Chapter 5

Freely dissolved concentrations of PAHs in soil pore water: measurements via solid phase extraction and consequences for soil tests

Thomas L. ter Laak, Stanley O. Agbo, Arjan Barendregt and Joop L. M. Hermens
IRAS - Institute for Risk Assessment Sciences, Utrecht University

Submitted

Abstract

Freely dissolved pore water concentrations are difficult to assess in complex matrices like soils or sediments. In this study a negligible-depletion partitioning based sampling technique was applied to measure freely dissolved pore water concentrations. A PDMS (poly(dimethylsiloxane)) coated glass fiber was exposed to a slurry of a soil spiked with several PAHs at concentrations ranging from 2 to 2000 mg/kg. Concentrations in the PDMS coating increased linear with the total soil concentration until a certain maximum was reached. Freely dissolved pore water concentrations were calculated using PDMS-water partition coefficients, and the observed maximum pore water concentrations were very similar to aqueous solubility of the tested PAHs. Estimated detection limits of pore water concentrations of the PAHs are very low and range from 0.2 to 10 ng/L. The sampling technique can measure pore water concentrations over a broad range of soil concentrations. Freely dissolved pore water concentrations are an important dose parameter for the exposure of organisms in soil. Therefore, increasing soil concentrations above a certain level, where aqueous solubility is reached in the pore water, might be irrelevant, and must at least be noticed.
Sorption coefficients that were calculated from the freely dissolved concentrations were slightly higher that estimates based on the octanol water partition coefficient. These differences are discussed in relation to the effects of dissolved organic matter in soil pore water on the determination of sorption coefficients.

Introduction

The freely dissolved concentration in soil pore water is an environmentally relevant parameter for various processes in soil, including for example evaporation to air, biological and chemical degradation and accumulation in soil-biota (*3, 47, 161*). These freely dissolved concentrations, however, are difficult to determine, especially when the compounds are very hydrophobic and sorb strongly to the soil and the dissolved organic matter in the pore water. Information about freely dissolved concentrations is also relevant for the accurate measurement of sorption coefficients, as the presence of dissolved organic matter (DOM) may affect the measured values (*14, 108*). Various techniques have been developed to determine (freely dissolved) pore water concentration in soils and sediments. The simplest method is to separate the solid and liquid phase by centrifugation, and subsequently extract the liquid phase with a solvent and measure the concentration. Centrifugation, however, does not remove dissolved organic matter (DOM) from the pore water. Since DOM has a high affinity for hydrophobic chemicals, free pore water concentrations can also be overestimated by this method (*66*). The freely dissolved pore water concentration can be estimated from the pore water extract by correcting for the DOM-associated compounds (*15, 108, 162-164*). In that case,

information is needed for the DOM concentration in the pore water and the sorption coefficient to the specific DOM material. Another option is to remove the DOM by flocculation (*16*). Flocculation might, however, disturb the equilibrium of the compounds between the DOM and the aqueous phase, thereby biasing the measurement of the free concentration. The DOM and water can also be separated passively by a dialysis membrane (*17*), since freely dissolved compounds will diffuse through this membrane while it is impermeable for the soil matrix and DOM. However, large amounts of water inside the dialysis membrane are necessary to detect and quantify very hydrophobic chemicals, and the technique is rather laborious. Additionally, various passive sampling techniques based on gas purging (*23, 24*) semi-permeable membrane devices (SPMD (*20, 22, 26*)), poly(oximethylene) sheets (POM (*28*)), polymer coated glass surfaces (*29*) or solid phase microextraction (SPME) fibers (*27, 30*) have been applied to assess pore water concentrations in complex matrices like soil or sediment. SPME has been developed by Pawliszyn and co-workers, as a very useful sampling technique (*21, 25*). These samplers only sense the freely dissolved concentration, and if they are equilibrated with the matrix without depleting the system, their equilibrium concentration can directly be used to calculate the freely dissolved concentration (*27, 33, 34*). The dimensions, properties, and agitation of the exposure vessel and the size of the passive sampler can be adjusted to sample detectable amounts of a test compound in a practical time span.

