Evaluation of drying agents for off-line supercritical fluid extraction

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ABSTRACT

Of 21 potential drying agents investigated, five (anhydrous and monohydrated magnesium sulfate, molecular sieves 3A and 5A and Hydromatrix) were able successfully to prevent restrictor plugging by water during off-line supercritical fluid extraction (*e.g.*, 400 atm CO_2 at 60°C) by retaining the majority of the water (but generally not the analytes of interest) in the extraction cell. Increasing the extraction temperature (*e.g.*, to 150°C) or adding a polar modifier [10% (v/v) methanol] to the CO_2 extraction fluid greatly reduced the amount of water the drying agents retained. However, when 10% (v/v) toluene was used for the extraction, the drying agents were able to retain the majority of the water (*ca.* 80% w/w). Polar and non-polar pollutants were quantitatively extracted from the wet drying agents (*i.e.*, water present), but nearly all of the drying agents selectively retained at least one of the polar analytes if used dry (*i.e.*, no water present), thus demonstrating the need for a spike recovery study to determine the potential for analyte loss. The successful drying agents eliminated restrictor plugging when used with moderately wet [*ca.* 20% (w/w) water at a 1:1 reagent-to-sample ratio] and very wet [*ca.* 90% (w/w) water at 4:1 reagent-to-sample ratio] samples without the need to heat the restrictor or the collection solvent.

INTRODUCTION

Supercritical fluid extraction (SFE) has become a popular alternative to conventional liquid solvent extraction methods. However, major emphasis has been placed on the application of SFE to various analytes and matrices, and less consideration has been given to some of the more practical aspects of SFE such as maintaining the supercritical fluid flow during the extraction of real samples. Most native samples contain water, and the presence of even a small amount of water (>1%, w/w) can prove problematic in SFE, as water freezes in the restrictor tip due to the Joule–Thomson cooling effect of the expanding extraction fluid at the restrictor outlet. As the cooling of the restrictor and collection solvent are very rapid (*i.e.*, the collec-

tion solvent temperature can drop to <0°C within 60 s [1]), plugging due to frozen water can occur within minutes of commencing an extraction. Maintaining the extraction flow is even more difficult with very wet samples [*ca.* >40% (w/w) water] such as fresh plant material and biosludges, as the sample matrix may compact down into the outlet of the extraction cell causing the outlet frit (rather than the restrictor) to block within seconds of pressurizing the extraction cell.

To avoid plugging from water, a number of techniques have been employed. The water can be removed from the sample prior to the extraction by air drying, oven drying (*ca.* 100°C) or freeze-drying. However, such dehydration techniques are time consuming and increase the possibility of the loss of volatile and semi-volatile analytes. Further, the presence of water can in some instances increase the extraction efficiency of the analytes from the sample matrix [2–4] so

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that removal of the water prior to SFE can be disadvantageous.

Instead of removing the water from the sample, dispersants such as sand or glass beads can be mixed with the sample matrix to stop the sample compacting and forming an impervious plug, or dispersants and/or filter-paper can be placed between the sample and the outlet frit to eliminate frit plugging [5–7]. The collection solvent can be maintained above 0°C or the restrictor can be heated to stop water freezing at the tip of the restrictor, but low collection efficiencies for volatile compounds have been associated with both techniques [1].

A recent development has been the use of drying agents to retain the water inside the extraction cell [8–14]. There are several advantages to using drying agents in that they are both sample dispersants and sorbent traps for water. As water potentially remains sorbed to the drying agent in the extraction cell during SFE, the need to control the collection solvent or restrictor temperature may be eliminated. A number of drying agents have been used in SFE with varying success, namely, Hydromatrix (pelletized diatomaceous earth) [8–10], magnesium sulfate [10,14], sodium sulfate [10–13], calcium chloride [13], molecular sieve [11] and silica [11]. However, at present a detailed survey has not been undertaken to evaluate the applicability of the drying agents with several real samples. In this study we investigated a number of conventional drying agents in terms of water retention, analyte retention and extraction flow characteristics under various SFE conditions and with several real samples.

