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Supercritical Fluid Extraction of Polycyclic
Aromatic Hydrocarbons from Soil Matrices

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Mark S. Ruppel

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of the requirements for

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SUPERCRITICAL FLUID EXTRACTION OF POLYCYCLIC AROMATIC HYDROCARBONS FROM SOIL MATRICES

By

Mark S. Ruppel

A THESIS

Submitted to
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ABSTRACT

SUPERCRITICAL FLUID EXTRACTION OF POLYCYCLIC AROMATIC HYDROCARBONS FROM SOIL MATRICES

By

Mark S. Ruppel

The primary objective of this thesis is to analyze the effectiveness of supercritical fluid extraction (SFE) for the determination of polycyclic aromatic hydrocarbons (PAHs) in soil. Ottawa Sand is used as an inert material to extract PAHs from using CO2 modified with 1% methylene chloride. Soil A is a clean, horizon A soil that is used to test the extractability of spiked PAHs. Soil C was obtained from an actual coal tar contaminated site and is used to test the extractability of native PAHs. It was found that the solubility of PAHs in the supercritical fluid was not a limiting factor. The extraction of the native PAHs in Soil C was most likely due to diffusion limitations. Addition of an organic modifier had a large effect on the recovery of native PAHs from Soil C.

Dedicated to:

my high school chemistry teacher, Mr. Schultz

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TABLE OF CONTENTS

APPENDICES

REFERENCES:

LIST OF TABLES

LIST OF FIGURES

CHAPTER 1

INTRODUCTION

The environmental industry relies on dependable quantitative analytical methods to allow for the successful protection and remediation of the environment. When determining contaminant levels, some of the most difficult matrices with which to work are soil, sediment, and other solid material. Conventional methods, such as sonication and soxhlet extraction techniques, are commonly used for the analysis of solid material, and have been approved by the United States Environmental Protection Agency (US EPA) for this use. Another method that has been gaining attention is supercritical fluid extraction (SFE). SFE is an analytical tool that can potentially save time, reduce hazardous waste, and be less expensive than conventional extraction techniques. There is a need for more research concerning SFE to determine the potential and limitations of this technique for applications in the environmental industry.

1.1 Objective

The primary objective of this thesis is to analyze the effectiveness of SFE for the determination of polycyclic aromatic hydrocarbons (PAHs) in soil. PAHs can be spiked onto soil to develop extraction methods for the quantitation of contaminants, but they may not have the same matrix interaction characteristics as native PAHs. The term native refers to the contaminants present in the soil as a result of environmental pollution. In the case of coal tar contamination, the native PAHs have had time to interact with the soil matrix and are likely to be more difficult to extract quantitatively than fresh spike PAHs. Extraction of spiked PAHs will usually result in an overestimation of the extraction method efficiency. The research results should indicate whether or not the SFE information gained from spiked PAH extractions can be used as a template for SFE method development for native PAHs. The above objective was obtained by the following:

- Extracting PAHs from soil using different SFE temperatures and pressures
- Extracting PAHs from various types of soil
- Investigating the aging process and its effect on extraction efficiency
- Conducting experiments which may indicate whether solubility or diffusivity are limiting physical parameters
- Comparing the efficiency of SFE vs. conventional methods for the analysis of native PAHs is soil

1.2 Organization of Thesis

The first part of this thesis describes the general background and technical information. Chapter 2 provides information on various applications of supercritical fluids and research that has been done to investigate the parameters that influence SFE efficiency.

The second part of the thesis describes the experiments performed and summarizes the experimental findings. The experimental methods and equipment used for this research are described in Chapter 3. The extraction of PAHs from Ottawa Sand using SFE is described in Chapter 4. Chapter 5 contains information about the SFE of PAHs that were spiked onto a clean soil. The SFE of native PAHs from a coal tar contaminated soil is described in Chapter 6. Chapter 7 summarizes the experimental findings and presents recommendations for further work.

CHAPTER 2

BACKGROUND

The applications of supercritical fluid technology have been expanding rapidly into many new areas. There have been new and continuing developments in the application of SFE for pharmaceuticals, polymers, chromatography, extractions, coal and petroleum processing, and environmental remediation (Kiran, 1993). In the environmental area, the applications for supercritical fluids range from the actual clean-up of contaminated soils to the laboratory analysis of contaminants (1-45). Supercritical fluid chromatography (SFC) and supercritical fluid extraction (SFE) have been used both independently and together as tools for analytical analyses. SFE is a technique that can save both time and money while reducing the amount of waste generated in the analytical laboratory.

A supercritical fluid is a pure substance that is above its critical temperature and pressure. When a substance is above its critical point it exhibits physical properties of both a liquid and a gas. In choosing the appropriate solvent for an extraction process two important points have to be considered (Engelhardt, 1991):

- *Selectivity*: In order to avoid additional separation processes, the solvent should extract only the components of interest.
- *Suitability*: The extractor should be removable without loss of the components of interest. In other words, there should be large differences in the volatility of the

extract and the extractor. Also, there should be no thermal strain, excessive heating, applied to the extracted solvents.

Both of the above criteria can be met by application of supercritical fluids. A table of physical properties of commonly used substances for the extraction of analytes from various matrices are shown in Table 2.1. One of the most common supercritical fluids used for analytical applications is carbon dioxide (CO_2). The critical temperature and pressure for CO_2 are 31 °C and 73 atm. Supercritical fluid extraction (SFE) takes advantage of the solvating power of substances that are in a supercritical state. The properties of viscosity and solute diffusivity are more typical of gases than liquids and contribute to improved mass transfer for solutes in the supercritical state, and therefore, increase extraction rates (Snyder, 1992).

SFE can be used as an effective analytical tool for the analysis of environmental contaminants in soil matrices, but care must be taken in developing the SFE method. The difficult part of applying SFE as an analytical tool is determining how the variables will effect the extraction performance. The control parameters must be modified to accommodate the analyte of interest and the matrix. Temperature and pressure are two important parameters that need to be optimized in order for SFE to be effective. Changing the temperature and pressure will change the density of the supercritical fluid, which affects the solvating power of the fluid, thereby affecting the performance of SFE (Langenfeld et al., 1993). There has been a great deal of research done to investigate the parameters that effect the performance of SFE. Research investigations on other parameters such as the matrix, extraction modifiers, and analyte collection have also been conducted (1-45). The rest of this chapter contains information about research done on these parameters. The research done for this thesis addresses these parameters and how they affect the SFE of PAHs from soil.

Table 2.1

Physical Properties of Commonly Used Supercritical Fluids

	T_c (°C)	P_c (atm)	Dipole Moment (debyes)	Extracted Analytes	Matrices
CO_2	31	73	0	Fat, PCBs, pesticides, PAHs, vitamins, polymer additives, hydrocarbons, halocarbons, aromatics	Meats, soil, Tenax, PUF, polymers, air particulates, sedimentatry rock
CO_2/MeOH	41*	74*	---	Pesticides, priority pollutants (semivolatiles)	Soils, plant tissue, sludge
N_2O	37	72	0.2	Amines, dioxin, dibenzofurans	Soils, fly ash
$CHClF_2$	96	49	1.4	PCBs, PAHs	Sludge, soils
C_2H_6	32	48	0	PAHs	Sediment, fly ash, air particulates

Tc - Critical Temperature Pc - Critical Pressure * - Modified with 5% (v/v) methanol
Table of information taken from Snyder, 1992

2.1 Temperature and Pressure

Changes in temperature and pressure during SFE will result in variations in the density of the supercritical fluid, thereby affecting the solvation power of the supercritical fluid. The solubility of a solute in a supercritical fluid will increase with increasing density (Snyder, 1992). An increase in pressure will increase the density of a supercritical fluid and it is possible to achieve densities that are higher than those of some liquids (Snyder, 1992). Temperature has the opposite effect on density; increasing the temperature will decrease the density, therefore decreasing the solvation power. However, higher temperatures may be appropriate for analytes with low vapor pressures (Snyder, 1992). There has been a significant amount of research done on how temperature and pressure affect the extraction of analytes during SFE. Various results have been reported on a variety of compounds and matrices.

Lopez-Avila et al. (1992) used a standard reference soil, (SRS103-100) purchased from Fisher Scientific, for experiments to determine the effect of temperature and pressure on the recovery of TPH. The SFE conditions tested included three pressures, 150, 250, and 350 atm, and two temperatures, 55°C and 90°C; results are shown in Table 2.2. The recovery of total petroleum hydrocarbons (TPH) from the PAH contaminated soil increased with an increase in pressure. However, the recovery appears to be inversely dependent on temperature (Lopez-Avila et al., 1992).

Table 2.2

Hydrocarbon Recovery from Soil SRS103-100

	150 atm	250 atm	350 atm
55 °C	77%	96%	98%
90 °C	---	85%	82%

* 150 atm and 55 °C - extraction time 120 minutes
* All other conditions - extraction time 60 minutes

The higher pressure resulted in a shorter extraction time required to obtain quantitative results, however the increase in temperature actually lowered the extraction efficiency of the TPHs from the test soil.

Ho and Tang (1992) conducted SFE experiments using a 2^3 factorial design to test the effect of three factors at two levels. The three factors tested were temperature, pressure, and extraction time. The two levels tested for each factor were temperatures of 40 °C and 80 °C, pressures of 100 atm and 300 atm, and extraction times of 10 minutes and 40 minutes. There was also a middle value tested: 60 °C, 200 atm, and 20 minute extraction time. A total of 29 compounds, including PAHs and organochlorine pesticides, was extracted from a commercially available liquid-solid extraction cartridge. The extraction cartridge contained 500 mg of silica gel with chemically bonded C18 octadecyl groups. Pressure was found to be the most influential variable that affected the recoveries of all the compounds. An increase in the pressure resulted in an increase in recovery of the analytes. At moderate pressure conditions from 350 to 400 atm and extraction time from 20 to 35 minutes, most of the analytes were quantitatively recovered from the C18 cartridge (Ho et al., 1992). Temperature affected the recoveries of only a few compounds. It had a positive effect on compounds such as chrysene, benzo[a]anthracene, and phenanthrene. Table 2.3 contains a list of 12 PAHs and the specific variables that had a statistically significant effect on the recovery of each individual PAH.

Table 2.3

Experimental Conditions for the Low and High Recoveries of 12 PAHs

Compound	Amount Fortified (µg)	Low and High Mean Recovery (%)	Experimental Conditions (P,T_e,T_i)*
Naphthalene	25	low 19.97	LHL
		high 83.5, 84.7	HLH, MMM
Acenaphthylene	25	low 0.26	LHL
		high 94.56	HLH
Acenaphthene	25	low 0.3, 0.5	LLL, LHL
		high 101	MMM
Fluorene	25	low 0.10, 0.18	LLL, LHL
		high 100	HLL,HHL,HLH,HHH,MMM
Phenanthrene	25	low 0.24, 0.25	LLL, LHL, LHH
		high 93.8	MMM
Anthracene	25	low 0.0	LLL, LHL, LHH
		high 93.7	MMM
Fluoranthene	25	low 0.0	LLL, LHL, LHH
		high 85.0	HHL, HHH, MMM
Pyrene	25	low 0.0	LLL, LHL, LHH
		high 89.04	HHH
Chrysene	25	low 0.03, 0.25	LLL, LHL, LHH
		high 74.99	HHH
Benzo[a]anthracene	50	low 0.0	LLL, LLH, LHH
		high 70.03	HHH
Benzo[b]fluoranthene	25	low 0.0	LLL, LHL, LHH
		high 87.4	HHH
Benzo[a]pyrene	50	low 0.0	LLL,LLH,LHL,LHH,HLL,HHL
		high 31.0	HHH

*For PT_eT_i (LHL) represents low level for pressure; high level for temperature; and low level for extraction time, and (MMM) represents middle level for all factors.

**Table of data from Ho and Tang (1992)

Extractions were performed by Langenfeld et al. (1993), on three different materials to recover PCBs from river sediment and PAHs from soil and air particulates. The extractions were carried out using two different temperatures, 50 °C and 200 °C. At 50 °C, pressures of 350 atm and 650 atm were tested, and at 200 °C, pressures of 150 atm, 350 atm, and 650 atm were tested. At 50 °C, raising the pressure from 350 atm to 650 atm had no effect on the recovery efficiency for any of the samples. The river sediment (NIST SRM 1939) was contaminated with PCBs and efficient extractions were obtained only when the temperature was raised to 200 °C. Quantitative extractions were obtained in 40 minutes, regardless of the temperature, for a soil highly contaminated with PAHs ("U.S. EPA Certified" PAH Contaminated Soil, Lot No. AQ103, Fisher Scientific). In contrast, a PAH contaminated air particulate sample (NIST SRM 1649) showed an increase in recovery when both higher temperatures and pressures were used. Their results indicate that temperature is more important for achieving high extraction efficiencies than pressure when the pollutant molecule interaction with the sample matrix is strong. In contrast to the urban air particulates, PAHs from the highly contaminated soil were quantitatively extracted using mild temperatures (50 °C), indicating that these PAHs are not strongly bound to this sample matrix and very little energy is required to desorb them from the sample matrix (Langenfeld, 1993). There was a large difference in the concentration of PAHs in the air particulate sample and the soil sample. The PAHs on the soil were 33 to 200 times more concentrated than those on the air particulates. When there is a low concentration of PAHs, the percentage of PAHs bound to strong reactive sites will be high. This is because the reactive sites outnumber the PAHs available. A lower percentage of PAHs will be bound to strongly reactive sites when a high concentration of PAHs is present. In this case the PAHs outnumber the available reactive sites. Also, the processes by which the two samples were contaminated are different. The PAHs were spilled onto the soil, while the air particulates and PAHs are likely to be formed together with soot during the combustion of vehicular fuels (Langenfeld et al., 1993).

Levy et al. (1993), came to a similar conclusion about analyte association with the matrix and the variables that affect efficient recovery using SFE. They state that, "SFE strategies are dependent on the solubility or kinetic limitations, which relates to the level of association of the target analytes with the respective heterogeneous sample matrices". SFE was performed on a soil contaminated with PAHs. The temperature held constant at 75 °C during SFE. Three different pressures (250 atm, 350 atm, and 450 atm) were investigated to determine their affect on analyte recovery. All three pressures gave acceptable results for the lower molecular weight PAHs, but the highest pressure resulted in the best efficiency for extracting all of the PAHs.

Levy et al. (1993) investigated the affect of temperature on extraction efficiencies. Marine sediment and a river sediment contaminated with PAHs were used for this experiment. Pressure was held constant at 475 atm while the temperature used for the extractions was varied. The marine sediment was extracted at 65 °C, 80 °C, and 150 °C. The river sediment was extracted at 40 °C, 100 °C, and 150 °C. The best recoveries were obtained using the highest temperature of 150 °C for both the marine and river sediments.

It was determined from the results that raising the extraction temperature and pressure will increase efficiencies for the extraction of PAHs from soils and sediments.

Hawthorne and Miller (1986) investigated the extraction efficiency for PAHs on Tenax-GC traps. They were able to quantitatively recover PAHs from Tenax-GC traps using CO_2 with a 15 minute extraction time, a temperature of 45 °C, and a pressure of 200 atm. Myer et al. (1992) investigated the effect of temperature on the extraction efficiency of PAHs from soil. The pressure was held constant at 350 atm and the total extraction time was 120 minutes. The extract was collected in three fractions; the first after 30 minutes at 90 °C, the second after another 30 minutes at 90 °C, and the final fraction after 60 minutes at 120 °C. The more volatile PAHs were recovered during the first of the three fractions collected, at 85-125% efficiency. The higher molecular weight PAHs required longer extraction time, but were all quantitatively recovered (>95%) by the end of the second fraction, except benzo[a]pyrene. Furton and Rein (1992) noted that SFE of small quantities of analytes from solid matrices may be diffusion controlled rather than solubility limited, but the relative importance of diffusion and solubility are highly dependent on the analyte and matrix being extracted.

The conclusion drawn from the literature reviewed is that the effects of temperature and pressure are highly dependent on the matrix from which the analyte is being extracted and on the type of analyte being extracted. TPHs were extracted efficiently from soil SRS103-100 at low temperatures and high pressures. PAHs can be extracted from sand, glass beads, Tenax, and similar non-reactive materials with moderate temperatures and high pressures. In this case, the limiting factor is the solubility of the

PAHs, if present at high concentrations. If the PAHs are on soil, river sediment, or air particulates it may be necessary to increase the temperature of the extraction to desorb the analyte from the matrix, because in this case the PAHs may be diffusion limited.

2.2 Matrix Effects

The matrix from which the sample is being extracted plays a critical role in the development of an effective SFE method. To accomplish a rapid extraction, the optimization process should consist of two steps. First, the extraction conditions of pressure, temperature, and/or co-solvent need to be optimized to ensure that retention of the target analytes on the matrix does not occur; this is termed the elution process (Pawliszyn, 1993). The key to this first step is to ensure that the solubility of the analyte in the supercritical fluid is high compared to that in the matrix. This will prevent the analyte from partitioning back onto the matrix at the fluid-matrix interface. Second, further optimization must be made to ensure that rapid mass transfer of the analytes from the matrix to the fluid can occur; this is termed the desorption process (Pawliszyn, 1993). The analyte must be desorbed from the matrix and brought into contact with the fluid-matrix interface. The more that is known about the analyte interactions with the matrix the easier it will be to make an educated estimate about the optimization procedure.

Consideration must be made as to how the analyte is associated with the matrix. If the analyte is at or near the surface of the matrix, the supercritical fluid will have adequate contact with the analyte and solubility will most likely be the limiting factor. If the analyte is entrained within crevices, voids, or adsorbed within the matrix, the extraction may take more time and be dependent upon desorption. When analytes are encapsulated within the matrix only a small fraction of solvent will reach these sites and the sample may need to be broken up or disrupted so that the solvent can reach the analyte (Pipkin, 1992). Soil and air particulate matter contaminated with PAHs are good examples of how an analytes association with the matrix is dependent on the type of media that is contaminated. PAH contamination of a soil most likely occurs by spilling or placing the contamination on the existing soil resulting in the PAHs being associated with the surface of the soil particle. As air particulates form they are contaminated by PAHs from vehicular exhaust and the PAHs are entrained within the particle and on the surface (Langenfeld, 1993).

The presence of water also affects the extraction of analytes. Water may help or hinder the process depending on whether the analyte is polar or nonpolar. Water will help bring polar compounds into solution by competing for active matrix sites, but water may hinder the extraction of nonpolar compounds by sheathing the surface of the matrix and acting as a barrier to supercritical fluid penetration (Pipkin, 1992). The affect of water is also dependent on whether the water is acting as a wetting or non-wetting fluid, which is dependent on the surface of the matrix.

