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A sediment extraction and cleanup method for wide-scope multitarget screening by liquid chromatography–high-resolution mass spectrometry

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Abstract Previous studies on organic sediment contaminants focused mainly on a limited number of highly hydrophobic micropollutants accessible to gas chromatography using non-polar, aprotic extraction solvents. The development of liquid chromatography–high-resolution mass spectrometry (LC–HRMS) permits the spectrum of analysis to be expanded to a wider range of more polar and ionic compounds present in sediments and allows target, suspect, and nontarget screening to be conducted with high sensitivity and selectivity. In this study, we propose a comprehensive multitarget extraction and sample preparation method for characterization of sediment pollution covering a broad range of physicochemical properties that is suitable for LC–HRMS screening analysis. We optimized pressurized liquid extraction, cleanup, and sample dilution for a target list of 310 compounds. Finally, the method was tested on sediment samples from a small river and its tributaries. The results show that the combination of 100 °C for ethyl acetate–acetone (50:50, neutral extract) followed by 80 °C for acetone–formic acid (100:1, acidic extract) and

methanol–10 mM sodium tetraborate in water (90:10, basic extract) offered the best extraction recoveries for 287 of 310 compounds. At a spiking level of 1 μg mL^{-1}, we obtained satisfactory cleanup recoveries for the neutral extract—(93 ± 23)%—and for the combined acidic/basic extracts—(42 ± 16)%—after solvent exchange. Among the 69 compounds detected in environmental samples, we successfully quantified several pharmaceuticals and polar pesticides.

Keywords Sediments · Liquid chromatography · High-resolution mass spectroscopy · Multitarget screening · Pressurized liquid extraction

Introduction

Aquatic environments are contaminated with several thousand chemicals, including pesticides, biocides, pharmaceuticals, personal care products, and industrial chemicals, together with numerous transformation products [1]. Organic pollutants enter water bodies through different pathways, such as wastewater treatment plant (WWTP) effluents and runoff from urban areas and agricultural fields [2], and they are distribute among different compartments according to their physicochemical properties. In particular, sediments are a well-known sink not only for highly hydrophobic chemicals [3–5] but also for ionic and zwitterionic compounds (i.e., pharmaceuticals, personal care products, or surfactants [6–8]) since they can form strong electrostatic interactions with sediment organic matter and clay minerals [9]. Although sediments are mainly considered as a sink, they may also act as sources for a large variety of pollutants that may impact the aquatic ecosystems and human health [10].

For decades, analyses of organic contaminants in sediments have been focused on highly hydrophobic chemicals

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such as polycyclic aromatic hydrocarbons, polychlorinated biphenyls , and other persistent organic pollutants that are accessible to gas chromatography (GC) [11–13]. In this context, many extraction methods have been developed based on nonpolar, aprotic solvents such as hexane, dichloromethane, or mixtures that use Soxhlet extraction, pressurized liquid extraction (PLE), or ultrasound-assisted extraction [14–17]. Chemical analyses were typically focused on the detection and quantification of specific compound classes by optimization of recoveries and detection limits for these chemicals as well as application of specific cleanup steps (e.g., sulfuric acid based cleanup for polychlorinated biphenyls and dioxins/furans [18]). The analysis of comprehensive chemical profiles in sediments was only rarely within the scope of research, and GC–mass spectrometry (MS) screening methods were mostly applied for the identification of novel and unexpected contaminants in sediments [19–21].

In the last two decades, liquid chromatography (LC)–MS offered the opportunity to expand the compound spectrum toward more polar compounds, and many studies have proved the presence of polar pesticides, pharmaceuticals, and industrial chemicals not only in water but also in sediments [22–24]. These methods used a wide range of extraction methods such as liquid–liquid extraction, ultrasonication, microwave-assisted extraction, and PLE normally followed by dispersive matrix extraction or solid-phase extraction for cleanup [6, 8, 25–27]. Polar or protic solvents or buffers were preferred for the extraction of more hydrophilic compounds [6, 23, 25, 26, 28].

With the availability of LC coupled with high-resolution MS (HRMS), it has become possible to conduct target, suspect, and nontarget chemical screening at high sensitivity and selectivity [29], which were mainly used for the analyses of wastewater and surface water [30–34]. In contrast, the application of broad, generic screening methods to characterize sediment pollution is still limited [24, 35]. In particular, there is currently a lack of adequate sample preparation methods allowing the screening of substances with a wide range of polarities, as most methods address mainly nonionizable compounds. A general problem in generic sample preparation procedures is to balance sufficient cleanup of crude extracts to make them amendable to analysis against a loss of compounds during such cleanup steps. Therefore, we aimed to develop and optimize a multitarget method for sediments to cover ionic and nonionic compounds with a broad range of physicochemical properties that are suitable for LC–HRMS screening analysis, expanding the PLE method for neutral compounds of Chiaia-Hernandez et al. [24] and aiming at including ionic compounds by additional extraction steps.

While for the neutral extract a clean-p procedure based on normal-phase (NP) column chromatography was evaluated (leaving as an option the exploitation of a nonpolar fraction by GC–MS), the ionic extracts were subjected only to solvent exchange. We also studied the impact of matrix load (i.e., organic matter load in the extracts) on LC–HRMS analysis to find the optimum dilution factor for the extracts. Finally, we demonstrate the applicability of the method developed on sediment samples from a small river and its tributaries receiving wastewater effluents and agricultural runoff.

