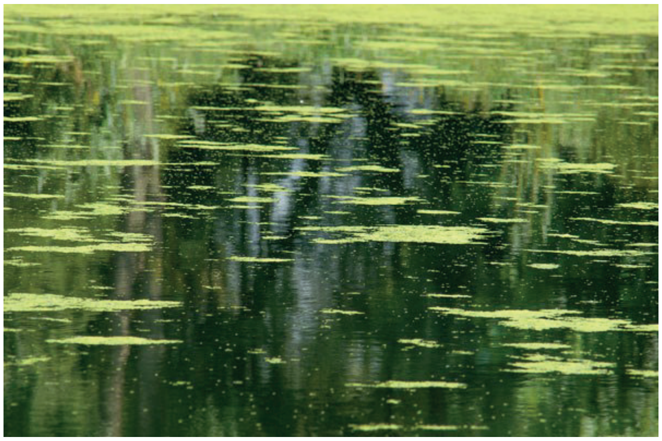
APPLICATION NOTE

Automated Solid Phase Extraction of PAH Compounds Utilizing the SPE-DEX 5000



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Key Words

PAH, polycyclic aromatic hydrocarbons, EPA 8270, water extraction, SPE, SPE disks

Introduction

Polycyclic aromatic hydrocarbons (PAHs) can be found in a number of water types, such as drinking water, surface water, ground water and wastewater. There are serval types of extraction techniques that can be implemented to remove them from water for measurement including liquid-liquid extraction (LLE), continuous liquid-liquid extraction (CLLE) and solid phase extraction (SPE). With LLE and CLLE there is little selectivity within the methods for difficult matrices or other compound interferences. Complex matrices can add time to the extraction process due to the creation of emulsions, which may require additional effort to break. Solid phase extraction avoids the formation of emulsions, making extraction of a wide variety of sample matrices more predictable. In addition, SPE is more customizable through selection of a disk sorbent to target extraction to the range of analytes desired. Less solvent is used with SPE, minimizing the evaporation time and reducing the solvent potentially released into the atmosphere.

Disk SPE is more suitable for larger volumes of water, such as the one-liter sample specified in many environmental methods. In addition, a disk essential when the sample contains particulates. Particulates can clog cartridge SPE quickly, but the ability to use a larger surface area and add prefilters maintains the flow through the disk and allows the particulates to be extracted with the solvent. This provides a faster preparation and includes the whole water sample in the extraction, an important component in an accurate analysis. This work describes the use of the SPE-DEX® 5000, a system for automation of disk SPE. Three different disk media types will be evaluated to better understand the performance of each for a wide range of PAH compounds. The media evaluated includes C18 high-capacity, DVB and HLB-M.

Shown: SPE-DEX 5000 Automated Disk Extraction System

C18 is a common, inexpensive sorbent

- Octadecyl bonded silica
- Good in moderate pH ranges (2-7)
- Effective for many compound classes and cost effective
- Must keep the disk wet during all precondition or "prewet" steps in order to effectively retain compounds onto the disk

Divinylbenzene sorbent is polymer-based

- Lipophilic compound
- Very effective for many compounds
- Can go completely dry during prewets and still be as effective without sacrificing the retention of compounds

Hydrophillic-Lipophillic Balanced (HLB) reversed-phase polymeric sorbent

- Combination of n-vinylpyrrolidone and divinylbenzene
- Good for a wide range of pH levels
- 3x the retention capacity of traditional silica-based sorbents
- Can go completely dry during prewets and still be as effective without sacrificing the retention of compounds

The performance of the system will be demonstrated by comparing three sets of three laboratory control samples (LCS) spiked at mid-level with three different Atlantic® SPE disk medias. In addition to the LCS study a matrix spike (MS) and matrix spike duplicate (MSD) study was carried in the same manner to demonstrate the effect of a matrix. The MS and MSD will be done using the Fast Flow Disk Holder. The Fast Flow disk holder was used with the pond water samples since sand and debris were observed in the samples. The Fast Flow disk holder uses the same SPE disk, but allows prefilters to be added above the disk to prevent disk clogging and retain the particulate material so that it can be included in the extraction.

Experimental

The instrumentation and consumables used in this work are listed next.