EXPERIMENTAL

Samples and standards

Several drying agents were evaluated, including molecular sieves, water-soluble polymers, diatomaceous earth (Hydromatrix), alumina, silica and metal salts. The drying agents carboxymethylcellulose (Aldrich, Milwaukee, WI, USA), xanthan gum (Aldrich), guar gum (Aldrich), polyacrylamide gel (Aldrich), molecular sieves 3A, 4A, 5A and 13X (Alltech, Deerfield, IL, USA), 100–200-mesh (150–75 μm) chro-

matographic-grade alumina (Fisher Scientific, Fair Lawn, NJ, USA), 100–200-mesh chromatographic-grade silica gel (Fisher Scientific) and 60–100-mesh (250–150 μm) Florisil (Fisher Scientific) were used as received. The drying agents, Hydromatrix (diatomaceous earth received from ISCO, Lincoln, NE, USA), anhydrous sodium sulfate (Fisher Scientific), anhydrous magnesium sulfate (Fisher Scientific), anhydrous calcium sulfate (Fisher Scientific), anhydrous copper sulfate (Aldrich), anhydrous calcium oxide (Fisher Scientific), anhydrous boron trioxide (Fisher Scientific), anhydrous potassium carbonate (Fisher Scientific) and anhydrous calcium chloride (Fisher Scientific) were sieved and the 30–80-mesh (600–180 μm) fractions collected for the study. Monohydrated magnesium sulfate was made by selective dehydration of heptahydrated magnesium sulfate (Fisher Scientific) at 160°C for 2 h. The heptahydrated magnesium sulfate was initially mixed with 2-mm diameter silanized glass beads (Fisher Scientific) to prevent the reagent from forming a hard mass during dehydration. After oven drying, the drying agent was ground in a pestle and mortar and sieved to obtain the 30–80-mesh fraction used in this study.

Standards (2 mg/ml each) of eleven compounds from the Environmental Protection Agency (EPA) semi-volatile target compound list (chosen for their range of polarities) were used to determine the potential retention of analytes on the drying agents (see Table IV). The eleven compounds were prepared in methylene chloride and stored at −10°C until used. The spiking level for the semi-volatile pollutants was 20 μg of each compound.

Three real samples with different water contents and a history of extraction cell outlet frit and restrictor plugging problems were chosen to investigate the suitability of the drying agents for SFE. Petroleum waste sludge was obtained from a commercial refinery and contained ca. 6% (w/w) water (based on oven drying at 105°C for 2 h) and a high concentration (ca. 20% w/w) of extractable hydrocarbons including a complex mixture of n-alkanes (C_{13}–C_{40}) and polycyclic aromatic hydrocarbons (PAHs). All extractions were performed on the sample as received. The lake sediment soil was collected in the Red River

Valley, North Dakota (USA) and contained *ca.* 20% (w/w) water (as determined by oven drying at 105°C for 2 h) and <0.05% (w/w) extractable hydrocarbons. Before extraction, the sample was sieved through a 2-mm screen to remove sticks and other debris. The biosludge was obtained from a commercial refinery. Prior to extraction, the sample was centrifuged at 3000 rpm (*ca.* 1250 g) for 30 min to remove the suspended particles from the aqueous media. The aqueous supernatant was then discarded and the wet precipitate collected and stored at −10°C until used. The precipitate contained *ca.* 90% (w/w) water (as determined by oven drying at 105°C for 2 h) and *ca.* 0.5% (w/w) extractable hydrocarbons, the majority of which were C_{13}–C_{30} *n*-alkanes.

A fourth real sample was used to investigate the volatile analyte losses which might occur when the sample is air or oven dried or mixed with an exothermic drying agent. A wet petroleum waste sludge obtained from a commercial refinery was chosen as it contained a sufficient amount of water [*ca.* 30% (w/w) as determined by oven drying] to generate an appreciable amount of heat when mixed with magnesium sulfate. The sample also contained both volatile and semi-volatile analytes [*ca.* 4% (w/w) extractable hydrocarbons] including a complex mixture of *n*-alkanes (C_8–C_{32}), alkylbenzenes and phenols. The sample was used as received.