Burford et al. (1993) studied four different spiking procedures to try to mimic the native PAHs in a real world sample, but none of the procedures were completely successful. The interactions of the spiked PAHs and the native PAHs with the matrix were different because of the aging process and solvents used to introduce the spiked PAHs. Although the spiking procedures do not mimic the real world sample, extracting spiked analytes from a matrix can provide useful information. To determine whether the limiting process is elution or desorption a standard solution of selected deuterated PAHs (d-PAHs) can be spiked onto a sample matrix and be extracted along with the native PAHs (Levy, 1992). The native PAHs undergo both the desorption and elution processes, because the contaminants have had time to adsorb and interact with the active sites of the matrix. The spiked d-PAHs will only undergo the elution process. Levy et al. (1992) determined through the above process that extraction efficiencies of PCBs from river sediment and low molecular weight PAHs from urban air particulate matter are primarily limited by the slow desorption process of the analytes from the matrix. The extraction efficiencies of the PAHs and PCBs showed a dependence on the modifier identity, which indicates that either the desorption was enhanced or readsorption was prevented, possibly by competing for active matrix sites. Work conducted by Langenfeld et al. (1993) using the same samples as used by Levy et al. (1992) showed that increasing temperature was much more important than increasing pressure. This supports the argument that desorption is the rate limiting step, because an increase in the temperature would allow the chemical to overcome the energy barrier of desorption. The increase in pressure would have increased the solubility of the analytes, but the pressure increase was not as important as the increase in temperature for improving the extraction efficiency.

2.3 Supercritical Fluids and Modifiers

The main limitation of the most often used supercritical fluid, CO_2, is its limited ability to dissolve polar analytes even at very high densities (Levy, 1992). Supercritical fluid CO_2 is compatible and miscible with a large range of organic modifiers. The addition of organic modifiers to CO_2 can overcome solubility limitations. The simplest method of using modifiers is to add the modifier directly to the sample in the extraction cell. Modifiers can also be mixed into the supercritical fluid by using an additional pump, which may mean mechanically altering the SFE apparatus. An alternative is to buy SFE CO_2 with the modifier already mixed in the tank of CO_2 by the supplier. Scott Specialty Gases, Inc. is one company that will make special order CO_2 tanks with modifiers. The limitation with this last alternative is that the modifier type and concentration cannot be changed.

It is important to ensure that the modifier type and concentration will be suitable for the analysis that is being performed. This will depend on both the analyte of interest and the matrix from which the analyte is being extracted. A common mistake in selecting a solvent modifier for SFE occurs when the selection is based on prior experience with conventional extraction techniques (Pipkin, 1992). A popular modifier that has been used in many SFE experiments is methanol, but this is not considered to be the best modifier for all analytes and matrices. Development of the SFE method should take into account the option of a variety of modifiers that are available for use and the effect of interactions with the matrix that occur with the analyte and modifier. In addition, derivatization reagents are available that can be used to increase extraction efficiencies by competing with the analyte for active sites on the matrix. There is also the option to choose a supercritical fluid besides CO_2 that may be better suited for the extraction process.

Hawthorne and Miller conducted experiments to determine the length of time required to recover PAHs quantitatively from SRM 1650 diesel exhaust particulates using CO_2 and CO_2 modified with 5% MeOH (v/v). The results showed that the supercritical

CO_2/5% MeOH extraction of the PAHs was essentially complete during the first 30 minutes (based on the lack of detectable PAH in the 30 to 60 minute sample), while the extraction using supercritical CO_2 required 90 minutes (Hawthorne, 1986). Ho and Tang showed poor recoveries of high molecular weight PAHs such as chrysene, benzo[a]pyrene, benzo[a]anthracene, and dibenzo[a,h]anthracene when pure supercritical CO_2 was used at moderate pressures for extraction from a C18 cartridge (Ho, 1992). The same high molecular weight PAHs were extracted from an inert matrix (Celite) at pressures of 300 and 400 atm with pure CO_2 and quantitative recoveries were obtained in 15 minutes. They concluded that matrix effects were the cause of the low recoveries of the high molecular weight PAHs from the C18 cartridges. After adding various amounts of methanol to the extraction cartridge the recoveries were greatly improved and satisfactory quantitative recoveries were obtained (Ho and Tang, 1992).

Burford et al., did sequential extractions on samples with pure CO_2 for 30 minutes, followed by CO_2 modified with 10% methanol (v/v) for 30 minutes, and finally by sonication in methylene chloride for an additional 14 hours (Burford, 1993). They found that the alkanes from the samples were extracted almost completely with the pure CO_2, while the native PAHs were found in the pure CO_2, CO_2/methanol, and methylene chloride extracts. The three samples used in this experiment were petroleum waste sludge, SRM 1649 urban air particulate matter, and railroad bed soil. The CO_2/methanol extraction yielded recoveries >85% of the native PAHs from the air particulate matter and railroad bed soil, but not from the petroleum waste sludge. PAH solubility was not the major limiting factor in the extraction process, as the majority of the spiked d-PAHs could be quantitatively recovered (>85%) from the petroleum waste sludge and urban air particulate matter using pure CO_2, whereas only about 25-75% of the native PAHs were recovered (Burford, 1993). It was concluded that the modification of the CO_2 with methanol increased the extraction rates by increasing the polarity of the supercritical fluid and by increasing its ability to interact with the matrix to compete with the analyte for matrix active sites. Langenfeld et al. (1994) tested nine different CO_2 and organic modifier mixtures to recover PAHs from urban air particulate matter. The type of modifier proved to be more important than the modifier concentration for increasing extraction efficiency. Shown in Table 2.4 are the results of the extraction efficiencies of PAHs from air particulate matter for two of the modifiers tested at different modifier concentrations. Although there is no definite theory that explains modifier selection for SFE, it appears that modifiers should be selected on the basis of matrix characteristics and the target analyte (Langenfeld et al., 1994).

The option of using a derivatization agent may be attractive when the analyte of interest is bound tightly to the matrix from which it is being extracted. The derivatization agent has the capability to displace the analyte from the matrix. Hills and Hill added commercially available derivatization reagents to two standard reference materials prior to SFE with CO_2 to evaluate for the analytical determination of PAHs. The addition of hexamethyldisilane (HMDS) and trimethylchlorosilane (TMCS) as a 2:1 mixture was found to enhance the extraction efficiency of PAHs from National Institute of Standards and Technology (NIST) standard reference material (SRM) 1649 and National Research Council of Canada (NRCC) SRM HS-3 (Hills, 1993). Hills and Hill used four sample

Table 2.4

Supercritical Fluid Extraction Efficiencies Using Different Modifiers and Modifier Concentrations

PAH	cert. conc. µg/g	1% CH_2Cl_2	10% CH_2Cl_2	1% toluene	10% toluene
phenanthrene	4.5 (7)	85 (4)	90 (4)	89 (3)	100 (2)
fluoranthene	7.1 (7)	79 (6)	83 (5)	82 (4)	96 (2)
pyrene	7.2 (7)	64 (6)	70 (5)	66 (3)	81 (2)
benz[a]anthracene	2.6 (12)	63 (7)	78 (3)	67 (11)	94 (4)
chrysene + triphenylene	5.3 (5)	62 (10)	77 (2)	59 (6)	90 (4)
benzo[b,k]fluoranthene	8.2 (5)	56 (11)	87 (8)	60 (3)	108 (8)
benzo[a]pyrene	2.9 (17)	18 (16)	41 (4)	20 (8)	60 (8)
benzo[ghi]perylene	4.5 (24)	6 (8)	18 (9)	6 (3)	26 (11)
indeno[123-cd]pyrene	3.3 (15)	9 (9)	39 (13)	10 (5)	57 (13)

*Results shown as percent recovery of certified concentration

**The number in parentheses indicates the percent relative standard deviation (%RSD) compounds, phenanthrene, fluoranthene, pyrene, and chrysene, and found that the reactive modifier increased the extraction efficiency by six times over that of pure CO_2 and by two times over that of CO_2 modified with 10% methanol.

Hawthorne and Miller tested a variety of supercritical fluids to determine their effectiveness for extracting PAHs from three common environmental solids; urban dust, fly ash, and river sediment. Supercritical N_2O with 5% methanol modifier gave the best recoveries of PAHs from all three samples when compared to supercritical CO_2 with 5% methanol, N_2O, CO_2, and ethane (Hawthorne, 1987). The poorest recoveries were obtained when either ethane or pure CO_2 were used for the extractions. N_2O without modifier provided better recoveries than ethane or CO_2, but the addition of methanol as a modifier increased the extraction efficiencies when CO_2 or N_2O were used. In most cases, 30-60 minute extractions of the river sediment and fly ash with supercritical N_2O/5% methanol gave better recovery of the deuterated PAH spikes than the recoveries obtained by using 4 hours of sonication or 8 hours of Soxhlet extraction with either benzene or methylene chloride (Hawthorne, 1987).

In SFE, matrix interactions with both solvent and analyte exert a powerful influence on the extraction process and must always be considered when selecting a solvent (Pipkin, 1992). The effect that the modifier has on the supercritical fluid must also be considered, especially when working at moderate temperatures and pressures. Depending on the modifier identity, the nature of the analytes of interest, as well as the type of sample matrix, there is a need to apply an equilibration period when using modifiers in SFE (Levy, 1992). If the modifier does not have sufficient contact time with the matrix and analyte the full effect of the modifier may not be achieved. This can especially be a problem when adding modifiers directly to the extraction cell instead of mixing the modifier into the supercritical fluid. The modifier can be displaced from the extraction vessel by the supercritical fluid and end up in the collection vial before it can interact with the matrix.

2.4 Analyte Collection

When using the SFE on-line with another instrument there is no sample handling, but if using the SFE in the off-line mode there are many options for sample collection. After optimizing the SFE to extract the analyte from the sample, the next step for off-line analysis, is to make sure that the method of sample collection is efficient. The analyte collection techniques that will be discussed below include collection in an empty vial, a vial with organic solvent, and a cryogenically cooled trap. The analyte of interest, the flow rate of CO_2 during extraction, and the type of analysis to be used after collecting the sample are some of the variables that will have a bearing on which collection method is employed.

Miller et al. (1993) conducted experiments to determine the effectiveness of using an empty vial for the collection of a variety of compounds. They developed a method that does not use a flow restrictor by collecting the analytes in an empty vial after a static extraction. Once the static extraction was complete the CO_2 was depressurized through a 178 μm i.d. stainless steel tube into a capped vial that had a single hole punctured in the septum with a 1/16 inch stainless steel rod. The compounds that were tested included alkanes (C_6-C_{30}), PAHs, PCBs, gasoline, and diesel fuel. The alkanes > C_{10} were recovered at 96-99% and the C_6-C_9 alkanes had recoveries of 39, 70, 88, and 94%, respectively. The PCBs recoveries for the di- through hepta-chloro congeners ranged from 90-97%. Efficient recoveries of the PAHs proved to be more difficult than the alkanes and PCBs, but it was mainly due to the PAHs not being extracted from the matrix. Initially, the low molecular weight PAHs were recovered at >90%, but the larger molecular weight PAHs chrysene and benzo[b]fluoranthene showed recoveries of 60 and 74%. When the spiked sand was extracted sequentially three times, the recoveries of the two higher molecular weight PAHs were increased to 84 and 82%. The recovery of gasoline hydrocarbons (TPH) was 73% and recovery of diesel fuel hydrocarbons (TPH) was 92%.

Studies were done by Lopez-Avila et al. (1992) on the use of organic solvents for the off-line collection of petroleum hydrocarbons from various soil samples after SFE. The organic solvents tested included Freon-113, perfluorohexane, hexafluorobenzene, Flon S-316, carbon tetrachloride, and PCE. The collection solvent for the extracted material must be compatible with the IR determination, and it should have the solubilizing properties of Freon-113, be nonflammable, nontoxic, have a boiling point higher than 100 °C (so that little will be lost through volatilization during the carbon dioxide depressurization step), and a freezing point below -30 °C (which is the typical temperature of the collection solvent during the 30 minute extraction when no heat is applied to the collection vial) (Lopez-Avila, 1992). The perfluorohexane had poor solubilizing properties for the analytes of interest compared to Freon-113, but it was a good solvent because 80% remained after bubbling carbon dioxide through it for 30 minutes at a flow rate of 1 ml/min (as liquid). The hexafluorobenzene cost was too high so it was eliminated immediately, hexafluorobenzene is $1780/L whereas Freon-113 is only $38/L. Flon S-316 (a tetrachloro-hexafluorobutane) had only 14% volatilization after carbon dioxide had been bubbled through it for 60 minutes at the same flow rate mentioned above. It is a good solvent for hydrocarbons, but it is about five times more expensive than Freon-113. Carbon tetrachloride worked well as a collection solvent, but it is listed by the EPA as a probable carcinogen, and thus its use is generally not recommended. PCE is an even better solvent than Freon-113 for petroleum hydrocarbons containing 30 or more carbon atoms. It proved to be a good collection solvent and was compatible with the IR analysis as well. As a result of the study, Lopez-Avila et.al., recommended PCE as a collection solvent for TPHs.

Ashraf-Khorassani et al. (1992) carried out studies comparing an empty vial, a vial with solvent, and a cryogenic trap as methods of collection after SFE. Initially, they extracted a mixture of phenanthrene and pyrene and collected the analytes with each of the three methods for off-line collection. The collection efficiency using the empty vial was very low, but recoveries were improved when methylene chloride was added to the vial. Others have reported good collection efficiencies using organic solvents, but it should be noted that the flow rates were relatively low (< 1 ml/min). The flow rate in this study was 2 ml/min and it is thought that the loss of analyte in the solvent filled vial was due to the formation of an aerosol. Collection efficiencies of greater than 90% were observed when a cryogenically cooled trap was used. The high percent recovery obtained with a cryogenically cooled adsorbent trap is due to two factors: (a) freezing the analytes in the trap upon decompression, and (b) retaining the analytes on a bed of silanized glass beads. Two wash solvents used to rinse the analytes from the cryogenic trap were also compared. Lower percent recoveries were obtained when methanol was used as compared to the recoveries obtained when methylene chloride was used as the wash solvent. The effect of the temperature of the cryogenic trap on collection efficiency of aliphatic hydrocarbons was also evaluated. A temperature of -45 °C compared to -20 °C increased the efficiency of decane trapping, but the collection efficiency of the rest of the test compounds remained unchanged. For volatile compounds the trap temperature is an important parameter that will affect collection efficiencies. Overall, the cryogenic trap was much more effective than the empty vial or solvent filled vial. Also, the flowrate of CO_2 through the extraction vessel did not have any effect on the collection efficiency of the cryogenic trap.

CHAPTER 3

METHODOLOGY

3.1 SFE Apparatus

A Suprex PrepMaster^{TM} and Accutrap^{TM} SFE with cryogenic collection purchased from the Suprex Corporation, Pittsburgh, PA, USA, was used throughout this research project. The SFE had the capability of reaching pressures of 500 atm and temperatures of 150 °C for the extraction of samples. A 3 ml stainless steel extraction vessel was used for all of the extractions. The restrictors used were stainless steel and were kept at 150 °C to prevent plugging. The SFE CO_2 used had helium headspace and 1.0% $MeCl_2$ modifier, purchased from Scott Specialty Gases, Inc., Plumsteadville, PA, USA. The nitrogen used for purging the cryogenic collection trap was purchased from AGA Gas Inc., Cleveland, OH, USA. Also purchased from AGA was CO_2 with a siphon, which was used to control the collection trap temperature.

3.2 Quantitative and Qualitative Analysis

Two instruments were used for the qualitative and quantitative analysis of the samples collected from the SFE. A Hewlett-Packard GC/MS was used for qualitative analysis, Hewlett-Packard, Palo Alto, CA, USA. The GC was a 5890 Series and the MS was a 5972 Series. The mass spectrometer was operated in the scan mode and a mass range of 10 to 500 atomic mass units (amu) was scanned. The column used was a HP-5 M.S. (Hewlett-Packard, Palo Alto, CA, USA) that was 30 m long and had a 0.25 mm ID and a 0.25 μm film thickness. The initial column temperature was 60 °C and the final column temperature was 300 °C. The temperature program ramped the column temperature from 60 °C to 300 °C at a rate of 5 °C/min. The column was held for 2.0 minutes at 60 °C before the temperature ramp was begun and also held for 2.0 minutes at 300 °C at the end of the temperature ramp. The injector was held at a constant temperature of 250 °C.

Varian 3400 GC/FID with a Varian 8100 Autosampler was used for the quantitative analysis (Varian, Walnut Creek, CA, USA). The column used was a Supelco SPB^{TM}-5 that was 30 m long and had a 0.53 mm ID and 0.50 μm film thickness (Supelco, Inc., Bellefonte, PA, USA). The initial column temperature was 60°C and the final column temperature was 300°C. The temperature program ramped the column temperature from 60°C to 300°C at a rate of 8°C/min. There was no initial hold time at 60°C, but the column was held at the final temperature of 300°C for 20 minutes. The injector had an initial temperature of 30°C and was held initially for 0.2 minutes before the 100°C/min temperature ramp program started. The final injector temperature was 320°C and was held at that temperature for 46.9 minutes. The FID was held at a constant temperature of 320°C.

3.3 Reagents

Solvents used with the SFE were methylene chloride (GC Resolv) and methanol (HPLC Grade) and were obtained from Fisher Scientific, Pittsburgh, PA, USA. Solvents used for the Soxhlet extractions included the same grade methylene chloride as mentioned above as well as cyclohexane (HPLC Grade) and pentane (Capillary GC/GC-MS Solvent) both of which were purchased from Baxter Diagnostics Inc., Mc Gaw Park, IL, USA. The extraction thimbles used were Whatman 43 mm x 123 mm single thickness cellulose extraction thimbles (Whatman International, Ltd, Maidstone, England). The Soxhlet extraction clean-up procedure required the use of sodium sulfate (Anhydrous Granular 12-60 mesh, J.T. Baker Inc., Phillipsburg, NJ, USA) and silica gel (Davidson 923 100/200 mesh, Supelco, Bellefonte, PA, USA). The PAH standard used for GC calibration curves was the Supelco TCL Polycyclic Aromatic Hydrocarbons Mix with 2000 µg/ml of each PAH in methylene chloride:benzene (50:50). PAHs used to spike soils were naphthalene, phenanthrene, pyrene, and benzo[a]pyrene. The naphthalene (99%) and benzo[a]pyrene (98%) were purchased from Sigma Chemical Company, St. Louis, MO, USA and the phenanthrene (98%) and pyrene (99%) were purchased from Aldrich Chemical Company, Inc., Milwaukee, WI, USA.

3.4 Soil Characteristics

Three materials were used in this study: Ottawa Sand, Soil A, and Soil C. The mechanical characteristics of the soils are shown on Table 3.1. The Ottawa Sand was used as an inert material from which PAHs could be extracted. Soil A was a clean, uncontaminated horizon A from a site near Collins Road and Forest Road in Lansing, MI, USA. The Soil A was used to test the extractability of PAHs from a matrix that was more representative of a real world matrix than the Ottawa Sand. There were two methods used to spike known amounts of PAHs onto the soil: (1) PAHs were spiked onto the soil just prior to extraction (fresh spike), and (2) PAHs were spiked onto the soil and were then allowed to age for over one year (aged spike). The spiking procedures are described in the next section of this chapter. Soil C was a soil from a coal tar contaminated site, which was contaminated during the time period from the late 1800's until approximately the late 1940's. Soil C was monitored for 16 PAHs. Appendix A contains physical characteristics and structures of the sixteen PAHs. The presence of native PAHs in the soil was significant for this research, but in some cases additional PAHs were spiked onto the soil; both fresh spike and aged spike.