Material and methods

Standards and reagents

For method development, a set of 310 compounds was selected, including pesticides, pharmaceuticals, personal care products, steroids, and other compounds known or likely to occur in aquatic systems and sediments. These cover a wide range of physicochemical characteristics (at pH 7 log D between -6 and 8, 205 neutral, 49 anionic, 56 cationic). A standard mixture of all compounds was prepared at a concentration of 1 μg mL^{-1} in methanol. A mixture of 19 isotope-labeled internal standards was also prepared in methanol at 1 μg mL^{-1}. Details on the target compounds and other chemicals used are provided in the electronic supplementary material (section S1 and Tables S1, S2).

Method development

PLE optimization

On the basis of the study of Chiaia-Hernandez et al. [24], a mixture of ethyl acetate and acetone (EtAC; 50:50 v/v) was chosen for the first extraction step targeting more hydrophobic, neutral compounds (neutral extract). As the second step, acetone–formic acid (AcFA; 100:1 v/v) was tested for the extraction of anionic compounds after protonation at low pH (acidic extract), and as a third step, methanol–10 mM sodium tetraborate in water (MeWA; 90:10, v/v) was tested to extract cationic compounds after (partial) deprotonation (basic extract).

To test the effect of temperature (i.e., mainly thermal degradation) on recovery and the distribution of compounds in the different extraction steps, first the extraction recoveries of compounds added to diatomaceous earth were assessed, followed by a comparison with a spiked test sediment collected from a lowly polluted stretch of the river Saale near Calbe (Germany).

For the first experiment, clean diatomaceous earth (Hydromatrix, Restek) was transferred to stainless steel PLE cells containing a 27-mm glass fiber filter (Dionex) at the bottom and spiked with 100 μL of analyte mixture. The cell contents were then extracted by PLE (ASE 200, Dionex) with the three different solvents in the order EtAC, AcFA, and MeWA. The extraction was performed at three different

temperatures (80, 100, and 120 °C) to assess the thermal degradation of the compounds., The pressure was set to 103.4 bar in all the experiments, as no significant effect on extraction was expected [36]. Further details of the PLE method are given in Table S3.

PLE extracts were collected separately as one fraction for each solvent, evaporated close to dryness with a rotary evaporator (60 rpm, 40 °C), redissolved in 1 mL of HPLC-grade methanol, and filtered with a cellulose filter (0.45 µm, Macherey-Nagel). Samples were stored at -20 °C until analysis. Experiments were performed in triplicate, and a process blank was run with each analysis. Recoveries were calculated by our comparing the peak areas of the spiked samples extracted by PLE with those of a standard solution prepared in pure solvent.

Optimization of the neutral PLE extract cleanup

The separation of those compounds detected in the neutral extract by NP chromatography was assessed. To this end, 1.5 g of activated silica gel was spiked with 1 µg of the analytes, and the solvent was evaporated to dryness with a rotary evaporator. This "loaded" silica was added on top of 1 g of activated aluminum oxide into a prefilled glass column. The analytes were eluted with a sequence of four solvents with increasing polarity: *n*-hexane (11 mL), *n*-hexane–dichloromethane (3:1, 7.5 mL), dichloromethane (15 mL), and methanol (10 mL). Each fraction was collected separately. Relative recoveries of the whole procedure were calculated in an additional experiment at two spiking levels (0.1 and 1 µg of the analytes) by our mixing the four solvents after cleanup. Eluted fractions were evaporated close to dryness, redissolved in 100 µL of HPLC-grade methanol, and filtered through a polytetrafluoroethylene syringe filter (0.45 µm, Macherey-Nagel). Recoveries were calculated by our comparing the peak areas of mixture components loaded on silica and chromatographed on aluminium oxide with those of a standard solution prepared in pure solvent. Experiments were performed in duplicate with a process blank. Detailed information about the preparation of the NP adsorbents is provided in the electronic supplementary material (section S1).

Solvent exchange of acidic and basic PLE extracts

The recoveries of the compounds detected in the acidic and basic extracts were determined by spiking 15 mL of AcFA with 1 and 10 ng of these analytes. The solvents were evaporated close to dryness with a rotary evaporator and mixed with 15 mL of MeWA extract spiked with 1 and 10 ng of these analytes. The pH was adjusted to 7 with 7 N ammonia in methanol. The samples were evaporated close to dryness, redissolved in 100 µL of HPLC-grade methanol, and passed through a polytetrafluoroethylene syringe filter (0.45 µm,

Macherey-Nagel). Recoveries were calculated by our comparing the peak area of spiked and processed extraction solvents with those of a standard solution prepared in pure methanol.

Optimal sample dilution to minimize the matrix effect

We determined the dilution factor (i.e., the ratio of the final solvent volume in the autosampler vial to the mass of sediment organic carbon extracted) that gives a good compromise between low signal suppression and high signal intensity. To this end, sediment extracts corresponding to increasing concentrations of extracted organic matter (20, 50, 100, 250, and 500 mg organic carbon per milliliter) with a correspondingly increasing concentration of a mixture of pesticides and pharmaceuticals (40, 100, 200, 500, and 1000 ng mL^{-1}) were combined to yield a constant analyte concentration of organic carbon per milliliter, but at different dilution levels. Organic matter was obtained by extraction of the Calbe test sediment by the optimized three-step PLE and processing of the neutral extract by NP chromatography and the acidic and basic extract mixture by solvent exchange. Matrix effects on recovery were calculated according to the equation:

$$Matrix\ effect\ on\ recovery$$
$$= \frac{Peak\ area\ of\ analyte\ with\ matrix}{Peak\ area\ of\ analyte\ without\ matrix}.$$

LC–HRMS analysis

For method development, target compounds were analyzed with an Agilent 1200 series ultraperformance LC system coupled to a hybrid linear ion trap–Orbitrap mass spectrometer (LTQ Orbitrap XL, Thermo Scientific) equipped with a heated electrospray ionization source. The samples from the river Holtemme were analyzed with a Thermo Scientific UltiMate 3000 LC system coupled via a heated electrospray ionization source to a quadrupole–Orbitrap mass spectrometer (QExactive Plus, Thermo Scientific). Detailed LC and MS settings are provided in the electronic supplementary material (section S2 and Tables S4, S5). The method detection limits (MDLs) for method development were determined on the basis of the US EPA guidelines [37]. Additional information is provided in the electronic supplementary material (section S3).