The objective of this study was to measure soil pore water concentrations of a series of PAHs at increasing concentrations in soil. We were interested in the trend of pore water concentration at high concentrations in soil as these are used for example in soil toxicity testing. Sorption coefficients were calculated as well and the linearity of the sorption process was analyzed. Moreover, potential effects of the presence of dissolved organic matter (DOM) in soil pore water on the measurements of sorption coefficients are discussed.

Negligible depletion SPME was applied to measure these pore water concentrations and another objective was to determine the detection limits of this technique. Disposable glass fibers with a 28.5 μm poly(dimethylsiloxane) (PDMS) coating were exposed to a soil, separately spiked with five PAHs, at a wide range of concentrations. The PDMS coated fibers were equilibrated with the soil slurry, and free pore water concentrations were calculated from PDMS-water partition coefficients. The results are also discussed from the perspective of soil (toxicity) testing.

Experimental Section

Soil, chemicals, fibers and solvents

Clean sandy agricultural soil (Borris-2) was collected in Denmark in 2001. The soil was stored at 4°C. Before use, the soil was dried to constant weight (at 25 ± 1°C), gently homogenized with a mortar, sieved (1 mm mesh size) and stored at room temperature.

Table 1 shows some properties of the test soil. Phenanthrene (Phe), pyrene (Pyr), benz[a]anhracene (BaA) benzo[b]fluoranthene (BbF) and benzo[ghi]perylene (BghiP), used for spiking the soils and making standard series, were all purchased at Sigma Aldrich Chemie BV (Zwijndrecht, The Netherlands). Glass fibers with a core diameter of 110 μm and a 28.5 μm poly(dimethylsiloxane) (PDMS) coating (volume 12.4 μl/m) were obtained from Poly Micro Industries (Phoenix, AZ, USA). Acetonitril, ethylacetate and acetone (Lab-Scan, Dublin, Ireland) used were of analytical grade, and highly pure water (R ≥18 MΩ) was prepared by a Millipore purification system equipped with organic free kit (Millipore waters, Amsterdam, The Netherlands).

Table 1: Soil properties of the Borris-2 soil (obtained from ref (*135*)).

pH	6.7
Sand (50-2000 μm, g/100g)	69.8
Silt (2-63 μm, g/100g)	20.5
Clay (<2 μm, g/100g)	6.9
Total organic carbon (g/100g)	1.67
Total organic matter (g/100g)	2.80
Dissolved organic carbon in pore water (mg/L)	20.9

Spiking Borris-2 soil

10 gram aliquots of air dried soil were put in 50 mL erlenmeyers and spiked with 2, 5, 10, 20, 50, 100, 200, 500 and 1000 mg/kg soil of Phe, Pyr, BaA, BbF and BghiP, separately. Additionally, Phe was spiked at 2000 mg/kg, and Pyr and BbF at 1500 mg/kg soil. All concentrations were spiked with 1 mL of acetone except for BghiP, where 2, 5 and 10 mL of acetone were necessary to dissolve and spike 200, 500 and 1000 mg/kg, respectively. The soil aliquots with acetone-spike were closed for 1 hour to let the acetone disperse, and reopened to let the acetone evaporate overnight at room temperature under a gentle stream of N_2. Subsequently, 1.25 mL water (~60% of the water holding capacity), with 10 mM sodium azide (NaN_3, Merck, Amsterdam, The Netherlands) to inhibit bacterial degradation, was added to the soil, and the soil was incubated for 28 days at 4°C.

Measurement of partition coefficients to PDMS

Fiber water partitioning were determined with a relatively new method that is based on the depletion of pre-loaded fibers with selected amounts of water (*137*). The advantage of this method is that it avoids difficulties that are often encountered when spiking aqueous solutions with hydrophobic chemicals. The PDMS coated fibers were cut into 5.0 or 3.0 cm pieces and thermally cleaned at 275°C for 16 hr under a constant helium flow of 30-35 mL/min. Fibers were stored in millipore water until use. Clean fibers were "loaded" by exposing them to a 1:1 methanol-water mixture (~6.2 μL PDMS in 5 mL methanol-water) spiked with Phe, BaA and BghiP separately at six concentration levels.

