

SUPERCRITICAL FLUID EXTRACTION OF ORGANIC COMPOUNDS FROM VARIOUS SOLID MATRICES

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INTRODUCTION

Supercritical fluid extraction (SFE) utilizes compounds such as carbon dioxide at supercritical temperatures and pressures. The supercritical fluid has properties of a liquid (solvating power) and the properties of a gas (low viscosity and high diffusivity).

The analysis of soil and sediment samples are routinely performed using either Soxhlet extraction or sonication. Both procedures utilize large quantities of organic solvents (methylene chloride, hexane, and acetone). These solvents are expensive to purchase and dispose of properly. In addition, these procedures are time consuming and tedious. Supercritical fluid extraction is rapid (minutes as opposed to hours), and uses very little solvent (a few mLs as opposed to hundreds of mLs). This technique employs small quantities of harmless gases, e.g., carbon dioxide, to extract the sample and collect target compounds in small volumes of solvent (about 10 mLs).

Pawliszyn of the University of Waterloo in Ontario, Canada¹ concludes that two steps are necessary to accomplish rapid extraction. First, the conditions need to be such that the analytes are not retained on the matrix. This can be done by increasing their solubility in the extraction fluid as compared to the matrix. Secondly, the rapid transfer of the analytes from the matrix to the fluid must be ensured. This transfer can be accomplished during static extraction.

Once the analytes are extracted from the matrix, the choice of trapping media can play a significant role in the accuracy and precision of the results. Different trapping approaches are commonly used. These include cryogenic, absorbent bed, and liquid trapping.

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The SFE technique has been available for several years. The Agency's use of this technique has been hampered by the extensive scope of work necessary to develop this technique for use in the many possible environmental matrices, and for use in extracting the many target analytes mandated by the Agency's various programs. An Office of Research and Development Leopard Team (a group of specialists) outlined a "Strategy Plan" for the development of SFE (October 31, 1992). Section 4.18 of this plan indicates that a method protocol for the extraction of Base/Neutral/Acidic compounds from soils would be developed and that this project is planned to be conducted in FY '95. In discussions with the Methods Research Branch, USEPA EMSL-Las Vegas, the complexity and extensive nature of the effort involved to overcome the development obstacles could benefit from a coordination of research efforts between ORD and EPA'S Regional laboratories (EMSL-LV and CRL Region III). Work on the extraction of Acidic and Base/Neutral compounds done by CRL could serve as the framework upon which EMSL-LV could build additional complementary capacities (more target compounds) and further improvements.

This work was part of a general effort by the USEPA Central Regional Laboratory (Region III) to minimize the solvent necessary for extraction of semi-volatile compounds. A series of experiments were conducted to optimize various operating conditions for SFE. These conditions included varying the pressure, flow, extraction modifiers, time, and temperature to optimize the recovery of the target compounds. One of the goals was to determine if there were a set of conditions that would extract all semi-volatile compounds (NPDES, Superfund) which vary widely in physical and chemical characteristics. Extractions were first attempted at low temperatures and pressures. These parameters were increased during the progress of this study in order to improve extraction recoveries. The initial phase of this work involved the recovery of the target compounds spiked onto diatomaceous earth. This material has become a commonly used matrix in published work on SFE. It is inexpensive, clean and absorbent. Modifier was added and extractions were performed at various temperatures and pressures. The extractions were directly performed off-line (not coupled to a detector). GC/FID, GC/ECD or GC/MS analyses were performed to determine efficiency of recovery as part of the method validation. Spiked cocktails containing a variety of target compounds were employed. During this on-going process various modifications were made in the extraction procedures to determine their effect on the efficiency of recovery. Work was also done on recovering spikes from soils and similar materials, as well as the extraction of reference soils (known PNA contaminant levels) for comparison to Soxhlet extractions. Generally, compounds that were difficult to extract from diatomaceous earth exhibited the same properties during extraction from the contaminated soil used in the study. Other researchers have found that native analytes present on solid matrices have

different extraction rates than those spiked onto the matrices.² The authors are aware that some matrices may be more difficult to extract than others and that spiked sample recoveries may not reflect those of native analytes. However, reference soils are not available for the great majority of semi-volatile target compounds.

DISCLAIMER

Although the research described in this document has been supported by the U.S. Environmental Protection Agency and is awaiting Agency wide review, it does not necessarily reflect the views of the Agency, and no official endorsement should be inferred. Mention of trade names or commercial products in this report is for illustrative purposes and does not constitute endorsement or recommendation.

EXPERIMENTAL

MATERIALS AND APPARATUS

THERMOLYNE MUFFLE FURNACE was used to clean glassware at a temperature of 450 °C for 8 hours.

CONCENTRATION OF EXTRACTS: used 10 mL concentrator tubes with a 19/22 joint (Labglass, Vineland N.J.) and 3 ball micro Snyder columns.

COLLECTION VIALS USED: Screw cap vials with teflon faced silicone septa, 1.8 mL, used to store references and extracts (Supelco #2-3277; screw cap vials with hole caps and septa, 5 mL (Supelco #2-3249) and 7.4 mL (Supelco #2-3218) used to collect extracts; and Environmental Concentrating Autosampler vials (Supelco #3-3255) used in analysis.

DRUMMOND WIRETROL disposable micropipettes; 10 uL, 50 uL, and 100 uL were used to dispense spike material, add modifiers, and make reference solutions.

CLASS A volumetric flasks: 1.0 mL, 5.0 mL and 10 mL were used to prepare standards and bring extracts to volume.

BRANSONIC 52 sonicator was used to clean extraction vessels and frits.

SUPERCritical FLUID EXTRACTION SYSTEM [Figure 1] The system used was a Suprex Model MPS 225 (Multipurpose SFE-SFC system). The SFC was not employed during this work. The system consisted of: a pump module containing a 250 mL syringe pump, phase valves etc.; control

module, containing electronic circuitry, CRT, keyboard necessary to control the system; and an oven module containing a GC oven, extraction vessel and two valves, a 10 and a 4 port valve. The modules were connected by a mobile phase fluid line from the pump module to the oven module and electronic control and power cables. The extraction vessels used were one and one-half mL stainless steel Suprex EX62010, maximum pressure of 500 atm. In the initial phase of this work a restrictor was employed to collect the extracted target compounds in methylene chloride. The restrictor was 40 μ m ID fused silica approximately 40 cm in length. The extracted material was collected by insertion of the restrictor through the septum of a 7.4 mL screw cap vial (Supelco #2-3218) into 5 mL of methylene chloride below the solvent surface. Methylene chloride was used as a collection solvent in all extractions.

FIGURE 1

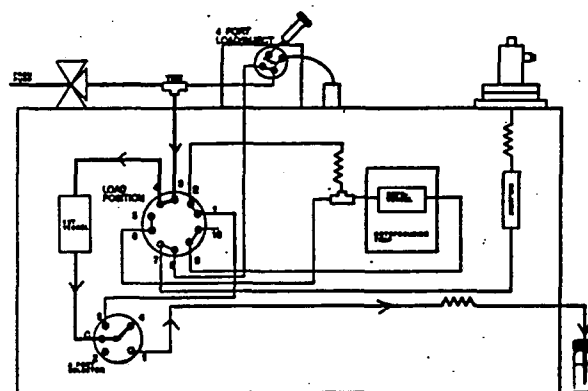


DIAGRAM OF SFE FLOW SYSTEM

In a later phase of this work a Suprex AccutrapTM was added to the system [Figure 2]. This unit consisted of: a variable restrictor of PEEK^R tubing, a heated restrictor module, a cryogenic collection trap and a collection system. During the dynamic extraction phase, the variable restrictor, heated to 50°C, allowed the extraction gases to pass through the SFE system at flow rates of 0.5 to 2 mL per minute. The cryogenic trap, cooled to -48°C with carbon dioxide, captured the target compounds onto glass beads. The trap was heated to 30°C during the desorption phase and was flushed with a measured amount of methylene chloride, which passes into a collection vial. The trap was then purged with nitrogen to clear out any remaining solvent.

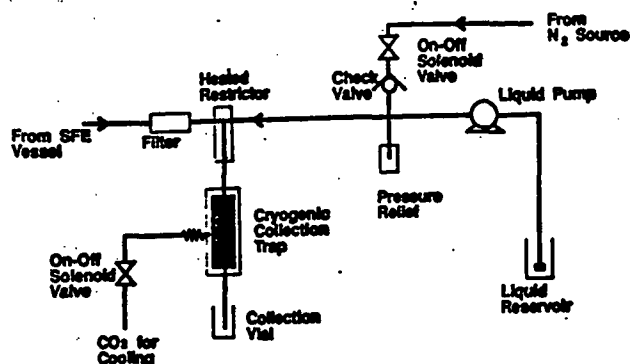


FIGURE 2 **DIAGRAM OF FLOW WITH CRYOGENIC TRAP**

The AccutrapTM system as provided by the manufacturer was altered by extending the stainless steel delivery tube further into the collection vessel below the solvent surface [Figure 3].