Table 3.1

Mechanical Characteristics of Soils used in Experiments

	Soil A	Soil C
Organic Matter (%)	10.8	7.8
Sand (%)	81.3	58.6
Silt (%)	12.4	24.0
Clay (%)	6.4	17.4
Soil Type	Loamy sand	Sandy loam

3.5 Spiking Procedure

As stated earlier naphthalene, phenanthrene, pyrene, and benzo[a]pyrene were used as the model PAH compounds for the spiking procedures. The two spiking procedures, fresh spike and aged spike, were used in this research to introduce the PAHs onto the soil matrices. The procedure for the fresh spike was to weigh out the soil sample, place it in the extraction vessel, and then spike the soil sample with the PAHs. An amount of 200 μl of a stock solution containing 1000 μg/ml of each of the four PAHs in methylene chloride was added to the soil sample in the extraction vessel. The extraction vessel was then immediately connected to the SFE and the extraction procedure carried out. An amount of 4.0 grams of Ottawa Sand was used for the fresh spike extractions of PAHs from the sand matrix. For both Soil A and Soil C, an amount of 2.0 grams of freeze dried soil was used for the fresh spike extractions.

The aged spike consisted of adding the four model PAHs to 100 grams (dry weight) of soil to obtain a concentration of 200 μg of each PAH/g of dry soil. The stock solution used to spike the soil contained approximately 1000 μg/ml of each PAH in a 50:50 (v/v) mixture of methylene chloride:benzene. The concentrations of the PAHs in the stock solution were determine by GC analysis and the results are shown in Table 3.2. After adding the stock solution to the soil, the methylene chloride and benzene were allowed to volatilize from the soil over a 16 hour time period. The samples were stored in glass bottles, placed in a refrigerator at 4 °C, and protected from exposure to light. The first month the samples were shaken once a week to mix the soil with the spiked PAHs.

The samples were shaken once every two or three months for the remainder of the aging process. The samples were not used for any extraction procedures until over 15 months of aging time. For both Soil A and Soil C aged spike samples, an amount of 2.00 grams of freeze dried soil was weighed out, placed in the extraction vessel, and the extraction process carried out.

Table 3.2

Concentration of PAHs Added to Soil A and Soil C for the Aged PAH Spike

PAH	Concentration (µg of PAH/g of dry soil)
Naphthalene	220 ± 15
Phenanthrene	220 ± 14
Pyrene	220 ± 16
Benzo[a]pyrene	200 ± 11

3.6 SFE Procedure

The SFE conditions used for the extractions are listed in Table 3.3. The pressure, temperature, and extraction time were variables that changed depending on the experiment being conducted. The pressures used for the extractions ranged from 200 to 400 atm and the temperatures ranged from 60 to 150 °C. The dynamic extraction times ranged from 5 to 45 minutes. Specific information about the pressure, temperature, and extraction time will be provided along with the results for each of the experiments conducted. The extractions were performed in triplicate and standard deviations are provided with the results.

Table 3.3

SFE Conditions used for the Extraction of PAHs from the Soil Matrices

Prepmaster Method	
Pressure	varied
Temperature	varied
Extraction Time	varied
Extraction Vessel Size	3 ml
Accutrap Method	
Collection Temperature	-20 °C
Desorption Temperature	15 °C
Desorption Rinse	3 ml of methanol at 1.0 ml/min
Flush Temperature	15 °C
Flush Rinse	5 ml of methanol at 1.0 ml/min
Restrictor	150 °C 3.2 ml/min flowrate

In some cases, the organic modifier concentration was increased by adding methylene chloride directly to the soil sample in the extraction vessel. When additional organic modifier was added to fresh spike extractions an amount of 500 μl of methylene chloride was added. This resulted in a total of 700 μl of methylene chloride being added to the soil; 200 μl of methylene chloride from the PAH stock solution plus the additional 500 μl of methylene chloride. When organic modifier was added to the aged spike samples, an amount of 700 μl of methylene chloride was added to the soil. It was assumed that the solvent used to spike the PAHs onto the soil had volatilized from the aged spike soil samples. This assumption was based on the fact that the solvents were allowed 16 hours to volatilize from the soil after spiking the PAHs onto the soil.

Ottawa Sand was used as an inert material from which PAHs could be extracted using SFE with CO_2 modified with 1% methylene chloride. The extractions carried out with the sand were useful for determining whether the SFE was functioning properly. The extraction efficiencies of the PAHs from the sand, at specific operating parameters, were used as a QA/QC check on the SFE system. The QA/QC extractions were done at various points during the research to ensure that the results obtained were not a result of the SFE functioning improperly. The reason for starting a QA/QC protocol was because of problems encountered at the beginning of the research project. Extraction recoveries of PAHs from the Ottawa sand were low at first. There were four problems found with the SFE that needed to be addressed before any data could be collected.

First, the nitrogen purge line for the cryogenic trap was fixed, because it was not hooked up correctly. The ferrule on the nitrogen inlet line to the SFE was not seated properly and the rinse solvent in the cryogenic trap was not being expelled into the collection vial. Second, the vent line on the SFE that releases the pressure from the system between the restrictor and the extraction vessel was plugged. When the dynamic extraction is completed, the manifold switches the SFE back to the static mode. There continued to be a volume of pressurized CO2 behind the restrictor that was trying to exit the system and the only path for it to take was through the cryogenic trap. This resulted in excessive gas flow into the collection vial while the rinse solvent was being dispensed and caused some of the rinse solvent, and PAHs, to enter the vent line of the collection vial. It may also have contributed to the volatilization of naphthalene, which showed the poorest recovery.

Third, the temperature of the cryogenic trap during the desorption and flushing stage was lowered from 40 °C to 15 °C. This was done after talking to a representative at Suprex about the problems encountered with the recovery efficiencies. The representative mentioned that the restrictor temperature (150 °C in this case) may cause the solvent to vaporize. If the cryogenic trap temperature is not low enough to cool the solvent back down then it may still be in the vapor form as it goes through the trap. Finally, the rinse solvent was changed from methylene chloride to methanol. The amount of methylene chloride being dispensed during the trap rinsing was not precise because the seal on the pump was not compatible with methylene chloride. The pump had to be taken apart and the methylene chloride evaporated from the seal. This allowed the pump to function properly again, although only for a limited time. Methanol will not cause any problems with the pump, but the solubility of PAHs in methanol is lower than it is in methylene chloride. This may or may not cause problems in the future when trying to rinse analytes from the trap, and should be taken into consideration if recoveries are low.

3.7 Soxhlet Extraction Procedure

A general description of the Soxhlet extraction procedure is described below. A more detailed, step-by-step set of instructions are given in Appendix B. Soil samples were weighed out, placed in extraction thimbles, and freeze dried for 12 to 16 hours. The Soxhlet extraction of the samples was conducted for 16 to 20 hours using 300 ml of methylene chloride and were done in triplicate. After the extraction was complete, an amount of 200 ml of cyclohexane was added, and the methylene chloride was boiled off. The cooled sample was then poured through a drying column containing silica gel and sodium sulfate. The column of silica gel was rinsed with 60 ml of pentane to displace the cyclohexane from the column. The analytes were then removed from the silica gel using a 60:40 (v/v) mixture of pentane:methylene chloride and collected in a flask. The concentration step was then carried out using a water bath at 65 - 70 °C to boil off the solvents. Methylene chloride was then added to a specific volume, the sample transferred to GC vials, and stored at 4 °C until the analysis could be performed.

CHAPTER 4

PAHs EXTRACTED FROM OTTAWA SAND

4.1 Temperature and Pressure Effects

The next set of extractions carried out, using Ottawa Sand as the media, tested the effect of using different temperatures and pressures on the extraction efficiency for PAHs. The total dynamic extraction time was 10 minutes. An amount of 4.0 grams of Ottawa Sand was placed in the 3 ml extraction vessel and spiked with 200 µl of PAH stock solution. The recoveries from the sand were compared to the concentration of the PAH stock solution, both of which were determined by GC analysis. Temperatures of 120 °C and 150 °C were tested with three different pressures of 200, 300, and 400 atm (Table 4.1 and Table 4.2). A pressure of 300 atm was used to test the effect of four temperatures: 60, 90, 120, 150 °C (Table 4.3).

The results of varying the pressure during the extraction at 120 °C showed that changing the pressure does not have a large effect on recovery efficiency for this matrix. All of the PAHs were recovered with efficiencies > 89%, with the exception of the benzo[a]pyrene when the SFE pressure was 200 atm. The recovery of benzo[a]pyrene was only 70% at 200 atm, but the standard deviation between the three extractions was relatively high compared to the rest of the results.

Table 4.1

Percent Recovery with Varying Pressure at a Constant Temperature of 120 °C

Temp. (°C)	120		120		120	
Pressure (atm)	200		300		400	
	Average Recovery	Standard Deviation	Average Recovery	Standard Deviation	Average Recovery	Standard Deviation
Naphthalene	93	10	94	5	89	14
Phenanthrene	96	3	101	3	99	6
Pyrene	97	4	103	3	100	5
Benzo[a]pyrene	70	37	105	4	103	14

Table 4.2

Percent Recovery with Varying Pressure at a Constant Temperature of 150 °C

Temp. (°C)	150		150		150	
Pressure (atm)	200		300		400	
	Average Recovery	Standard Deviation	Average Recovery	Standard Deviation	Average Recovery	Standard Deviation
Naphthalene	84	6	88	19	93	15
Phenanthrene	98	1	99	5	104	7
Pyrene	98	4	103	4	105	8
Benzo[a]pyrene	81	10	113	5	102	14

Table 4.3

Percent Recovery with Varying Temperature at a Constant Pressure of 300 atm

Temp. (°C) Pressure (atm)	60 300 Avg. Recovery ± Std. Dev.	90 300 Avg. Recovery ± Std. Dev.	120 300 Avg. Recovery ± Std. Dev.	150 300 Avg. Recovery ± Std. Dev.
Naphthalene	97 ± 9	101 ± 18	94 ± 5	88 ± 19
Phenanthrene	102 ± 1	103 ± 2	101 ± 3	99 ± 5
Pyrene	104 ± 2	103 ± 1	103 ± 3	103 ± 4
Benzo[a]pyrene	102 ± 6	109 ± 7	105 ± 4	113 ± 5

The recovery of the PAHs increased slightly when the temperature was increased to 150 °C. A statistical analysis using the Student t test with a 95% confidence interval was done and there was no statistical difference in the recoveries obtained at pressures of 300 or 400 atm pressure. Both of the higher pressures resulted in a statistically significant increase in the recovery of benzo[a]pyrene compared to the recovery obtained when 200 atm was used.

When the pressure was held constant at 300 atm and the temperature was varied there was no change in recovery of any of the PAHs. Recoveries were all >94%, with the exception of the naphthalene recovery being 88% when the conditions were 300 atm and 150 °C. However, there was no statistical difference in the recovery of naphthalene obtained when either 120 °C or 150 °C were used.

These results indicate that the PAHs are not limited by solubility at the concentrations that were spiked onto the sand. An amount of 4.0 grams of sand was used for the extractions and 200 µl of PAH stock solution was added to the sand. The stock solution concentration contained 1000 µg/ml of each of the four model PAHs, therefore 200 µg of each PAH was extracted using CO2 modified with 1% methylene chloride.

CHAPTER 5

PAHs EXTRACTED FROM SOIL A

5.1 Extractions of Fresh Spike using 90 °C and 300 atm

PAHs were extracted from Soil A after adding PAHs to this matrix using the fresh spike technique. The SFE conditions are shown in Table 5.1. Note that the collection of the analyte was fractionated in order to plot the rate of analyte recovery versus time (Figure 5.1). The overall PAH recovery was >87% for naphthalene, phenanthrene, and pyrene, but the recovery of benzo[a]pyrene was only 66% (Table 5.2). The predominant difference in the recovery of the PAHs from Soil A as compared to that observed with Ottawa Sand is that the benzo[a]pyrene was not as easily extracted in Soil A. It only took 10 minutes to quantitatively recover the benzo[a]pyrene from the Ottawa Sand, but after a 30 minute extraction of Soil A the benzo[a]pyrene was recovered at less than 70%. Since solubility was not a problem, it is believed that the slow desorption of the benzo[a]pyrene from Soil A limits the extraction efficiency.

5.2 Extractions of Fresh Spike using 150 °C and 300 atm

The temperature of the extraction was increased to 150 °C to try to the efficiency at which benzo[a]pyrene can be extracted from Soil A. All other experimental conditions

Table 5.1

SFE Conditions used for the Extraction of Fresh Spike PAHs from Soil A

Prepmaster Method	
Pressure	300 atm
Temperature	90 °C
Extraction Time	1 min Static 5 min Dynamic 5 min Dynamic 10 min Dynamic 10 min Dynamic
Extraction Vessel Size	3 ml
Accutrap Method	
Collection Temperature	-20 °C
Desorption Temperature	15 °C
Desorption Rinse	3 ml of methanol at 1.0 ml/min
Flush Temperature	15 °C
Flush Rinse	5 ml of methanol at 1.0 ml/min
Restrictor	150 °C 3.2 ml/min flowrate

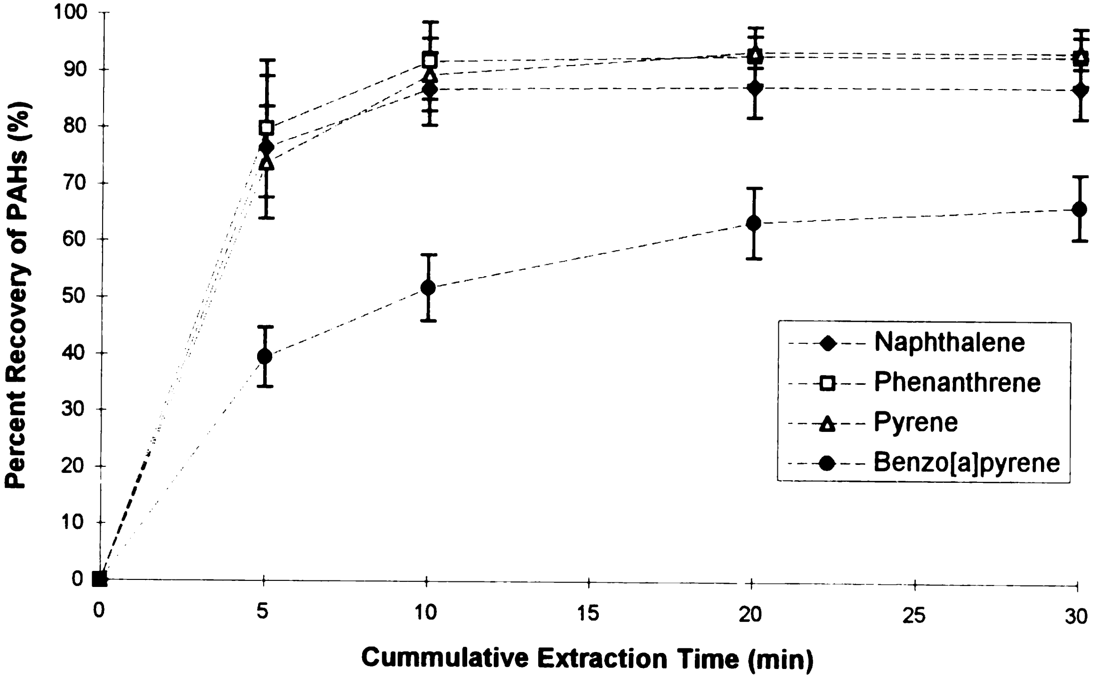


Figure 5.1
Fractionated Collection During SFE of Fresh PAH Spike from Soil A
with SFE at 90 °C and 300 atm

Table 5.2

Percent Recovery of PAHs from Soil A with the SFE at 90 °C and 300 atm

Cummulative Extraction Time (min)	5	10	20	30
Naphthalene	77 ± 13	87 ± 6	87 ± 5	87 ± 5
Phenanthrene	80 ± 12	92 ± 7	93 ± 5	93 ± 5
Pyrene	74 ± 10	89 ± 6	94 ± 3	94 ± 3
Benzo[a]pyrene	40 ± 5	52 ± 6	64 ± 6	66 ± 6

were the same as listed in Table 5.1. The result was that the naphthalene, phenanthrene, and pyrene were recovered at efficiencies of >94%, but the recovery efficiency for benzo[a]pyrene remained only 62% (Table 5.3). The standard deviation between the triplicate samples decreased for all of the compounds except benzo[a]pyrene. The rate of extraction was increased slightly for all the compounds except benzo[a]pyrene (Figure 5.2).

5.3 Extractions of Fresh Spike using an Organic Modifier

Since the benzo[a]pyrene was not extracted satisfactorily by increasing the temperature, an organic modifier was added to the extraction cell and the temperature was reduced to 90 °C. The supercritical fluid CO_2 already had 1% methylene chloride mixed into the gas cylinder and methylene chloride was used as a solvent for the PAHs spiked into the soil. Therefore, methylene chloride was chosen as the organic modifier to be

Table 5.3

Percent Recovery of PAHs from Soil A with the SFE at 150 °C and 300 atm

Cummulative Extraction Time (min)	5	10	20	30
Naphthalene	92 ± 3	97 ± 3	97 ± 2	97 ± 2
Phenanthrene	88 ± 6	95 ± 3	95 ± 3	95 ± 3
Pyrene	80 ± 8	92 ± 5	94 ± 3	94 ± 3
Benzo[a]pyrene	40 ± 10	51 ± 11	59 ± 11	62 ± 9

added directly to the extraction cell. An amount of 500 µl of methylene chloride was added immediately after the 200 µl PAH spike was added to the soil. The extraction cell was then attached to the SFE for extraction. The result was that the benzo[a]pyrene was extracted at a recovery of 99% (Table 5.4). The phenanthrene and pyrene were recovered at >100% and the naphthalene was recovered at 89%. The rates of recovery were also improved greatly when the organic modifier was added to the extraction cell (Figure 5.3). The phenanthrene and pyrene were completely recovered after the first 5 minute dynamic extraction step. The naphthalene and benzo[a]pyrene had 89% and 94% recoveries after a total of only 10 minutes of dynamic extraction time.

5.4 Extractions of Aged Spike using 90 °C and 300 atm

In experiments in which the PAHs were to be extracted from the aged spiked Soil A the SFE conditions were the same as those listed in Table 5.1. An amount of 2.0 grams of the aged spike Soil A was placed in the extraction cell for the analysis. The recoveries of phenanthrene and pyrene were 80% and 87%, respectively. Naphthalene and benzo[a]pyrene were not recovered very efficiently with only 33% and 38% recoveries, respectively (Table 5.5). Also, the rate of extraction was slower for the aged spike than for the fresh spike (Figure 5.4).