The Phe, BaA and BghiP loaded fibers were exposed to 6.2, 38 and 102 mL water (10 mM sodium azide) respectively, thereby creating volume water-volume PDMS ratios of 11000, 102000 and 272000. The flasks were shaken for 27 d which is sufficient to reach equilibrium (*165*) in the dark at 20 ± 1°C. Loaded and water-exposed fibers were extracted with various volumes of acetonitrile (200 µL to 20 mL, depending on expected concentrations in the coating) for at least one day. The initial concentrations in the PDMS coating ranged from 1.7 to 4500 mg/L, 0.09 to 600 mg/L and 0.09 to 16.6 mg/L, for Phe, BaA, and BghiP respectively. The assumption behind this method is that the mass balance (Equation 1) was 100% and the validity of this assumption has been proved in (*165*),

$$C_{f\,(initial\ )} * V_f = C_f * V_f + C_{aq} * V_{aq} \qquad (1)$$

where $C_{f\,(initial)}$ is the initial concentration in the loaded fiber, C_f is the concentration in the exposed fiber, V_f is the PDMS volume, C_{aq} is the aqueous concentration and V_{aq} is the aqueous volume. The fiber-water partition coefficient (K_f) can then be calculated from:

$$\frac{C_f}{C_{f\,(initial)}} = \frac{1}{1 + \frac{V_{aq}}{V_f * K_f}} \qquad (2)$$

Measurement of total and pore water concentrations in soil

Total soil concentrations were determined by a soxhlet extraction according to Szolar et al. (2002) (*100*). 2 grams of soil were sampled (triplicate per concentration) and extracted with 45 mL ethylacetate for 16 hours. The extract was diluted with acetonitrile (6 to 1000 times depending on soil concentration) and no further cleanup treatments were necessary. Parallel to the soxhlet extractions, 2 grams soil (triplicate per concentration), 2 mL of water (10 mM sodium azide), 2 thermally cleaned fibers of 5 cm were put in a 7.4 mL amber vial for SPME analysis (*30, 163*). The soil slurry was shaken for 48 hours on a "rock and roller" shaker (Snijders Scientific, Tilburg, The Netherlands). Earlier studies have shown that 48 hours rock and rolling is sufficient to reach ≥95% of the equilibrium for the selected compounds (*30, 166*). After exposure, the fibers were sampled and cleaned with a moist tissue and extracted in various volumes of acetonitrile (200 µL to 10 mL), depending on the expected concentrations in the fibers. Freely dissolved pore water concentrations (C_{aq}) in the soil were calculated using the concentrations in the fiber coating (C_f) and PDMS-water partition coefficients (K_f) according to Equation 3.

$$C_{aq} = \frac{C_f}{K_f} \quad (3)$$

Analysis of samples

The concentrations in the soil- and fiber-extracts were determined by HPLC-fluorescence detection. The system consisted of a Shimadzu DGU 14A degasser (Den Bosch, The Netherlands), a Varian Prostar 420 autosampler (Bergen op Zoom, The Netherlands), a Gynkotek P580 HPG HPLC pump (Gemering, Germany), and a Jasco FP-920 fluorescence detector (Maarssen, The Netherlands). Separation was performed using a Supelcosil (Supelco, Bellefonte, CA, USA) LC-PAH column (length 100 mm, ø 4.6 mm, particles 3 μm) that was operated at 26°C. All analyses were performed isocratically with a flow rate of 1.0 mL/min and an injection volume of 20 μL. Phe was eluted with a acetonitrile-H_2O ratio of 70%:30% Pyr was eluted at 80%:20%, BaA at 85%:15% and BbF at 90%:10%, while BghiP was eluted with 100% acetonitrile. The excitation and emission wavelengths (nm) of Phe were 255 and 355, Pyr was analyzed at 274/400, BaA at 280/390, BbF at 260/420 and BghiP at 295/415. Chromatograms were analyzed using Chromcard version 1.21 (Milan, Italy), and corrected by hand if necessary. Detection limits (peaks ≥5 times background noise) ranged from 0.05 to 0.2 μg/L for the selected PAHs.

