay et al., 1997) in which sorption affinity was generally expressed as K_{oc} . The relevant physicochemical properties compared or used for calculation in this study are listed in Table 2.

3. Results and discussion

3.1. Relative distribution dynamics of PHE and PYR in soil and humic fractions

The dynamics of solutes in water and the distribution in soil humic fractions were investigated using soil B and the results are depicted in Fig. 3. Relative contents of PHE and PYR were used for comparison. The data points were presented as single points showing the averaged values of replicates. The relative standard errors were below 10%. Due to an instrumental failure, data of PHE at 12 h and PYR at 72 h were missing, which hopefully had no significant effect on the overall results. As indicated in Fig. 3 (PHE-water, PYR-water), the changes of PHE and PYR in water phases can be distinguished into a fast and a slow sorption phase while an apparent equilibrium was reached around 48 h. The relative contents of PHE or PYR in FA/HA and HM generally increased with time during the first 48 h, with a clear fluctuation for FA/HA within 48 h

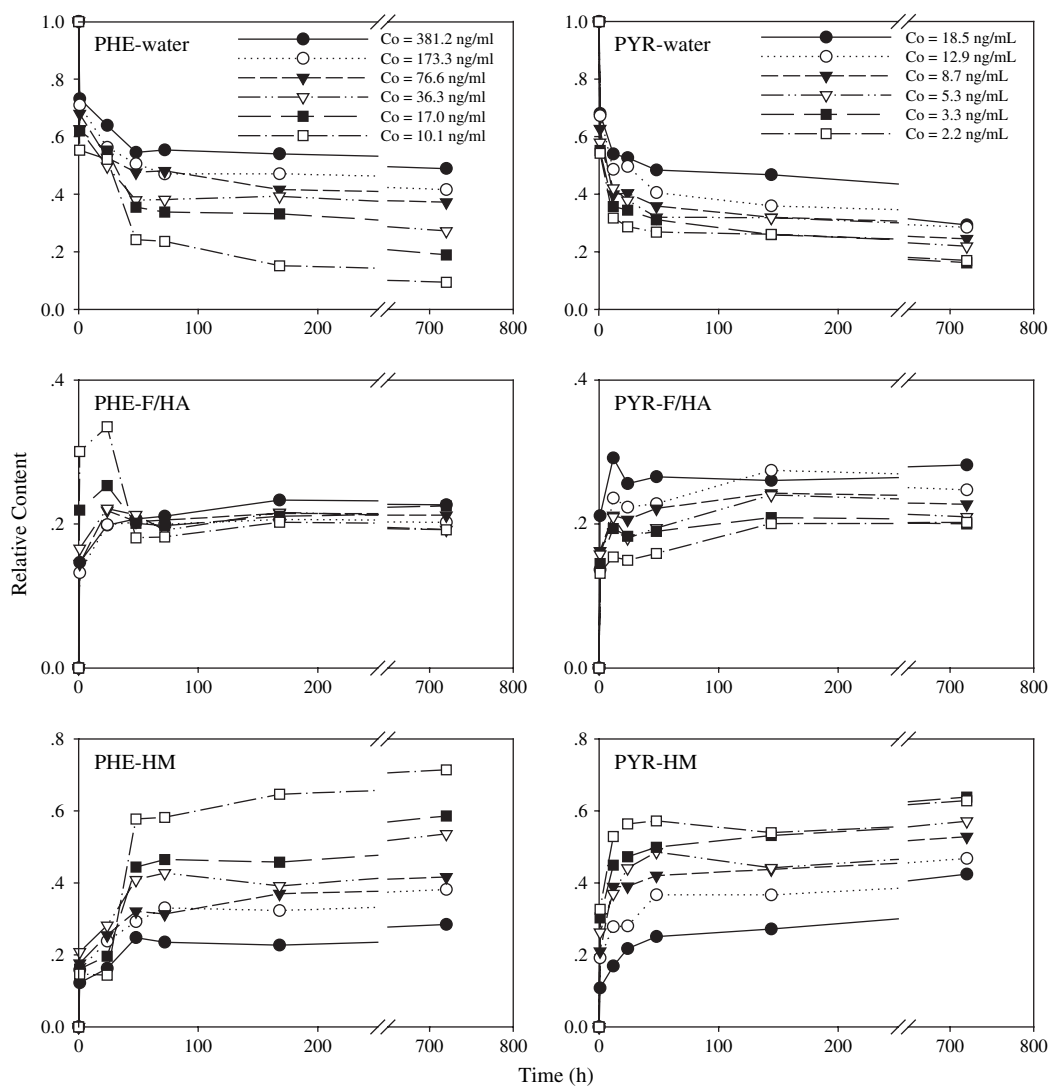


Fig. 3. Relative contents of PHE and PYR in water (top), PHE and PYR in FA/HA (middle), and PHE and PYR in HM (bottom) fractions. The time range from 250 to 650 h was omitted in order to better display the early time points. C_0 stands for the initial concentrations (ng/mL).