The naphthalene recovery may be better than the calculated value, because the percent recovery was based on the amount of naphthalene spiked into the soil and not the actual amount of naphthalene left in the soil after one year. After it was spiked with the PAHs, the soil sample was left open to the atmosphere overnight to allow the solvent to evaporate from the soil. It is likely that some of the naphthalene volatilized and was lost from the sample. It is also possible that some of the naphthalene was degraded during the aging process by microorganisms present in the soil. An indication that the actual

Table 5.4

Percent Recovery of PAHs from Soil A with the SFE at 90 °C and 300 atm with 500 µl of Methylene Chloride Added as an Organic Modifier

Cummulative Extraction Time (min)	5	10	20	30
Naphthalene	87 ± 3	89 ± 4	89 ± 4	89 ± 4
Phenanthrene	101 ± 1	104 ± 2	104 ± 2	104 ± 2
Pyrene	101 ± 2	107 ± 1	107 ± 1	107 ± 1
Benzo[a]pyrene	86 ± 7	94 ± 5	98 ± 4	99 ± 4

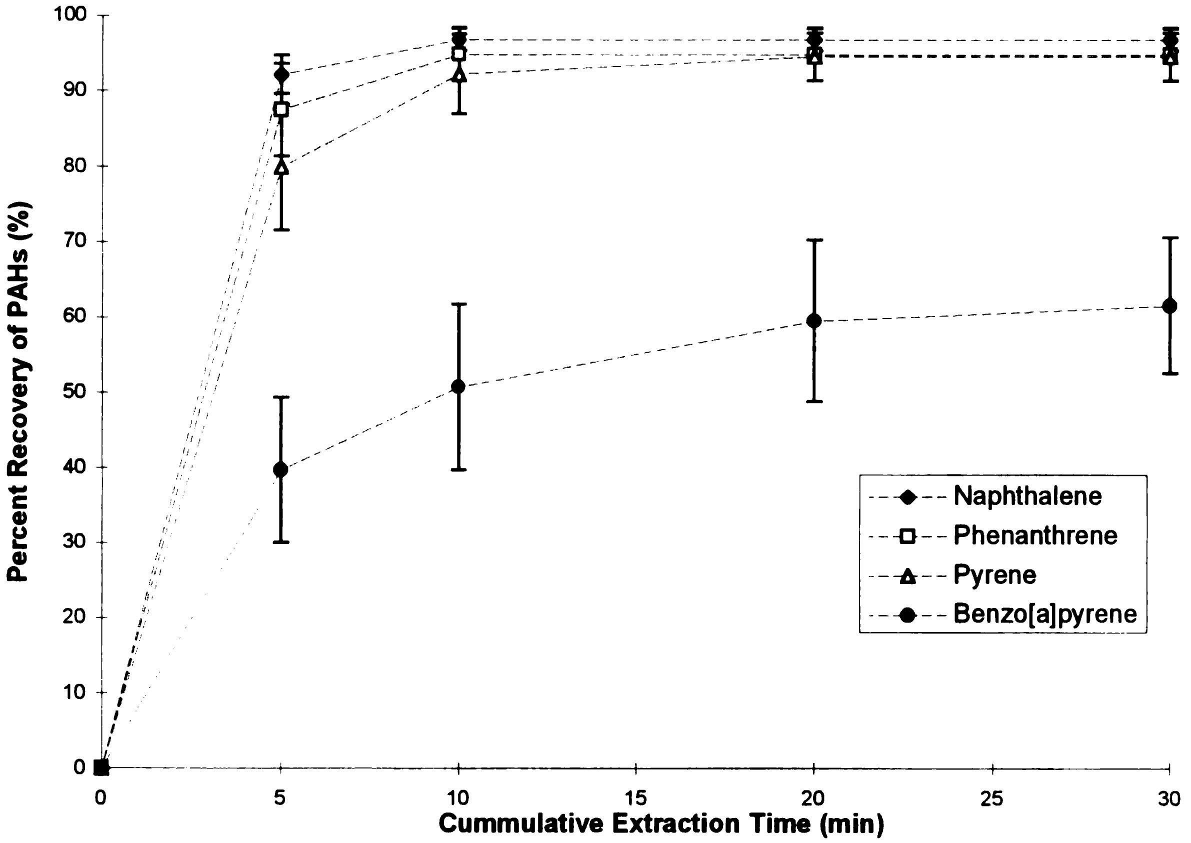


Figure 5.2
Fractionated Collection During SFE of Fresh PAH Spike from Soil A with SFE at 150 °C and 300 atm

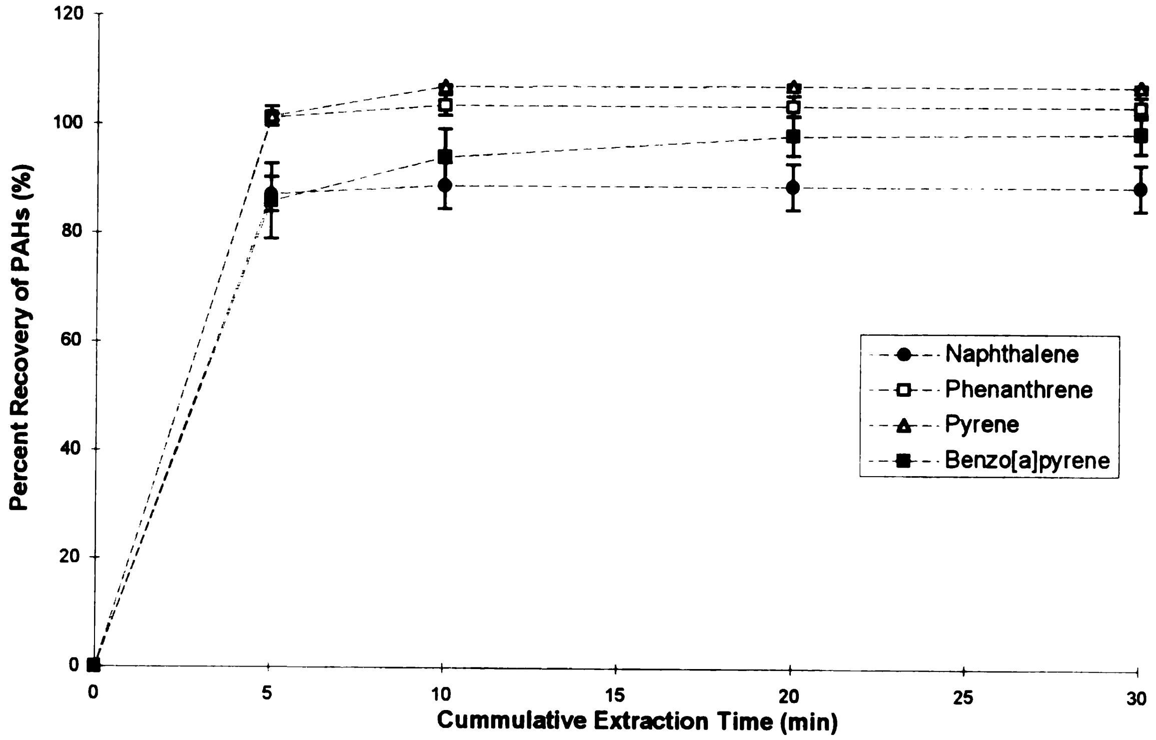


Figure 5.3
Fractionated Collection During SFE of Fresh PAH Spike from Soil A
with 500 µl Methylene Chloride Modifier and SFE at 90 °C and 300 atm

Table 5.5

Percent Recovery of PAHs from Aged Spike Soil A with SFE at 90 °C and 300 atm

Cummulative Extraction Time (min)	5	10	20	30
Naphthalene	25 ± 2	30 ± 1	32 ± 1	33 ± 1
Phenanthrene	60 ± 5	74 ± 4	79 ± 4	80 ± 4
Pyrene	59 ± 5	79 ± 4	86 ± 4	87 ± 4
Benzo[a]pyrene	13 ± 2	23 ± 3	34 ± 4	38 ± 5

concentration may be lower than actually noted is the flattening of the extraction rate curve for naphthalene in Figure 5.4. In the first 10 minutes of the dynamic extraction it appears that all of the naphthalene was extracted from the sample. The other compounds, phenanthrene, pyrene, and benzo[a]pyrene could have been subject to biodegradation as well, but based on the recoveries and extraction rates it was less likely to be as important of a factor as it was for naphthalene. The sample was kept in a dark place so that photodegradation should not have been a factor.

The aged spike Soil A was then analyzed using a Soxhlet method (Appendix B), which provided a comparison for the SFE method to something other than the theoretical concentrations of PAHs. The results of the Soxhlet extraction of the one year aged PAH spike Soil A are shown in Table 5.6. The results indicate that the naphthalene was much lower than the theoretical value of 220 µg/g of dry soil. The concentrations for the phenanthrene, pyrene, and benzo[a]pyrene are close to the threoretical values of 220, 220,

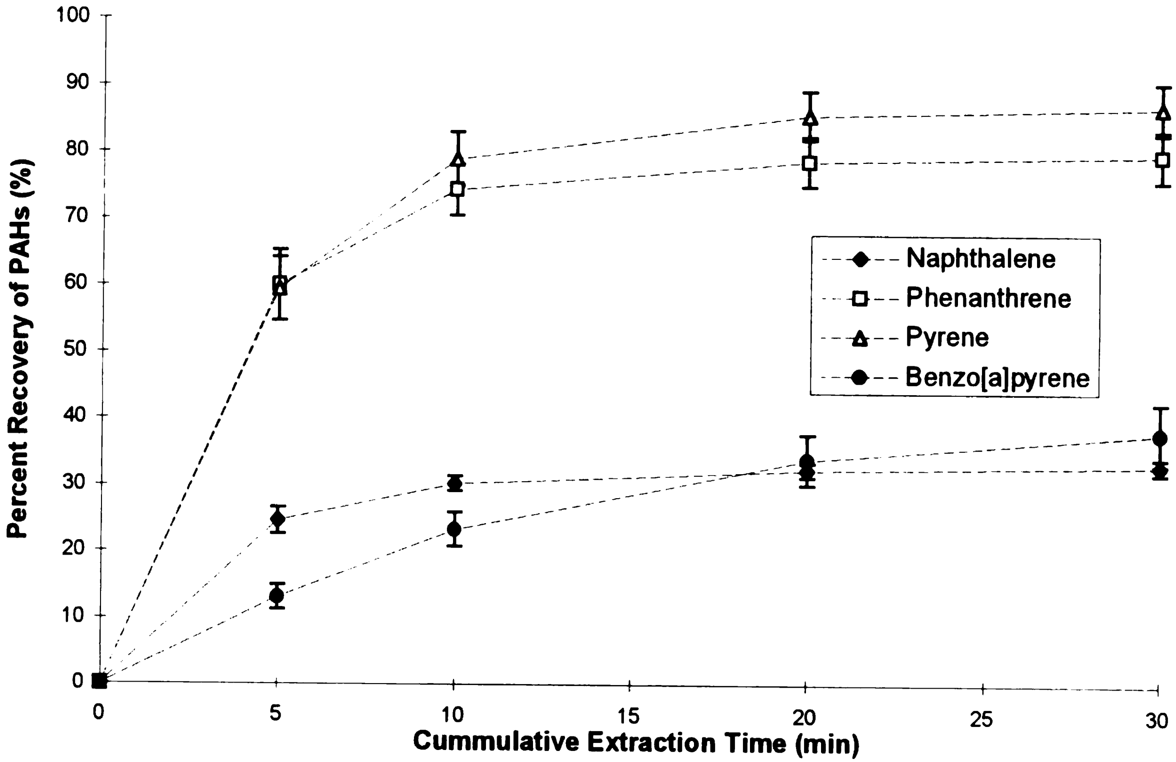


Figure 5.4
Fractionated Collection During SFE of Aged PAH Spike from Soil A with SFE at 90 °C and 300 atm

Table 5.6

Concentration of PAHs Determined by Soxhlet Extraction in Aged PAH Spiked Soil A

PAH	Concentration (µg of PAH/g of dry soil)
Naphthalene	92 ± 15
Phenanthrene	173 ± 25
Pyrene	177 ± 27
Benzo[a]pyrene	152 ± 20

and 200 ug/g of dry soil, respectively. While the small differences in the concentration values of phenanthrene, pyrene, and benzo[a]pyrene as determined using Soxhlet extraction and the theoretical values may be due to inefficiencies of the Soxhlet method, this is unlikely to be true for naphthalene (Table 5.7). If the SFE recoveries are compared to the Soxhlet recoveries the extraction efficiencies with the SFE at 90 °C and 300 atm are greatly improved over those calculated by comparing the SFE recoveries to the theoretical values (Table 5.8). The benzo[a]pyrene recovery remains low and needs to be improved. The rates of extraction of the PAHs by SFE (relative to the recoveries using Soxhlet extraction) are shown in Figure 5.5.

Table 5.7

Percent Recovery of PAHs by Soxhlet Extraction (18 hour extraction time) Based on Theoretical Concentrations in Aged PAH Spiked Soil A

PAH	Percent Recovery
Naphthalene	41 ± 7
Phenanthrene	79 ± 12
Pyrene	81 ± 12
Benzo[a]pyrene	77 ± 10

Table 5.8

Percent Recovery of PAHs from Aged Spike Soil A with the SFE at 90 °C and 300 atm Relative to the Soxhlet Extraction Recoveries

Cummulative Extraction Time (min)	5	10	20	30
Naphthalene	59 ± 5	73 ± 3	78 ± 3	80 ± 3
Phenanthrene	76 ± 7	95 ± 5	100 ± 5	101 ± 5
Pyrene	74 ± 6	98 ± 5	106 ± 5	108 ± 5
Benzo[a]pyrene	17 ± 2	30 ± 3	44 ± 5	49 ± 6

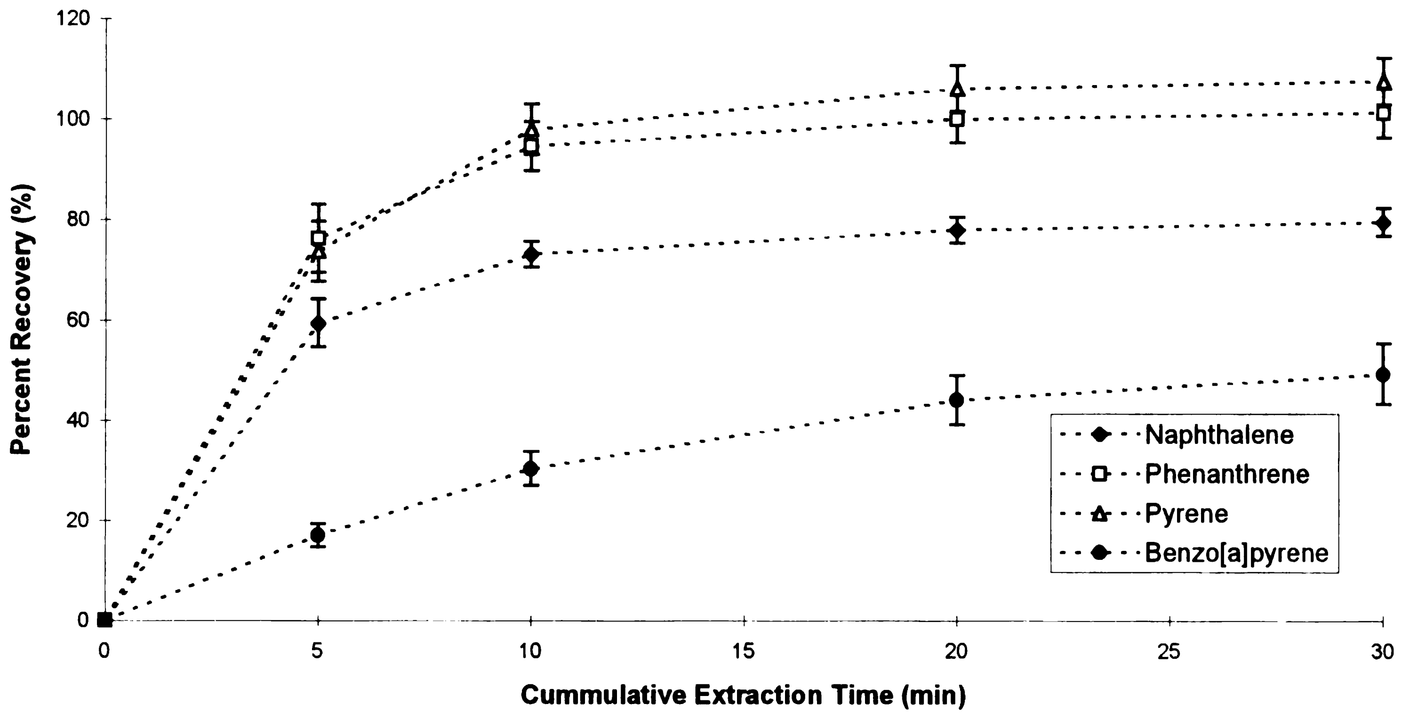


Figure 5.5
Fractionated Collection During SFE of Aged PAH Spike from Soil A
with SFE at 90 °C and 300 atm Relative to Soxhlet Extraction Results

5.5 Extractions of Aged Spike using an Organic Modifier

The addition of an organic modifier to the fresh spike Soil A resulted in an increase in the extraction efficiency of the benzo[a]pyrene. Therefore, the addition of an organic modifier was used to attempt to increase the extraction efficiency of benzo[a]pyrene from the aged spike Soil A. After the sample was placed in the extraction vessel, an amount of 700 µl of methylene chloride was added as an organic modifier. The SFE conditions were the same as those listed in Table 5.1, with the temperature at 90 °C and the pressure at 300 atm. While the addition of the organic modifier increased the recovery of benzo[a]pyrene, the recovery was still only 77% based on the soxhlet extraction results (Table 5.9). The extraction rates of the PAHs increased compared to the rates when no organic modifier was used at 90 °C and 300 atm (Figure 5.6).

Table 5.9

Percent Recovery of PAHs from Aged Spike Soil A with the Addition of 700 µl of Methylene Chloride as an Organic Modifier SFE at 90 °C and 300 atm Relative to the Soxhlet Extraction Recoveries

Cummulative Extraction Time (min)	5	10	20	30
Naphthalene	71 ± 6	83 ± 5	86 ± 6	86 ± 6
Phenanthrene	92 ± 6	110 ± 6	115 ± 6	115 ± 6
Pyrene	95 ± 7	115 ± 5	117 ± 5	117 ± 5
Benzo[a]pyrene	59 ± 5	70 ± 5	75 ± 4	77 ± 4

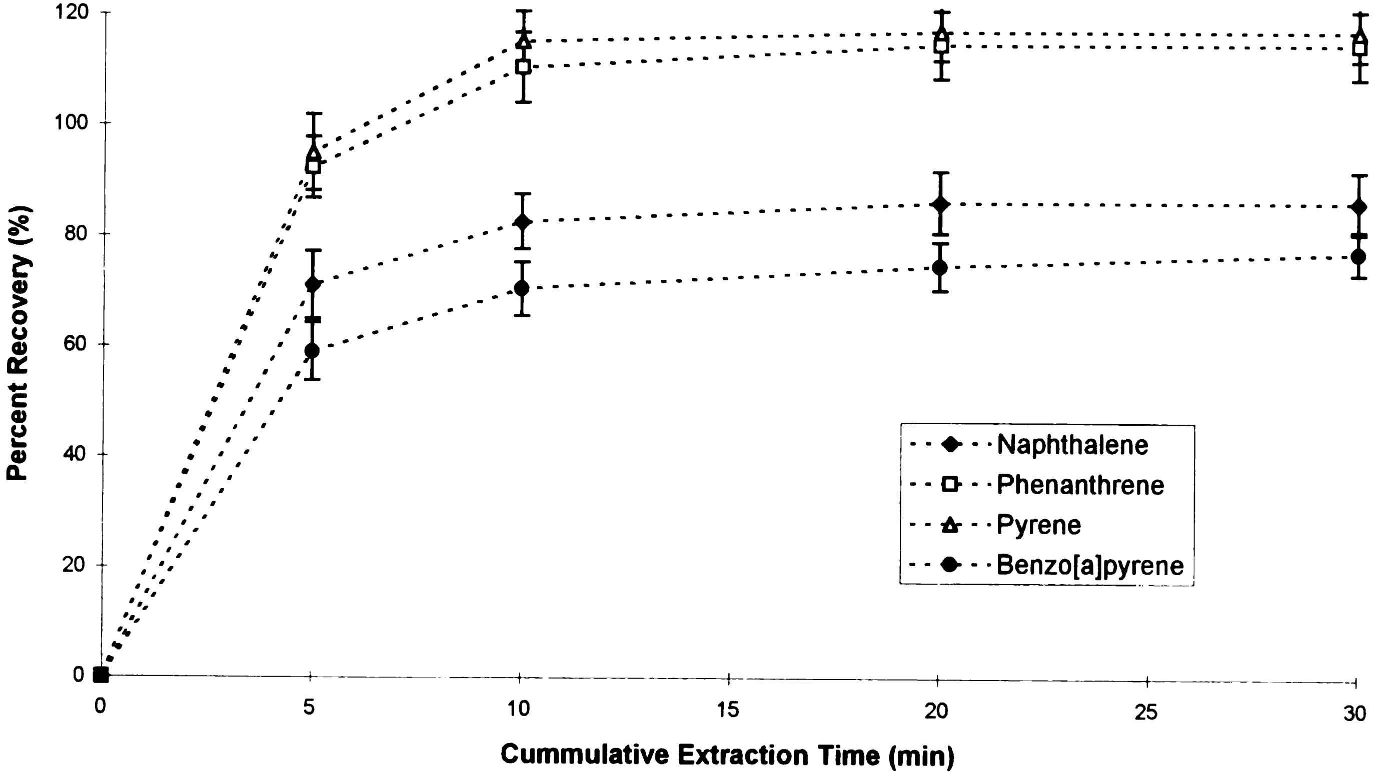


Figure 5.6
Fractionated Collection During SFE of Aged PAH Spike from Soil A
with 700 µl Methylene Chloride Modifier and SFE at 90 °C and 300 atm

5.6 Extractions of Aged Spike using an Organic Modifier and 150 °C

Since the benzo[a]pyrene recovery remained relatively low the extraction temperature was raised to 150 °C and methylene chloride was used as an organic modifier. The extraction conditions were the same as those listed in Table 5.1, with the exception of the 150 °C temperature. The results show that the benzo[a]pyrene recovery had increased to >90% (Table 5.10). The extraction rate curves are shown in Figure 5.7. In order to extract the PAHs from Soil A after one year of aging it was required to raise the temperature and add methylene chloride as an organic modifier.