Results and Discussion

Determining PDMS sorption isotherms

A Freundlich isotherm (Equation 4) was fitted to test the linearity of the relation between the concentration in the fiber coating (C_f, μg/L) and the aqueous phase (C_{aq}, μg/L).

$$C_f = K_f * C_{aq}^{\ n} \quad (4)$$

The K_f is the PDMS-water partition coefficient at an aqueous concentration of 1.0 μg/L, and n is the parameter describing the sorption linearity. The obtained n-values (1.01 ± 0.01, 1.01 ± 0.02 and 1.05 ± 0.03 for Phe, BaA and BghiP respectively) did not differ significantly from 1, so the sorption to the PDMS material can be considered linear, and a single concentration-independent K_f could be calculated. Figure 1 shows the concentrations in the fiber coating (C_f) plotted against the aqueous concentrations (C_{aq}). The lines represent the fit of Equation 4 with n fixed at 1. The obtained K_f-values are listed in Table 3. There has been some debate about the process of sorption to PDMS coated SPME fibers (*71, 73, 167*). Concentration-independent K_f-values over a broad range of concentrations up to 4 orders of magnitude, give strong evidence that the sorption to the PDMS polymer on the disposable fibers used is a partitioning process. A

similar conclusion was drawn by Mayer et al. (*70*), Poerschmann et al. (*73*) and Vaes et al. (*71*).

Table 2: Detection limits of pore water analysis using nd-SPME.

Comp.	*Detection limits in acetonitrile (µg/L)*	*Detection limits in pore water (ng/L)* [a]	*Corresponding organic carbon normalized concentration in the soil (µg/kg)* [b]	*Volume of pore water sampled by a 5 cm fiber (mL)* [c]	*Depletion of 2 grams of Borris-2 soil by the addition of 2*5 cm fiber (%)*
Phe	0.20	10	66	4.9	0.55
Pyr	0.30	3.7	95	15.9	0.53
BaA	0.1	0.51	31	37.4	0.14
BbF	0.30	0.51	97	118.1	0.14
BghiP	0.20	0.20	76	187.2	0.07

[a] A 5 cm fiber extracted in 200 µL acetonitrile and the analytical methods described in paragraph "Analysis of Samples" were used.
[b] Detection limits in field soils can be higher if sorption is higher than in spiked soils (*28, 166, 168*).
[c] Calculation with: $V_{fiber} * K_{fiber} = V_{aq}$.

Table 3: PDMS-water partition coefficients (K_f) and aqueous solubility, and soil sorption coefficients of the test compounds.

Comp.	*Log K_{OW}*	*Log K_f (SE, n)*	*Aq. sol. µg/L from lit.*	*Max. pore water conc. µg/L (SD, n)*	*Log K_{OC} at 1000 mg/kg OC (SE, n)*	*$n_{Freundlich}$ (SE)*	*Log K_{OC} from lit.* [h]	*Ratio between measured and estimated K_{OC}-values*
Phe	4.56[a]	3.82 (0.01, 15)[e]	1100 [g], 823 [c]	912 (39, 12)	4.65 (0.03, 22)	0.84 (0.01)	4.45	1.6
Pyr	5.22[b]	4.41 [f]	131 [g]	99.0 (4.0, 11)	5.26 (0.02, 24)	0.90 (0.02)	5.01	1.8
BaA	5.91[c]	4.78 (0.03, 17)[e]	13.0 [c]	7.76 (0.19, 8)	6.21 (0.01, 20)	0.89 (0.01)	5.70	3.2
BbF	6.20[d]	5.28 [f]	15.1 [g], 1.09 [c]	5.34 (0.53, 8)	6.72 (0.01, 20)	0.89 (0.01)	5.99	5.4
BghiP	6.85[b]	5.48 (0.03, 11)[e]	0.27 [g], 0.137 [c]	0.329 (0.03, 20)	7.18 (0.06, 20)	0.88 (0.05)	6.64	3.5

[a] Data from De Bruijn et al. (*104*).
[b] Data from Yalkowsky et al. (*139*).
[c] Data from De Maagd et al. (*169*).
[d] Data from Ma et al. (*140*).
[e] Data obtained from this study, Figure 1.
[f] Data from Ter Laak et al. (*165*).
[g] Data selected by Mackay et al. (*53*).
[h] Calculated from a QSAR of from Karickhoff et al. (*35*): $\log K_{OC} = \log K_{OW} - 0.21$.