- Horizon Technology, Inc.
  - SPE-DEX® 5000 Automated Disk Extractor
  - DryVap® Concentrator System
  - DryDisk® Separation Membranes
  - Atlantic C18 HC Disk, 47 mm
  - Atlantic DVB Disk, 47 mm
  - Atlantic HLB-M Disk, 47 mm
  - 47 mm disk holder
  - Fast Flow disk holder (FFDH)
  - 1 micron prefilter (used with FFDH)
  - 5 micron prefilter (used with FFDH)

- Agilent Technologies
  - 6890 GC
  - 5975C inert MSD
  - 7683 Autosampler

- Phenomenex®
  - Column: ZB SemiVolatiles, 30 m X 0.25 mm, 0.25 µm

The method summary is shown next and outlines the steps taken for the preparation of spiked reagent water and spiked pond water.

Reagent Water Analysis

1. Obtain nine 1-liter samples of reagent water.
2. Acidify each 1-liter water sample to pH <2 using concentrated HCl.
3. Spike PAH compounds into samples at 50 µg/L.
4. Setup the SPE-DEX 5000 extractor with 47 mm disk holders. (3 samples will be run with each of the following Atlantic SPE disks: C18 HC, DVB, and HLB-M)
5. Start extraction method shown in Table 1 for Atlantic C18 HC disks or extraction method shown in Table 2 for Atlantic DVB/HLB-M disks and collect extract (≈ 55 mL).
6. Add extract to the DryDisk holder and start automated drying and concentration process on the DryVap system (the DryVap system automatically dries and concentrates extract to 0.9 mL). Note: Vacuum pump setting for the DryVap is –8 psi, and Nitrogen pressure setting is 20 psi.
7. Quantitatively bring extract volume to 1.0 mL using methylene chloride
8. Add internal standard into the 1-milliliter extract.
9. Transfer the extract to a 2.0 mL GC vial.
10. Analyze by GC/MS.

Pond Water Analysis

1. Obtain nine 1-liter samples of surface water.
2. Acidify each 1-liter water sample to pH <2 using concentrated HCl.
3. Spike PAH compounds into nine of the samples at 50 µg/L.
4. Setup the SPE-DEX 5000 extractor with Fast Flow disk holders. (3 spike samples and 1 blank sample will be run with each of the following 47 mm Atlantic SPE disks: C18 HC, DVB, HLB-M)
5. Start extraction method shown in Table 3 for Atlantic C18 HC disks or extraction method shown in Table 4 from Atlantic DVB/HLB-M disks and collect extract (≈100 mL).
6. Add extract to the DryDisk holder and start automated drying and concentration process on the DryVap system (the DryVap system automatically dries and concentrates extract to 0.9 mL). Note: Vacuum pump setting for the DryVap is –8 psi, and Nitrogen pressure setting is 20 psi.
7. Quantitatively bring extract volume to 1.0 mL using methylene chloride.
8. Add internal standard into the 1-milliliter extract.
9. Transfer the extract to a 2.0 mL GC vial.
10. Analyze by GC/MS.

Table 1. 47 mm Disk Holder Extraction Method for Atlantic C18 HC Disks

Step	Solvent	Solvent Vol. (mL)	Purge Time (s)	Pump Rate (#)	Sat. Time (s)	Soak Time (s)	Drain Time (s)
Condition SPE Disk	$MeCl_2$	11	60	2	1	30	30
Condition SPE Disk	Acetone	11	60	2	1	30	30
Condition SPE Disk	Methanol	11	60	2	1	60	2
Condition SPE Disk	Reagent Water	9	30	2	1	30	5
Condition SPE Disk	Reagent Water	9	60	2	1	30	0

Step	Sample Flow Rate (#)	Done Loading Sample Delay (s)
Load Sample	2	45

Step	Dry Time (s)	Pump Rate (#)	N_2 Blanket
Air Dry Disk Timer	30	6	Off

Step	Solvent	Solvent Vol. (mL)	Purge Time (s)	Pump Rate (#)	N_2 Blan-ket	Sat. Time (s)	Soak Time (s)	Elute Time (s)
Elute Sample Container	Acetone	11	60	2	Off	1	180	45
Elute Sample Container	$MeCl_2$	11	15	2	Off	1	180	45
Elute Sample Container	$MeCl_2$	11	15	2	Off	1	60	45
Elute Sample Container	$MeCl_2$	11	15	2	Off	1	60	45
Elute Sample Container	$MeCl_2$	11	60	6	Off	1	60	60