Table 5.10

Percent Recovery of PAHs from Aged Spike Soil A with the Addition of 700 μl of Methylene Chloride as an Organic Modifier SFE at 150 °C and 300 atm Relative to the Soxhlet Extraction Recoveries

Cummulative Extraction Time (min)	5	10	20	30
Naphthalene	72 ± 4	85 ± 4	88 ± 4	88 ± 4
Phenanthrene	105 ± 3	116 ± 4	117 ± 4	117 ± 4
Pyrene	100 ± 5	113 ± 6	116 ± 7	117 ± 8
Benzo[a]pyrene	70 ± 7	85 ± 5	91 ± 5	93 ± 5

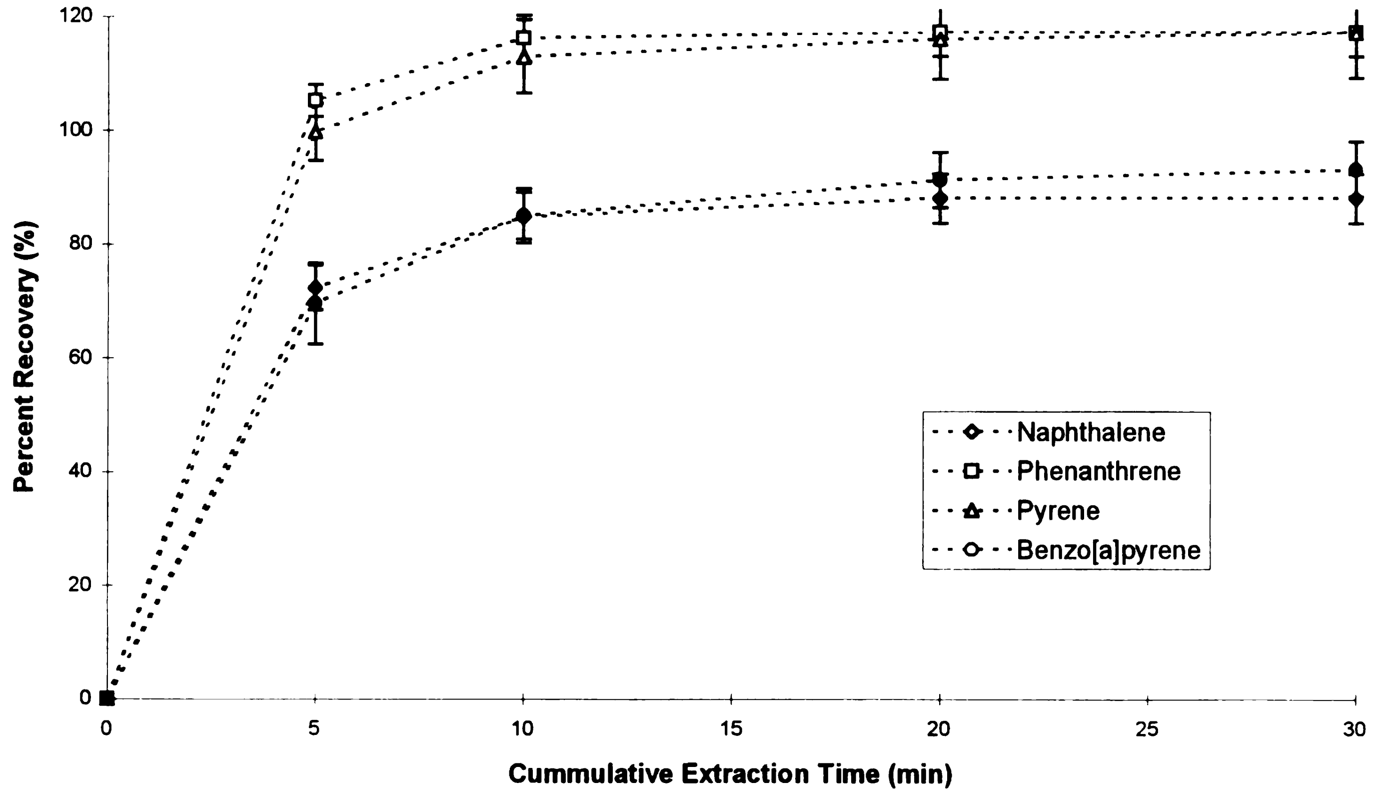


Figure 5.7
Fractionated Collection During SFE of Aged PAH Spike from Soil A with 700 µl Methylene Chloride Modifier and SFE at 150 °C and 300 atm

CHAPTER 6

PAHs EXTRACTED FROM SOIL C

Soil C was obtained from an actual coal tar contaminated site, but the specific site description is confidential. The contamination was the result of the disposal of the coal tar waste in open, unlined pits from a time period between the late 1800's to approximately the late 1940's. The term native PAHs is used to describe the contamination present in the soil as a result of the disposal of the coal tar. The relevence of Soil C for this research is that the PAHs have been in contact with the soil for 50 to 100 years. These native PAHs from the coal tar have had time to interact with the soil matrix and are likely to be more difficult to extract quantitatively than fresh spike PAHs. PAHs that are spiked onto soil to develop extraction methods for the quantification of contaminants may not have the same matrix interaction characteristics as native PAHs. Extraction of spiked PAHs will usually result in an overestimation of the extraction method efficiency. The advantage of using spiked PAHs is that the concentration of PAHs is known. If a soil with native PAHs is used to develop a SFE method the actual concentration of PAHs is not known. For this research, the native PAH contamination of Soil C was determined by Soxhlet extraction and the SFE method was compared to the Soxhlet results. The actual efficiency is not known for either method, because the true PAH concentration is not known.

6.1 Soxhlet Extraction

Soxhlet extraction was performed on Soil C using the method outlined in Appendix B. The results of triplicate samples are shown in Table 6.1. There were a total of 12 PAHs (out of the 16 PAHs included in the Supelco standard) at concentrations high enough for quantitation. The benzo[b]fluoranthene and benzo[k]fluoranthene peaks co-eluted and are reported as a total concentration of the the two compounds; the same was true for indeno[1,2,3-c,d]pyrene and dibenzo[a,h]anthracene. The higher standard deviations for phenanthrene, fluoranthene, and pyrene are most likely due to greater baseline noise near their retention times than in other areas of the chromatogram.

6.2 Fractionated Extraction with SFE at 90 °C and 300 atm

The SFE results from Soil A aged PAH spike showed that both organic modifier and increasing temperature resulted in better recoveries of PAHs. The SFE of native PAHs from Soil C were conducted using the same series of extraction conditions used to test Soil A. The series of conditions included 90 °C and 300 atm; 90 °C, 300 atm, and organic modifier; and 150 °C, 300 atm, and organic modifier. Results should indicate whether or not the SFE information from Soil A can be used as a template for SFE method development for Soil C.

The first SFE condition tested was 90 °C and 300 atm with no organic modifier (Table 6.2). Concentrations determined by SFE were reported as percent recovery based on the concentrations obtained by Soxhlet extraction (Table 6.3). The rates of extraction are shown in Figure 6.1. The recoveries of naphthalene and phenanthrene were well over

Table 6.1

Concentration of Native PAHs in Soil C Determined by Soxhlet Extraction

PAH	Concentration (µg of PAH/g of dry soil)
Naphthalene	11 ± 3
Phenanthrene	82 ± 20
Fluoranthene	75 ± 15
Pyrene	93 ± 22
Benzo[a]anthracene	57 ± 7
Chrysene	63 ± 6
Benzo[b]fluoranthene & Benzo[k]fluoranthene	89 ± 6
Benzo[a]pyrene	60 ± 8
Indeno[1,2,3-c,d]pyrene & Dibenzo[a,h]anthracene	59 ± 9
Benzo[ghi]perylene	55 ± 4

Table 6.2

SFE Conditions used for the Extraction of Native PAHs from Soil C

Prepmaster Method	
Pressure	300 atm
Temperature	90 °C
Extraction Time	1 minute Static 5 minutes Dynamic 5 minutes Dynamic 10 minutes Dynamic 10 minutes Dynamic
Extraction Vessel Size	3 ml
Accutrap Method	
Collection Temperature	-20 °C
Desorption Temperature	15 °C
Desorption Rinse	3 ml of methanol at 1.0 ml/min
Flush Temperature	15 °C
Flush Rinse	5 ml of methanol at 1.0 ml/min
Restrictor	150 °C 3.2 ml/min flowrate

Table 6.3

Percent Recovery of PAHs from Soil C with the SFE at 90 °C and 300 atm

Cummulative Extraction Time (min)	5	10	20	30
Naphthalene	125 ± 33	177 ± 36	194 ± 40	198 ± 42
Phenanthrene	88 ± 18	113 ± 22	126 ± 22	130 ± 22
Fluoranthene	52 ± 3	76 ± 6	94 ± 6	101 ± 5
Pyrene	54 ± 6	80 ± 8	102 ± 8	110 ± 7
Benzo[a]anthracene	20 ± 5	28 ± 10	42 ± 10	48 ± 11
Chrysene	18 ± 2	27 ± 6	43 ± 6	52 ± 5
Benzo[b]fluoranthene & Benzo[k]fluoranthene	8 ± 1	13 ± 4	22 ± 5	29 ± 6
Benzo[a]pyrene	7 ± 3	13 ± 2	20 ± 4	25 ± 6
Indeno[1,2,3-c,d]pyrene & Dibenzo[a,h]anthracene	2 ± 1	3 ± 1	5 ± 1	7 ± 1
Benzo[ghi]perylene	1 ± 1	2 ± 1	4 ± 1	6 ± 2

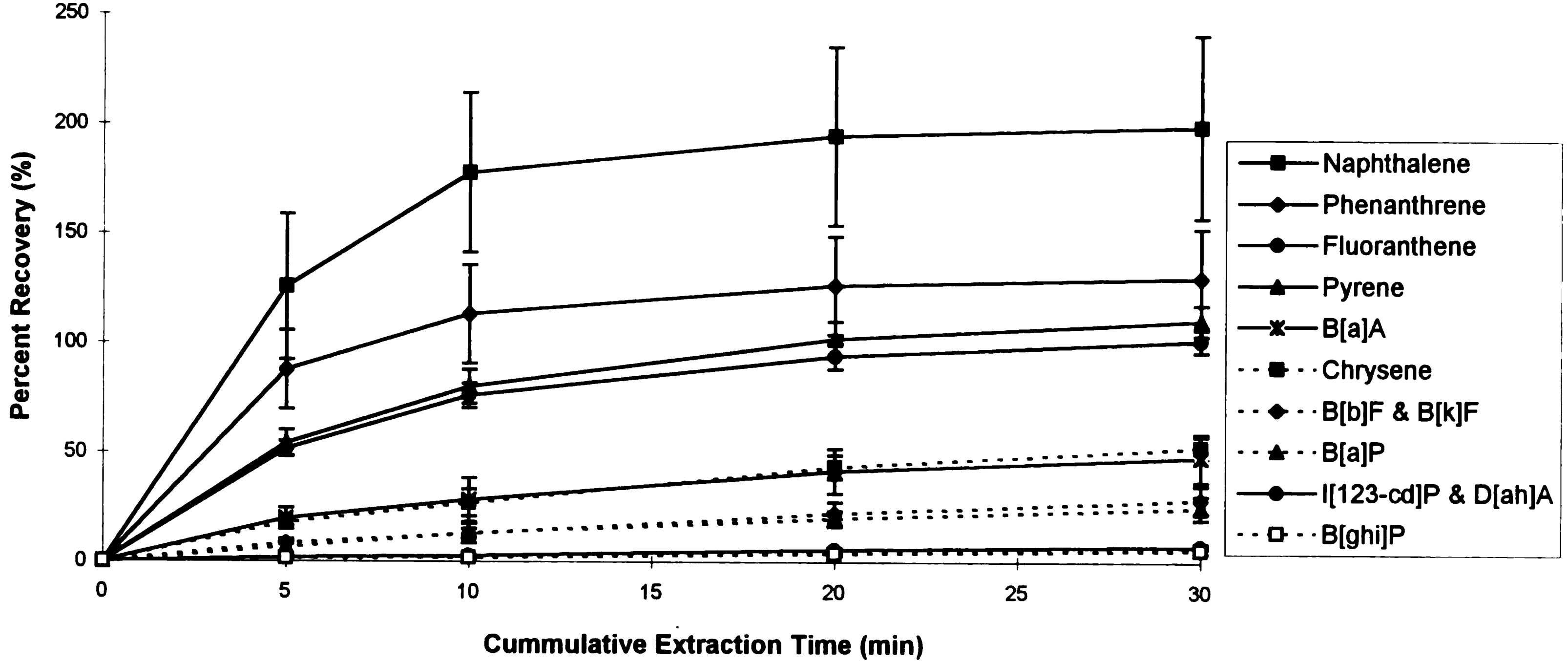


Figure 6.1
Fractionated Collection During SFE of Native PAHs from Soil C with SFE at 90 °C and 300 atm

100% of the Soxhlet values. The high standard deviation for naphthalene may be due to its volatility. The high standard deviation for the phenanthrene may be due to baseline noise in the region of the chromatogram were the peak elutes. Fluoranthene and pyrene were both extracted at 100% efficiency, but all other compounds had recoveries of <50%.

6.3 45 Minute Dynanmic Extration with SFE at 90 °C and 300 atm

The next set of extractions were performed using the same SFE conditions listed in Table 6.2, except that the collection of the analyte was not fractionated. Instead, one 45 minute dynamic extraction replaced the fractionated dynamic steps. This was done for two reasons: (1) fractionated collection of analytes makes the concentrations in the samples low and this was a concern with regard to GC analysis, and (2) the longer extraction time might increase recovery of the higher molecular weight PAHs. The results of the extraction are shown in Table 6.4. The results of the 45 minute dynamic extraction and the fractionated collection are compared graphically in Figure 6.2. It appears that the GC analysis of the fractionated samples was accurate, as the 45 minute dynamic recoveries were only slightly higher than the fractionated results. This was most likely the result of the extra 15 minutes of dynamic extraction time, which allowed a larger volume of CO_2 to be used as extractant for PAH recovery.

Table 6.4

Percent Recovery of PAHs from Soil C with the SFE at 90 °C and 300 atm

Cummulative Extraction Time (min)	45
Naphthalene	231 ± 50
Phenanthrene	140 ± 4
Fluoranthene	110 ± 20
Pyrene	129 ± 12
Benzo[a]anthracene	60 ± 5
Chrysene	61 ± 5
Benzo[b]fluoranthene & Benzo[k]fluoranthene	37 ± 3
Benzo[a]pyrene	27 ± 2
Indeno[1,2,3-c,d]pyrene & Dibenzo[a,h]anthracene	11 ± 1
Benzo[ghi]perylene	9 ± 1

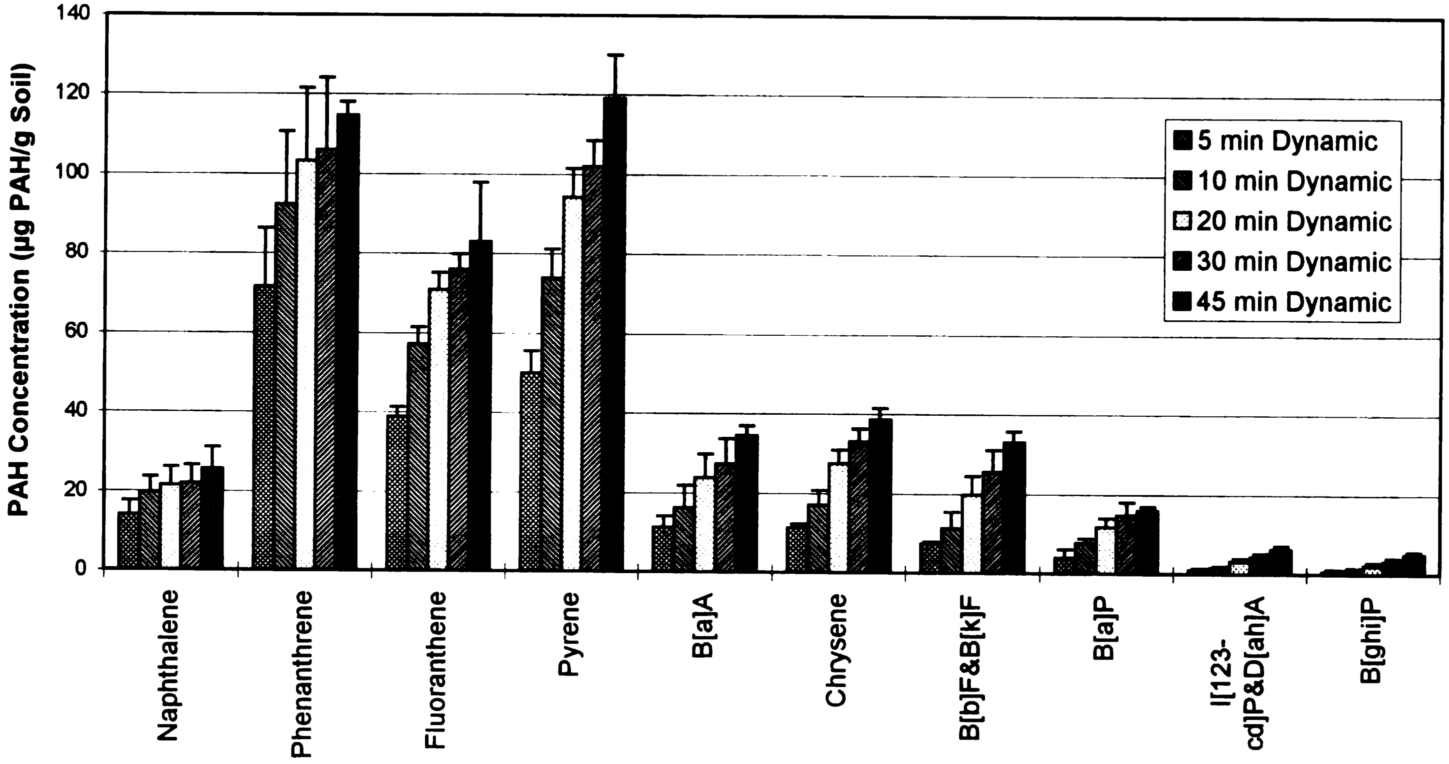


Figure 6.2

PAH Concentrations Determined by Fractionated Collection and 45 Minute Dynamic

6.4 Fractionated Extraction using an Organic Modifier

The results of the fractionated extraction using methylene chloride as an organic modifier with the SFE at 90 °C and 300 atm are shown in Table 6.5. The SFE conditions were the same as those listed in Table 6.2. The Soil C sample was placed in the extraction vessel and 700 µl of methylene chloride was added directly to the soil. The recoveries of the higher molecular weight PAHs were increased compared to the recoveries obtained without methylene chloride as a modifier. The rates of extraction are depicted in Figure 6.3.