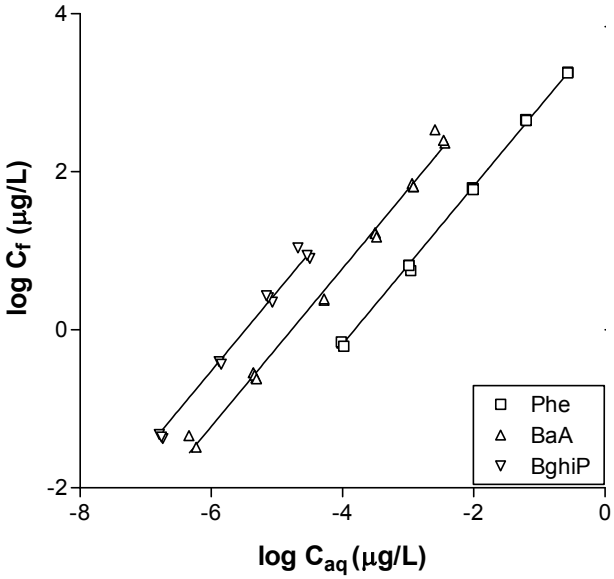

Figure 1: The concentrations in the fiber coating (log C_f) vs. the aqueous concentration (log C_{aq}) of three PAHs. Aqueous concentrations are calculated using a 100% mass balance approach.

Measuring aqueous concentrations in soil pore water and determining soil sorption isotherms

The large partition coefficients, and clean fiber extracts lead to low detection limits of aqueous concentrations in soil pore water (*33*). Table 2 shows the detection limits using the current analytical set-up, using 5 cm fibers extracted with 200 µL acetonitrile, and an injection volume of 20 µL. The exposed fibers did not deplete the soil slurry as they sampled 0.55% or less of the total amount in the system (Table 2). This is important information because, negligible depletion is a critical assumption behind the measurement of freely dissolved pore water concentrations (*34*). Increasing the fiber volume, decreasing the acetonitrile volume and improving analytical conditions and equipment could lower the detection limits even further. Pore water concentrations could be detected in the ng/L-range, with only using 2 grams of soil, and without any cleanup. Five to 187 mL pore water should be extracted and transferred into 200 µL acetonitrile to obtain similar detection limits from pore water extractions. Collecting these volumes of pore water requires large amounts of soil and the procedure for the collection of pore water is rather laborious and time consuming. Another complicating factor in the analysis of pore water concentrations in soil is the presence of dissolved organic matter (DOM). A more detailed quantitative discussion of the effects of DOM is given below.

Concentrations of DOM can range from single milligrams per liter to hundreds of milligrams per liter, depending on the soil type and soil pH. The sorption of hydrophobic compounds to DOM can lead to an overestimation of the freely dissolved concentration in exhaustive liquid-liquid extractions (*14, 17, 40, 51, 108*). Generally, the more hydrophobic the compound, the higher the sorption coefficients to the DOM, and therefore the larger the overestimation of the free concentration (*66*). The overestimation of the aqueous concentration would be a factor of 1.1 for Phe to 11.3 for BghiP for a soil

with 10 mg/L dissolved organic carbon (DOC) in the pore water (log K_{DOC}-values are based on the octanol-water partition coefficient and a QSAR specific for pore water-DOC (*40*)). The removal of DOM without disturbing the equilibrium of the compounds in the pore water is difficult (*1*). Flocculation (*16, 168*) can remove the DOM from the aqueous phase, but the addition of e.g. aluminum potassium sulfate, and adjusting the pH might affect the equilibrium of the freely dissolved and sorbed compounds (*74, 90, 91*). Passive separation by a dialysis membrane might be a better option, but both techniques still need large aqueous volumes to obtain sufficient sensitivity.