Analysis of river Holtemme sediment samples

Sediment samples were collected in October 2014 from the river Holtemme and its tributaries. While the upper stretch of the river is surrounded by forest, its lower stretches are characterized by discharges from two WWTPs and intensive agricultural areas. Surface sediments were collected in three different sedimentation spots (at a weir on the main

river downstream of the two WWTPs, and two tributaries). Details on sampling, water content, and total organic carbon (TOC), total inorganic carbon, and black carbon contents of the sediments are provided in the electronic supplementary material (sections S4 and S5 and Table S6). For sample quantification, two sets of method-matched calibration standards were processed; one for the neutral extract by our spiking 1.5 g of activated silica with analyte standards of different levels and processing it through the NP column, and one for the acidic/basic fraction by our spiking 15 mL of AcFA and MeWA with analyte standards of different levels, correcting the pH to 7 with 7 N ammonia, evaporating the fraction and redissolving it in HPLC-grade methanol. In both cases the final concentrations in the vial were 0.1, 1, 5, 10, and 25 ng mL^{-1}. The extraction from sediment was not included in the method-matched calibration, as it is not possible to adequately mimic the extractability of native analytes by artificial spiking of sediments. An internal standard mixture containing 40 isotope-labeled compounds was added before injection to each vial at a final concentration of 100 ng mL^{-1}. Internal calibration was used, assigning one internal standard to each analyte on the basis of the closest retention time. Details are given in Table S1. For peak integration, compound calibration, and compound quantification the software program TraceFinder 3.2 (Thermo Scientific) was used. The full-scan extracted ion chromatograms (within ±3.5-ppm mass accuracy) were used for the detection and quantification of target compounds, whereas the match of isotope patterns and the presence of one to three HRMS/MS fragment ions were used for the confirmation of the analytes. Software settings are given in Table S7.

Results and discussion

PLE optimization

Temperature optimization and solvent selection

Temperature and solvents were expected to have the major effect on the extraction efficiency because of their impact on mass-transfer kinetics and solubility [38]. In the temperature and solvent selection experiment, the combination of 100 °C for EtAC followed by 80 °C for AcFA and MeWA gave the best results, with 223 of 310 compounds recovered at 30% or more. About 200 of these compounds had been recovered in the EtAC fractions. The lowest recoveries were obtained at 120 °C for all three solvents, probably due to thermal degradation of compounds at higher temperatures [36]. Extraction recoveries at different temperatures are provided in Tables S8 and S9.

Extraction recoveries and distribution among sequential solvents

The sequential extraction using EtAC at 100 °C (neutral extract) followed by AcFA at 80 °C (acidic extract) and MeWA at 80 °C (basic extract) was evaluated for spiked Hydromatrix and sediments. In the recovery experiment, 279 of 310 compounds were extracted from Hydromatrix with an average relative recovery of 72%, with 50% of compounds being extracted with an average relative recovery of between 59% and 88% (Fig. 1). For the spiked reference sediment we achieved lower recoveries (64% on average) and fewer compounds were recovered (208), with 50% of compounds being extracted with an average relative recovery of between 29% and 64%. In both analyses, compounds with recoveries lower than 10% where considered lost.

For Hydromatrix, the highest number of compounds (245) was extracted in the first step with EtAC, but the recoveries of 51 compounds were increased with the additional AcFA step, such as the pharmaceutical ibuprofen, the recovery of increased from 33% to 83%. The pharmaceuticals bosentan, lorazepam, and losartan could be extracted only with AcFA. The third extraction step (MeWA) recovered nine compounds otherwise lost. Also in the reference sediments the highest number of compounds (164) was extracted with EtAC, followed by AcFA (46). Moreover, it was possible to recover 12 compounds only with the third extraction step with an average recovery of 31%.

The distribution of 40 compounds among the different extracts showed remarkable differences between sediment and Hydromatrix (Fig. 2). Whereas from both sediment and Hydromatrix the largest number of nonionizable compounds were extracted with EtAC, cationic compounds (e.g., amantadine, mebeverine, and metoprolol) were found in larger fractions in the MeWA extract of sediments as compared with Hydromatrix. Anionic compounds (e.g., propoxycarbazone and mefenamic acid) showed higher recoveries in AcFA in sediment as compared with Hydromatrix.