6.5 45 Minute Dynanmic Extration using Organic Modifier

The results of the 45 minute dynamic extraction with methylene chloride as a modifier and the SFE at 90 °C and 300 atm are shown in Table 6.6. SFE conditions were the same as those listed in Table 6.2, with the exception of a 45 minute dynamic extraction replacing the fractionated extraction steps. The results of the 45 minute dynamic extraction and the fractionated collection are compared graphically in Figure 6.4. Also, a graphical comparison of the 45 minute dynamic extraction with and without organic modifier is shown in Figure 6.5. The modifier had the largest effect on the recovery of the higher molecular weight PAHs.

Table 6.5

Percent Recovery of PAHs from Soil C using an Organic Modifier

Cummulative Extraction Time (min)	5	10	20	30
Naphthalene	150 ± 51	171 ± 53	179 ± 53	179 ± 54
Phenanthrene	129 ± 15	144 ± 14	150 ± 13	150 ± 14
Fluoranthene	76 ± 9	86 ± 10	93 ± 9	96 ± 9
Pyrene	90 ± 12	102 ± 12	112 ± 11	116 ± 11
Benzo[a]anthracene	51 ± 6	56 ± 6	60 ± 6	60 ± 7
Chrysene	46 ± 4	52 ± 4	59 ± 3	65 ± 3
Benzo[b]fluoranthene & Benzo[k]fluoranthene	36 ± 7	41 ± 7	46 ± 7	49 ± 8
Benzo[a]pyrene	32 ± 5	35 ± 8	39 ± 9	41 ± 9
Indeno[1,2,3-c,d]pyrene & Dibenzo[a,h]anthracene	24 ± 5	25 ± 5	25 ± 6	26 ± 6
Benzo[ghi]perylene	17 ± 5	18 ± 5	19 ± 5	20 ± 5

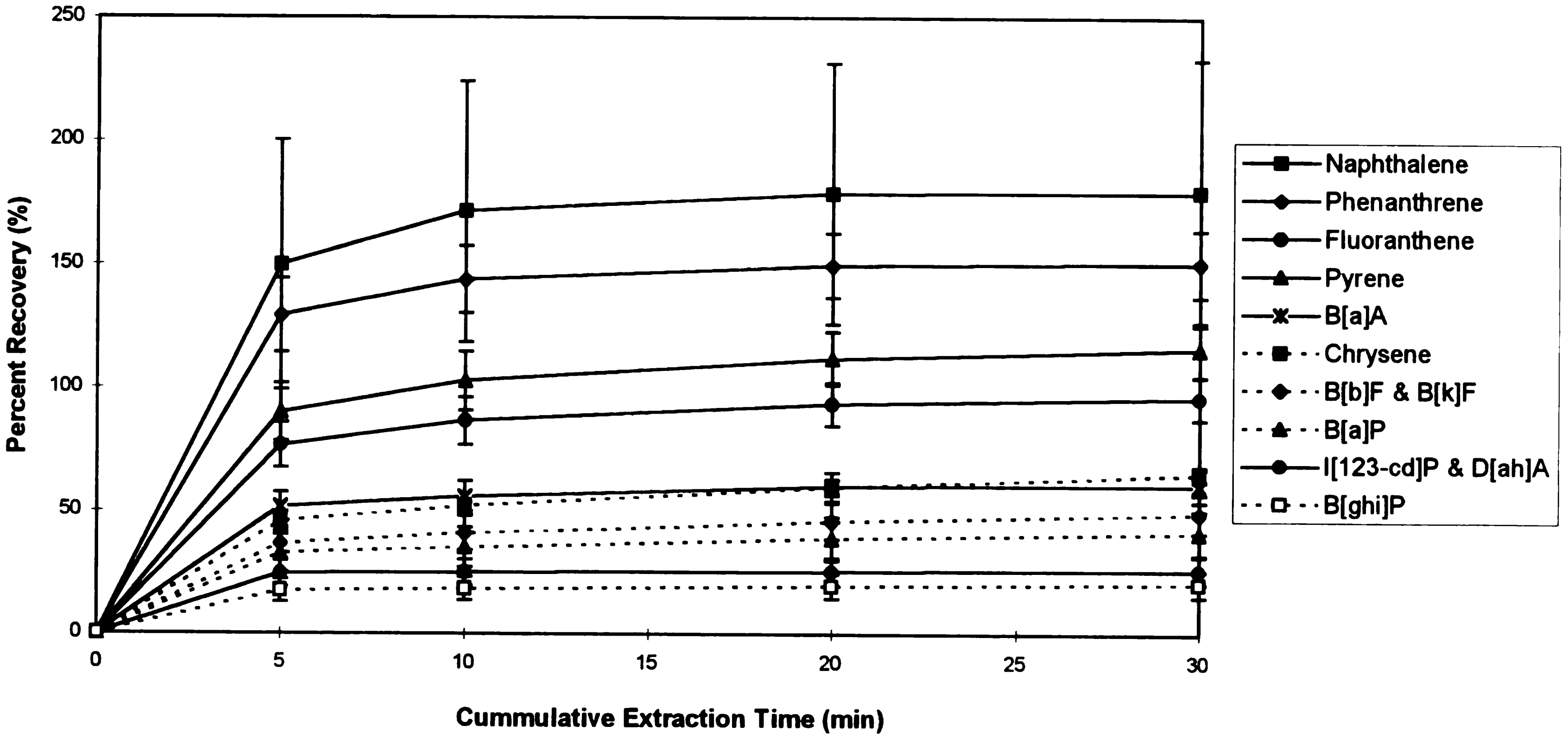


Figure 6.3
Fractionated Collection During SFE of Native PAHs from Soil C with
700 µl Methylene Chloride Modifier and SFE at 90 °C and 300 atm

Table 6.6

Percent Recovery of PAHs from Soil C using an Organic Modifier

Cummulative Extraction Time (min)	45
Naphthalene	202 ± 25
Phenanthrene	162 ± 2
Fluoranthene	109 ± 9
Pyrene	129 ± 6
Benzo[a]anthracene	68 ± 4
Chrysene	71 ± 4
Benzo[b]fluoranthene & Benzo[k]fluoranthene	57 ± 4
Benzo[a]pyrene	52 ± 3
Indeno[1,2,3-c,d]pyrene & Dibenzo[a,h]anthracene	33 ± 4
Benzo[ghi]perylene	26 ± 1

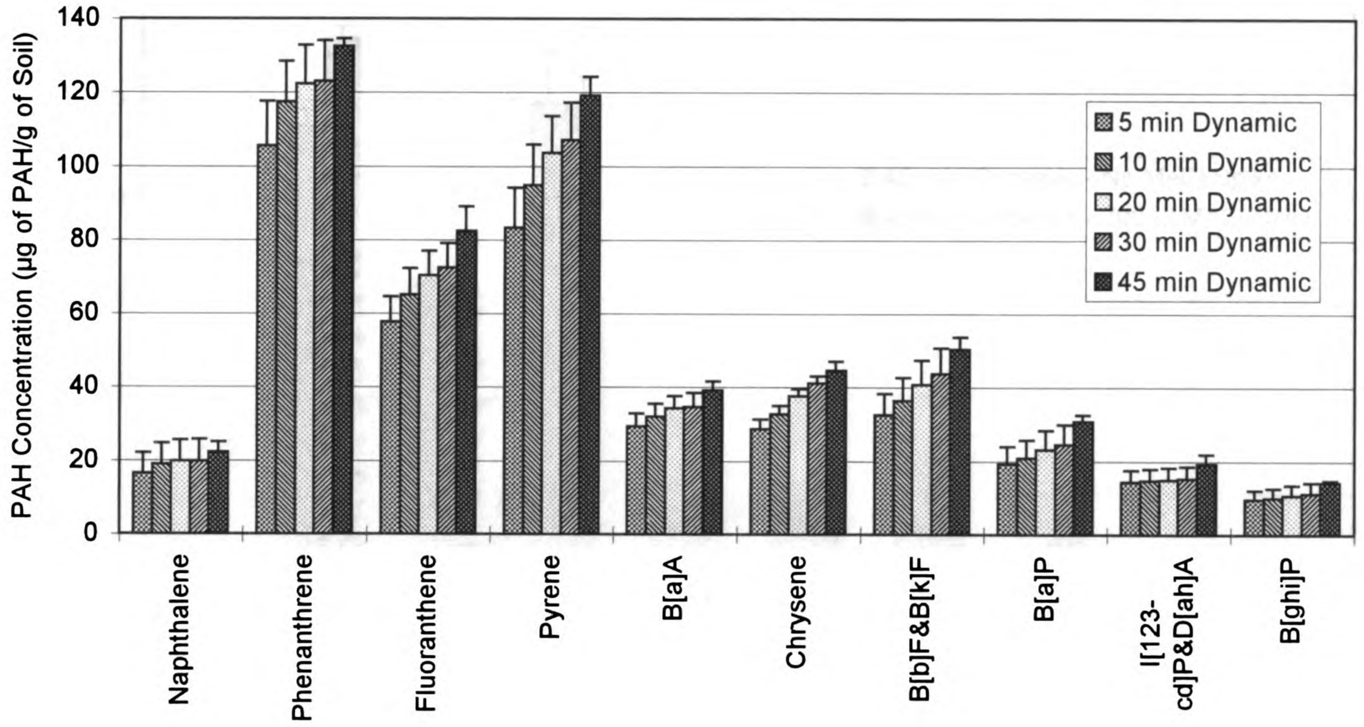


Figure 6.4
PAH Concentrations Determined by Fractionated Collection and 45 Minute Dynamic with 700 µl Methylene Chloride Modifier

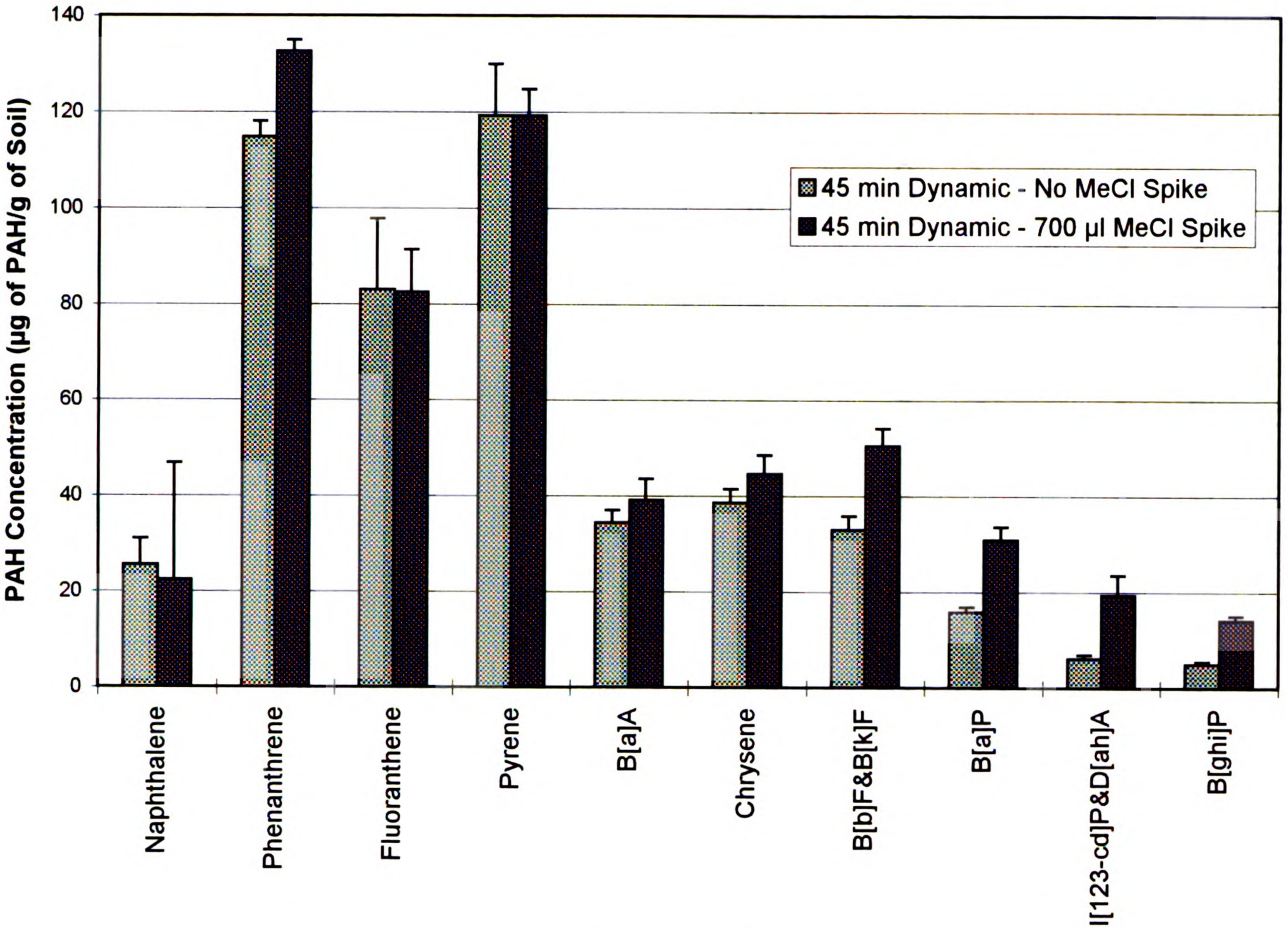


Figure 6.5
Concentration of PAHs Recovered from Soil C
with and without Methylene Chloride Modifier

6.6 Fresh Spike Recovery using an Organic Modifier

The low recoveries of some of the PAHs could be a result of solubility limitations of the PAHs in the supercritical fluid. To determine if solubility is a limiting factor a fresh PAH spike was added to Soil C. The fresh PAH spike of 200 µl was added to the soil sample in the extraction vessel and then 500 µl of methylene chloride was added as organic modifier. The sample was extracted with the same SFE conditions as shown in Table 6.2, with the exception that the dynamic extraction time was 45 minutes, instead of the analyte collection being fractionated. The results are shown in Table 6.7. The recoveries indicated that the solubility of the PAHs was not a factor affecting the efficient extraction of PAHs.

The recoveries were calculated by subtracting the PAH concentrations determined by SFE of the native PAHs from the concentrations of the spiked plus native PAHs in Soil C. The SFE conditions were the same for the extraction of the native PAHs and for the spiked plus native PAHs. The calculation was done for all PAH compounds that were quantitated. Only four of the PAHs were in the fresh spike solution. The results show that the concentration of the remaining native PAHs were within ± 4 µg/g for the two SFE extractions performed (Table 6.8).

Table 6.7

Percent Recovery of Fresh Spiked PAHs from Soil C using an Organic Modifier

Cummulative Extraction Time (min)	45
Naphthalene	80 ± 11
Phenanthrene	113 ± 21
Pyrene	110 ± 21
Benzo[a]pyrene	81 ± 6

Table 6.8

Difference in Native PAH and Spiked plus Native PAH Concentrations

	Native (µg/g of soil)	Spiked + Native (µg/g of soil)	Difference (µg/g of soil)
Naphthalene	22 ± 3	85 ± 10	63 ± 11
Phenanthrene	133 ± 2	214 ± 21	81 ± 21
Fluoranthene	82 ± 7	80 ± 7	-3 ± 9
Pyrene	119 ± 5	203 ± 20	84 ± 21
Benzo[a]anthracene	39 ± 3	43 ± 6	4 ± 7
Chrysene	45 ± 3	44 ± 7	-1 ± 7
Benzo[b]fluoranthene & Benzo[k]fluoranthene	50 ± 3	51 ± 5	1 ± 6
Benzo[a]pyrene	31 ± 2	89 ± 6	58 ± 6
Indeno[1,2,3-c,d]pyrene & Dibenzo[a,h]anthracene	19 ± 3	23 ± 1	3 ± 3
Benzo[ghi]perylene	14 ± 1	17 ± 1	3 ± 1

6.7 Solvent Rinse of Cryogenic Trap

The other parameter that might have had an impact on extraction efficiencies was the type of solvent used to rinse the cryogenic trap. Methanol had been used because it was compatible with the delivery pump. Methylene chloride may be a better rinse solvent for PAHs, because PAH solubility is higher in methylene chloride than in methanol. The results of using methylene chloride and methanol as rinse solvents are shown in Table 6.9. Extractions were performed using an organic modifier with the SFE conditions the same as in Table 6.2, except a 45 minute dynamic extraction was used instead of fractionated steps. A statistical analysis using the Student t test was performed with a 95% confidence interval and there was no statistical difference in PAH recovery between the two rinse solvents.

Table 6.9

Percent Recovery of PAHs from Soil C using an Organic Modifier and Different Cryogenic Trap Rinse Solvents

Cryogenic Trap Rinse Solvent	MeOH	$MeCl_2$
Extraction Time (min)	45	45
Naphthalene	202 ± 25	174 ± 31
Phenanthrene	162 ± 2	154 ± 10
Fluoranthene	109 ± 9	112 ± 10
Pyrene	129 ± 6	123 ± 6
Benzo[a]anthracene	68 ± 4	75 ± 6
Chrysene	71 ± 4	68 ± 5
Benzo[b]fluoranthene & Benzo[k]fluoranthene	57 ± 4	53 ± 4
Benzo[a]pyrene	52 ± 3	52 ± 5
Indeno[1,2,3-c,d]pyrene & Dibenzo[a,h]anthracene	33 ± 4	31 ± 7
Benzo[ghi]perylene	26 ± 1	28 ± 6

6.8 Aged Spike Recovery using an Organic Modifier

The fresh PAH spike extraction from Soil C indicated that solubility was not a limiting factor affecting the SFE efficiency for the PAHs. The extraction process could instead be limited by the diffusion of the analytes from the matrix. The aged PAH spiked Soil C was used to show any indications that diffusion limitations may possibly be the cause of the low recoveries of the native PAHs. The extractions were performed using both SFE and Soxhlet extraction techniques. The SFE conditions were the same as those listed in Table 6.2, with the exception that the dynamic extraction time was 45 minutes and the sample collection was not fractionated. The results were compared to the theoretical PAH concentrations determined from the amount of PAHs spiked onto the soil (Figure 6.6). The results were obtained by subtracting the native PAH concentrations from the native plus aged spike concentrations, as determined by both SFE and Soxhlet extraction (Table 6.10). The results indicate that diffusion may be a limiting factor for the SFE conditions used as the ring size and molecular weight of the PAHs increase. The Soxhlet extraction results show less of a dependence on ring size and molecular weight, but the Soxhlet extraction has a higher deviation in the triplicate samples extracted than the SFE.