The used negligible depletion passive sampler is a good alternative to determine free aqueous concentrations in complex matrices like soil or sediment, as has also been shown by others (*27, 30, 170*). The technique does not need separation of the aqueous and matrix phase. It can also detect very low aqueous concentrations, since the partition coefficient increases with increasing hydrophobicity and decreasing solubility of the test compounds. Detection limits are in the range of 0.2 to 10 ng/L in pore water, corresponding with ~1 to 10 μg/kg in a soil with 2% organic carbon and sorption coefficients estimated from the octanol-water partition coefficient and a QSAR of Karickhoff et al. (*35*). In addition, the extracts are very clean and, therefore, no clean up steps are needed, contrary to what is often the case in soil analysis. The only requirements are: no depletion of the system by the passive sampler, known partition coefficients between the passive sampler and water, measurements performed at equilibrium and no substantial fouling on the fiber surface. The selection of an appropriate set-up and dimensions of the system can easily meet the first three requirements, but fouling may affect uptake kinetics or increase the sorption capacity (*171*). Several studies have shown that this does not occur to an extent that it influences the measurements (*48, 79, 172*). In addition, disposable fibers used in this study are exposed and extracted only once, so the disturbing fouling effects are probably less than for the repetitive exposure and thermal desorption of commercial SPME fibers (e.g. at 275°C).

The relation between the organic carbon normalized soil concentration (C_{OC}) and the concentration in the PDMS coating of the fiber (C_f) is shown in Figure 2. It can be observed that the concentration in the fiber increase with the soil concentration until a certain threshold-value. This threshold might be an effect of saturation of the PDMS phase or the aqueous phase. The compounds are thought to partition into the "rubbery liquid" (*173*) PDMS material (*25, 70, 71, 73*). The sorption to the PDMS is linear over a broad range of concentrations, so this assumption is probably correct. In true partitioning processes, the ratio of the solubility of a compound in phase A (PDMS) and phase B (water) should be equal to their partition coefficient.

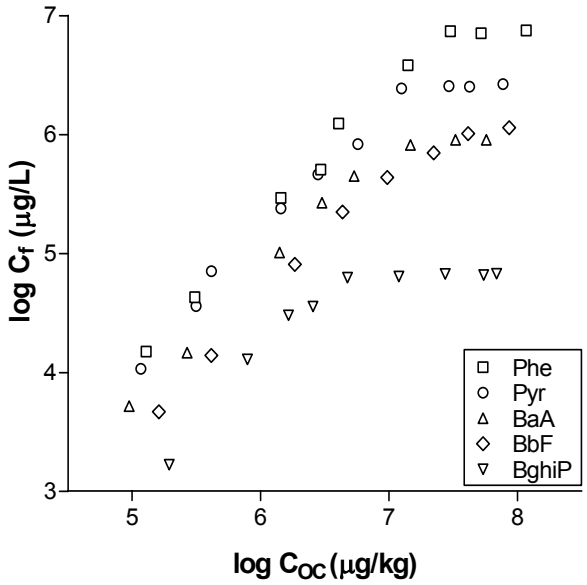

Figure 2: The logarithm the PAH-concentration in the fiber coating (log C_f) plotted against the logarithm of the organic carbon normalized PAH-concentration in the soil (log C_{OC}).

Figure 3 shows the relation between the soil concentrations (C_{OC}) and the free pore water concentration (C_{aq}), calculated from fiber concentrations (C_f) and fiber partition coefficients (K_f) using Equation 3.