Supercritical fluid extraction

Two ISCO Model 260D syringe pumps were used for the SFE extractions, one containing pure SFC-grade CO_2 and the other premixed CO_2–10% (v/v) methanol or CO_2–10% (v/v) toluene (Scott Gases, Plumsteadville, PA, USA). Each pump was filled and operated independently to avoid any possible carryover of modifier. The SFE pumps were connected to 0.5- or 2.5-ml extraction cells with 1/16 in. (1.6 mm) O.D. stainless steel tubing and "Slip-free" finger-tight connectors (Keystone Scientific, Bellefonte, PA, USA). A 1-m coil of 1/16 in. (1.6 mm) O.D. tubing (placed before the extraction cell to prewarm the fluid to the extraction temperature) and the extraction cell were placed in a thermostated tube heater to maintain the extraction temperature.

The flow-rate of the supercritical fluid through the extraction cell was controlled by a 10 cm × 32 μm I.D. × 145 μm O.D. restrictor cut from fused-silica tubing (Polymicro Technologies, Phoenix, AZ, USA). However, when using CO_2–methanol as the extraction fluid it was found that the restrictor became brittle and broke within a few minutes of commencing the extraction. To eliminate the problem of restrictor breakages, the restrictor was secured inside a stainless-steel tube [10 cm long × 0.02 in. (0.5 mm) I.D. × 1/16 in. (1.6 mm) O.D.] as described elsewhere [15].

Extracted analytes were collected by inserting the outlet of the restrictor into a 7.4-ml vial (48 mm height × 14 mm I.D. neck) containing 5 ml of Fisher Optima-grade acetone. The collection vial was either free standing in air or placed in a container of water (60 ml of water in a 100-ml beaker) initially at room temperature. The collection solvent volume was maintained by small additions of solvent during SFE.

Initial experiments were performed to determine the amount of water required to plug a 10 cm × 32 μm I.D. capillary restrictor. Increasing amounts of water (HPLC grade, Fisher Scientific) were injected on to the top of 600 mg of silanized 70–80-mesh glass beads placed inside a 0.5-ml extraction cell. The cell was then immediately sealed and extracted with CO_2 at 60°C and 400 atm. A 150-μl volume of water was found to block the restrictor even when the collection vial was placed in a beaker of water to reduce the cooling of the collection solvent. As previous work had shown that maintaining the collection solvent temperature above 0°C improved the extraction flow for wet samples [16], the initial evaluation of the drying agents was carried out with the collection vial in a beaker of water. However, subsequent experiments demonstrated that a beaker of water was no longer required when the "successful" drying agents which passed the initial evaluation were used with a high temperature (*e.g.*, 150°C) or organic-modified supercritical fluid. Further, a continuous extraction flow could be obtained with pure low-temperature CO_2 (60°C) and a cooled collection solvent by increasing the amount of the "successful" drying agent used.

A 150-μl volume of water was used throughout the study to evaluate the ability of the drying agents to retain water. For consistency, all the drying agents were packed into the extraction cell and water was added to the reagents in the following manner. Silanized 70–80-mesh-glass beads (ca. 150 mg) were initially placed at the bottom of a 0.5-ml extraction cell to minimize plugging of the extraction cell outlet frit with fines from the drying agents. The extraction cell was then filled with drying agent (ca. 150–250 mg depending on the density and physical packing characteristic of the drying agent). In some instances (i.e., magnesium sulfate) glass beads were also mixed with the drying agent to avoid forming a hard plug in the extraction cell during SFE. The 150-μl volume of water was injected on to the top of the reagents. The cell was then immediately sealed and placed inside the tube heater, equilibrated for 10 min and extracted for 10 min at either 60 or 150°C with pure CO_2 at 400 atm, or modified CO_2 [i.e., CO_2–10% (v/v) methanol at 400 atm and 60°C or CO_2–10% (v/v) toluene at 400 atm and 80°C]. At the end of the extraction, the water content in the collection solvent was determined by Karl-Fischer titration [17].

