DETERMINATION OF ORGANIC PRIORITY POLLUTANTS AND EMERGING COMPOUNDS IN WASTEWATER BY MICRO SOLID PHASE EXTRACTION COUPLED TO GCMS

Prieto A[1], Schrader S[1], Moeder M[1], Lahoutifard N[2]

[1]Helmholtz-Center for Environmental Research UFZ, Department of Analytical Chemistry, Permoserstr. 15, D-04318 Leipzig; [2]SGE Europe, 1 Potters Lane, Kiln Farm, Milton Keynes, MK11 3LA United Kingdom

Introduction

A new fully automated procedure for the simultaneous determination of 41 multi-class priority and emerging organic pollutants in water samples is presented which combines micro solid phase extraction (MEPS) with large volume injection–GC-MS. Priority hazardous substances are recognized on the basis of their toxic effects, persistence, accumulation potential, and widespread in environment. In compliance with national and international directives, regular environmental monitoring is demanded which requires appropriate analytical methods for fast and sensitive detection of relevant compounds such as the priority pollutants polychlorinated biphenyls (PCBs), polycyclic aromatic hydrocarbons (PAHs) or endocrine disrupting compounds. Generally, automated multi-residue methods including analyte enrichment are preferred allowing high sample throughput at low labour effort. Miniaturization, particularly of included sample preparation, is a key feature in the development of automated analytical protocols and in this context, MEPS is emerging as a novel and promising tool. The list of target compounds included PAHs, PCBs, phthalate esters (PEs), nonylphenols (NPs), bisphenol A (BPA) and selected steroid hormones. The performance of the new at-line microextraction-LVI–GC-MS protocol was compared to standard solid-phase extraction (SPE) and LVI–GC-MS analysis. The developed methods were applied to the determination of the target analytes in snow and wastewater samples.

Materials and methods

MEPS Procedure

The microextraction was carried out with a MEPS device delivered by SGE Europe (Milton Keynes, United Kingdom). The 100 µL gas-tight syringe is equipped with a small container incorporated into the needle (see Fig 1). The MEPS syringe was used in connection with a large volume injector type KAS 4 (Gerstel, Mühlheim an der Ruhr, Germany) and the samples were fully automatically processed by a Multi Purpose Sampler MPS 2 (Gerstel) and controlled by the Maestro software of Gerstel.

Figure 1. The MEPS syringe and SPE cartridge or BIN "Barrel Insert and Needle" filled with 2 mg of sorbent commonly used for SPE.

Prior to each sample extraction, the MEPS-BIN was conditioned using ten 100 µL portions of hexane/ethyl acetate (50:50 v/v) mixture and three 100 µL portions of both MeOH and bidistilled water. All portions were discarded into the waste vials. The extraction was realized in 100 µL aspiring steps at a speed of 10 µL s^{-1}. After sample extraction the BIN was dried by 5 cycles of drawing and pressing air through the sorbent at a rate of 10 µL s^{-1}. Subsequently, two portions, first of them of 50 µL and the second one of 25 µL of ethyl acetate/hexane mixture (50:50, v/v) were drawn through the BIN and each portion injected at 2.5 µL s−1 of injection speed directly into the large volume injector of the GC-MS instrument. After the extraction/elution process, ten wash cycles, each with 100 µL of elution solvent mixture, were used to clean the sorbent in order to avoid carryover effect.

Standard SPE was performed with a Visiprep SPE manifold (Supelco, Bellefonte, PA, USA) using 200 mg of C-18 sorbent (polar plus® C-18 bonded phase from J.T. Baker, Phillipsburg, NJ, USA) in 2 ml cartridges.

Large volume injection and GC-MS analysis
Instrument: GC-MSD instrument (Agilent Technologies, San José, CA, U.S.A.) that consists of an Agilent 6890 series gas chromatograph equipped with a programmed temperature vaporizer (PTV) injector (KAS 4, Gerstel) and an Agilent 5973 Mass Selective Detector. The PTV was operated in solvent vent mode and used an empty baffled deactivated liner. During injection in split mode at a rate of 2.5 µL s^{-1} the PTV was set at 50 °C (inlet temperature) and at 87.6 kPa (vent pressure). The solvent mixture ethyl acetate/hexane (50:50, v/v) was purged out with a vent flow of 70 mLmin^{-1} within 0.7 min (vent time), then, splitless mode was programmed for 1.5 min while the temperature increased at 720 K min^{-1} to 300 °C and held to 300 °C during 5 min.
Capillary: HP-5MS (30 m×0.25 mm, 0.25 µm, Agilent Technologies), Oven temperature program:50 °C for 2 min with 15 K min^{-1} to 100 °C with 10 K min^{-1} to 290 °C for 15 min. The temperatures of the transfer line, ion source and quadrupole analyser were 300, 230 and 150 °C respectively.
Carrier gas: helium at constant flow conditions of 1.5 mLmin^{-1}. Mass analysis after electron impact ionization (70eV) used selected ion monitoring (SIM) mode.