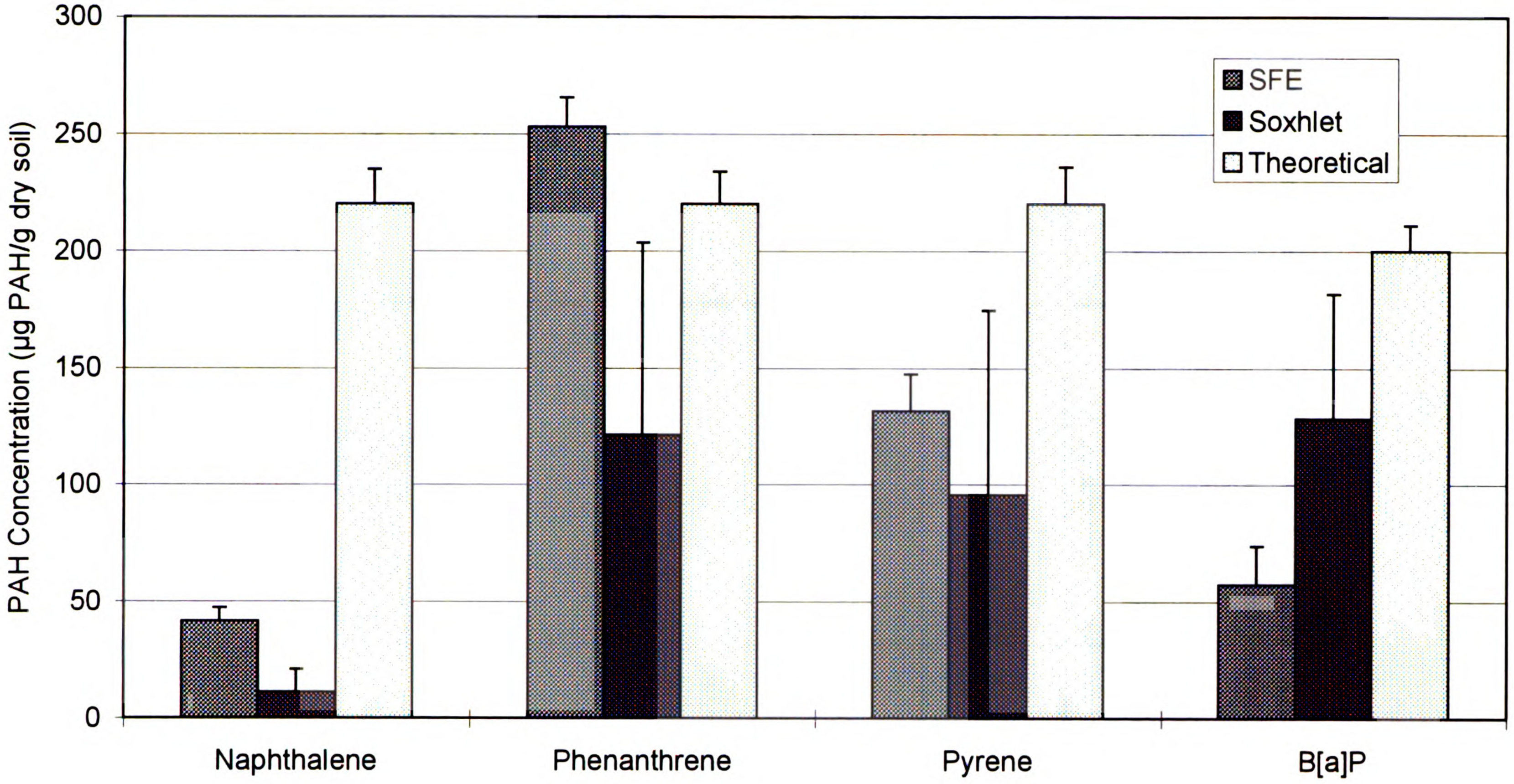


Figure 6.6
Aged PAH Spike Recovery from Soil C by SFE and Soxhlet Extraction Methods

Table 6.10

Difference in Native PAH and Spiked plus Native PAH Concentrations as Determined by SFE and Soxhlet Extraction

	SFE (µg/g of soil)	Soxhlet Extraction (µg/g of soil)
Naphthalene	41 ± 6	11 ± 10
Phenanthrene	253 ± 12	121 ± 82
Fluoranthene	2 ± 11	-4 ± 29
Pyrene	132 ± 16	96 ± 79
Benzo[a]anthracene	-1 ± 5	-12 ± 18
Chrysene	-3 ± 4	-11 ± 20
Benzo[b]fluoranthene & Benzo[k]fluoranthene	-7 ± 9	-11 ± 18
Benzo[a]pyrene	57 ± 17	128 ± 54
Indeno[1,2,3-c,d]pyrene & Dibenzo[a,h]anthracene	-2 ± 4	-7 ± 14
Benzo[ghi]perylene	-2 ± 3	-10 ± 12

6.9 Fractionated Extraction using an Organic Modifier and 150 °C

The indication that diffusion may possibly be a limiting factor for the extraction of the native PAHs led to the decision to increase the extraction temperature. Increasing the extraction temperature could be beneficial for a system that is limited by diffusion. The higher temperature could provide the energy necessary to overcome the diffusion energy barrier that may exist. TheSFE conditions were the same as those listed in Table 6.2, with the exception of the temperature being 150 °C instead of 90 °C. The results of the fractionated collection are shown in Table 6.5 and Figure 6.7 and are based on the PAH concentrations obtained from the Soxhlet extraction of Soil C. The increase in the extraction temperature did not satisfactorily increase the recovery of the native PAHs from Soil C compared to the results obtained at 90 °C.

Table 6.11

Percent Recovery of PAHs from Soil C using an Organic Modifier and Temperature of 150 °C

Cummulative Extraction Time (min)	5	10	20	30
Naphthalene	144 ± 17	207 ± 15	230 ± 17	234 ± 17
Phenanthrene	122 ± 20	146 ± 7	153 ± 7	153 ± 7
Fluoranthene	69 ± 18	83 ± 10	93 ± 7	95 ± 6
Pyrene	79 ± 17	97 ± 9	111 ± 5	115 ± 5
Benzo[a]anthracene	49 ± 13	56 ± 10	63 ± 8	66 ± 8
Chrysene	44 ± 12	53 ± 8	64 ± 7	69 ± 4
Benzo[b]fluoranthene & Benzo[k]fluoranthene	37 ± 11	42 ± 8	49 ± 8	53 ± 7
Benzo[a]pyrene	36 ± 9	40 ± 7	45 ± 6	47 ± 5
Indeno[1,2,3-c,d]pyrene & Dibenzo[a,h]anthracene	24 ± 6	25 ± 5	26 ± 5	26 ± 5
Benzo[ghi]perylene	21 ± 7	22 ± 6	24 ± 6	25 ± 6

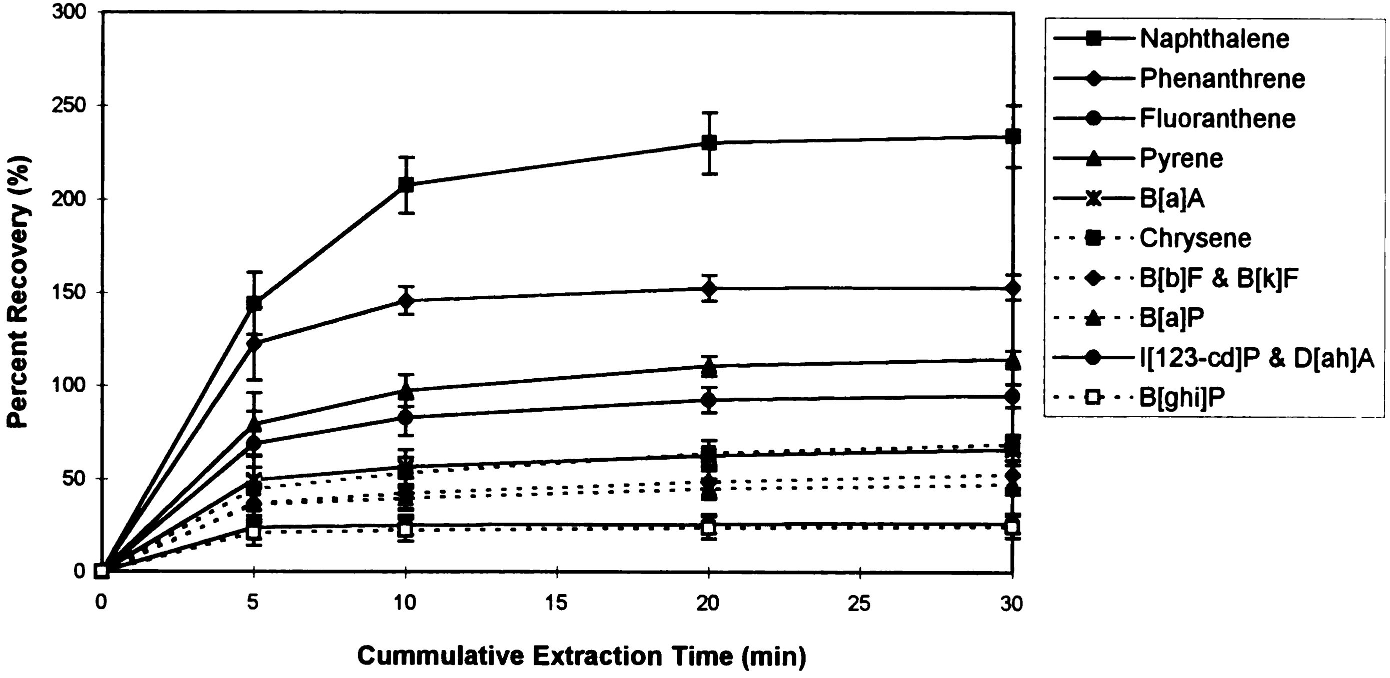


Figure 6.7
Percent Recovery of Native PAHs from Soil C using Organic Modifier with SFE at 150 °C and 300 atm

6.10 CO2 and Modifier Contact Time

The addition of methylene chloride modifier had a large effect on the recovery of native PAHs from Soil C. The largest difference in recovery was noticed in the first five minutes of the extraction process. The methylene chloride added to the extraction cell was most likely flushed from the cell during the first 5 minute dynamic extraction. The contact time of the methylene chloride was changed by changing the static extraction time prior to the dynamic extraction to try to increase the recovery of the native PAHs. The results of changing the static and dynamic extraction times are shown in Figure 6.8 and Figure 6.9. There was a statistically significant increase in the recovery of phenanthrene, fluoranthene, pyrene, benzo[a]anthracene, chrysene, benzo[b]fluoranthene, and benzo[k]fluoranthene when the static extraction time was increased from 1 minute to 5 minutes with a 5 minute dynamic extraction time. Again, the Student t test with a 95% confidence interval was used to test the data. There was no a statistically significant increase in the recovery of the PAHs over that obtained by the 5 minute static/5 minute dynamic when the dynamic extraction time was increased to 10 minutes.

The addition of methylene chloride to the 5 minute static/5 minute dynamic extraction was then compared to the 45 minute dynamic extractions with and without methylene chloride added (Figure 6.10). As the molecular weight of the PAHs increases, there was a larger affect noticed from the addition of methylene chloride modifier. The 5 minute static/5 minute dynamic extraction with methylene chloride modifier showed a statistically significant increase in the recovery of benzo[a]pyrene, indeno[1,2,3-c,d]pyrene

& dibenzo[a,h]anthracene, and benzo[ghi]perylene when compared to the recovery obtained using a 1 minute static/45 minute dynamic without methylene chloride modifier added to the extraction cell. When compared to the 1 minute static/45 minute dynamic with methylene chloride modifier added, the 5 minute static/5 minute dynamic had statistically lower recoveries for all the PAHs except naphthalene and indeno[1,2,3-c,d]pyrene & dibenzo[a,h]anthracene.

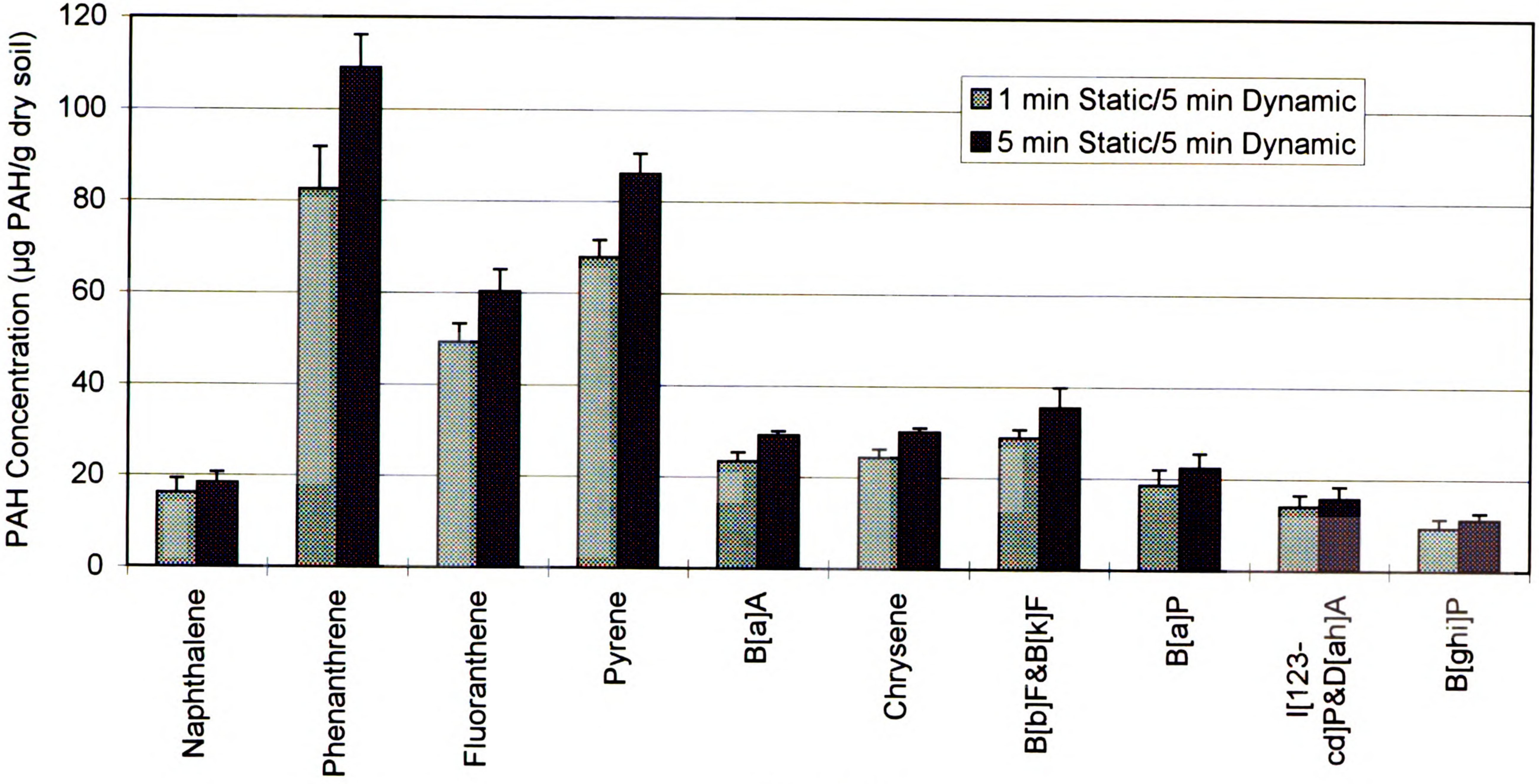


Figure 6.8
Concentration of PAHs Recovered from Soil C with Change in Static Extraction Time

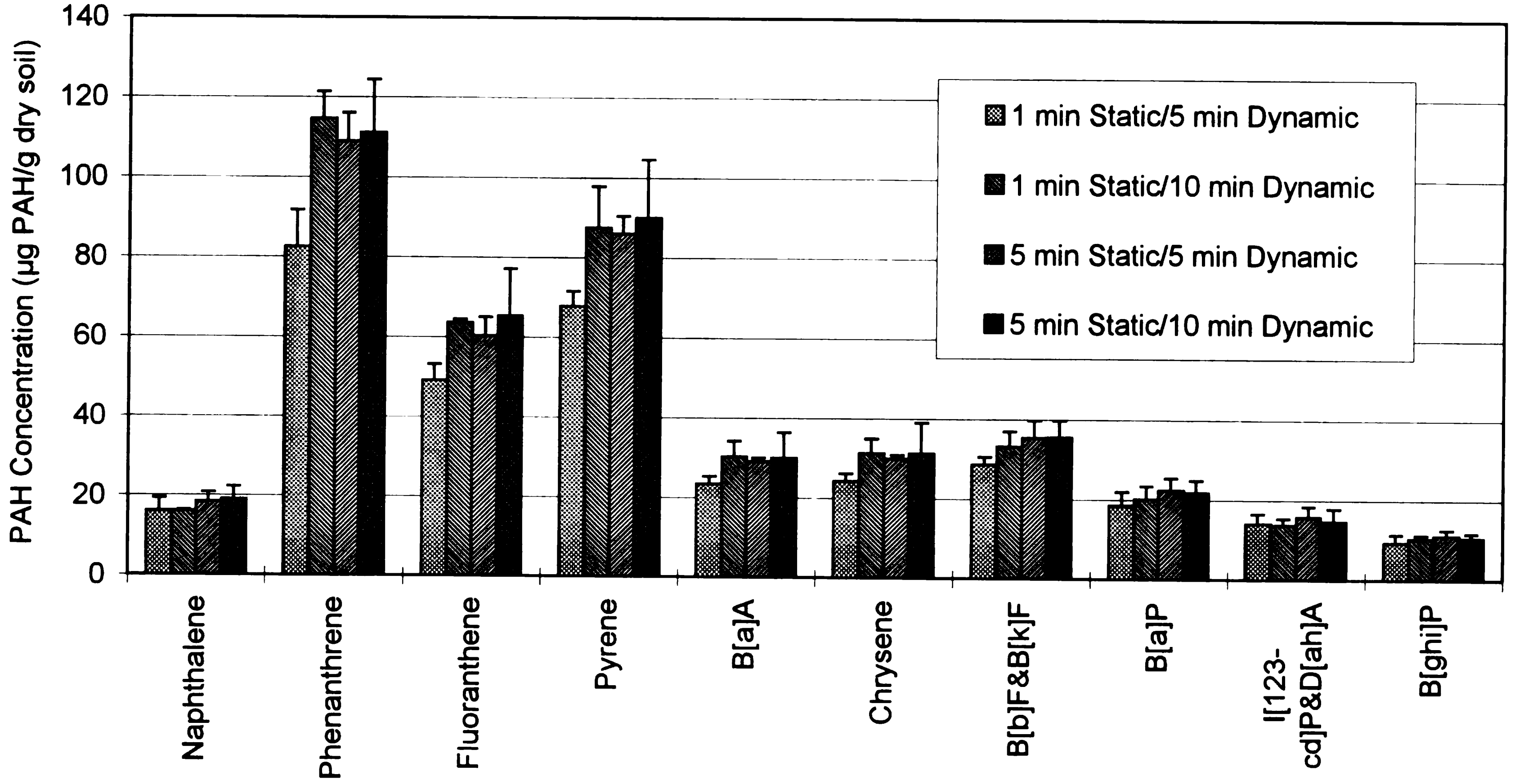


Figure 6.9
Concentration of PAHs Recovered from Soil C with Change in Static and Dynamic Extraction Time