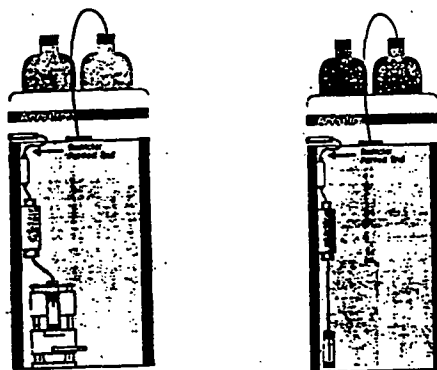


FIGURE 3 **COMPARISON OF COLLECTION SYSTEMS**

GAS CHROMATOGRAPH. An HP 5890 Series II gas chromatograph was equipped with: an electron capture detector; a flame ionization detector; an HP 7673 Automatic sampler; and an HP 3365 Chemstation data system. The FID was equipped with a Supelco SPB-5 fused silica capillary column, 60 meters in length, 0.32 mm ID with a film thickness 0.25 μm (catalogue # 2-4050). The ECD was equipped with a Supelco SPB-608, fused silica capillary column of 30 m, 0.53 mm ID with a film thickness of 0.50 μm . For FID analysis, the GC was programmed from 50°C to a final temperature of 280°C at 5°C/minute with a final holding time of 10 to 25 minutes. The GC temperature program for the pesticides (ECD) was from 150°C to 280°C at 10 degrees /minute with a final holding time of 10 minutes.

GAS CHROMATOGRAPH-MASS SPECTROMETER. The Finnigan MAT 4500 was equipped with: a quadrupole analyzer and EI source; an HP 7673 automatic sampler and an Incos data system. The FSCC column was a DB-5, J&W Scientific, 30m x 0.32 mm with a film thickness of 1 μm . The GC temperature program was: 30°C for 2 minutes, ramped to 300°C at 10°C/minute.

STANDARDS and SPIKING MATERIAL. Analytical reference standards for the Base/Neutral Extractables, Pesticides, Toxic Substances and Phenol Mixtures were obtained from AccuStandard, Inc., New Haven, CT. The following compounds are now available through EMSL-CI, but were originally obtained through the EPA Quality Assurance Materials Bank (Analytical Reference Standards, Las Vegas, Nevada): Toxaphene; Chlordane; Naphthalene; PCBs; 2,4,6- Tribromophenol; p-Terphenyl-d₁₄; Organic surrogate mix; 1,4- Dichlorobenzene; N-Nitrosodi-n-propylamine; 2,4-Dinitrotoluene; Di-n-butyl Phthalate; Acenaphthene; 1,2,4-Trichlorobenzene; Pyrene; 4-Nitrophenol; 2-Chlorophenol; Phenol; 4-Chloro-3-methylphenol; Pentachlorophenol; Benzo(a)pyrene; Fluoranthene; Indeno(1,2,3-c,d)pyrene; Bis(2-chloroiso-propyl) ether; and 2-Chloroethyl vinyl ether.

SUPERCRITICAL FLUIDS. SFC grade carbon dioxide, with helium headspace and a dip tube, was obtained from Scott Specialty Gases, Inc. of Plumsteadville, Pa. SFC grade carbon dioxide with ten percent methanol obtained through Scott was also used in some extractions.

SAMPLE MATRICES. Matrices used included: diatomaceous earth, PAH-contaminated soil SRS 103-100 (Fisher Scientific), NIST SFE Round Robin Exercise sediments and a test soil containing a large percentage of clay which had been dried, ground and sieved.

EXTRACTIONS OF DIATOMACEOUS EARTH WITH FUSED SILICA RESTRICTOR

All extractions were performed by adding spike and modifier to approximately 0.2 g. diatomaceous earth in the bottom of the extraction vessel. The solvent was not allowed to evaporate before the modifier was added and the spike solvent probably acted as an additional modifier. The sample was then covered with a glass fiber filter (to eliminate any possible dead space and to prevent any diatomaceous earth from passing through the frit into the restrictor). The supercritical fluid containing the extracted material passed through the restrictor and bubbled into the methylene chloride solvent in the collection vial. Upon completion of the extraction, the collection solvent was adjusted to 1.0 mL. in class "A" volumetric flasks either by evaporation using a micro Kuderna-Danish apparatus and/or by diluting with methylene chloride.

Compound spike recoveries were computed against the response of a reference standard prepared the same day that the samples were extracted. The reference standards and extracts were analyzed on the same GC/FID or GC/ECD.

The temperatures and pressures used in initial extractions were similar to those used by others as cited in the literature in similar extractions. The densities obtained from each temperature/pressure combination were displayed as part of the run

parameters during the extraction. Information was obtained that provided Hildebrand solubility parameter data on organic solvents and extraction conditions for supercritical carbon dioxide (temperature, pressure and resulting density) that emulated the solubility parameters or solvent strengths of various organic solvents. Later extractions used temperature and pressure combinations that gave carbon dioxide densities that were similar to the solvent strengths of various organic solvents (Table 1).

TABLE 1
SUPERCritical CARBON DIOXIDE EXTRACTION CONDITIONS

EXTRACTION TEMPERATURE/PRESSURE	ACTUAL CARBON DIOXIDE DENSITY	THEORETICAL CARBON DIOXIDE DENSITY	CORRESPONDING SOLVENT
40 ⁰ C./150 atm	0.78 g/mL	0.75 g/mL	heptane
40 ⁰ C./210 atm	0.85 g/mL	0.86 g/mL	hexane
40 ⁰ C./390 atm	0.95 g/mL	0.96 g/mL	cyclohexane
60 ⁰ C./300 atm	0.83 g/mL	0.82 g/mL	pentane
80 ⁰ C./150 atm	0.44 g/mL		unlisted
80 ⁰ C./210 atm	0.62 g/mL		unlisted
80 ⁰ C./300 atm	0.75 g/mL	0.75 g/mL	heptane
80 ⁰ C./330 atm	0.85 g/mL	0.86 g/mL	hexane

PROTOCOLS USED IN EXTRACTIONS

The APPENDIX lists composition and concentration of standards and spiking solutions.

The **Acid/Base Matrix** spike extractions were begun at 40° C and 150 atm, continued at 210 atm for 7.5 min (static) and 15 minutes dynamic extraction using carbon dioxide. No modifier was added.

Base/Neutral Mix 2 - MO01E was extracted under a variety of conditions as indicated in Tables 2 & 3. All extractions were done with 50 uL of 1:1 MeCl₂-MeOH as the modifier with an initial pressurization at 150 atm for 1 minute, followed by a static extraction for 7.5 minutes. Pressure, temperature and time were varied. Extractions were run under three sets of conditions: at 80°C with a dynamic extraction for 45 minutes at 210 atm; at 80°C with a dynamic extraction at 250 atm for 45 minutes; and at 100°C for 45 minutes at 210 atm.

Base/Neutral Mix 1-MOO1D, MOO1G/Naphthalene, Mix 3 MOO1F, Mix 4 - the Phenol Mix - MOO1P as well as Chlordane, Toxaphene and PCB 1260 were extracted with 50 uL of 1:1 MeCl₂-MeOH modifier at 80°C, with an initial pressurization at 150 atm for one minute, followed by a static extraction at 210 atm for 7.5 min and dynamic extraction at 210 atm for 45 minutes. Collection was done using a 40 micron fused silica restrictor with methylene chloride as collection solvent. The spikes of Phenol mix MOO1P were also analyzed using the Suprex Accutrap™.

Spikes of Toxic Substances Mixes 2- Z-014E/Z-014F were extracted at 80°C with 50 uL of 1:1 MeCl₂-MeOH modifier. Initial pressurization took place at 150 atm for one minute, followed by static extraction at 210 atm for 7.5 minutes and dynamic extraction at 210 atm for 15 minutes. The compounds were collected in methylene chloride with the 40 micron fused silica restrictor. Spikes of this mix were also analyzed using the Suprex Accutrap™.

Pesticides Mix MOO1H/M-608-1 and the Surrogate Mix were extracted at 80°C, with initial pressurization at 150 atm for one minute, static extraction at 210 atm for 7.5 min and dynamic extraction at 210 atm for both 30 and 45 minutes. A modifier of 50 uL of 1:1 methanol-methylene chloride was used. Collection was in methylene chloride using a 40 micron fused silica restrictor.

PCBs: 1260, 1254, 1248, 1242, 1232, 1221, and 1016 were extracted using 50 uL of 1:1 MeCl₂-MeOH as modifier at 80°C. The extraction included an initial pressurization at 150 atm for one minute, followed by static extraction at 210 atm for 7.5 minutes and dynamic extraction at 210 atm for 30 minutes.