The recovery of the compounds from sediment and the distribution among the extracts with different solvents could hardly be related to physicochemical properties or different classes of the compounds, which was anticipated when we were designing the three-step extraction procedure. In the EtAc extract, many cationic compounds also showed high recoveries, while the recoveries of anionic compounds were in general lower. However, there were many neutral as well as cationic and anionic compounds that could not be extracted from the spiked sediment. These substances might undergo specific interactions with the sediment organic matter, among them covalent bonding, which has been shown to occur nearly instantaneously with soil organic matter for certain compounds such as aromatic amines [39–41]. The AcFA extraction step
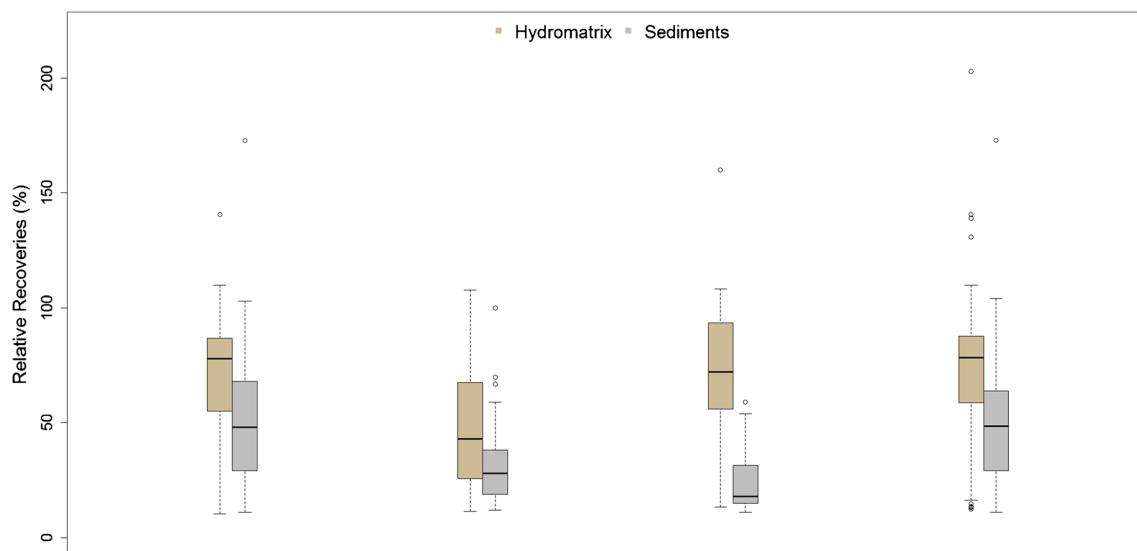
Fig. 1 Recoveries in spiked Hydromatrix and test sediments of target compounds with a sequential extraction using ethyl acetate–acetone (EtAC) at 100 °C, acetone–formic acid (AcFA) at 80 °C, and methanol (MeWA)–10 mM sodium tetraborate in water at 80 °C. Compounds with recoveries lower than 10% were considered lost. Whiskers represent the 9th and the 91st percentiles. Dots are considered outliers.

showed high recoveries of some acidic but also neutral and cationic compounds not extracted in the first step. Overall, the data suggest that the extractability of compounds with polar functional groups is governed by a complex interplay of sediment and compound properties. Additional information about recoveries and distribution is provided in Fig. S1.

Recoveries of the sample extract processing steps

NP chromatography of the neutral extract

We tested the recoveries of the 164 compounds detected in the neutral extract after NP chromatographic cleanup with a sequence of four solvents with increasing polarity: *n*-hexane, *n*-hexane–dichloromethane (3:1), dichloromethane, and methanol. Most of the target compounds in our study were partly found in the *n*-hexane–dichloromethane and methanol fractions with a relative recovery of more than 90% for 155 compounds at both concentrations tested and an overall method precision of 14% for all concentrations. Anionic compounds present in the neutral fractions showed lower recovery than neutral and cationic compounds, $(60 \pm 33)\%$. Amitriptyline, benzophenone-3, and temazepam were recovered only at the higher spiking level (1000 ng mL^{-1}). Moreover, five pharmaceuticals were completely lost during NP cleanup (pioglitazone, sulfamethazine, bisoprolol, sulfadimethoxine, and diphenhydramine). The distribution of compounds among the fractions depended on the number of hydrogen-bond donors and acceptors present in the molecule. While compounds with

no or a low number of hydrogen-bond donors/acceptors such as fenthion and iminostilbene were eluted with the *n*-hexane–dichloromethane fraction, those with a high number such as thiamethoxam were recovered only in the methanol fraction. The distribution of the compounds among the fractions is given in Table S10 and Fig. S2.

*Processing of the acidic and basic extract*

The recoveries of the compounds detected in the protic PLE fractions after evaporation, pH adjustment, and combination were $(52 \pm 25)\%$ at a spiking level of 100 ng mL^{-1} and $(42 \pm 16)\%$ at a spiking level of 1000 ng mL^{-1}. Previous studies on recoveries of pesticides and biocides [32, 42] showed comparable results after similar solvent exchange steps [i.e., solid-phase extraction and quick, easy, cheap, effective, rugged, and safe (QuEChERS) extraction], with recoveries ranging from 42% to 103%.

The overall recoveries after sample preparation (without extraction) for neutral, acidic, and basic extracts are shown in an overview in Fig. 3 for a spiking level of 100 ng mL^{-1}. An overview on the average relative recoveries and relative standard deviation for both procedures at different concentrations is given in Tables S11 and S12.