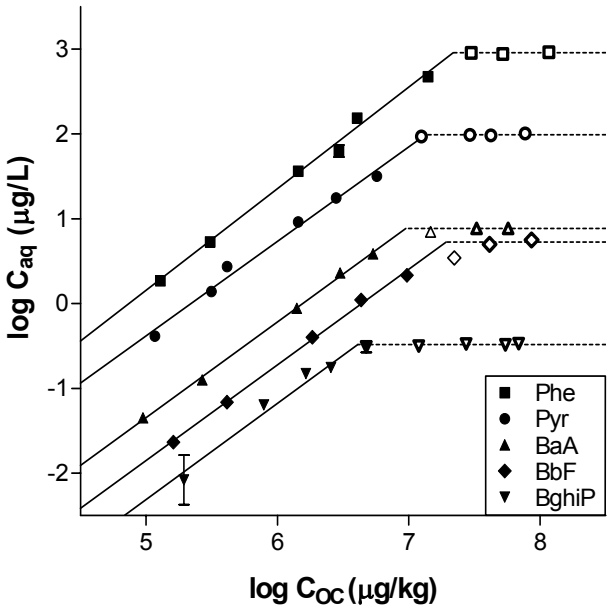

Figure 3: The logarithm of the freely dissolved concentration in the pore water (log C_{aq}) plotted against the logarithm of the organic carbon normalized concentration in the soil (log C_{OC}). The solid lines represent the Freundlich isotherms, and are fitted on the solid symbols. The maximum freely dissolved concentrations measured in the pore water (broken lines), were determined with the thick open symbols (the maximum range of selected values was 0.10 log units). Error bars represent the standard deviations, and are in most cases too low to be visible.

Freely dissolved pore water concentrations increase with increasing soil concentration up to a certain maximum (threshold). The observed maximum in soil pore water concentrations are very similar to aqueous solubility data obtained from literature (*53, 99*).

The observed threshold-value is very likely determined by the aqueous solubility of the PAHs (Table 3, Figure 4). As a spin off of this study, we suggest that the fiber method can also be used as a tool to estimate aqueous solubility as long as; the large amounts of PAHs (up to 7.0 g/L PDMS) do not affect the properties of the PDMS and all compounds extracted from the PDMS coating are dissolved (no crystals in the PDMS material or on its surface).

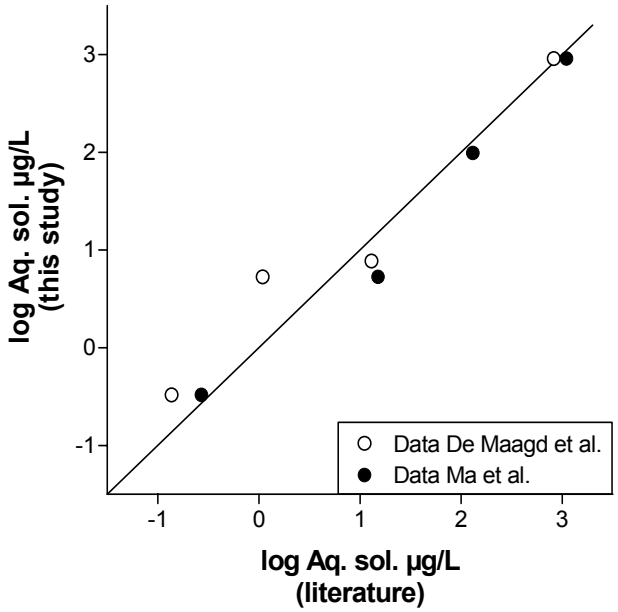

Figure 4: Estimated aqueous solubility of the tested PAHs by maximum observed concentrations in the PDMS coating, plotted against aqueous solubility determined by De Maagd et al. (*99*) and selected by Ma et al. (*140*). The line represents a 1:1 relationship.

Figure 3 also shows a Freundlich isotherm (Equation 5) that describes the relation of the organic carbon normalized soil concentration (C_{OC}) and the freely dissolved aqueous concentrations (C_{aq}). Only values below the threshold level were used.

$$C_{OC} = K_{OC} * C_{aq}^{\ n} \qquad (5)$$

The K_{OC} is the organic carbon normalized sorption coefficient at an aqueous concentration of 1.0 µg/L, and n is the Freundlich linearity parameter. Table 3 displays the calculated sorption coefficients at a concentration of 1000 mg/kg organic carbon, and the $n_{Freundlich}$. The sorption is close to linearity ($n_{Freundlich}$ ranges from 0.84 to 0.90),

suggesting that a non-specific hydrophobic absorption process into relatively amorphous "soft" natural organic matter (*7*) is the most important sorption process**.** The rather linear sorption isotherm could be expected, since the Borris-2 soil was freshly spiked and does not contain large amounts of (xenobiotic) organic materials like soot, coal or tar that could have led to nonlinear sorption isotherms due to slow absorption and strong adsorption interactions (*7, 112, 113, 174*).