GENERAL RESULTS AND DISCUSSION

Technical problems associated with the restrictor technique that have been quoted by SFE researchers include: vaporization and increased loss of solvents due to heated restrictors and loss of volatile compounds due to high flow rates. We encountered these problems and many of the difficulties cited by Engelhardt and Haas and others. These included: clogged restrictors; poor reproducibility; and variable flow rates with changing restrictor size. This was especially true with mixes that contained high molecular weight compounds such as late-eluting PNAs. Apparently, the compounds were not passing entirely through the restrictor and were ultimately causing it to plug. There were many unsuccessful attempts to solve this problem. Base/Neutral mix 2 (MOO1E) proved to be the ultimate challenge as it contained both volatile dichlorobenzenes and late-eluting dibenzo(a,h)anthracene. Believing that the compounds were plating out when the gases left the restrictor and entered the cooler methylene chloride in the collection vial, the collection vial was placed in a beaker of warmed water. This resulted only in increased evaporation of the collection solvent. Clipping the end of the restrictor after each

use did not eliminate the problem either. Another approach was to put the restrictor in a teflon tube in such a way that only the teflon tube entered the collection solvent; with the restrictor not extending past the cap of the collection vial. The clogging was reduced, but the compounds plated out on the teflon tube. Table 2 compares these extractions with those obtained when the restrictor was placed directly in the collection solvent. Recoveries were comparable, but the standard deviation increased greatly when using the teflon tube.

TABLE 2
COMPARISON OF MOOIE EXTRACTIONS
USING TEFLON TUBE AND FUSED SILICA RESTRICTOR
FROM DIATOMACEOUS EARTH

Compounds	Teflon tube		Fused silica	
Conditions	7.5/45/80/210 *		7.5/45/80/210 *	
Ave = %, Recovery N = 4	Average	s	Average	s
1,3-Dichlorobenzene	97.8	5.7	93.1	1.3
1,2-Dichlorobenzene	98.4	5.2	93.5	1.4
Bis(2-chloroethoxy)methane	100.4	3.6	96.3	1.8
Naphthalene	99.3	3.8	94.4	1.3
Hexachlorobutadiene	99.8	4.4	94.3	1.1
Acenaphthene	97.8	3.8	95.6	1.2
2,4-Dinitrotoluene	93.4	4.5	97.5	1.5
Diethylphthalate	98.0	5.0	101.7	3.8
Fluorene	96.8	4.7	96.1	1.6
Anthracene	91.5	5.7	95.5	0.9
Hexachlorobenzene	87.4	5.6	93.8	1.4
Pyrene	88.0	7.0	90.8	0.5
Benzo(a)anthracene	82.0	12.5	86.3	1.3
Chrysene	81.7	17.8	83.2	2.6
Dibenzo(a,h)anthracene	68.5	39.8	53.8	5.1

* static time(min)/dynamic time(min)/temperature(°C)/pressure(atm)

Extracting at higher temperatures accelerated the rate of evaporation of collection solvent, causing some of the extracts to go to dryness. Adding more solvent in mid-run was difficult and undesirable. Extending the time of the dynamic extraction from 15

minutes to 45 minutes resulted in less plugging and better recoveries. Increasing the pressure also resulted in better recoveries. Table 3 compares the recoveries with increased temperature and pressure. Overall, recoveries decreased and standard deviation increased when the temperature was increased. While increasing the pressure did increase the standard deviation, the recovery of dibenzo(a,h)anthracene increased by 20% when compared to the recovery obtained at lower atmospheric pressure. However, other researchers have reported better extraction efficiencies when the extraction temperature is increased. They also found that for sample matrices having tightly bound analytes, temperature was more important than pressure in achieving better extraction recoveries.

TABLE 3

**COMPARISON OF MOOIE EXTRACTIONS FROM DIATOMACEOUS EARTH
VARYING TEMPERATURE AND PRESSURE**

Compounds	Increased Temperature	Increased Pressure
Conditions	7.5/45/100/210 *	7.5/45/80/250 *
Ave = % recovery N = 4	Average s	Average s
1,3-Dichlorobenzene	83.1 12.3	80.5 8.5
1,2-Dichlorobenzene	83.5 12.1	81.1 8.4
Bis(2-chloroethoxy)methane	84.7 13.3	86.5 7.9
Naphthalene	84.0 13.2	82.9 7.4
Hexachlorobutadiene	85.0 12.1	81.8 7.2
Acenaphthene	76.0 21.2	83.0 2.1
2,4-Dinitrotoluene	67.1 21.9	81.5 0.9
Diethylphthalate	72.2 21.2	85.5 1.6
Fluorene	72.7 21.3	86.7 6.1
Anthracene	63.4 23.7	94.8 25.8
Hexachlorobenzene	60.9 24.3	81.7 5.1
Pyrene	61.6 24.5	77.8 2.3
Benzo(a)anthracene	85.6 28.2	76.2 3.5
Chrysene	56.6 32.6	78.7 2.2
Dibenzo(a,h)anthracene	49.5 34.2	77.0 8.2

* static time(min)/dynamic time(min)/temperature(°C)/pressure(atm)

Table 4 summarized the results of the ninety-seven compounds that were extracted from spikes onto diatomaceous earth using a fused glass restrictor. It should be noted that five different sets of extraction conditions were used during efforts to optimize the maximum recoveries. Pesticides and PCBs were analyzed by GC/ECD. All others were analyzed by GC/FID. To compare these recoveries with an approved EPA method, the QC parameters for Method 625 are included. Thirteen of the compounds had a recovery of 100-107%, forty-seven fell between 90 and 100%, thirty-two between 80 and 90%, and seven between 70 and 80%, and one below 70%. Fifteen of the averages had a standard deviation of greater than 10%. The lowest recovery was that of benzo(g,h,i)perylene at 68.8% and a standard deviation of 11.86. This compares favorably with the 625 method QC limits of Detected-195.0 percent recovery and a standard deviation of 58.9. This set of extractions was performed at a pressure of 210 atm., possibly a similar increase in recovery similar to that of dibenzo(a,h)anthracene could have been obtained if extracted at 250 atmospheres. Five of the seven compounds with recoveries between 70 and 80% were also late eluting PNAs. The volatile compound 2-chloroethyl vinyl ether was recovered at 80%. Failure to recover a greater percent of benzidine and 3,3'-dichlorobenzidine may be a function of their very basic nature.

TABLE 4

COMPARISON OF SPIKE RECOVERIES FROM DIATOMACEOUS EARTH WITH METHOD 625 QC

	D.E.		Method 625 QC		Conditions	Mix
Target Compounds Recovery = %	Average	s n = 4	Average	s	s/d/t/p	
Alpha-BHC	91.1	2.7			7.5/45/80/210	Pesticides, MOO1H
Delta-BHC	92.3	3.1			7.5/45/80/210	Pesticides, MOO1H
Gamma-BHC	91.8	3.2			7.5/45/80/210	Pesticides, MOO1H
Beta-BHC	88.8	5.9			7.5/45/80/210	Pesticides, MOO1H
Heptachlor	85.9	11.7	D-172.2	37.2	7.5/45/80/210	Pesticides, MOO1H
Aldrin	88.7	1.4	7.2-152.2	39.0	7.5/45/80/210	Pesticides, MOO1H
Heptachlor epoxide	91.3	3.5	70.9-109.4	54.7	7.5/45/80/210	Pesticides, MOO1H
Endosulfan I	91.9	3.6			7.5/45/80/210	Pesticides, MOO1H
4,4'-DDE	88.1	1.8	19.2-119.7	32.0	7.5/45/80/210	Pesticides, MOO1H
Dieldrin	96.4	5.7	44.3-119.3	30.7	7.5/45/80/210	Pesticides, MOO1H
Endrin	90.0	5.1			7.5/45/80/210	Pesticides, MOO1H
4,4'-DDD	106.7	4.4	D-134.5	31.0	7.5/45/80/210	Pesticides, MOO1H
4,4'-DDT	91.4	3.8	D-170.6	61.6	7.5/45/80/210	Pesticides, MOO1H
Endrin aldehyde	94.4	6.8	D-188.8	32.5	7.5/45/80/210	Pesticides, MOO1H
Endosulfan sulfate	91.6	5.5	D-103.5	16.7	7.5/45/80/210	Pesticides, MOO1H
Endosulfan II	92.8	2.0			7.5/45/80/210	Pesticides, MOO1H

s/d/t/p = static extraction(min)/dynamic extraction(min)/temperature(⁰C)/pressure(atm)
 Analysis of pesticides and PCBs by GC/ECD, all others by GC/FID.