Optimal extract dilution for minimizing the matrix effect

Increasing concentrations of TOC in the analyzed extracts resulted in a decrease of the signal and consequently lower recoveries of the target compounds because of increasing ion

Fig. 2 Distribution of selected neutral, cationic, and anionic target compounds (at pH 7) in spiked Hydromatrix and reference sediments among the pressurized liquid extraction fractions. Formal charge at pH 7 was calculated with the calculator plugin JChem version 17.4.3.0 (ChemAxon). AcFA acetone–formic acid, EtAC ethyl acetate–acetone, MeWA methanol–10 mM sodium tetraborate in water
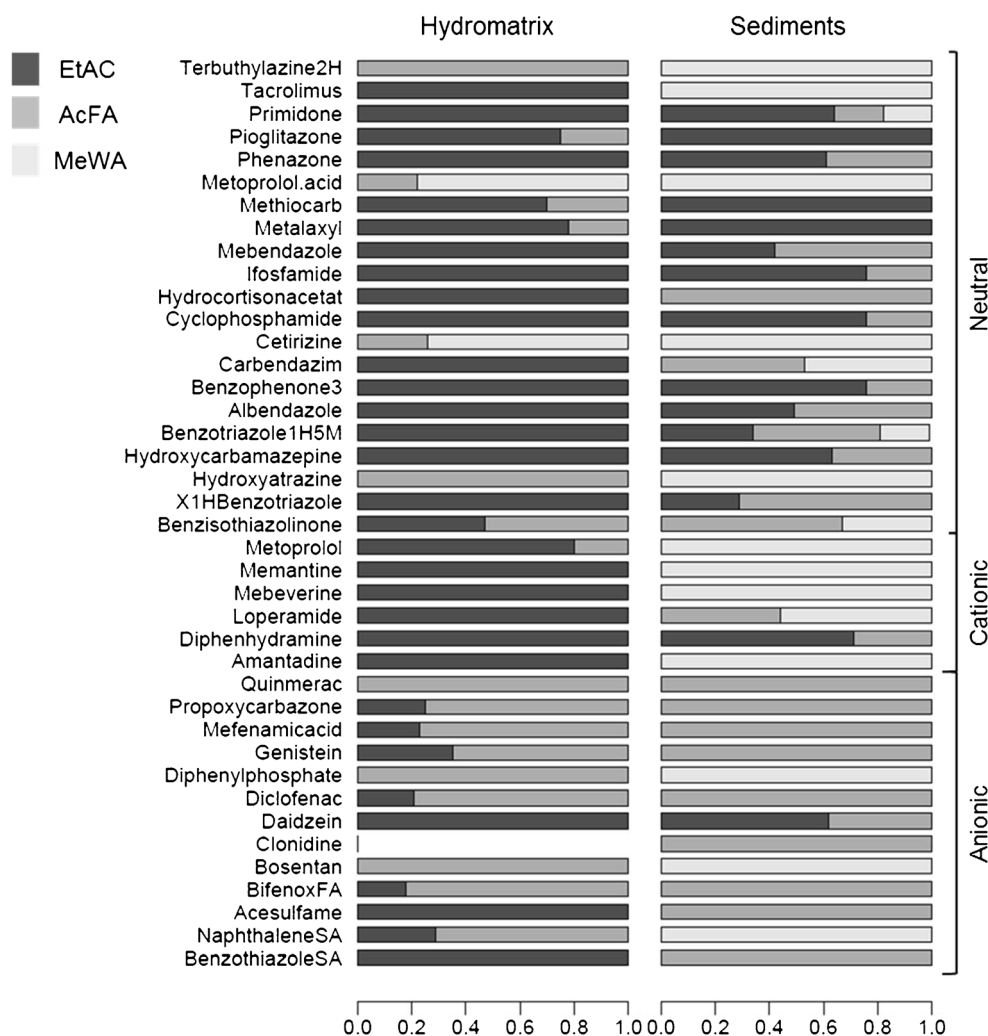
suppression (Table 1). TOC concentrations above 200 ng mL^{-1} resulted in poor performance of the HRMS analysis, because the ion transfer capillary became partially blocked and signal intensities of control standards decreased after five to ten sample injections.

This finding is in good agreement with the findings of previous studies on matrix effects [24]. To keep an acceptable balance between the amount of matrix injected and the signal intensity, 50 mg of TOC per milliliter in the vial (average recovery more than 60%) was selected as the best amount for the samples.

Optimized workflow

The final workflow resulting from the optimization of PLE, extract processing, and dilution is as follows. A total amount of 50 mg of sediment TOC equivalent (grain size 63 μm or less) was sequentially extracted by PLE using EtAc (50:50 v/v) at 100 °C (neutral extract), AcFA (100:1 v/v) at 80 °C (acidic extract), and MeWA (90:10 v/v) at 80