(Fig. 3, PHE-FA/HA, PYR-FA/HA). This phenomenon reflected difference in sorption kinetics of various humic fractions and redistribution of solutes among the fractions as sorption proceeded. The unevenly distributed sorption sites may contribute to the redistribution of solutes. As concluded by White et al. (1999), HM fraction exhibited strong affinity for organic pollutants and the adsorbed solutes were strongly resistant to desorption. Because HM tends to be buried under FA/HA (Gunasekara and Xing, 2003), it is reasonable to speculate that HM competes for solute molecules with FA/HA when the solute passed through FA/HA layer. This competition was confirmed by the decrease of the solute contents in FA/HA as shown in Fig. 3 (PHE-FA/HA, PYR-FA/HA). As stated earlier, the solutes in FA/HA fraction may include those desorbed from HM-bound fraction. Although desorbed amount during the humic fractionation could not be accurately quantified, it has been reported that the sorption of HOCs by HM is very strong and highly irreversible (White et al., 1999). Thus it is useful to investigate the distribution of solutes in humic fractions using the experimental design of this study.

The sorption reached its apparent, not final though, equilibrium at 48 h equilibrium time. After that, the concentrations of both PHE and PRY in water decreased very slowly. The change of relative content of solutes was not obvious in FA/HA after 48 h, but was in HM, indicating that slow sorption occurred primarily in HM fraction.

3.2. Concentration dependence of sorbed PHE and PYR in humic fractions

As presented in Fig. 3 (PHE-water, PYR-water), the relative contents of PHE and PYR in water were lower for low initial concentration experiments than those for high initial concentration ones, suggesting nonlinear sorption, which resulted in higher K_{oc} , consequently lower relative content in water at lower concentrations. Such a concentration-dependent distribution of both solutes in soil humic fractions can also be clearly illustrated by Fig. 4, in which distribution of PHE and PYR between FA/HA and HM at apparent equilibrium of 48 h as a function of initial concentrations is shown. Although mineral fraction is included in HM fraction by operational

definition, sorption by this fraction is small enough to be neglected (Schwarzenbach and Westall, 1981).

Apparently, lower percentage of PHE or PYR was bound in HM at higher initial concentrations. Similar results were reported in the literature. For instance, Chung and Alexander (1999) found that lower initial concentration resulted in decrease of solvent extractability and biodegradation. They discussed nonlinear sorption based on their data and concluded that a declining percentage of organic chemicals was sequestered as the concentration increased. Macleod and Semple studied the effect of aging on extractability, distribution among soil organic fractions and biodegradation of PYR at either high (100 mg PYR/kg) or low (32.5 μ g PYR/kg) concentrations using the same experimental design and soil samples (Macleod and Semple, 2000, 2003). By comparing the data collected in the two studies, significantly higher percentage of nonextractable residues in both whole soil ($p < 0.01$) and HM ($p < 0.01$) was reported for the soil that received lower concentration of PYR. Such a concentration-dependent behavior of HOCs in soils suggested stronger sequestration at lower concentrations.

3.3. Sorption of PHE and PYR characterized by phase distribution relationships (PDRs)

The concentration dependence of the behavior of various chemicals in soil was reported previously in the literature. However, since the results were often presented in two or three concentration levels (Chung and Alexander, 1999; Macleod and Semple, 2000, 2003), or the systems were complicated including biodegradation (Mu and Scow, 1994; Gan et al., 1996) and other competitive chemicals (Mu and Scow, 1994), mathematical fitting was not applicable in these investigations. In current study, however, the concentration dependence of PHE and PYR sorption in soil humic fractions was examined at six concentration levels that made isotherm fitting possible. Therefore, Freundlich equation was used to describe PDRs (Weber and Huang, 1996) of PHE and PYR in soil humic fractions at different equilibration time. Sorption data and model fitting were shown in Fig. 5, and the parameters calculated are presented in Table 3. As could be seen in Table 3, most fitting coefficients (r^2) of all equations were higher than 0.99. In