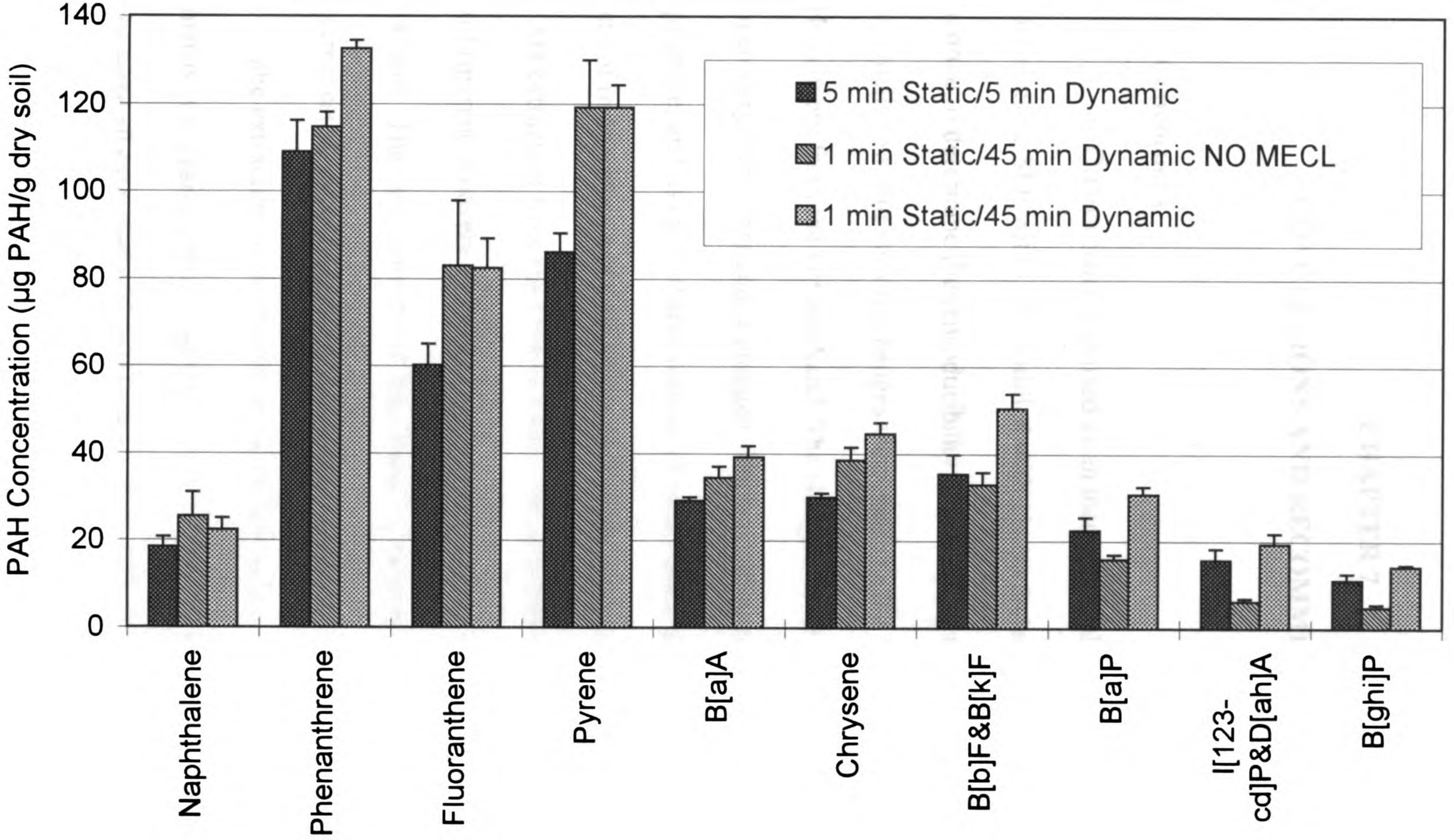


Figure 6.10
Concentration of PAHs Recovered from Soil C after Different Extraction Times

CHAPTER 7

CONCLUSIONS AND RECOMMENDATIONS

7.1 Conclusions

Ottawa Sand was used as an inert material from which PAHs could be extracted using SFE with CO_2 modified with 1% methylene chloride. Initially, extractions were done to determine the reproducibility of the SFE extraction efficiencies. Recoveries were initially low, but various changes were made to the SFE apparatus that increased the PAH recovery from the Ottawa Sand. The changes made to the SFE included fixing the nitrogen purge line, changing a plugged vent line, lowering the cryogenic trap desorption temperature, and using methanol instead of methylene chloride as a rinse solvent. A variety of temperature and pressure combinations were then tested to determine the effect on PAH extraction from the Ottawa Sand. The only effect noticed was the increase in benzo[a]pyrene recovery when the pressure was increased from 200 atm to either 300 atm or 400 atm. The recoveries for all four compounds were >85% at all temperatures and pressures tested, except when 200 atm pressure was used.

The extraction of the freshly spiked PAHs proved to be a little more difficult when the matrix was changed from Ottawa Sand to Soil A. Naphthalene, phenanthrene, and pyrene were all extracted with recoveries > 85% at 90 °C and 300 atm, but benzo[a]pyrene had a recovery of only 66%. Increasing the temperature did not increase the benzo[a]pyrene recovery. The addition of methylene chloride as an organic modifier did increase the recovery of benzo[a]pyrene, with the temperature at 90 °C and pressure at 300 atm. The recovery of the PAHs from Soil A that had aged for over a year was affected by both the organic modifier and temperature. The addition of methylene chloride as an organic modifier increased the recovery of benzo[a]pyrene from 49% to 77%. Increasing the extraction temperature from 90 °C to 150 °C and adding methylene choride increased the benzo[a]pyrene recovery from 49% to 93%.

The native PAHs in Soil C proved to be much more difficult to extract than the freshly or aged spiked PAHs in Soil A. The SFE recoveries were compared to the Soxhlet extraction recoveries. Not only was the overall recovery of PAHs increased when methylene chloride was added as an organic modifier, but the rate of extraction was also increased. In the first ten minutes of extraction with an organic modifier, all of the recoveries were at or above the recoveries obtained after 30 minutes of extraction without methylene chloride being added to the extraction vessel.

7.2 Recommendations

The development of a SFE method for PAHs in soils will be dependent upon the matrix from which the PAHs are extracted. The difficult part of applying SFE as an analytical tool is determining how the variables will effect the extraction performance. In many cases known concentrations of analytes are spiked onto the matrix and extracted to determine recovery efficiencies. This method can give a false recovery efficiencies, which will be higher in most cases than the recovery efficiencies of the native PAHs.

Future work could include trying to determine a method by which extraction efficiencies could be determined. This might consist of obtaining a variety of soil samples from contaminated sites and performing SFE analysis to determine contaminant concentrations. A database of information about the mechanical characteristics of the soil, the age of the contamination, the SFE variables, and other conditions that affect the recovery of analytes from the soil would be useful. This database could be accessed when a SFE method is needed to analyze a contaminated soil sample. It would serve as a starting point for the method development and also provide an indication of what type of recoveries should be expected. Comparison to other extraction methods, such as soxhlet or sonication, would also be a helpful addition to this database.

Although comparing SFE to conventional extraction methods can be beneficial, one of the reasons for using SFE is to eliminate the use of conventional methods that produce large amounts of organic waste. The SFE results could be compared to toxicity information instead of being compared to conventional extraction methods. The basis of cleaning up contaminated soils is that there is a hazard due to the toxic effects of the pollutants present. If a link between SFE extraction recoveries and toxicity data could be developed it would prove to be a helpful database of information.

Finally, there could be more work done to determine the effect of modifiers on the extraction efficiencies obtained with SFE. There was a large affect seen in this research when methylene chloride modifier was added to the extraction cell. The financial limitations made it impractical to order more than one concentration of methylene chloride mixed into the tank of SFE CO_2. It would be interesting to see how the increase in methylene chloride concentration in the SFE CO2 tank would affect the recovery efficiencies by having a continual supply of the modifier at a higher concentration. The type of modifier that is used for the extraction could be investigated as well. The modifier seemed to play the most important role for increasing the extraction efficiencies of the soils tested in this research.

APPENDICES

APPENDIX A

APPENDIX A

PHYSICAL CHARACTERISTICS AND STRUCTURES OF PAHs

Table A

Physical Characteristics and Structures of PAHs

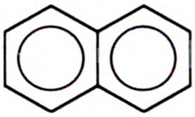
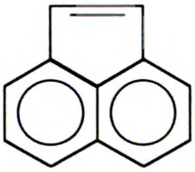
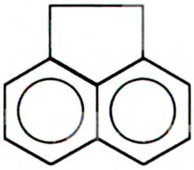
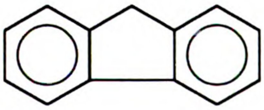
Compound	Empirical Formula	Molecular Weight (g/mol)	Melting Point (°C)	Boiling Point (°C)	Structure
Naphthalene	**$C_{10}H_8$**	**128.16**	**80.2**	**217.9**	
Acenaphthylene	**$C_{12}H_8$**	**152.20**	**92-93**	**265-275**	
Acenaphthene	**$C_{12}H_{10}$**	**154.21**	**95**	**279**	
Fluorene	**$C_{13}H_{10}$**	**166.21**	**116-117**	**295**	

Table A (cont'd)

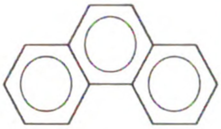
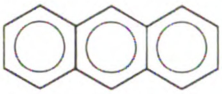
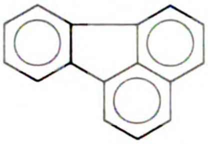
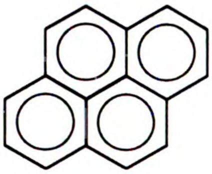
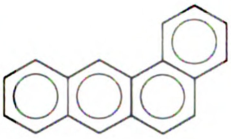
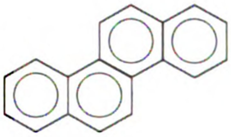
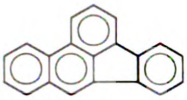
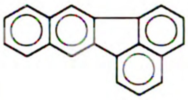
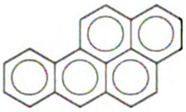
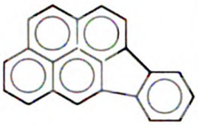
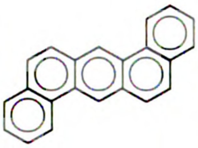
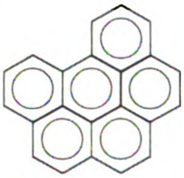
Compound	Empirical Formula	Molecular Weight (g/mol)	Melting Point (°C)	Boiling Point (°C)	Structure
Phenanthrene	$C_{14}H_{10}$	178.22	100	340	
Anthracene	$C_{14}H_{10}$	178.22	218	342	
Fluoranthene	$C_{16}H_{10}$	202.26	NA	375	
Pyrene	$C_{16}H_{10}$	202.24	156	404	
Benz[a]anthracene	$C_{18}H_{12}$	228.28	162	435	
Chrysene	$C_{18}H_{12}$	228.28	254	448	

Table A (cont'd)

Compound	Empirical Formula	Mol. Weight (g/mol)	Melting Point (°C)	Boiling Point (°C)	Structure
Benzo[b]fluoranthene	$C_{20}H_{12}$	252.32	163-165	NA	
Benzo[k]fluoranthene	$C_{20}H_{12}$	252.32	215-217	NA	
Benzo[a]pyrene	$C_{20}H_{12}$	252.30	179-179.3	310-312	
Indeno[123-cd]pyrene	$C_{22}H_{12}$	276.34	NA	NA	
Dibenz[ah]anthracene	$C_{22}H_{14}$	278.35	266-267	524	
Benzo[ghi]perylene	$C_{22}H_{12}$	276.34	277-279	>500	

APPENDIX B

APPENDIX B

SOXHLET EXTRACTION PROCEDURE FOR PAH CONTAMINATED SOIL

I. Extraction
II. Clean-up
III. Concentration

I. Extraction Procedure for PAH Contaminated Soil

Extraction

1. Pour 300 ml of methylene chloride ($MeCl_2$) into the round bottom flask.
2. Add a few boiling chips to the flask.
3. Place the thimble containing the soil sample in the soxhlet extractor and connect glassware.
4. Turn heating blocks on high and turn cold water on to the condenser.
5. Extract samples about 24 hours.

Solvent Exchange

1. After the extraction is complete, remove the thimble from the soxhlet extractor and discard the $MeCl_2$ contained in the thimble holder. Allow the $MeCl_2$ containing the analyte in the round bottom flask to cool before disconnecting the thimble holder. Squirt methanol on the outside of the glassware in order to assist the cooling process of the vapors in the round bottom.
2. Distill the remaining $MeCl_2$ in the round bottom flask until there is approximately 100 ml left. Avoid concentrating the liquid to a level where solids form or attach to the walls of the flask.
3. Remove the unit (cooling the glassware again before disconnecting any joints) and add 200 ml of cyclohexane to the flask. Distill off the rest of the $MeCl_2$.
4. Remove the unit (cool the glassware) and place a ground glass stopper on the flask.
5. The Clean-up Procedure can be started.

II. Clean-up Procedures for PAH Extraction Samples

Before starting this procedure, turn the water bath on

1. Pour the cyclohexane containing the analyte from the round bottom flask into the drying tube (2 cm x 20 cm) containing 30 g of freshly activated silica gel and 15 g of sodium sulfate. The sodium sulfate is placed on top so that the cyclohexane comes in contact with it before reaching the silica gel. Use glass wool to plug the bottom of the drying tube.
2. Rinse the silica gel column with pentane, approximately 60 ml. The last few drops of pentane should be clear, if not, then add more pentane until the eluate is clear.
3. Rinse the silica gel with 60% pentane/40% $MeCl_2$ (v/v) to remove the PAHs. Collect the liquid in a clean flask. Use 200 ml of the 60/40 mixture to rinse the PAHs from the silica gel.
4. Take the flask containing the PAHs in the 60/40 mixture of pentane and $MeCl_2$ and proceed with the concentration operation.

III. Concentration Operation for PAH Extraction Samples

1. Assemble the concentrator tube and the boiling flask using a teflon sleeve at the ground glass joint.
2. Pour the sample into the boiling flask and add a small boiling chip. Rinse the sample flask out with $MeCl_2$ and pour into the boiling flask.
3. Attach the Snyder column, pour 10 ml of $MeCl_2$ down the snyder column, and mount the assembly above the water bath clamping onto the neck of the flask.
4. Lower the assembly into the water bath and secure with clamps.
5. Once the evaporation/concentration is complete (to a few mls), remove the assembly and wipe off the excess water at the base of the flask and around the concentrator tube.
6. Squirt methanol on the glassware to assist in cooling and then remove the Snyder column and add 1 or 2 ml of $MeCl_2$ to the flask and spin. This should rinse down the sides of the flask.
7. Rinse the joint between the flask and tube with $MeCl_2$ and then remove the flask. Be sure to remove the teflon sleeve also.
8. Add $MeCl_2$ to the sample to obtain the desired volume. For most purposes the total volume will usually be 6 ml or 10 ml, note the volume used.
9. Fill two or three GC vials with the sample and discard the rest into the waste bottle. Store the GC vials in the freezer until they can be analyzed on the GC.

APPENDIX C

APPENDIX C

COST COMPARISON OF SFE AND SOXHLET EXTRACTION METHODS

The costs for consumable items of both the SFE and Soxhlet extraction methods are listed in Table B and Table C. Included in the costs are only those items which are non-reusable. For instance, neither extraction method has the cost of glassware included in the cost analysis. Also, labor costs and waste disposal cost have not been included. The cost of the Soxhlet extraction would increase significantly more than the SFE method if waste disposal were included.

The SFE cost is a conservative number and may be actually lower for most samples. This is due to assumptions made about the amount of SFE CO_2 used per sample, the number of frits used per sample, and the number of restrictors used per sample. The two most expensive items for the SFE analysis are the restrictors and SFE CO_2. The assumption made was that the dynamic extraction time would be 45 minutes with a flow rate of 2.2 ml/min. This is highly dependent on the type of matrix and analyte of interest. The number of samples that can be processed using the same restrictor is also dependent on the sample type. The restrictors need to be replaced if and when they become plugged. There may or may not be waxes and other materials within the sample that can plug the restrictor.

Table B

Soxhlet Extraction Consumables Cost

Material	Price	Amount Used per Sample	Cost per Sample
Extraction Thimble, Cellulose, 43 x 123, 1.5 mm wall thickness ‡	$60.00/box of 25 thimbles	1 thimble	$2.40
Methylene Chloride, 99.9%, HPLC Grade †	$155.00/16 L	380 ml	$3.68
Pentane, 99.9%, Capillary GC Grade †	$266.15/16 L	200 ml	$5.32
Cyclohexane, 99.9%, HPLC Grade †	$120.80/8 L	200 ml	$3.02
Sodium Sulfate, 99+%, granular, ACS reagent †	$16.35/500 g	15 g	$0.50
Silica Gel, 99+%, Davidsil grade 644, 100-200 mesh †	$110.50/kg	30 g	$3.32
Total Cost	----------	----------	**$18.24**

‡ Supelco, Bellefonte, PA, USA
† Aldrich, Milwaukee, WI, USA

Table C

SFE Consumables Cost

Material	Price	Amount Used per Sample	Cost per Sample
Septa, heavy duty, 0.01” PTFE bonded over 0.09” silicone †	$19.80/pack of 100	1 septa	$0.20
Restrictors, Stainless Steel ‡	$354.60/pack of 6	1/25 samples	$2.36
Extraction Cell Frits ‡	$80.10/pack of 12	1/25 samples	$0.27
SFE CO_2 w/1% $MeCl_2$ Helium Headspace ✞	$494.00/17.69 kg	70 g	$1.95
Bone Dry CO_2, 99.8% ✞	$35.00/27.22 kg	270 g	$0.35
Nitrogen, 99.99%, High Purity ✞	$36.00/Cylinder Size K	1 tank/200 samples	$0.18
Methanol, 99.9+%, ACS HPLC Grade *	$109.20/16 L	8 ml	$0.06
Total Cost	----------	----------	**$5.30**

† Fisher Scientific, Pittsburgh, PA, USA
‡ Suprex, Pittsburgh, PA, USA
✞ Scott Specialty Gases, Plumsteadville, PA, USA
* Aldrich, Milwaukee, WI, USA

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