	D.E.		Method 625 QC		Conditions	Mix
Target Compounds Recovery = %	Average	s n = 4	Average	s	s/d/t/p	
Bis(2-chloroethyl)ether	89.7	6.9	42.9-126.0	55.0	7.5/45/80/210	MOO1D
1,4-Dichlorobenzene	88.0	6.6	37.3-105.7	32.1	7.5/45/80/210	MOO1D
Bis(2-chloroisopropyl)ether	90.7	6.8	62.8-138.6	43.6	7.5/45/80/210	MOO1D
Nitrobenzene	91.6	7.3	54.3-157.6	39.3	7.5/45/80/210	MOO1D
Dimethylphthalate	99.3	10.7	D-100.0	23.2	7.5/45/80/210	MOO1D
2,6-Dinitrotoluene	98.1	11.4	68.1-136.7	29.6	7.5/45/80/210	MOO1D
Acenaphthylene	96.7	8.4	53.5-126.0	40.2	7.5/45/80/210	MOO1D
4-Bromophenyl phenyl ether	100.0	10.7	64.9-114.4	23.0	7.5/45/80/210	MOO1D
Di-n-butyl phthalate	104.5	11.1	8.4-111.0	16.7	7.5/45/80/210	MOO1D
3,3'-Dichlorobenzidine	88.9	17.9	8.2-212.5	71.4	7.5/45/80/210	MOO1D
Bis(2-ethylhexyl)phthalate	106.4	11.2	28.9-136.8	41.1	7.5/45/80/210	MOO1D
Benzo(b)fluoranthene	94.9	17.5	42.0-140.4	38.8	7.5/45/80/210	MOO1D
1,2-Dichlorobenzene	80.5	8.5	48.6-112.0	30.9	7.5/45/80/250	MOO1E
1,3-Dichlorobenzene	81.1	8.4	16.7-153.9	41.7	7.5/45/80/250	MOO1E
Bis(2-chloroethoxy)methane	86.5	7.9	49.2-164.7	34.5	7.5/45/80/250	MOO1E
Naphthalene	82.9	7.4	35.6-119.6	30.1	7.5/45/80/250	MOO1E
Hexachlorobutadiene	81.8	7.2	37.8-102.2	26.3	7.5/45/80/250	MOO1E
Acenaphthene	83.0	2.1	60.1-132.3	27.6	7.5/45/80/250	MOO1E

s/d/t/p = static extraction(min)/dynamic extraction(min)/temperature(⁰C)/pressure(atm)
 Analysis of pesticides and PCBs by GC/ECD, all others by GC/FID.

	D.E.		Method 625 QC		Conditions	Mix
Target Compounds Recovery = %	Average	s n = 4	Average	s	s/d/t/p	
2,4-Dinitrotoluene	81.5	0.9	47.5-126.9	21.8	7.5/45/80/250	MOO1E
Diethyl phthalate	85.5	1.6	D-100.0	26.5	7.5/45/80/250	MOO1E
Fluorene	86.7	6.1	71.6-108.4	20.7	7.5/45/80/250	MOO1E
Anthracene	94.8	25.8	43.4-118.0	32.0	7.5/45/80/250	MOO1E
Hexachlorobenzene	81.7	5.1	7.8-141.5	24.9	7.5/45/80/250	MOO1E
Pyrene	77.8	2.3	69.6-100.0	25.2	7.5/45/80/250	MOO1E
Benzo(a)anthracene	76.2	3.5	41.8-133.0	27.6	7.5/45/80/250	MOO1E
Chrysene	78.7	2.2	44.1-139.9	48.3	7.5/45/80/250	MOO1E
Dibenzo(a,h)anthracene	77.0	8.2	D-199.7	70.0	7.5/45/80/250	MOO1E
Hexachloroethane	99.6	3.2	55.2-100.0	24.5	7.5/45/80/210	MOO1F
N-Nitroso-dipropylamine	100.8	2.5	13.6-197.9	55.4	7.5/45/80/210	MOO1F
Isophorone	102.5	5.2	46.6-180.2	63.3	7.5/45/80/210	MOO1F
1,2,4-Trichlorobenzene	100.8	2.7	57.3-129.2	28.1	7.5/45/80/210	MOO1F
Hexachlorocyclopentadiene	100.7	1.4			7.5/45/80/210	MOO1F
2-chloro-naphthalene	102.7	3.3	64.5-113.5	13.0	7.5/45/80/210	MOO1F
1,2-Diphenylhydrazine	101.5	3.5			7.5/45/80/210	MOO1F
N-Nitrosodiphenylamine	102.7	3.4			7.5/45/80/210	MOO1F
Phenanthrene	102.0	4.1	65.2-108.7	20.6	7.5/45/80/210	MOO1F

s/d/t/p = static extraction(min)/dynamic extraction(min)/temperature(⁰C)/pressure(atm)
 Analysis of pesticides and PCBs by GC/ECD, all others by GC/FID.

	D.E.		Method 625 QC		Conditions	Mix
Target Compounds Recovery = %	Average	s n = 4	Average	s	s/d/t/p	
Fluoranthene	102.0	5.6	42.9-121.3	32.8	7.5/45/80/210	MOO1F
Butyl benzyl phthalate	104.4	6.6	D-139.9	23.4	7.5/45/80/210	MOO1F
2-chloroethyl vinyl ether	80.0	14.0			7.5/45/80/210	MOO1G/Naphthalene
N-Nitrosodimethyl amine	85.7	9.8			7.5/45/80/210	MOO1G/Naphthalene
Naphthalene	88.8	7.4	35.6-119.6	30.1	7.5/45/80/210	MOO1G/Naphthalene
4-Chlorophenylphenyl ether	82.5	6.1	38.4-144.7	33.4	7.5/45/80/210	MOO1G/Naphthalene
Benzidine	83.5	9.7			7.5/45/80/210	MOO1G/Naphthalene
Di-n-octyl phthalate	90.4	6.4	18.6-131.8	31.4	7.5/45/80/210	MOO1G/Naphthalene
Benzo(k)fluoranthene	85.4	15.9	25.2-145.7	32.3	7.5/45/80/210	MOO1G/Naphthalene
Benzo(a)pyrene	81.3	8.8	31.7-148.0	39.0	7.5/45/80/210	MOO1G/Naphthalene
Indeno(1,2,3-cd) pyrene	80.4	10.5	D-150.9	44.6	7.5/45/80/210	MOO1G/Naphthalene
Benzo(g,h,i)perylene	68.8	11.9	D-195.0	58.9	7.5/45/80/210	MOO1G/Naphthalene
Phenol	92.5	4.2	16.6-100.0	22.6	7.5/30/80/210	MOO1P
2-Chlorophenol	91.2	3.8	36.2-120.4	28.7	7.5/30/80/210	MOO1P
2-Nitrophenol	93.2	3.9	45.0-166.7	35.2	7.5/30/80/210	MOO1P
2,4-Dichlorophenol	92.6	4.1	52.5-121.7	26.4	7.5/30/80/210	MOO1P
2,4-Dimethylphenol	93.2	4.2	41.8-109.0	26.1	7.5/30/80/210	MOO1P
4-chloro-3-methyl phenol	95.0	4.4	40.8-127.9	37.2	7.5/30/80/210	MOO1P

s/d/t/p = static extraction(min)/dynamic extraction(min)/temperature(⁰C)/pressure(atm)
 Analysis of pesticides and PCBs by GC/ECD, all others by GC/FID.

	D.E.		Method 625 QC		Conditions	Mix
Target Compounds Recovery = %	Average	s n = 4	Average	s	s/d/t/p	
2,4,6-Trichlorophenol	93.4	4.2	52.4-129.2	31.7	7.5/30/80/210	MOO1P
2,4-Dinitrophenol	84.3	14.7	D-172.9	49.8	7.5/30/80/210	MOO1P
4-Nitrophenol	97.8	5.5	13.0-106.5	47.2	7.5/30/80/210	MOO1P
2-Methyl-4,6-dinitrophenol	89.6	9.8	53.0-100.0	93.2	7.5/30/80/210	MOO1P
Pentachlorophenol	91.1	5.0	36.1-151.8	48.9	7.5/30/80/210	MOO1P
Toxaphene	85.5	6.9			7.5/45/80/210	CAS:8001-35-2
Chlordane (alpha isomer)	92.1	2.3			7.5/45/80/210	CAS:57-74-9
Chlordane (gamma isomer)	91.4	1.5			7.5/45/80/210	CAS:57-74-9
PCB 1260	95.9	2.9	19.3-121.0	54.2	7.5/30/80/210	CAS:11096-82-5
PCB 1254	96.5	7.1			7.5/30/80/210	CAS:11097-69-1
PCB 1248	93.9	2.3			7.5/30/80/210	CAS:12672-29-6
PCB 1242	87.0	7.5			7.5/30/80/210	CAS:53469-21-9
PCB 1232	94.5	7.3			7.5/30/80/210	CAS:11141-16-5
PCB 1221	92.5	2.9			7.5/30/80/210	CAS:11104-28-2
PCB 1016	97.3	9.2			7.5/30/80/210	CAS:12674-11-2
Phenol	89.7	9.5	16.6-100.0	22.6	7.7/15/40/210	Matrix
2-Chlorophenol	89.2	9.5	36.2-120.4	28.7	7.7/15/40/210	Matrix
1,4-Dichlorobenzene	89.0	9.6	37.3-105.7	32.1	7.7/15/40/210	Matrix

s/d/t/p = static extraction(min)/dynamic extraction(min)/temperature(⁰C)/pressure(atm)
 Analysis of pesticides and PCBs by GC/ECD, all others by GC/FID.