°C (basic extract). The extraction was performed with two static cycles of 5 min. For the cleanup of the neutral extract, deactivated silica (1.5 g) and aluminum oxide (1 g) were used. The sorbents were suspended in n-hexane and poured into a glass column with silica at the bottom and aluminum oxide on top of the silica. In a round-bottom flask, 0.75 g silica was mixed with the EtAC extract and evaporated to dryness with a rotary evaporator. This "loaded" silica was added to the column onto the aluminum oxide. This procedure allows the transfer of a broad range of organic compounds onto the column without the need to dissolve them in the first NP column elution solvent (n-hexane). The analytes were eluted sequentially with n-hexane (11 mL), n-hexane–dichloromethane (3:1, 7.5 mL), dichloromethane (15 mL), and methanol (10 mL). Half of the n-hexane–dichloromethane eluate was mixed with half of the methanol eluate and evaporated to 500 μL under a gentle stream of nitrogen. Samples were filtered (pore size 45 μm) and stored at -20 °C until analysis. Acidic extracts were evaporated close to dryness with
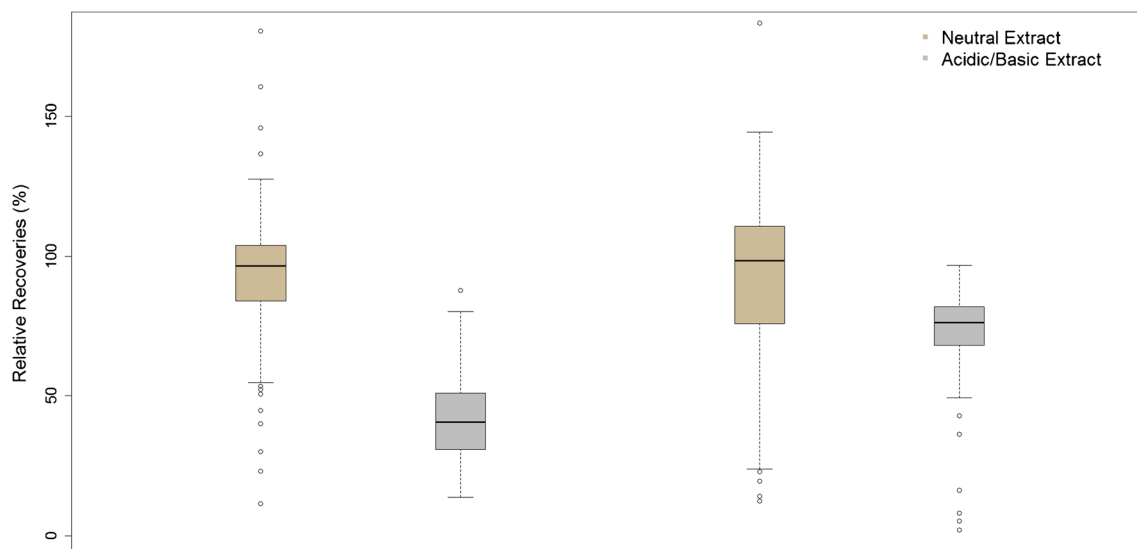
Fig. 3 Overall recoveries of 171 compounds for neutral and acidic/basic extracts at two spiking levels (100 and 1000 ng mL^{-1}). Whiskers represent the 9th and the 91st percentiles. Dots are considered outliers.

a rotary evaporator, and the solvent was changed to 15 mL of methanol. After mixing with the basic extract, the pH was adjusted to 7 with 7 N ammonia in methanol. The sample was evaporated close to dryness, redissolved in 1 mL of methanol, and filtered (pore size 45 μm). Samples were stored at -20 °C until analysis. A flowchart of the final method is given in Fig. 4.

With this optimized method, MDLs ranging from 0.016 to 12.8 μg g^{-1} TOC were achieved for 185 compounds. This range is comparable with the ranges in previous studies on screening of organic pollutants in sediments [23, 24, 43]. MDL data are provided in Fig. S3.

Analysis of river sediment samples

The performance of the method was demonstrated by our performing screening for 310 compounds in sediment samples collected from the river Holtemme and its tributaries. In total, 69 compounds were detected in a range of concentrations between 1.36 and 1327 μg g^{-1} TOC (Table 2, Fig. S5). Method-matched calibration curves for quantified compounds

had R^2 between 0.91 and 0.998 for the neutral fraction (mean $R^2 = 0.99 \pm 0.02$) and 0.82 and 0.99 for the acidic plus basic fractions (mean $R^2 = 0.93 \pm 0.03$). Twenty-eight false positives were detected in the processed blanks along with the samples and were excluded from the results. Details on quantification and quality control are given in Tables S13 and S14.

Distribution of compounds across extracts

The recoveries of 30 compounds were increased by the additional acidic and basic extraction steps in Holtemme sediments. Sixteen compounds that were extracted from spiked sediments with EtAc were completely or partly detected in the acidic and basic extracts of the native sediments (e.g., diphenhydramine, bisoprolol, and propranolol). It is common practice to spike sediments or soils with the target compounds to evaluate the extraction efficiency [23, 24, 43]. Thus, our results suggest that the recovery of analytes with different PLE solvents may be different for spiked and native sediment contaminants. The differences may be related to the mode of compound introduction, the contact time, and maybe the

Table 1 Effect of sample matrix (given as the concentration of TOC in the vial) on recoveries of pharmaceuticals and pesticides in spiked neutral and basic/acidic extracts mimicking their dilution of organic carbon (n = 2)