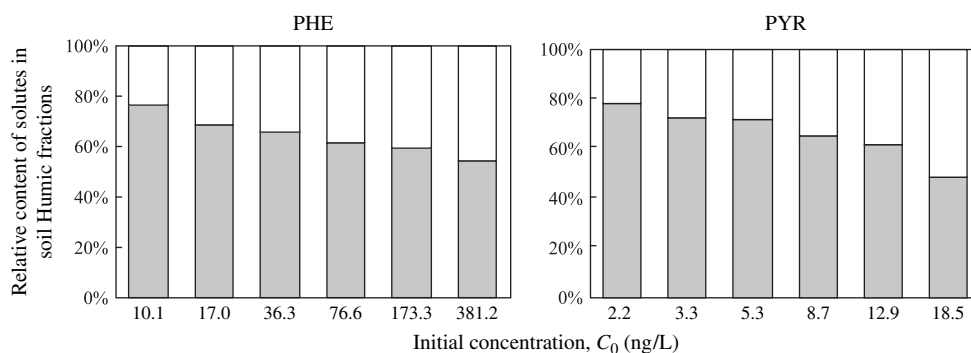


Fig. 4. Distribution of PHE and PYR in soil B after 48 h contact time. Open areas (\square) are the relative contents of PHE or PYR in FA/HA and gray areas (\blacksquare) are the relative contents of PHE or PYR in HM.

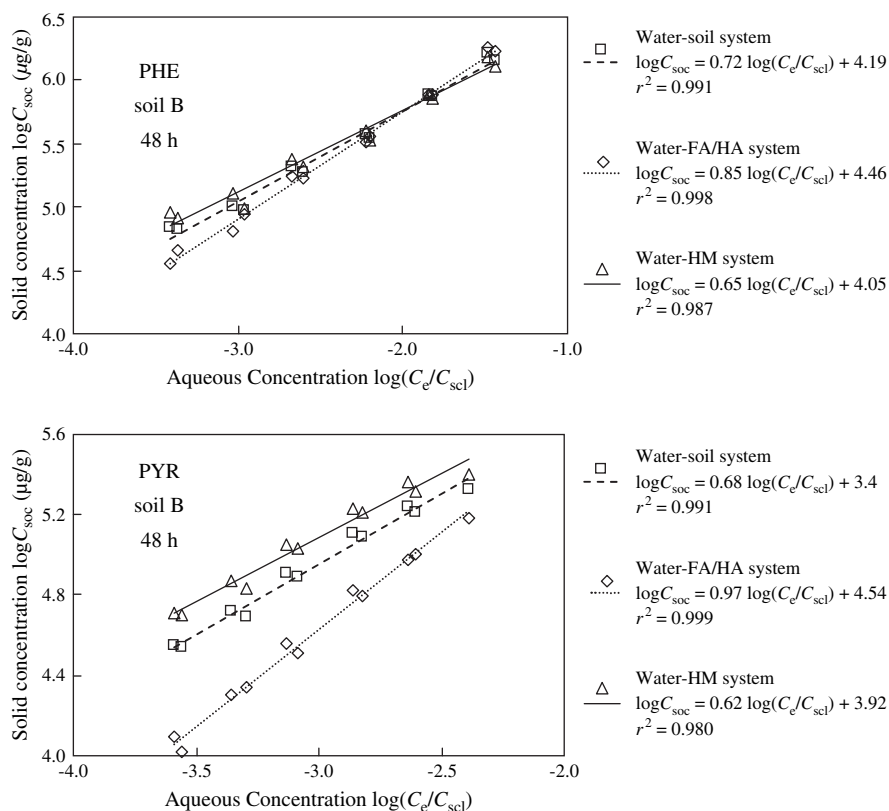


Fig. 5. Freundlich equation fitting of phase distribution relationships of PHE or PYR in soil fractions. The distributions of PHE and PYR in soil B at 48 h were plotted as examples.

general, the r^2 values for water–HM system were lower than others, which could be caused by the additional error induced by subtraction calculation. The three systems of water–soil, water–FA/HA, and water–HM were discussed separately as follows.

3.3.1. Water–soil system

The parameter of n in Freundlich equation represents the degree of curvature of sorption isotherm: the greater the deviation of n from 1, the greater the nonlinearity. In this case, nonlinear sorption was observed for all water–soil systems indicated by n values ranging from 0.59 to 0.89 for PHE and 0.64 to 0.73 for PYR. Moreover, the n values decreased as the contact time increased. Other researchers have observed the similar trend and believed that the sorption sites in soil were not evenly distributed and different sorption mechanisms or domains predominated at different contact times (Xing and Pignatello, 1996; Weber and Huang, 1996).