	D.E.		Method 625 QC		Conditions	Mix
Target Compounds Recovery = %	Average	s n = 4	Average	s	s/d/t/p	
N-Nitroso-di-n-propylamine	89.0	10.4	13.6-197.9	55.4	7.7/15/40/210	Matrix
1,2,4-trichlorobenzene	89.2	9.6	57.3-129.2	28.1	7.7/15/40/210	Matrix
4-Chloro-3-methyl phenol	90.5	6.6	40.8-127.9	37.2	7.7/15/40/210	Matrix
Acenaphthene	89.0	9.8	60.1-132.3	27.6	7.7/15/40/210	Matrix
4-Nitrophenol	88.1	11.4	13.0-106.5	47.2	7.7/15/40/210	Matrix
2,4-Dinitrotoluene	87.8	9.6	47.5-126.9	21.8	7.7/15/40/210	Matrix
Pentachlorophenol	96.1	28.1	38.1-151.8	48.9	7.7/15/40/210	Matrix
Di-n-butylphthalate	83.8	12.4	8.4-111.0	16.7	7.7/15/40/210	Matrix
Pyrene	87.2	11.0	69.6-100.0	25.2	7.7/15/40/210	Matrix
2-Fluorophenol	81.5	2.0			7.5/30/80/210	Surrogates
D5-Phenol	83.7	1.3			7.5/30/80/210	Surrogates
D5-Nitrophenol	83.7	1.2			7.5/30/80/210	Surrogates
2-Fluoro-1-1'-biphenyl	85.6	1.3			7.5/30/80/210	Surrogates
2,4,5-Tribromophenol	99.9	4.6			7.5/30/80/210	Surrogates
D14-Terphenyl	88.2	4.7			7.5/30/80/210	Surrogates
Alpha BHC	91.0	4.5			7.5/30/80/210	Pesticides
Delta BHC	92.8	2.9			7.5/30/80/210	Pesticides
Gamma BHC	91.5	2.8			7.5/30/80/210	Pesticides

s/d/t/p = static extraction(min)/dynamic extraction(min)/temperature(⁰C)/pressure(atm)
 Analysis of pesticides and PCBs by GC/ECD, all others by GC/FID.

	D.E.		Method 625 QC		Conditions	Mix
Target Compounds Recovery = %	Average	s n = 4	Average	s	s/d/t/p	
Beta BHC	92.4	4.3			7.5/30/80/210	Pesticides
Heptachlor	90.5	15.2	D-172.2	37.2	7.5/30/80/210	Pesticides
Aldrin	92.0	3.5	7.2-152.2	39.0	7.5/30/80/210	Pesticides
Heptachlor epoxide	92.6	3.4	70.9-109.4	54.7	7.5/30/80/210	Pesticides
Endosulfan I	93.2	3.0			7.5/30/80/210	Pesticides
4,4'-DDE	90.4	4.6	19.2-119.7	32.0	7.5/30/80/210	Pesticides
Dieldrin	97.0	3.2	44.3-119.3	30.7	7.5/30/80/210	Pesticides
Endrin	92.9	2.6			7.5/30/80/210	Pesticides
4,4'-DDD	106.7	4.5	D-134.5	31.0	7.5/30/80/210	Pesticides
4,4'-DDT	92.6	4.3	D-170.6	61.6	7.5/30/80/210	Pesticides
Endrin aldehyde	95.2	2.9	D-188.8	32.5	7.5/30/80/210	Pesticides
Endosulfan sulfate	93.3	1.2	D-103.5	16.7	7.5/30/80/210	Pesticides
Endosulfan II	96.1	1.6			7.5/30/80/210	Pesticides
Aniline	95.8	1.0			7.5/15/80/210	Z014E, Z014F
Benzyl alcohol	96.4	1.2			7.5/15/80/210	Z014E, Z014F
4-Chloroaniline	95.7	1.2			7.5/15/80/210	Z014E, Z014F
2-methylnaphthalene	96.1	1.4			7.5/15/80/210	Z014E, Z014F
2-Nitroaniline	95.6	1.4			7.5/15/80/210	Z014E, Z014F

s/d/t/p = static extraction(min)/dynamic extraction(min)/temperature(⁰C)/pressure(atm)
 Analysis of pesticides and PCBs by GC/ECD, all others by GC/FID.

	D.E.		Method 625 QC		Conditions	Mix
Target Compounds Recovery = %	Average	s n = 4	Average	s	s/d/t/p	
3-Nitroaniline	94.2	1.2			7.5/15/80/210	Z014E, Z014F
Benzidine	70.4	13.5			7.5/15/80/210	Z014E, Z014F
3,3'-Dichlorobenzidine	71.8	17.8			7.5/15/80/210	Z014E, Z014F

s/d/t/p = static extraction(min)/dynamic extraction(min)/temperature(⁰C)/pressure(atm)
 Analysis of pesticides and PCBs by GC/ECD, all others by GC/FID.

EXTRACTION OF DIATOMACEOUS EARTH USING THE ACCUTRAPTM

With the installation of the AccutrapTM cryogenic trap, it was decided to test the new system with the standard mix, MO01E, which includes a variety of compounds. Initial extractions suggested that a CO₂ flow rate of 2-3 mLs per minutes increased the recoveries of dibenzo(a,h)anthracene, but resulted in poor or no recovery of the two volatile dichlorobenzenes in the mix.

We reprogrammed the extraction (using the micro processor controlled multiport valve) so that during the first minute the flow was directed through a 50 micron glass restrictor into a vial containing methylene chloride. The remainder of the dynamic flow period was directed to the cryogenic trap. This was the reverse of the configuration used by Richards and Campbell while extracting priority pollutants in soil. They installed a cryogenic trap after the collection flask to improve the recoveries of volatile compounds. Analysis showed that the dichlorobenzenes were being extracted during the first minute, but were not being captured in the cryogenic trap.

Consequently, we reconfigured the collection system again as shown previously in Figure 3 by installing a piece of 1/16 inch stainless steel tubing after the trap in place of the AccutrapTM collection system. The tubing passed through the rubber septum of a 7.5 mL screw cap collection vial and down into five mL. of methylene chloride. The carbon dioxide would now bubble through the solvent during the dynamic extraction period, capturing the volatile compounds that had eluted from the cryogenic trap. It was also necessary to restrict the flow of the carbon dioxide through the solvent during the first few minutes of the dynamic phase of the extraction to allow the more volatile compounds to be trapped more efficiently in the solvent. It is planned to replace this restrictor with 1/32 inch ID stainless steel tubing which should reduce the size of the bubbles and perhaps enhance recoveries because of the greater area for exchange between the carbon dioxide and solvent. The smaller tubing should also eliminate the necessity to vary the flow through the restrictor during the dynamic stage. This is tedious and difficult to reproduce the flows with each run. The most optimum extraction conditions using the AccutrapTM proved to be 80°C at 300 atmospheres with a static extraction of 10 minutes and a dynamic extraction of 45 minutes. These conditions produced a density emulating that of pentane. For the first three minutes, the flow was restricted and for the balance of the extraction, the flow was set at approximately 1.0 mL/min. A comparison of the recoveries achieved from extraction under the original configuration as provided by the manufacturer with those obtained with the modified collection system are shown in Table 5.

The Phenol mix MO01P was also extracted. Recoveries were about 10% lower than those achieved using the configuration where the fused silica restrictor bubbled through the collection solvent.

The Combined Toxic Substances mix Z-014E and Benzidines mix Z014F were extracted using the same parameters. The recovery of the toxic substances were generally down by about 9% from those obtained using the fused silica restrictor configuration, while the benzidines were up by about 10%.

TABLE 5

COMPARISON OF RECOVERIES FROM DIATOMACEOUS EARTH OF COMPOUNDS FROM MO01E USING DIFFERENT RECOVERY CONFIGURATIONS OF THE ACCUTRAP™

Compounds Recovery in % Original n = 2 Altered n = 4	Original System * Average	Altered System * Average	Altered System s
1,3 Dichlorobenzene	16.8	80.1	3.2
1,2 Dichlorobenzene	22.2	88.6	2.0
Bis(2-chloroethoxy)methane	33.6	92.9	1.8
Naphthalene	35.3	89.1	1.7
Hexachlorobutadiene	31.8	91.3	1.6
Acenaphthene	53.8	92.7	2.7
2,4-Dinitrotoluene	81.8	91.2	6.3
Diethylphthalate	81.8	92.5	5.6
Fluorene	69.6	91.4	4.6
Anthracene	85.1	100.2	18.8
Hexachlorobenzene	85.8	91.6	12.3
Pyrene	89.8	91.1	8.0
Benzo(a)anthracene	97.4	88.9	10.6
Chrysene	105.6	88.8	13.4
Dibenzo(a,h)anthracene	108.4	94.6	14.2

* at 80°C/300 atm

This new configuration achieved recoveries of greater than 80% for all compounds, both semi-volatile and late-eluting PNAs. The recovery of the last four late-eluting PNAs increased from an average of 77% to an average of 100%.

EXTRACTION OF SOILS USING FUSED SILICA RESTRICTOR

Attempts at extraction of Fisher PAH Contaminated Soil (SRS 103-100) proved trying to both the analysts and extraction apparatus. This soil is widely used in SFE studies and is one of the few "real-world" reference materials that is available at the necessary concentrations. An acceptance range of recoveries for each of the compounds is provided by Fisher.