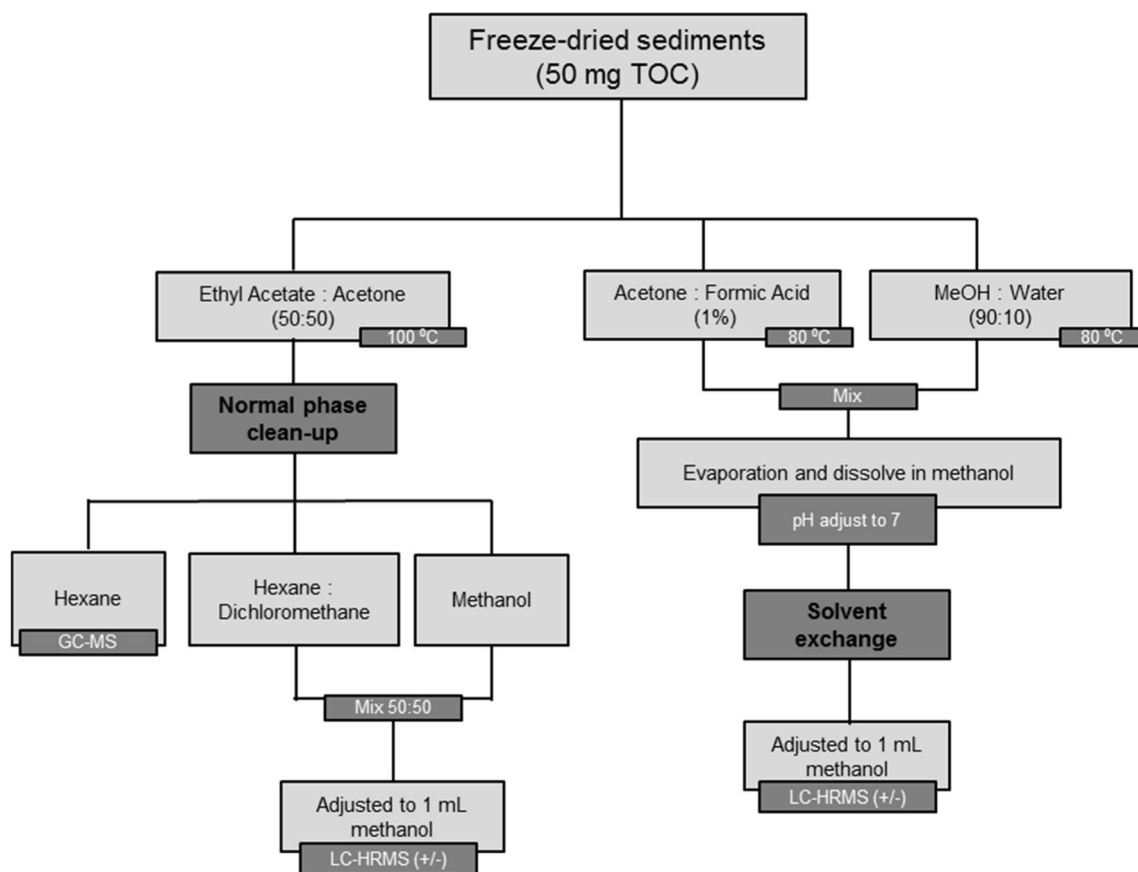| TOC (mg mL^{-1})	Compound (ng mL^{-1})	Average recovery ± standard deviation (%)	
		Pesticides (*n* = 38)	Pharmaceuticals (*n* = 51)
20	40	72 ± 2	73 ± 3
50	100	61 ± 2	63 ± 5
100	200	54 ± 2	59 ± 6
200	500	44 ± 4	53 ± 10
500	1000	35 ± 1	39 ± 4

Fig. 4 The whole extraction and cleanup procedure. GC-MS gas chromatography–mass spectrometry, LC-HRMS liquid chromatography–high-resolution mass spectrometry, TOC total organic carbon

composition of organic and mineral matter. It has been demonstrated that "aging" of chemicals (i.e., migration of a compound from a more accessible site to less accessible sites) has an influence on the extractability, which depends on the contact time and physicochemical properties of the compound and the sorbent [44]. As consequence, extraction efficiency results obtained during method development with spiked sediments should be carefully checked on field-contaminated sediments.

An overview of the distribution of the compounds detected among the different PLE extracts is shown in Fig. S6. Several cationic pharmaceuticals and other compounds could not be extracted by the first step with EtAC, but could be extracted with the protic extraction solvents. Nineteen anionic and cationic compounds were detected mainly in the acidic/basic extracts. Among the cationic compounds, three beta blockers (metoprolol, propranolol, and pindolol) and melperone (at high concentrations up to 1371 ng g^{-1} TOC) were found in sediments despite their low log D values (-0.9 to 0.02). This shows that even relatively hydrophilic compounds may significantly accumulate in sediments because of electrostatic interactions with the negatively charged surfaces of organic matter or clay minerals.

Compounds occurring in the river Holtemme sediments

As expected, we mainly detected compounds that are moderately lipophilic and fairly persistent in the environment. Our results are in accordance with those of a multicompartment study recently performed in the Holtemme area by Inostroza et al. [45]. They prepared a neutral extract with PLE cleaned up with NP chromatography in agreement with the present study. Fungicides were the most common pesticide class detected with altogether 12 compounds, with propiconazole, spiroxamine, dodemorph, and fenpropimorph being the most prominent in terms of concentration and frequency (Table 2). Propiconazole is often detected in environmental samples, and its relative high log K_{ow} translates into a high affinity for organic matter [46]. It is one of the most widely used fungicides in modern agriculture, and it was found at a concentration of 35–107 ng g^{-1} TOC at two sites. Other azole fungicides detected included prothioconazole-desthio (active transformation product of prothioconazole), epoxiconazole, and tebuconazole. The presence of spiroxamine in sediment samples is not surprising as it was already documented in several studies [47, 48] and it