3.3.2. Water–FA/HA system

The n values obtained again showed nonlinearity and a generally decreasing trend of both solutes in FA/HA (Table 3), suggesting the unevenly distributed sorption sites in this fraction. Xing and Chen (1999) provided nuclear magnetic resonance (NMR) evidence demonstrating that HAs were more condensed from mineral horizons than those from surface organic horizons. More solutes would be adsorbed by

condensed domains of FA/HA as sorption continued. As a result, the percentage of hole filling (or adsorption) fraction increased, causing lower n values consequently.

After apparent equilibrium reached at 48 h, the n values were significantly ($p < 0.01$) lower than 1 for both solutes in water–FA/HA system. In preparing HA, it was first dissolved in alkaline solution and then precipitated in acid. Thus, the sorption sites of separated HA may not represent those in soil aggregates. Many studies have clarified that the sorption properties of HA in aqueous and HA–mineral complexes were significantly different and the complexes displayed nonlinear isotherms and competitive adsorption behaviors (Murphy and Zachara, 1995) and had relatively higher sorption affinity with HOCs (Terashima et al., 2003). In this study, although the alkaline fractionation may somewhat alter the distribution of solutes in FA/HA and HM, the results could provide more direct sorption characteristics of PHE and PYR by soil humic fractions.

3.3.3. Water–HM system

The distribution of PHE and PYR in HM is also presented in Table 3. The n values after 48 h were 0.53–0.65 for PHE and 0.62–0.71 for PYR, revealing strong nonlinear sorption in this fraction. Although organic matter in HM is bound tightly with mineral particles and is normally embedded under FA/HA, the sorption capacity is very high even at the start of sorption. Chang et al. (1997) examined the diffusion of organic

Table 3
Freundlich parameters as functions of time (soil B)

Equilibration time (h)	Water-soil system				Water-FA/HA system				Water-HM system								
	N^a	n^b	$\log K_{Foc}$ ^b ($\mu\text{g}/(\text{g of OC})$)	r^2	$\log K_{oc}$ (mL/g) $C_e = 0.1S$	$C_e = 0.01S$	n	$\log K_{Foc}$ ($\mu\text{g}/(\text{g of OC})$)	r^2	$\log K_{oc}$ (mL/g) $C_e = 0.1S$	$C_e = 0.01S$	n	$\log K_{Foc}$ ($\mu\text{g}/(\text{g of OC})$)	r^2	$\log K_{oc}$ (mL/g) $C_e = 0.1S$	$C_e = 0.01S$	
PHE	1	12	0.81 ± 0.02	4.02 ± 0.04	0.998	3.56	3.76	0.75 ± 0.06	2.06 ± 0.13	0.977	1.71	1.96	0.89 ± 0.05	4.09 ± 0.12	0.986	3.50	3.61
	24	12	0.89 ± 0.05	4.40 ± 0.11	0.990	3.80	3.90	0.84 ± 0.03	4.38 ± 0.06	0.996	3.87	4.04	0.98 ± 0.11	4.48 ± 0.26	0.953	3.73	3.75
	48	12	0.72 ± 0.03	4.19 ± 0.09	0.991	3.89	4.17	0.85 ± 0.02	4.46 ± 0.05	0.998	3.93	4.08	0.65 ± 0.04	4.05 ± 0.10	0.987	3.86	4.22
	72	12	0.71 ± 0.04	4.19 ± 0.09	0.990	3.90	4.19	0.85 ± 0.02	4.46 ± 0.06	0.997	3.93	4.08	0.64 ± 0.04	4.04 ± 0.11	0.983	3.87	4.23
	168	12	0.65 ± 0.06	4.10 ± 0.15	0.968	3.90	4.25	0.79 ± 0.05	4.37 ± 0.13	0.984	3.95	4.16	0.58 ± 0.06	3.93 ± 0.16	0.957	3.86	4.28
48 (purified HA and HM)	720	12	0.59 ± 0.04	4.07 ± 0.10	0.984	3.93	4.38	0.72 ± 0.04	4.27 ± 0.11	0.987	3.97	4.26	0.53 ± 0.04	3.97 ± 0.10	0.983	3.97	4.44
								0.81 ± 0.02	4.42 ± 0.05	0.993	3.96	4.15	0.68 ± 0.02	4.20 ± 0.06	0.986	3.95	4.27
PYR	1	12	0.73 ± 0.03	3.74 ± 0.07	0.995	4.01	4.28	1.02 ± 0.08	4.32 ± 0.21	0.978	3.94	3.92	0.62 ± 0.04	3.55 ± 0.10	0.987	4.05	4.43
	12	12	0.67 ± 0.04	3.88 ± 0.12	0.985	4.26	4.59	1.01 ± 0.03	4.61 ± 0.08	0.998	4.24	4.23	0.56 ± 0.06	3.67 ± 0.18	0.956	4.31	4.74
	24	11	0.64 ± 0.03	3.79 ± 0.08	0.992	4.26	4.62	0.96 ± 0.02	4.43 ± 0.07	0.998	4.19	4.23	0.55 ± 0.04	3.67 ± 0.11	0.983	4.32	4.76
	48	11	0.68 ± 0.04	3.40 ± 0.11	0.989	4.36	4.68	0.97 ± 0.02	4.54 ± 0.05	0.999	4.27	4.30	0.62 ± 0.04	3.92 ± 0.13	0.980	4.44	4.82
	168	11	0.69 ± 0.05	4.05 ± 0.16	0.977	4.41	4.72	0.93 ± 0.03	4.52 ± 0.10	0.996	4.34	4.41	0.72 ± 0.03	4.28 ± 0.09	0.994	4.57	4.86
720 and HM)	12	12	0.69 ± 0.02	4.23 ± 0.07	0.995	4.59	4.90	0.86 ± 0.03	4.42 ± 0.09	0.995	4.40	4.54	0.65 ± 0.02	4.24 ± 0.07	0.995	4.68	5.04
								0.89 ± 0.03	4.54 ± 0.13	0.989	4.46	4.57	0.59 ± 0.02	3.84 ± 0.08	0.986	4.41	4.81