Results and discussion:
Optimization of the SPE methodology
Sample fill and injection speed, elution/injection volume, drying step and carry over effects were all evaluated as these have been found as critical steps for the MEPS analysis[1]. A C-18 BIN applied together with the hexane:ethyl acetate as elution mixture was the basis for optimizing the MEPS procedure. Several elution volumes (25–100µL) injected in one or two portions of 25 or 50µL were tested (see Fig. 2). 75µL (1×25µL+1×50µL) was chosen not only as consensus elution volume but also in order to avoid peak splitting observed for the most volatile compounds when total volumes higher than 75µL were injected consecutively (2×50µL) into the PTV.

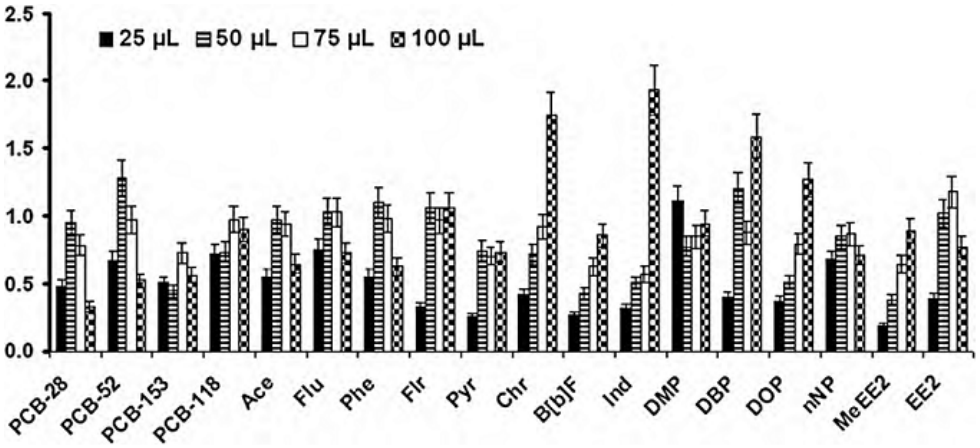

Figure 2. Comparison of several elution/injection volumes (25–100 µL) for selected analytes extracted with MEPS

The number of extraction steps (2, 4, 6, 8 and 10×100µL) was also evaluated (see Fig. 3). A reduction in the response of most of the compounds was obtained when ten 100µL portions of sample were loaded on the MEPS BIN. Thus, eight 100µL portions of sample were finally chosen as optimum. In order to optimize the way of sample loading, a multiple draw/eject cycle mode was tested compared to an extract-discard mode where each sample aliquot was pumped only once through the MEPS-BIN and the sample portion extracted was discarded into waste before the next aliquot from the sample was pumped. This procedure was applied to the extraction of 800µL of sample with an analyte concentration of 2.5 ngmL^{-1} (see Fig. 4.). The responses obtained with the extract-discard mode were similar or even higher than obtained by the multiple draw-eject procedure. Therefore, this procedure was selected for further experiments.
Re-using an SPE device is not often recommended due to both the cost of washing and the risk of carry-over. However, the miniature format of MEPS makes washing a feasible and cost-effective solution as long as carry over is eliminated. In order to evaluate possible carryover problems two, four, six, eight and ten wash-discard

cycles each with 100 µL of the elution solvents mixture were passed through the syringe after the extraction and elution steps of the target analytes. The results obtained using the different five protocols were compared. The carryover effect checked after the washing procedure using 10 portions of 100 µL of elution solvents mixture was reduced to 0.002–3.9% of the initial extracted analyte amounts. In the case of the two, four, six and eight wash-discard cycles, higher carryover values (>10%) were obtained for some analytes. Thus, ten wash-discard cycles each with 100 µL of elution solvents mixture was selected as optimum washing protocol.

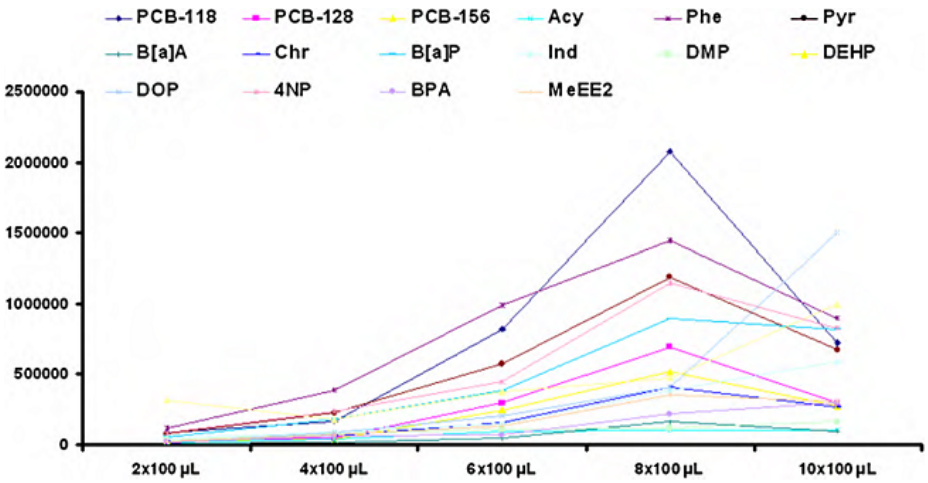

Figure 3. Influence of the number of extraction steps on the responses of some analytes using MEPS