Effects of dissolved organic matter on soil sorption coefficient determination

The obtained sorption coefficients are higher (a factor 1.6 to 5.4) than estimated according to a QSAR of Karickhoff et al. (*35*) (see Table 3). Generally, this factor increases with the hydrophobicity of the PAH. The difference might be an effect of slight overestimations of the concentration in the aqueous phase, due to DOM sorption, in the original sorption studies. Other explanations are related to potential differences in sorbent properties between the two studies or differences in the concentration level at which the sorption studies were performed. The potential effect of dissolved organic matter on the measurement of sorption coefficients has been recognized earlier by Schrap et al. (*14*). The ratio of the actual freely dissolved concentration in soil pore water (C_{aq}) and the total concentration in solution (C_{total}, including the DOM bound fraction) can be estimated from Equation 6.

$$\frac{C_{aq}}{C_{total}} = \frac{1}{1 + K_{DOC} * C_{DOC}} \qquad (6)$$

Where K_{DOC} is the sorption coefficient and C_{DOC} is the dissolved organic carbon concentration. Exact DOC or DOM concentrations and specific sorption coefficients are unknown in our experiments. Therefore, the effect of DOC is illustrated, using a pore water specific log K_{OW}-based relationship of Burkhard (*40*) and a relatively low DOC concentration of 10 mg/L (Table 4).

Table 4: Potential overestimation of aqueous concentrations by sorption to DOC.

Compound	*Log K_{OW}*	*Log K_{DOC}* [e]	*Fraction freely dissolved at 10 mg/L DOC*	*Overestimation-factor of pore water concentration at 10 mg/L DOC*
Phe	4.56 [a]	3.93	0.92	1.1
Pyr	5.22 [b]	4.53	0.75	1.3
BaA	5.91 [c]	5.16	0.41	2.4
BbF	6.20 [d]	5.42	0.27	3.6
BghiP	6.85 [d]	6.01	0.09	11.3

[a] Data from De Bruijn et al. (*104*).
[b] Data from Yalkowski et al. (*139*).
[c] Data from De Maagd et al. (*169*).
[d] Data from Ma et al. (*140*).
[e] Calculated from a pore water-DOC specific QSAR (*40*): $\log K_{DOC} = 0.91 * \log K_{OW} - 0.22$.

It is obvious from this simulation that measurement of total concentrations in pore water can be highly overestimated, leading to systematic errors in the determination of soil sorption coefficients. The estimated fractions listed in Table 4 are in the same range as observed in our experiment (Table 3). These systematic errors can be avoided by actual measurements of free concentrations.

Relevance for soil testing

The free concentration in soil pore water is a relevant entity for all kinds of processes in soil, including evaporation to air, (bio)degradation and accumulation in biota living in the soil or sediment (*3, 47, 159, 161, 175*). The freely dissolved concentration of a compound is generally thought to be decisive in determining the internal concentration and subsequent toxic effects in small soil dwelling deposit feeders (*47, 163, 170, 176, 177*). Due to their limited aqueous solubility, the larger PAHs (≥4 rings) can't evoke lethal body burdens (LBB) in the organisms, and therefore, these chemicals are generally found to be non-toxic (*178-180*). In soil toxicity tests, however, organisms are generally exposed to a series of increasing concentrations in soil, even above the aqueous solubility in the pore water. Effects are then usually expressed on basis of total soil concentrations, or sometimes to estimated pore water concentrations (*176, 179, 180*). Freely dissolved concentrations in the pore water are hardly ever measured, and the saturation of the aqueous phase is often disregarded. Results from this study show that the aqueous phase can get saturated in a soil toxicity test set-up at concentrations of 80 to 400 mg/kg (at an organic carbon content of 2%). Negligible depletive passive samplers like PDMS coated fibers and also other partition based sampling methods might be applied to monitor freely dissolved concentrations (and saturation of the aqueous phase) in soil and sediment toxicity set-ups. Information on these pore water concentrations may be extremely useful in interpreting the outcome of these soil tests.

Aknowledgements

This work was funded by the LIBERATION project, EVK1-CT-2001-00105. We would like to thank the partners of the LIBERATION project for supplying the soil and data on the soil. Furthermore, we would like to thank Kai-Uwe Goss for fruitful discussions on solubility issues.