^a Number of data points for fitting.

^b Average ± standard error.

chemicals in pressed humic acid disks and concluded that the sorption of organic chemicals on humic acid was a physical process, thus the penetration through it would not be a limiting slow process. Therefore, the solutes may adsorb directly on exposed HM, or penetrated fast through FA/HA and sorbed by HM.

3.4. Sorption of PHE and PYR by individually separated humic fractions

A separate sorption experiment using isolated humic fractions was conducted to compare with the fractionation results after the sorption experiments. As listed in Table 3, at the 48 h equilibration time, the sorption nonlinearity was in the following order: isolated HA < soil < isolated HM, consistent with the fractionation experiment. In previous studies on extracted SOM fractions, the commonly reported result was that the nonlinearity for PAHs sorption by HM was distinctly stronger than HA (Xing and Pignatello, 1997; Chiou et al., 2000; Gunasekara and Xing, 2003; Kang and Xing, 2005).

The K_{oc} values were also calculated and listed in Table 3. Although the K_{oc} of different systems varied, the values were generally compatible with those in literature (Table 2). The K_{oc} for both extracted HA and HM was slightly higher than that in the fractionation experiment at the corresponding equilibration time, but comparable. As discussion on sorption dynamics above, 48 h were not be the final equilibrium time. Compared to the well-organized soil aggregates in the original soil samples, the sorption sites of extracted HA and HM might be more accessible to solutes and thus shortened the kinetics process. From the K_{oc} comparison (Table 3) between the fractionation experiments and sorption after humic fractionation, it is fairly safe to say that desorption of HM-bound solutes during alkaline fractionation would be relatively low.

In all the systems examined, the K_{oc} values at $C_e = 0.01S$ were significantly higher than those at $C_e = 0.1S$ ($p < 0.01$) after 48 h, consistent with nonlinear sorption. Furthermore, the K_{oc} values for PHE were significantly lower than PYR ($p < 0.01$).

3.5. Importance of HM

SOM had been proved to be an important component affecting sequestration of HOCs in soil. A number of studies have reported that a substantial percentage (up to 93%) of nonextractable organic pollutants resided in HM (Guthrie and Pfaender, 1998; Guthrie et al., 1999; Nam and Kim, 2002; Macleod and Semple, 2003). This is also true in this study. As shown in Fig. 4, the solutes resided in HM accounted for 55–76% of PHE and 49–78% of PYR in soil depending on initial concentrations, and the percentages were rising as slow sorption proceeded (Fig. 3, PHE-HM, PYR-HM). According to the results of a paired *t*-test ($p < 0.05$) based on the data after 48 h (Table 3), the *n* values of water-HM system for both solutes were significantly lower than other systems examined, indicating a wider distribution of sorption energy than other systems (Yuan and Xing, 1999). Similar differences were