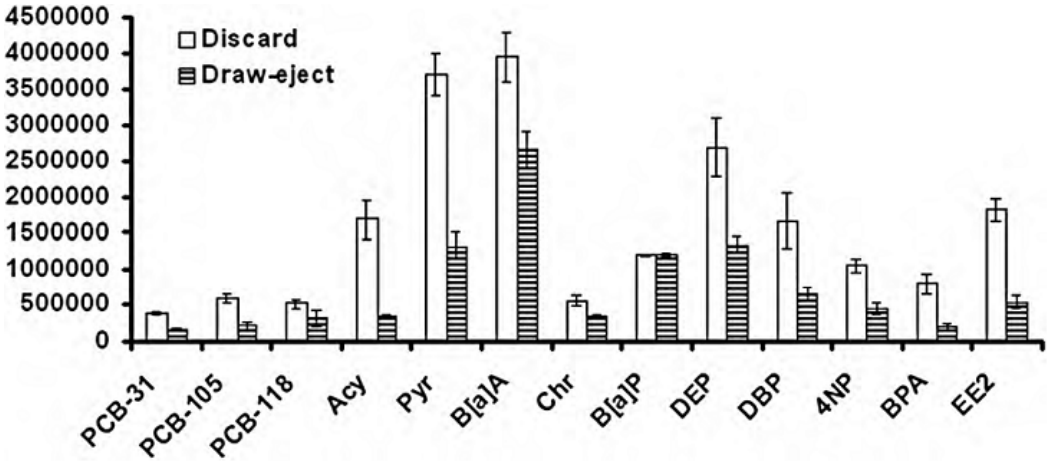

Figure 4. Evaluation of the way of sample loading (multiple draw-eject cycle mode vs. extract-discard mode) in the case of some selected compounds shown with their chromatographic responses.

Performance of the Optimised Method.

The developed MEPS-LVI-GC-MS and SPE-LVI-GC-MS chromatographic procedures exhibited excellent linearity (R2 > 0.99) for the majority of compounds (PAHs, PCBs, phthalate esters and nonylphenols[1]). The results demonstrated the high sensitivity of the MEPS procedure in comparison to the commonly applied SPE methodology. Furthermore, MEPS allows the extraction of only 800µL of sample volume to detect the target compounds at ng L^{-1} concentration level.

The optimized and validated methodologies were applied to real samples influent and effluent samples of a wastewater treatment plant at Leipzig (Germany) and snow samples taken at Leipzig in February 2010. The average analyte concentrations determined in snow by both MEPS and SPE protocols were determined as well as the results of the MEPS analyses of the wastewater samples with their corresponding uncertainties in Table 1.

Table 1: Average concentrations (ng L^{-1}) (n=4) and standard deviations determined by MEP-LVI-GC-MS in three different wastewater samples

Analyte	Inffluent	Effluent 1	Effluent 2
Acy	947±160	n.d	23±1
Ace	688±37	396±19	1898±179
Flu	25±1	< LOQ	31±2
Ant	n.d	n.d	n.d
B[a]A	27.2±0.3	23.1±0.4	n.d
Chr	45±1	43±1	n.d
B[b]F	58±2	26.0±0.4	n.d
B[k]F	57.4±0.2	55.2±2	n.d
B[a]P	44.3±0.4	44±1	n.d
Ind	78±3	63±2	n.d
DMP	1433±145	315±8	5435±221
DEP	9973±303	1773±48	3505±236
BBP	154±11	132±6	158±11
DEHP	4752±360	985±48	1172±30
DOP	166±20	153±12	131±22
n-NP	330±12	194±11	167±10
4-NP	40±3	49±2	52±3
MeEE2	57±4	52±5	56±4
EE2	751±16	< LOQ	151±12

Conclusions

- accurate multi-residue determination of 41 organic pollutants in water at low levels (ng L^{-1}) for sample volume of 800 µL
- use of isotopically labelled standards avoided standard addition even for wastewater samples
- fully automated multi-residue protocol saves time and solvent as compared to standard SPE methodology.
- in opposite to normal SPE cartridges the MEPS materials allow multiple use (life time depends on sample matrix and analytes)
- LODs for the MEPSs protocol: 0.2 and 266 ng L^{-1} (800 µL sample) comparable to those obtained by off line SPE (0.2 to 736 ng L^{-1} for 100 mL sample volume)
- recoveries (>75%) and precision of the methods (RSD) was below 21% for all compounds

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References:

1. Prieto A, Schrader S, Moeder M. (2010); *J Chrom A*, 1217: 6002–6011