Initial attempts at extraction of soil involved a three step program that increased both the temperature and pressure from 40°C/150 atm initially to 80°C at 350 atm. Problems encountered included: the inability to pressurize the extraction chamber past 300 atm (blown lines); clogged restrictors; and excessive evaporation of collection solvent. Much effort was focused on: the causes and possible solutions for restrictor clogging; increasing the recoveries of late eluting compounds; improved reproducibility; and accuracy. Numerous modifications included: shortening the restrictor length; increasing time and temperature in the extraction program; increasing the temperature of the collection solvent and use of various modifiers. Eventually, with the addition of 1:1 MeCl₂-MeOH modifier, multiple extractions were accomplished without restrictor clogging at a temperature of 80°C with dynamic extraction of 45 minutes at 210 atm. These were the conditions used in the extraction of Base/neutral compounds. The extracted compounds were collected by the insertion of the 40 micron restrictor through the septum of a 10 mL screw cap vial into approximately seven milliliters of methylene chloride. After extraction, the volume of the collection solvent was diluted to 5.0 mL volumetrically. Analysis was done by GC/MS. The results appear in Table 6 under Fused Silica extractions.

EXTRACTION OF PNA CONTAMINATED SOILS USING ACCUTRAP™

With the addition of the cryogenic trap and the availability of the SFE grade carbon dioxide with 10% methanol, the Fisher Soil was again extracted. A one mL extraction vessel was used so that the sample size could be increased to approximately one gram. This set of extractions was initially carried out at 80°C at 300 atmospheres, a density of 0.75 g/mL, with a static extraction for 10 minutes and a dynamic extraction of 45 minutes (restricting the flow during the first few minutes to insure that the volatile compounds were captured in the solvent). The collection vessel was a 20 mL screw top vial with approximately 10 mL of methylene chloride. The trap was cooled to -45°C, and was desorbed at 30°C with 3 mL of methylene chloride. The extract was diluted to 10 mL volumetrically. Interestingly, during the desorption process, a solid phase appeared in the collection vial which went back into solution upon warming to room temperature. Analysis was done by GC/MS.

Levy⁸ and Langenfeld⁹ reported greater recoveries than we were able to attain, Langenfeld by increasing pressures to as much as 650 atm at 200°C and Levy to a maximum of 450 atmospheres at 75°C. However, they did not report recoveries on all fifteen compounds present.

The second set of extractions took place at 40°C. and 390 atmospheres (a density of 0.95 g/mL, equivalent to cyclohexane).

Soxhlet results were established at the CRL using a 24 hour hexane-acetone extraction procedure and analyzed by GC/MS.

Comparison of the PAHs extracted using SFE extractions of 0.2 gm (fused silica restrictor), 1.0 gm¹⁰ extractions using CO₂-Methanol (AccutrapTM), those of Lopez-Avila¹⁰, and the Soxhlet extraction are shown in Table 6. It has been indicated in an EPA publication that higher recoveries than those listed in the table were achieved by Lopez-Avila using 2.5 grams¹¹.

TABLE 6

COMPARISON OF RECOVERIES OBTAINED BY VARIOUS METHODS REPORTED FOR SRS 103-100 REFERENCE SOIL
WITH CERTIFIED VALUES USING GC/MS ANALYSIS

Recoveries in mg/kg	Fused silica	Accutrap TM	Lopez-Avila ⁶	Soxhlet	Fisher Cert. Values	Fisher Range of recoveries
Weight extracted	0.2 g	1.0 g	6.0 g	5.0 g		
Compound						
Naphthalene	21.3	19.4	9.5	32.6	24.2-40.6	8.9-38.3
2-Methylnaphthalene	61.7	53.5	37.6	70.9	50.6-73.6	37.8-75.6
Acenaphthylene	13.2	9.0	5.3	14.5	14.7-23.5	8.3-24.3
Acenaphthene	635	370	616	665	527-737	516.3-665.5
Dibenzofuran	284	217	307	307	258-356	253.5-357.7
Fluorene	427	304	209	466	414-570	392.5-558.5
Pentachlorophenol	820	713	443	1660	591-1339	410.8-1357.2
Phenanthrene	1823	869	368	1646	1270-1966	995.6-1903.4
Anthracene	488	289	177	441	373-471	377.4-472.2
Fluoranthene	1406	766	584	1505	1060-1500	1021.3-1591.9
Pyrene	1140	769	592	1318	744-1322	643.7-1278.7
Benzo(a)anthracene	179	135	245	231	214-290	203.9-294.9
Chrysene	141	162	319	272	271-323	265-356
Benzo(b,k)fluoranthene	51.3	43.3	135	77.0	130-174	126.3-185.9
Benzo(a)pyrene	39.5	39.6	36.4	66.8	80.1-114	77.5-117.5

The extraction of 0.2 g of soil was with a static extraction of 7.5 minutes and a dynamic extraction of 45 minutes at 210 atmospheres at 80°C. A one gram sample was extracted using carbon dioxide with 10% methanol. The static extraction time was lengthened to 10 minutes, with a dynamic extraction of 45 minutes at 80°C and 300 atm. Lopez-Avila collected the extracted compounds by inserting the restrictor in a vial containing either methanol or methylene chloride. The recoveries are the sum of nine extractions over a time period of 270 minutes. The use of the Accutrap did not improve the recovery over that obtained using the fused silica restrictors. For most of the compounds, the highest recoveries were obtained by Soxhlet extraction. Low recoveries were obtained for late-eluting PNAs using the fused silica restrictor and the Accutrap™. More work needs to be done to improve the extraction recoveries of these compounds.

Initial extraction of NIST SFE Round Robin Sediment A exhibited the same problems as the Fisher Soil sample, i.e., restrictor clogging. Extraction of Sediments A and B and the Air Particulate sample did not result in restrictor plugging using the procedure for Base/neutral compounds.

EXTRACTION OF TEST SOIL USING ACCUTRAP™

Test soil (see sample matrices) was first extracted using the cryogenic trap and carbon dioxide with ten percent methanol to determine its chromatographic characteristics and suitability for spiking. The resulting extracts were relatively clear and had few chromatographic peaks. The soil was then spiked with 100 uL of MOO1E and extracted at 60°C at 330 atmospheres, a density of 0.83 g/mL or roughly that of pentane. The extract was adjusted to 2.0 mL for analysis with the Finnigan GC/MS. The results are summarized as part of Table 7.

The soil was also spiked with Z014GR, a mixture of 17 Polynuclear Aromatic Hydrocarbons, of which all but carbazole were analyzed. The extractions took place at 40°C at 390 atmospheres, a density of 0.95 g/mL, with a 10 minute static extraction and a 45 minute dynamic extraction. Due to the volatile nature of some of the compounds, flow was restricted during the first few minutes of dynamic extraction. The remainder of the dynamic extraction was at one mL/min. Analysis was by GC/MS.

In Table 7, the test soil extractions are compared with diatomaceous earth extractions obtained using the fused silica restrictor, a modifier of 1:1 methanol-methylene chloride and SFC grade carbon dioxide. Generally, test soil extraction recoveries were lower and standard deviations were higher when compared to the diatomaceous earth results. This is to be expected since diatomaceous earth is a "clean matrix".

TABLE 7

COMPARISON OF SPIKE RECOVERIES OF MOO1E AND Z014GR ON TEST SOIL USING ACCUTRAP™ WITH SPIKE RECOVERIES ON DIATOMACEOUS EARTH WITH FUSED SILICA RESTRICTOR

Matrix	Test soil		Test soil		Diatom. Earth		Diatom. Earth	
Compounds Avg in % recovery, n = 4	MOO1E ¹ Avg.	s	Z014GR ² Avg.	s	MOO1E ³ Avg.	s	MOO1E ⁴ Avg.	s
Acenaphthene	93.2	5.8	103	3.6	98.1	1.2	83.0	2.1
Acenaphthylene			106	9.1				
Anthracene	92.2	4.2	91.0	6.3	95.5	0.9	94.8	2.1
Benzo(a)anthracene	73.4	10.9	102	2.1	86.3	1.3	76.2	3.5
Benzo(a)pyrene			107	8.3				
Benzo(b,k)fluoranthene			106	24.0				
Benzo(g,h,i)perylene			109	11.0				
Chrysene	69.0	12.9	104	2.9	83.2	2.6	78.7	7.2
Dibenzo(a,h)anthracene	102	11.8	118	12.2	53.8	5.1	77.1	8.2
1,2-Dichlorobenzene	61.3	14.0			93.1	1.3	80.5	8.5
1,3-Dichlorobenzene	75.7	16.6			93.5	1.4	81.1	8.4
Diethyl phthalate	102	14.7			98.4	5.0	85.5	1.6
2,4-Dinitrotoluene	83.9	6.7			98.4	1.4	81.5	0.9

¹ Extracted at 10 min/45 min/60°C/330 atm with 10% Methanol enhanced CO₂.