Table 4
Freundlich parameters of other soils

Soil	N ^a	Water–soil system			Water–FA/HA system			Water–HM system									
		n ^b	log K _{Foc} ^b (µg/(g of OC))	r ²	log K _{oc} (mL/g) C _e = 0.1S C _e = 0.01S	n	log K _{Foc} (µg/(g of OC))	r ²	log K _{oc} (mL/g) C _e = 0.1S C _e = 0.01S	n	log K _{Foc} (µg/(g of OC))	r ²	log K _{oc} (mL/g) C _e = 0.1S C _e = 0.01S				
PHE	A	12	0.73 ± 0.06	4.39 ± 0.15	0.961	4.07	4.33	0.88 ± 0.09	4.59 ± 0.22	0.945	4.02	4.14	0.65 ± 0.07	4.29 ± 0.18	0.933	4.09	4.44
	B	12	0.72 ± 0.03	4.19 ± 0.09	0.991	3.89	4.17	0.85 ± 0.02	4.46 ± 0.05	0.998	3.93	4.08	0.65 ± 0.04	4.05 ± 0.10	0.987	3.86	4.22
	C	16	0.80 ± 0.03	4.45 ± 0.08	0.993	4.01	4.22	0.92 ± 0.02	4.66 ± 0.06	0.997	4.02	4.10	0.74 ± 0.03	4.37 ± 0.10	0.988	4.02	4.28
	D	12	0.78 ± 0.04	4.37 ± 0.11	0.989	3.95	4.16	0.86 ± 0.02	4.38 ± 0.06	0.997	3.84	3.98	0.75 ± 0.06	4.38 ± 0.15	0.979	4.01	4.26
PYR	A	13	0.76 ± 0.08	4.23 ± 0.25	0.948	4.42	4.66	1.25 ± 0.08	5.47 ± 0.26	0.979	4.58	4.34	0.61 ± 0.07	3.86 ± 0.23	0.933	4.40	4.79
	B	16	0.68 ± 0.04	4.00 ± 0.11	0.989	4.36	4.68	0.97 ± 0.02	4.54 ± 0.05	0.999	4.27	4.30	0.62 ± 0.04	3.92 ± 0.13	0.980	4.44	4.82
	C	16	0.72 ± 0.02	4.13 ± 0.05	0.997	4.42	4.70	0.83 ± 0.02	4.54 ± 0.08	0.995	4.59	4.77	0.63 ± 0.02	3.80 ± 0.07	0.994	4.27	4.64
	D	12	0.81 ± 0.05	4.31 ± 0.17	0.982	4.40	4.60	0.96 ± 0.10	4.72 ± 0.35	0.950	4.48	4.53	0.70 ± 0.05	4.01 ± 0.18	0.974	4.34	4.64

^a Number of data points for fitting.

^b Average ± standard error.

observed when the experiments were repeated using other three soil samples at 48 h (Table 4, soil B was also included in this table for the convenience of comparison). This trend was also confirmed in a separate sorption experiment using extracted HA and HM instead of whole soil samples (Table 3, this study) and previous studies (Xing and Pignatello, 1997; Chiou et al., 2000; Gunasekara and Xing, 2003; Kang and Xing, 2005).

Yuan and Xing reported that the sorbents with lower *n* values showed less desorption of PAHs, which indicated stronger retention of PAHs (Yuan and Xing, 2001). Their observation may help to explain sequestration mechanism. HM exhibited the most nonlinear sorption with PAHs either in the separate sorption experiments (Xing and Pignatello, 1997; Chiou et al., 2000; Gunasekara and Xing, 2003 and this work) or in distribution observation (this study), thus it may be the main region (or domains) for slowly irreversible binding or sequestration. Researchers have reported some special physical properties of HM, such as higher surface area and smaller average pore size (Malekani et al., 1997), the larger aggregates formed by crystalline and noncrystalline domains (Hu et al., 2000), and the enriched subnanometer-size pores (de Jonge and Mittelmeijer-Hazeleger, 1996; Gunasekara and Xing, 2003). However, relatively limited research has been conducted on HM compared to other humic fractions (Rice, 2001). Although several researchers have focused on the special sorption property of organo-mineral complex (Murphy and Zachara, 1995; Terashima et al., 2003; Wang and Xing, 2005), more research is needed in this area toward better understanding of the HOCs sequestration mechanisms in soils and sediments.