Table 2. 47 mm Disk Holder Extraction Method for Atlantic DVB and HLB-M Disks

Step	Solvent	Solvent Vol. (mL)	Purge Time (s)	Pump Rate (#)	Sat. Time (s)	Soak Time (s)	Drain Time (s)
Condition SPE Disk	$MeCl_2$	11	60	2	1	30	30
Condition SPE Disk	Acetone	11	60	2	1	30	30
Condition SPE Disk	Reagent Water	11	60	2	1	30	30

Step	Sample Flow Rate (#)	Done Loading Sample Delay (s)
Load Sample	2	45

Step	Dry Time (s)	Pump Rate (#)	N_2 Blanket
Air Dry Disk Timer	30	6	Off

Step	Solvent	Solvent Vol. (mL)	Purge Time (s)	Pump Rate (#)	N_2 Blan-ket	Sat. Time (s)	Soak Time (s)	Elute Time (s)
Elute Sample Container	Acetone	11	60	2	Off	1	180	45
Elute Sample Container	$MeCl_2$	11	15	2	Off	1	180	45
Elute Sample Container	$MeCl_2$	11	15	2	Off	1	60	45
Elute Sample Container	$MeCl_2$	11	15	2	Off	1	60	45
Elute Sample Container	$MeCl_2$	11	60	6	Off	1	60	60

Table 3. Fast Flow Disk Holder Extraction Method for Atlantic C18 HC Disks

Step	Solvent	Solvent Vol. (mL)	Purge Time (s)	Pump Rate (#)	Sat. Time (s)	Soak Time (s)	Drain Time (s)
Condition SPE Disk	$MeCl_2$	20	60	2	2	60	60
Condition SPE Disk	Acetone	20	60	2	2	60	60
Condition SPE Disk	Methanol	20	60	2	2	120	2
Condition SPE Disk	Reagent Water	18	30	2	2	60	5
Condition SPE Disk	Reagent Water	18	60	2	2	60	0

Step	Sample Flow Rate (#)	Done Loading Sample Delay (s)
Load Sample	2	45

Step	Dry Time (s)	Pump Rate (#)	N_2 Blanket
Air Dry Disk Timer	30	6	Off

Step	Solvent	Solvent Vol. (mL)	Purge Time (s)	Pump Rate (#)	N_2 Blanket	Sat. Time (s)	Soak Time (s)	Elute Time (s)
Elute Sample Container	Acetone	20	60	2	Off	2	180	180
Elute Sample Container	$MeCl_2$	20	15	2	Off	2	180	180
Elute Sample Container	$MeCl_2$	20	15	2	Off	2	120	120
Elute Sample Container	$MeCl_2$	20	15	2	Off	2	120	120
Elute Sample Container	$MeCl_2$	20	60	6	Off	2	120	180

Table 4. Fast Flow Disk Holder Extraction Method for Atlantic DVB and HLB-M Disks

Step	Solvent	Solvent Vol. (mL)	Purge Time (s)	Pump Rate (#)	Sat. Time (s)	Soak Time (s)	Drain Time (s)
Condition SPE Disk	$MeCl_2$	20	60	2	2	60	60
Condition SPE Disk	Acetone	20	60	2	2	60	60
Condition SPE Disk	Reagent Water	20	60	2	2	60	60

Step	Sample Flow Rate (#)	Done Loading Sample Delay (s)
Load Sample	2	45

Step	Dry Time (s)	Pump Rate (#)	N_2 Blanket
Air Dry Disk Timer	30	6	Off

Step	Solvent	Solvent Vol. (mL)	Purge Time (s)	Pump Rate (#)	N_2 Blanket	Sat. Time (s)	Soak Time (s)	Elute Time (s)
Elute Sample Container	Acetone	20	60	2	Off	2	180	180
Elute Sample Container	$MeCl_2$	20	15	2	Off	2	180	180
Elute Sample Container	$MeCl_2$	20	15	2	Off	2	120	120
Elute Sample Container	$MeCl_2$	20	15	2	Off	2	120	120
Elute Sample Container	$MeCl_2$	20	60	6	Off	2	120	180

The GC/MS conditions used for analysis are shown next.