² Extracted at 10 min/45 min/60°C/390 atm with 10% Methanol enhanced CO₂.

³ Extracted at 7.5 min/45 min/80°C/210 atm with SFC grade CO₂ fused silica restrictor.

⁴ Extracted at 7.5 min/45 min/80°C/250 atm with SFC grade CO₂ using teflon tube with restrictor.

Matrix	Test soil		Test soil		Diatom. Earth		Diatom. Earth	
Compounds Avg in % recovery, n = 4	MOO1E ¹ Avg.	s	Z014GR ² Avg.	s	MOO1E ³ Avg.	s	MOO1E ⁴ Avg.	s
Fluoranthene			96.0	4.6				
Fluorene	97.1	6.1	97.0	5.0	96.1	1.6	86.7	1.6
Hexachlorobenzene	93.1	6.5			93.8	1.4	81.7	5.1
Hexachlorobutadiene	76.5	5.8			94.3	1.1	81.9	7.2
Indeno(1,2,3-cd)pyrene			124	20.6				
Naphthalene	87.4	6.4	111	6.9	94.4	1.3	82.9	7.4
Bis(2-chloroethoxy)methane	87.0	4.8			96.3	1.8	86.5	7.9
Phenanthrene			95.0	7.1				
Pyrene	92.6	5.4	97.0	4.6	90.7	.8	77.8	2.3

¹ Extracted at 10 min/45 min/60°C/330 atm with 10% Methanol enhanced CO₂.

² Extracted at 10 min/45 min/60°C/390 atm with 10% Methanol enhanced CO₂.

³ Extracted at 7.5 min/45 min/80°C/210 atm with SFC grade CO₂ fused silica restrictor.

⁴ Extracted at 7.5 min/45 min/80°C/250 atm with SFC grade CO₂ using teflon tube with restrictor.

CONCLUSIONS

The use of diatomaceous earth as the matrix for our initial extractions proved very beneficial. As a "clean" simple substrate, it could be expected to yield the spiked compounds easily, as we did not have to deal with the complexities found in "native" pollutants¹². However, the problems that were encountered in extracting various high molecular weight compounds from diatomaceous earth were also encountered in the contaminated soil analyzed in this study.

The use of an unheated fixed diameter restrictor in conjunction with the extraction of the high molecular weight compounds generated problems with restrictor plugging. Attempts to overcome this included: lengthening the restrictor so that a measured amount could be cut off after each extraction; removing the clogged portion; placing the collection vial in warmed water; putting the restrictor in a copper tube which was heated with electrical heating tape; and placing the restrictor in a teflon tube. By increasing the extraction time, the useful life of the restrictor was increased.

FUSED SILICA RESTRICTOR

Generally, the 40 micron restrictor when bubbling into the collection solvent gave extremely good recoveries and standard deviations when using diatomaceous earth as shown. Problems appeared when extracting mixtures of various classes of semi-volatile compounds as shown in Table 7. The conditions which produced excellent recoveries and standard deviations for most compounds were not adequate to extract high-molecular weight compounds. Conditions which produced better recoveries for high molecular weight compounds resulted in lower recoveries and higher standard deviations for the other compounds.

CRYOGENIC TRAP

The cryogenic trap posed its own set of problems. The glass beads in the trap were not sufficiently adsorbent to capture the volatile compounds, such as the dichlorobenzenes, so they literally blew through the collection system. The system was not configured to have the carbon dioxide pass through a solvent to collect these compounds. However, the trap did hold the heavier compounds until desorption. The adjustable restrictor has proven difficult to operate as it works by crimping PEEK^R manually and this has not proven to be a reproducible method to regulate the flow rate. The flow rate was that from the pump and not the flow through the restrictor. After changing the collection system to incorporate the bubbling of the gases through a solvent, we discovered that the

stainless steel tube frosted up and there was a real chance that melting water would find its way through the septum during desorption. The water was trapped by an absorbent material placed around the stainless steel tubing.

GENERAL CONCLUSIONS

Results obtained using the fused silica restrictor collection system or AccutrapTM gave acceptable QC data when compared to EPA method 625. The results produced by the modified AccutrapTM seemed to produce the best overall recoveries when working with the complex matrix of soils. This configuration demonstrated the capability of recovering both volatile and high-molecular weight compounds which would be a necessary condition for routine laboratory soil analysis. It was not subject to the frequent clogging encountered with the fused silica restrictor. However, the fused silica restrictor gave better recoveries for certain compounds, but it was not as rugged as the AccutrapTM system. At this time, there is no single set of SFE conditions to extract the entire class of semi-volatile compounds tested. Avenues of future inquiry should explore methods that result in better recoveries of all classes of semi-volatile compounds in the extraction of soils. These include: increasing the density of the supercritical CO₂ to emulate the solubility parameters of other common solvents; mixing the soil with diatomaceous earth to determine if this allows better penetration of the carbon dioxide during static and dynamic extractions; using a different adsorbent in the cryogenic trap, such as C-18; continuing to investigate the roles of the size of the delivery tube (would the smaller ID stainless steel tube allow the recovery of volatile and high-molecular weight compounds, without the necessity to vary the flow rate during the extraction); investigating the use of different modifiers and increasing the sample size to be more reproducible and afford more sensitivity.

Supercritical fluid extraction has been shown to be accurate and precise in the extraction of semi-volatile compounds from solid matrices. More work needs to be done to improve the extraction process and test other types of solid matrices.

APPENDIX A STANDARD PREPARATION

ACID/BASE NEUTRAL MATRIX SPIKE SOLUTION

BASE/NEUTRAL SPIKE Combined vial 5000 ug/mL of:

1,4-Dichlorobenzene	CAS 106-46-7
N-Nitroso-di-n-propylamine	CAS 621-0-7
2,4-Dinitrotoluene	CAS 121-1402
Di-n-butyl Phthalate	CAS 84-74-2
Acenaphthene	CAS 83-32-9
1,2,4-Trichlorobenzene	CAS 120-82-1
Pyrene (1000 ug/mL-2 mL)	CAS 129-00-0

Diluted 1.5 mL to 15 mL with methanol, approx 10x dilution(500 ug/mL)

ACID SPIKE Combined 2 vials 5000 ug/mL of:

4-Nitrophenol	CAS 100-02-7
2-Chlorophenol	CAS 95-57-8
Phenol	CAS 108-95-2
4-Chloro-3-methyl phenol	CAS 59-50-7
Pentachlorophenol	CAS 87-86-5

Diluted contents of 2 vials of each to 15 mL with methanol, approximately a 5x dilution or 1000 ug/mL stock

REFERENCE SOLUTION: 100 uL of combined stock diluted to 1.0 mL volumetrically.

TARGET COMPOUND CONCENTRATION: 100 ug/mL for acid compounds. 50 ug/mL for base/neutral compounds, except for pyrene, which is 20 ug/mL.

SPIKE: 100 uL of combined stock solutions.

BASE-NEUTRAL MIX 1 MOOID ACCUSTANDARD

Conc. 100, 200 ug/mL in methanol.

REFERENCE SOLUTION: Dilute 100 ul to 1.0 mL volumetrically with MeCl₂.

TARGET COMPOUND CONCENTRATION:

10 ug/mL	Benzo-(b)-fluoranthene
20 ug/mL	Acenaphthylene
	4-Bromophenyl phenyl ether
	Bis(2-chloroethyl) ether
	Bis(2-chloroisopropyl) ether
	1,4-dichlorobenzene
	3,3-dichlorobenzidine
	Dimethyl phthalate
	Di-n-butyl phthalate
	2,6-dinitrotoluene
	Bis (2-ethylhexyl) phthalate
	Nitrobenzene

SPIKE: 100 uL of stock plus 50 uL 1:1 MeCl₂-MeOH.

BASE-NEUTRAL MIX 2 MOO1E ACCUSTANDARD

Conc. 100, 200 ug/mL in methanol

REFERENCE SOLUTION: Dilute 100 uL to 1.0 mL volumetrically with MeCl₂.

TARGET COMPOUND CONCENTRATION:

10 ug/mL	Benzo(a)anthracene
	Chrysene
	Dibenzo(a,h)anthracene
20 ug/mL	Acenaphthene
	Anthracene
	1,2-dichlorobenzene
	1,3-dichlorobenzene
	Diethyl phthalate
	2,4-dinitrotoluene
	Fluorene
	Hexachlorobenzene
	Hexachlorobutadiene
	Naphthalene
	Bis(2-chloroethoxy) methane
	Pyrene

SPIKE: 100 uL stock plus 50 uL 1:1 MeCl₂-MeOH

BASE-NEUTRAL MIX 3 MOO1F ACCUSTANDARD

Conc. 100, 200 ug/mL in methanol.

REFERENCE SOLUTION: Dilute 100 uL of stock to 1.0 mL volumetrically with MeCl₂.