4. Conclusions

Although recent studies have recognized soil as a multi-domain sorbent for hydrophobic organic chemicals (HOCs), direct observation of HOCs sorption characteristics in various soil humic fractions is lacking. In this study, contributions of fulvic–humic acids (FA/HA) and humin (HM) to sorption of phenanthrene (PHE) and pyrene (PYR) in a soil were differentiated by using a humic separation procedure after multi-concentration sorption experiments, which has been frequently used in fractionation of the bound residues of HOCs. The experiments were conducted at various concentrations and durations. It was found that the amount of solutes in FA/HA did not change significantly after 48 h, while that in HM increased continuously and slowly up to the end of the experimental period (720 h), indicating that HM was the main region for slow sorption. The amounts of both PHE and PYR associated with HM accounted for more than 50% of those in the soil after 48 h, and higher initial concentrations resulted in lower percentage of solutes in HM. In addition, the distributions of PHE and PYR in soil humic fractions exhibited significant nonlinearity. Based on the fitting results using Freundlich equation, it was found that nonlinearity of both solutes was greater in HM than in FA/HA, consistent with the sorption characteristics of individually extracted HA and HM in a separate experiment. The observed nonlinearity of the solute distribution was additionally

confirmed by using three other soil samples with organic carbon contents ranging from 0.7 to 7.9%.

Acknowledgements

Financial support was provided by the National Natural Science Foundation of China (Grant 40428005, 40332015, and 40021101), National Basic Research Program (Grant 2003CB415004), and a USDA Competitive Grant (2002-35107-12544).