Flow rate	9 psig helium which is ramped up with the oven temp to maintain a constant flow		
Temperature Ramp	Temp (°C)	Rate (°C/min	Hold (min)
	45	0	1.00
	270	15	0.00
	318	6.0	0.00
Total run time	24.00 min		
Injection Method	Spit, Ratio 1:10, 1.0µL injected		
	Temp (°C)	Rate (°C/min)	Hold (min)
	280	0	0.00

Results

Fifteen spiked water samples were extracted following the procedure in the method summary in this note, 9 using reagent water and 6 using pond surface water. The spiked aqueous samples had PAH compounds added at a concentration of 50 µg/L. Three of the pond water samples (one for each disk type) were run as blanks to show if any existing native PAH content was present. All of the pond water used in this study was collected at the same time from the same source.

The percent recovery of spikes are shown for the three different disk types in Table 5. The recoveries average 83.4% recovery or better for each run. The lowest compound average recovery is 69.4% for naphthalene on the C18 HC disk. The standard deviation of the three replicate preparations on each disk is excellent with HLB-M showing the best precision.

Table 5. Percent Recovery of Reagent Water Samples with 50 µg/L of PAH spike for Three Different Disk Media

	C18 HC 1	C18 HC 2	C18 HC 3	SD	DVB 1	DVB 2	DVB 3	SD	HLB-M 1	HLB-M 2	HLB-M 3	SD
Naphthalene	74.0	67.9	66.3	4.04	69.3	69.7	71.7	1.26	77.0	72.2	71.5	3.02
Acenaphthylene	89.6	85.9	85.5	2.24	93.7	92.2	88.1	2.90	92.3	88.8	88.5	2.10
Acenaphthene	87.2	83.5	82.7	2.41	90.6	88.8	84.8	2.95	89.0	87.2	86.1	1.47
Fluorene	95.2	90.7	90.7	2.59	99.8	96.8	92.4	3.72	98.6	94.5	93.1	2.84
Phenanthrene	93.2	89.5	89.0	2.30	99.3	95.3	91.5	3.90	97.9	93.3	92.3	2.96
Anthracene	93.7	90.7	89.9	2.03	101	96.1	92.8	3.99	98.2	95.0	94.0	2.20
Fluoranthene	97.3	93.6	91.9	2.79	104	98.8	95.9	4.26	101	98.6	95.6	2.55
Pyrene	96.5	93.5	92.7	1.96	104	98.6	96.5	3.68	101	100	95.7	2.88
Benz(a)anthracene	96.8	92.8	91.2	2.87	108	101	98.9	4.85	104	102	98.4	2.86
Chrysene	91.2	86.6	84.1	3.58	98.4	93.9	91.0	3.74	95.5	93.1	89.8	2.82

	C18 HC 1	C18 HC 2	C18 HC 3	SD	DVB 1	DVB 2	DVB 3	SD	HLB-M 1	HLB-M 2	HLB-M 3	SD
Benzo(b)fluoranthene	101	95.0	93.4	3.77	112	104	102	5.23	106	103	99.9	3.08
Benzo(k)fluoranthene	95.9	91.4	89.4	3.32	107	99.6	96.3	5.29	102	97.0	96.1	3.29
Benzo(a)pyrene	101	95.9	94.0	3.40	112	105	101	5.41	107	103	102	2.59
Indeno(1,2,3-cd)pyrene	102	99.0	96.0	3.08	116	107	104	6.19	110	106	104	3.20
Dibenz(ah)anthracene	101	96.8	93.3	3.79	111	103	99.5	5.72	107	101	99.9	3.91
Benzo(ghi)perylene	99.8	95.6	93.2	3.34	110	103	99.5	5.33	106	100	100	3.13

The percent recovery of spikes in pond water are shown in Table 6. In this case, the relative percent difference (RPD) between the spike and spike duplicate is shown, as generally required in a method such as US EPA 8270.[1,2] The RPD is slightly better for the HLB-M media, perhaps indicating that HLB is a better match with this suite of analytes and pond water matrix.