To determine if analytes were retained on the drying agents, 10 μl of the semi-volatile pollutant mixture were spiked on to the top of ca. 200 mg of wet (150 μl of water added) or dry (no water added) reagents. The 0.5-ml extraction cell was then immediately sealed to prevent any loss of the volatile spike components, placed inside the tube heater, equilibrated for 10 min and then extracted for 10 min with CO_2 at 400 atm and 60°C. The extracted analytes recovered in the collection solvent were determined by GC with flame ionization detection (FID).

Three samples (petroleum waste sludge, bio-sludge and soil) known to cause frit and restrictor plugging problems were used to test the ability of the drying agent to retain water from real samples. Silanized 70–80-mesh glass beads (ca. 100 mg) were placed at the bottom of a 0.5- or 2.5-ml extraction cell to prevent plugging of the extraction cell outlet frit from sample matrix material or from fines extracted of the drying agent. The drying agent [200 mg (0.5-ml cell) or 800 mg (2.5-ml cell)] was then placed inside the extraction cell as either a "bed" of drying agent with the environmental sample (200 mg) placed on top, or the drying agent was mixed with the environmental sample [1:1 or 4:1 (w/w) drying agent-to-sample ratio] and the mixture placed inside the extraction cell (0.5 or 2.5 ml, respectively). The extraction cell was then immediately sealed, placed inside the tube heater, equilibrated for 10 min and extracted for 30 min with CO_2 at 400 atm and 60°C.

To determine the potential loss of volatile analytes when drying an environmental sample prior to extraction, a wet [30% (w/w) water] petroleum waste sludge containing both volatile and semi-volatile analytes was air dried at room temperature for 18 h or oven dried at 105°C for 1 h. The possible losses of volatile components on mixing with an exothermic drying agent (anhydrous magnesium sulfate) were also determined by comparing the components recovered when the wet petroleum waste sludge was mixed with the magnesium sulfate with those recovered when the drying agent was used as a "bed" under the wet sludge sample. The air-dried, oven-dried, and "bed" or "mixture" of petroleum waste sludge was extracted with 400 atm CO_2 at 60°C for 30 min. The extracted components were determined by GC–FID.

Gas chromatographic analysis

All GC analyses were carried out with a Hewlett-Packard Model 5890 gas chromatograph with both FID and electron-capture detection (ECD). Hydrogen was the carrier gas and nitrogen was used as the detector make-up gas for ECD. The semi-volatile pollutant extracts and the petroleum waste sludge extracts were determined by GC–FID and the drying agent extracts by GC–FID and GC–ECD. The injections were performed in the split mode with a 10:1 splitting ratio (drying agent and semi-volatile pollutant extracts) into a wide-bore (25 m × 0.32 mm I.D., 0.17 μm film thickness) HP-5 fused-silica capillary column or a 40:1 splitting ratio (real samples) into a narrow-bore (50 m × 0.2 mm I.D., 0.33 μm film thickness) HP-5 fused-silica capillary column. The injector and detector temperatures were maintained at 300°C.

GC separations of the drying agent extracts were performed with the oven temperature initially at 40°C, then programmed at 12°C/min to 320°C. For the semi-volatile pollutant extracts, the oven temperature at injection was 50°C for 1 min, then programmed at 12°C/min to 280°C. The wet petroleum waste sludge extracts were analyzed with the oven temperature set at 40°C for 2 min, then programmed at 6°C/min to 300°C. The internal standards used for GC–FID analysis of the drying agent extracts, semi-volatile pollutant extracts and the wet petroleum waste sludge extracts were phenanthrene (8 μg), fluoranthene (30 μg) and 2-phenylnaphthalene (460 μg), respectively. The internal standard used for the GC–ECD analysis of the drying agent extracts was 1,2,4-trichlorobenzene (1 μg).

RESULTS AND DISCUSSION

As the presence of water in environmental samples can cause the restrictor to plug during SFE, the simplest solution would be to air or oven dry the sample prior to extraction. However, such drying techniques are not suitable as they can result in the loss of volatile and semi-volatile analytes. For example, as shown in Fig. 1, air drying a wet petroleum waste sludge completely removed the volatile C_8–C_{11} n-alkanes from the sample and also reduced the amount of semi-volatile analytes present [e.g., 60% of the phenol and 40% of the m- and p-cresol lost, Table I]. Oven drying the sample resulted in even greater analyte losses with only the high-molecular-mass C_{25}–C_{30} n-alkane recoveries remaining unaffected by the drying technique (Table I, Fig. 1).