Table 2 Target compounds detected in sediments from the Holtemme catchment

Compound	Site 14 (Silstedter Bach)	Site 36 (Weir Gross Quenstedt	Site 41 (Salzgraben)	Compound class
N-Acetyl-4-aminoantipyrine		35		Pharmaceutical TP
7-Amino-4-methylcoumarin	68	195	223	Industrial
Amiodarone		33		Pharmaceutical
Bendiocarb		32	76	Insecticide
Benzocaine	60	44	104	Pharmaceutical
Bifonazole		45	7.6	Pharmaceutical
Bisoprolol		63	74	Pharmaceutical
Boscalid			8.7	Fungicide
N-Butylbenzenesulfonamide		802		Industrial
Carbamazepine	20	146	122	Pharmaceutical
Carbendazim		7.2	38	Biocide/fungicide
N-Cyclohexyl-2-benzothiazolamine	66	10		Industrial
Crotamiton	31		7.2	Pharmaceutical
DEET	45		41	Insect repellent
Diazinon		150		Insecticide
7-Diethylamino-4-methylcoumarin	19	1085	1327	Industrial
Diphenhydramine	59			Pharmaceutical
Diphenyl phosphate		226	100	Flame retardant TP
Diuron	11	7.6		Herbicide
Dodemorph	20		323	Fungicide
Epoxiconazole	16	5.6		Fungicide
N-Ethyl-o-toluenesulfonamide	459	99	825	Industrial
Fenpropimorph	68	16	75	Fungicide
Fluconazole	175		46	Pharmaceutical
Flurtamone	17			Herbicide
N-Formyl-4-aminoantipyrine		47	44	Pharmaceutical TP
Hexa(methoxymethyl)melamine	18	9.2		Industrial
Hexazinone	16		2.2	Herbicide
2-Hydroxyatrazine			5.8	Herbicide TP
2-Hydroxycarbamazepine	21	4.1	8.7	Pharmaceutical TP
Imazalil	65			Fungicide
Irgarol	10	14		Herbicide
Ketoprofen	77			Pharmaceutical
Lidocaine			1.4	Pharmaceutical
Mebendazole	97			Pharmaceutical
Melperone	83	1371		Pharmaceutical
Memantine	45	4		Pharmaceutical
Metalaxyl	15			Fungicide
Metazachlor	8.5			Herbicide
4-Methyl-1H-benzotriazole and 5-methyl-1H-benzotriazole	278			Corrosion inhibitor
Metoprolol	172	798	499	Pharmaceutical
2(4-Morpholinyl)benzothiazole	39			Industrial
2-Octyl-4-isothiazolin-3-one	25	26		Biocide
Ondansetron	121		12	Pharmaceutical

Table 2 (continued)

Compound	Site 14 (Silstedter Bach)	Site 36 (Weir Gross Quenstedt	Site 41 (Salzgraben)	Compound class
Oxybutynin	18			Pharmaceutical
Pirimicarb	44			Insecticide
Propiconazole		35	107	Fungicide
Propranolol	59	79	127	Pharmaceutical
Propyphenazone	31			Pharmaceutical
Prothioconazole-desthio			5.7	Fungicide TP
Risperidone	237		14	Pharmaceutical
Spiroxamine	182	27	19	Fungicide
Sulfapyridine			124	Pharmaceutical
Tebuconazole			32	Fungicide
Terbinafine		9.6		Pharmaceutical
Terbuthylazine		12		Herbicide
Terbuthylazine-2-hydroxy	77	32	20	Herbicide TP
Terbutryn	19	90	52	Biocide/herbicide
Tetracaine	64			Pharmaceutical
Tetraglyme	253		129	Industrial
Thiabendazole	88		55	Biocide/fungicide
Thiacloprid	8			Insecticide
Triethyl phosphate	505	812	133	Flame retardant
Triisobutyl phosphate	90	240	18	Flame retardant
Tris(2-chloroethyl) phosphate	5.9			Flame retardant
Verapamil		344	294	Pharmaceutical

Concentrations are expressed in nanograms per gram of total organic carbon.

TP transformation product

was detected at all the sampling locations at concentrations between 19 and 182 ng g^{-1} TOC. The morpholine fungicides dodemorph and fenpropimorph were detected at two and three sites, respectively (up to 323 and 75 ng g^{-1} TOC, respectively). Also several fungicides approved only as biocides were found, namely, carbendazim and the preservatives imazalil and thiabendazole. Herbicides and transformation products (altogether eight compounds) were less frequently detected, and their concentrations were in general below 20 ng g^{-1} TOC, except for the terbutryn, used as a biocide, and terbuthylazine-2-hydroxy. Only four insecticides—diazinon, bendiocarb, thiacloprid, and pirimicarb—were found, with the legacy compound bendiocarb and diazinon (used in Germany only in veterinary treatment) showing the highest concentrations.

Many pharmaceuticals were detected, among them well-known water contaminants such as carbamazepine and its 2-hydroxy transformation product (all sites, up to 146 and 21 ng g^{-1} TOC, respectively), the beta blockers bisoprolol, metoprolol, and propranolol, 4-methylbenzotriazole and 5-methylbenzotriazole (one site), and DEET (two sites). The presence of different wastewater-derived compounds in the tributaries

Silstedter Bach (site 14) and Salzgraben (site 41) indicates direct wastewater inputs or the deposition of contaminated particulate matter from the river Holtemme during flood events. None of these streams are connected to any WWTPs, but both sampling spots are located within the main floodplain of the river Holtemme.

Conclusions

Our study demonstrates that wide-scope screening of sediments for polar and ionic compounds can be achieved if the extraction uses acidic and basic solutions in addition to neutral, nonpolar ones. The sequential use of different solvents for PLE and their separate collection allows the use of a cleanup step with NP chromatography for the hydrophobic sediment contaminants without losses of cationic and anionic compounds. By adjustment of the final extract to a defined quantity of organic carbon before injection, problems with ion source contamination and matrix effects during LC–HRMS analysis can be reduced. Overall, good recoveries and acceptable MDLs were achieved for many compounds. However, some compounds have low recoveries within such a broad

screening approach and would require more specific targeted extraction and cleanup methods. Using the river Holtemme as a case study, we demonstrated the good performance of the method and confirmed the presence of a large number of polar and ionic chemicals in river sediments, including many pesticides from agricultural and urban use.

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