Table 6. Percent Recovery of Pond Water Samples with 50 µg/ of PAH spike for Three Different Media

	C18 HC MS	C18 HC MSD	RPD (%)	DVB MS	DVB MSD	RPD (%)	HLB-M MS	HLB-M MSD	RPD (%)
Naphthalene	72.2	76.5	5.86	79.7	73.4	8.16	75.4	75.7	0.36
Acenaphthylene	88.4	92.7	4.81	99.3	95.0	4.49	91.2	92.1	0.99
Acenaphthene	85.4	88.5	3.56	91.8	88.5	3.68	87.4	86.7	0.76
Fluorene	90.8	94.1	3.58	101	97.1	3.67	93.0	92.4	0.63
Phenanthrene	89.8	92.4	2.91	99.0	94.6	4.60	92.7	93.3	0.58
Anthracene	90.6	92.1	1.70	98.5	93.2	5.58	92.6	93.2	0.60
Fluoranthene	93.3	95.7	2.60	104	99.1	5.24	95.5	95.5	0.00
Pyrene	93.6	96.1	2.58	104	98.8	4.80	94.9	95.2	0.34
Benz(a)anthracene	95.8	95.6	0.26	106	99.1	6.40	95.7	97.5	1.86
Chrysene	89.6	89.5	0.08	91.7	85.9	6.58	90.3	91.5	1.34
Benzo(b)fluoranthene	97.6	98.7	1.10	106	101	5.13	94.6	97.5	3.03
Benzo(k)fluoranthene	93.0	91.4	1.76	97.7	88.9	9.51	88.6	89.2	0.68
Benzo(a)pyrene	97.0	96.2	0.89	106	98.3	7.92	94.6	95.5	0.94
Indeno(1,2,3-cd)pyrene	99.5	97.2	2.33	110	101	7.91	92.8	92.7	0.03
Dibenz(ah)anthracene	90.5	87.9	2.93	102	85.8	16.8	87.8	88.7	1.05
Benzo(ghi)perylene	92.4	90.0	2.69	102	93.3	9.14	84.1	84.9	0.91

To be sure that native concentrations were not present in the pond water, blanks were measured on each type of disk, shown in Table 7. The measured blanks were quite low, all non-detected under 0.5 µg/L.

Table 7. Pond Water Sample Blanks, Measured with each Disk Type (µg/L)

Disk Type	C18 HC	DVB	HLB-M
Naphthalene	ND	ND	ND
Acenaphthylene	ND	ND	ND
Acenaphthene	ND	ND	ND
Fluorene	ND	ND	ND
Phenanthrene	ND	ND	ND
Anthracene	ND	ND	ND
Fluoranthene	ND	ND	ND
Pyrene	ND	ND	ND
Benz(a)anthracene	ND	ND	ND
Chrysene	ND	ND	ND
Benzo(b)fluoranthene	ND	ND	ND
Benzo(k)fluoranthene	ND	ND	ND
Benzo(a)pyrene	ND	ND	ND
Indeno(1,2,3-cd)pyrene	ND	ND	ND
Dibenz(ah)anthracene	ND	ND	ND
Benzo(ghi)perylene	ND	ND	ND

The three types of Atlantic disks (C18 HC, DVB, and HLB-M) all performed well for the spiked reagent water and pond water. All results were above 80% recovery for PAH compounds with the exception of naphthalene, which was above 70% recovery on the HLB-M disk and close to that on the other two disk types. The precision was good overall and the precision for HLB-M was slightly better for pond water than that of the other two media. Any of the three media choices would serve well for this application.

The Fast Flow disk holder was used with the pond samples because sand and debris was seen in the samples. The time for sample loading was approximately the same as the spiked reagent water. The total time for PAH methods (conditioning, loading and elution) on the SPE-DEX 5000 was 40-45 minutes for the reagent water and 55-60 minutes for the pond water. Using the SPE-DEX 5000 for automation of the process provides better precision between operators and less attention is required for good results.

References:

1. US EPA Method 8270E, available from https://www.epa.gov/sites/production/files/2017-04/documents/method_8260d_update_vi_final_03-13-2017_0.pdf.
2. US EPA Method 8000D, available from https://www.epa.gov/sites/production/files/2015-12/documents/8000d.pdf.

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