TARGET COMPOUND CONCENTRATION:

10 ug/mL	Fluoranthene
20 ug/mL	Butyl benzyl phthalate
	2-chloronaphthalene
	1,2-diphenylhydrazine
	Hexachlorocyclopentadiene
	Hexachloroethane
	Isophorone
	N-nitroso-di-n-propylamine
	N-nitrosodiphenylamine
	Phenanthrene
	1,2,4-Trichlorobenzene

SPIKE: 100 uL stock plus 50 uL 1:1 MeCl₂-MeOH.

COMBINED BASE NEUTRAL MIX 4 MOO1G/NAPHTHALENE ACCUSTANDARD

MOO1G Conc. 100, 200 ug/mL in 1:1 MeCl₂-MeOH

Naphthalene CAS 91-20-3 Conc. 5000 ug/mL

100 uL added to 2.0 mL to 2.0 mL MOO1G yielding a 250 ug/mL concentration.

REFERENCE SOLUTION: Diluted 100 uL of combined stock to 1.0 mL volumetrically with MeCl₂.

TARGET COMPOUND CONCENTRATION:

10 ug/mL	Benzo(k)fluoranthene
	Indeno(1,2,3-cd)pyrene
	Benzo(g,h,i)perylene
	Benzo(a)pyrene
20 ug/mL	Benzidine
	2-chloroethyl vinyl ether
	4-chlorophenyl phenyl ether
	Di-n-octyl phthalate
	N-nitrosodimethylamine
25 ug/mL	Naphthalene

SPIKE: 100 uL of combined stock plus 50 uL 1:1 MeCl₂-MeOH.

PHENOL MIX ACCUSTANDARD MOO1P

Conc. 500 TO 2500 ug/mL in methanol

REFERENCE SOLUTION: Diluted 50 uL to 1.0 mL volumetrically in MeCl₂.

TARGET COMPOUND CONCENTRATION:

25 ug/mL	2-Nitrophenol
	Phenol
	2-Chlorophenol
	2,4-dichlorophenol
	2,4-dimethylphenol
75 ug/mL	2,4-dinitrophenol
	2,4,6-Trichlorophenol
125 ug/mL	4-Chloro-3-methylphenol
	4-Nitrophenol
	2-Methyl-4,6-dinitrophenol
	Pentachlorophenol

SPIKE: 50 uL stock solution plus 50 uL of Methanol.

SURROGATE MIX EPA QUALITY ASSURANCE MATERIALS BANK

2,4,6-TRIBROMOPHENOL CAS 118-79-6 (5000 ug/mL) in
methanol (2 ml)

ORGANIC SURROGATE MIX (5000 ug/mL) in methylene
chloride (1 ml)

p-TERPHENYL-D14 CAS 1718-51-0 (5000 ug/mL) in THF (2 mL)

Diluted volumetrically to 5.0 mL resulting in a conc of
2000 ug/mL for 2,4,6-Tribromophenol and p-Terphenyl-
D-14 and 1000 ug/mL for the Organic Surrogate Mix.

REFERENCE SOLUTION: Diluted 100 uL stock to 1.0 mL.
volumetrically with MeCl₂.

TARGET COMPOUND CONCENTRATION

40 ug/mL	2-Fluorophenol
	D ₅ -Phenol
	D ₅ -Nitrobenzene
	2-Fluoro-1,1'-biphenyl
200 ug/mL	2,4,6-Tribromophenol
	D ₁₄ -p-terphenyl

SPIKE: 100 uL stock plus 50 uL 1:1 MeCl₂-MeOH.

COMBINED TOXIC SUBSTANCE MIX 2 ACCUSTANDARD Z-014E

Conc. 2000 ug/mL in methanol

BENZIDINES MIX ACCUSTANDARD Z-014F

Conc. 2.0 mg/mL (2000 ug/mL) in methylene chloride

REFERENCE SOLUTION: 100 uL of both Z-014E and Z-014F were
diluted volumetrically to 5.0 mL with methylene
chloride.

TARGET COMPOUND CONCENTRATION:

40 ug/mL	Aniline
	Benzyl alcohol
	4-Chloroaniline
	Dibenzofuran
	2-methylnaphthalene
	2-Nitroaniline
	3-Nitroaniline
	4-Nitroaniline
	Benzidine
	3,3'-Dichlorobenzidine

SPIKE: 100 uL of combined stock plus 50 uL of 1:1 MeCl₂-MeOH
diluted to 5.0 mL with MeCl₂ after extraction.

PAH MIX Z014GR

Conc. 2.0 mg/mL of each in MeCl₂-benzene (50:50)

REFERENCE SOLUTION: diluted 100 uL of stock solution to 2.0 mL volumetrically with methylene chloride.

TARGET COMPOUND CONCENTRATION

40 ug/mL Acenaphthene
Acenaphthylene
Anthracene
Benzo(a)anthracene
Benzo(a)pyrene
Benzo(b)fluoranthene
Benzo(g,h,i)perylene
Benzo(k)fluoranthene
Chrysene
Dibenzo(a,h)anthracene
Fluoranthene
Fluorene
Indeno(1,2,3-cd)pyrene
Naphthalene
Phenanthrene
Pyrene
Carbazole (not analyzed)

SPIKE: 100 uL stock with no modifier

TOXAPHENE CAS 8001-35-2

CONC 1000 ug/mL +/-100 ug/mL in methanol

REFERENCE SOLUTION: 200 uL of stock diluted to 1.0 mL volumetrically with 1:1 methylene chloride yields a 200 ug/mL solution.

SPIKE: 200 uL of stock plus 50 uL 1:1 MeCl₂-MeOH.

CHLORDANE CAS 57-74-9

Conc. 1000 ug/mL +/- 100 ug/mL

REFERENCE SOLUTION: 50 uL stock diluted to 1.0 mL volumetrically with methylene chloride yields 50 ug/mL solution.

SPIKE: 50 uL of stock plus 50 uL of 1:1 MeCl₂-MeOH.

PESTICIDE MIX ACCUSTANDARD MOO1H/M-608-1

Conc. 100 TO 600 ug/mL in methanol

INTERMEDIATE REFERENCE STANDARD: Diluted 100 uL to 1.0 mL volumetrically with methylene chloride.

TARGET COMPOUND CONCENTRATION (Intermediate):

10 ug/mL	Aldrin	
	alpha-BHC	
	beta-BHC	
	gamma-BHC	
	delta-BHC	
	Heptachlor	
	Heptachlor epoxide	
20 ug/mL	p,p'-DDE	
	Dieldrin	
		Endosulfan I
	Endosulfan II	
	Endrin	
60 ug/mL	p,p'-DDT	
	p,p'-DDD	
	Endosulfan sulfate	
	Endrin aldehyde	

SPIKE: 100 uL of intermediate reference solution plus 50 uL of 1:1 MeCl₂-MeOH diluted to 1.0 mL.

PCB 1260 CAS 11096-82-5

Conc. 1000 ug/mL +/-100 ug/mL in iso-octane

REFERENCE SOLUTION: Diluted 50 uL to 1.0 mL volumetrically with methylene chloride.

SPIKE: 50 uL of stock plus 50 uL of 1:1 MeCl₂-MeOH.

PCB 1254 CAS 11097-69-1

Conc. 1000 ug/mL in iso-octane

REFERENCE SOLUTION: 50 uL stock to 1.0 mL volumetrically with 1:1 MeCl₂-MeOH

SPIKE: 50 uL of stock plus 50 uL of methylene chloride

PCB 1248 CAS 12672-29-6
Conc 5000 ug/mL +/- 500 in iso-octane

REFERENCE SOLUTION: 10 uL of stock diluted to 1.0 mL
volumetrically with methylene chloride yields
50 ug/mL solution.

SPIKE: 10 uL of stock plus 50 uL of MeCl₂-MeOH.

PCB 1242 CAS 53469-21-9
Conc. 3000 ug/mL +/- 300 in iso-octane

REFERENCE SOLUTION: Diluted 20 uL of stock to 1.0 mL
volumetrically with methylene chloride yields
a 60 ug/mL solution.

SPIKE: 20 uL of stock plus 50 uL of 1:1 MeCl₂-MeOH.

PCB 1232 CAS 11141-16-5
Conc. 5000 ug/mL +/- 500 in methanol

REFERENCE SOLUTION: Diluted 10 uL to 1.0 mL volumetrically
with methylene chloride yielding a 50 ug/mL solution.

SPIKE: 10 uL of stock plus 50 uL of 1:1 MeCl₂-MeOH.

PCB 1221 CAS 11104-28-2
Conc. 1000 ug/mL +/-100 in iso-octane

REFERENCE SOLUTION: Diluted 50 uL to 1.0 mL volumetrically
with methylene chloride yielding a 50 ug/mL solution.

SPIKE: 50 uL stock plus 50 uL of 1:1 MeCl₂-MeOH.

PCB 1016 CAS 12674-11-2
CONC. 5000 ug/mL +/-500 in iso-octane

REFERENCE SOLUTION: Diluted 10 uL to 1.0 mL volumetrically
with methylene chloride yielding a 50 ug/mL solution.

SPIKE: 10 uL plus 50 uL of 1:1 MeCl₂-MeOH.

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