References

- Carmo, A.M., Hundal, L.S., Thompson, M.L., 2000. Sorption of hydrophobic organic compounds by soil materials: application of unit equivalent Freundlich coefficients. *Environmental Science and Technology* 34, 4363–4369.
- Chang, M.L., Wu, S.C., Chen, C.Y., 1997. Diffusion of volatile organic compounds in pressed humic acid disks. *Environmental Science and Technology* 31, 2307–2312.
- Chiou, C.T., Kile, D.E., Rutherford, D.W., Sheng, G.Y., Boyd, S.A., 2000. Sorption of selected organic compounds from water to a peat soil and its humic-acid and humin fractions: potential sources of the sorption nonlinearity. *Environmental Science and Technology* 34, 1254–1258.
- Chung, N., Alexander, M., 1999. Effect of concentration on sequestration and bioavailability of two polycyclic aromatic hydrocarbons. *Environmental Science and Technology* 33, 3605–3608.
- Gan, J., Becker, L., Koskinen, W.C., Buhler, D.D., 1996. Degradation of atrazine in two soils as a function of concentration. *Journal of Environmental Quality* 25, 1064–1072.
- Gunasekara, A.S., Xing, B.S., 2003. Sorption and desorption of naphthalene by soil organic matter: importance of aromatic and aliphatic components. *Journal of Environmental Quality* 32, 240–246.
- Guthrie, E.A., Bortiatynski, J.M., Van Heemst, J.D.H., Richman, J.E., Hardy, K.S., Kovach, E.M., Hatcher, P.G., 1999. Determination of [¹³C]pyrene sequestration in sediment microcosms using flash pyrolysis GC–MS and C-13 NMR. *Environmental Science and Technology* 33, 119–125.
- Guthrie, E.A., Pfaender, F.K., 1998. Reduced pyrene bioavailability in microbially active soils. *Environmental Science and Technology* 32, 501–508.
- Hu, W.G., Mao, J.D., Xing, B.S., Schmidt-Rohr, K., 2000. Poly(methylene) crystallines in humic substances detected by nuclear magnetic resonance. *Environmental Science and Technology* 34, 530–534.
- de Jonge, H., Mittelmeijer-Hazeleger, M.C., 1996. Adsorption of CO₂ and N₂ on soil organic matter: nature of porosity, surface area, and diffusion mechanisms. *Environmental Science and Technology* 30, 408–413.
- Karickhoff, S.W., Brown, D.S., Scott, T.A., 1979. Sorption of hydrophobic pollutants on natural water sediments. *Water Research* 13, 241–248.
- Kang, S.H., Xing, B.S., 2005. Phenanthrene sorption to sequentially extracted soil humic acids and humins. *Environmental Science and Technology* 39, 134–140.
- Kohl, S.D., Rice, J.A., 1998. The binding of contaminants to humin: a mass balance. *Chemosphere* 36, 251–261.
- Mackay, D., Shiu, W., Ma, K., 1997. Illustrated Handbook of Physical–Chemical Properties and Environmental Fate for Organic Chemicals. In: Pesticide Chemicals [M], vol. V. Lewis Publishers, Boca Raton, FL.
- Macleod, C.J.A., Semple, K.T., 2000. Influence of contact time on extractability and degradation of pyrene in soils. *Environmental Science and Technology* 34, 4952–4957.
- Macleod, C.J.A., Semple, K.T., 2003. Sequential extraction of low concentrations of pyrene and formation of non-extractable residues in sterile and non-sterile soils. *Soil Biology and Biochemistry* 35, 1443–1450.
- Malekani, K., Rice, J.A., Lin, J.S., 1997. The effect of sequential removal of organic matter on the surface morphology of humin. *Soil Science* 162, 333–342.
- Mu, D.Y., Scow, K.M., 1994. Effect of trichloroethylene (TCE) and toluene concentrations on TCE and toluene biodegradation and the population density of TCE and toluene degraders in soil. *Applied and Environmental Microbiology* 60, 2661–2665.
- Murphy, E.M., Zachara, J.M., 1995. The role of sorbed humic substances on the distribution of organic and inorganic contaminants in groundwater. *Geoderma* 67, 103–124.
- Nam, K., Kim, J.Y., 2002. Role of loosely bound humic substances and humin in the bioavailability of phenanthrene aged in soil. *Environmental Pollution* 118, 427–433.
- Northcott, G.L., Jones, K.C., 2000. Experimental approaches and analytical techniques for determining organic compound bound residues in soil and sediment. *Environmental Pollution* 108, 19–43.
- Pignatello, J.J., 1998. Soil organic matter as a nanoporous sorbent of organic pollutants. *Advances in Colloid and Interface Science* 76–77, 445–467.
- Rice, J.A., 2001. Humin. *Soil Science* 166, 848–857.
- Schwarzenbach, R.P., Westall, J., 1981. Transport of nonpolar organic compounds from surface water to groundwater: laboratory sorption studies. *Environmental Science and Technology* 15, 1360–1367.
- Terashima, M., Tanaka, S., Fukushima, M., 2003. Distribution behavior of pyrene to adsorbed humic acids on kaolin. *Journal of Environmental Quality* 32, 591–598.
- Todoruk, T.R., Langford, C.H., Kantzas, A., 2003. Pore-scale redistribution of water during wetting of air-dried soils as studied by low-field NMR relaxometry. *Environmental Science and Technology* 37 (12), 2707–2713.
- Wang, K.J., Xing, B.S., 2005. Structural and sorption characteristics of adsorbed humic acid on clay minerals. *Journal of Environmental Quality* 34, 342–349.
- Weber Jr., W.J., Huang, W.L., 1996. A distributed reactivity model for sorption by soils and sediments. 4. Intraparticle heterogeneity and phase-distribution relationships under nonequilibrium conditions. *Environmental Science and Technology* 30, 881–888.
- Weber Jr., W.J., McGinley, P.M., Katz, L.E., 1992. A distributed reactivity model for sorption by soils and sediments. 1. Conceptual basis and equilibrium assessments. *Environmental Science and Technology* 26, 1955–1962.
- White, J.C., Hunter, M., Nam, K.P., Pignatello, J.J., Alexander, M., 1999. Correlation between biological and physical availabilities of phenanthrene in soils and soil humin in aging experiments. *Environmental Toxicology and Chemistry* 18, 1720–1727.
- Wolf, D.C., Dao, T.H., Scott, H.D., Lavy, T.L., 1989. Influence of sterilization methods on selected soil microbiological, physical, and chemical properties. *Journal of Environmental Quality* 18, 39–44.
- Xing, B., 2001. Sorption of naphthalene and phenanthrene by soil humic acids. *Environmental Pollution* 111, 303–309.
- Xing, B.S., Chen, Z.Q., 1999. Spectroscopic evidence for condensed domains in soil organic matter. *Soil Science* 164, 40–47.
- Xing, B.S., Pignatello, J.J., 1996. Time-dependent isotherm shape of organic compounds in soil organic matter: implications for sorption mechanism. *Environmental Toxicology and Chemistry* 15, 1282–1288.
- Xing, B.S., Pignatello, J.J., 1997. Dual-mode sorption of low-polarity compounds in glassy poly (vinyl chloride) and soil organic matter. *Environmental Science and Technology* 31, 792–799.
- Yuan, G.S., Xing, B.S., 1999. Site-energy distribution analysis of organic chemical sorption by soil organic matter. *Soil Science* 164, 503–509.
- Yuan, G.S., Xing, B.S., 2001. Effects of metal cations on sorption and desorption of organic compounds in humic acids. *Soil Science* 166, 107–115.