Water retention of drying agents during extraction with CO_2 at 60 and 150°C

Table II summarizes the ability of several drying agents to eliminate restrictor plugging after adding 150 μl of water to 200 mg of reagent and extracting for 10 min with CO_2 at 400 atm and 60 or 150°C. [Note that in this initial survey (Table II) a beaker of water was required to help maintain a continuous extraction flow although, as discussed below, it was not required at high
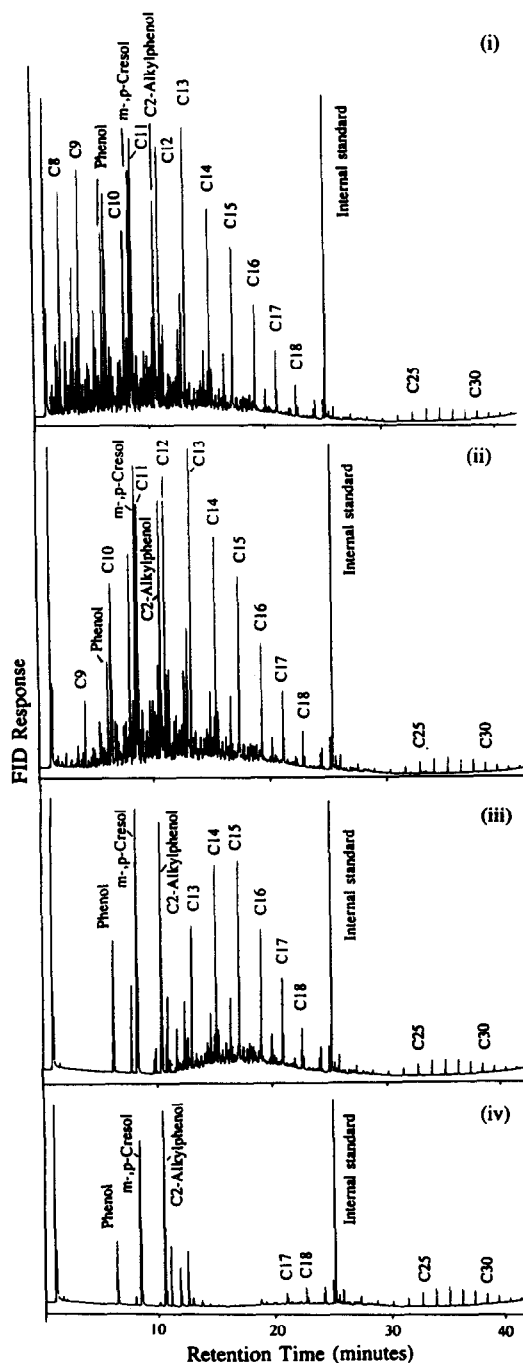

Fig. 1. GC–FID of the SFE extracts from a wet petroleum sludge: (i) wet sample placed on bed of anhydrous magnesium sulfate; (ii) wet sample mixed with anhydrous magnesium sulfate; (iii) sample air dried at room temperature for 18 h; (iv) sample oven dried at 105°C for 1 h. Extractions were performed with CO_2 at 400 atm and 60°C for 30 min.

TABLE I

LOSSES OF VOLATILE AND SEMI-VOLATILE ORGANICS FROM A WET PETROLEUM WASTE SLUDGE ON MIXING WITH $MgSO_4$ AND FROM AIR OR OVEN DRYING

Native analyte	Analyte concentration (μg/g)[a] (bed of drying agent + sample)	Recovery (%)[b]		
		Mixture of drying agent + sample (R.S.D., %)	Sample air dried for 18 h (R.S.D., %)[c]	Sample oven dried for 1 h at 105°C (R.S.D., %)[c]
n-Octane (C_8)	1499 ± 275	3 (49)	ND[d]	ND
m-, p-Xylene	510 ± 102	22 (51)	ND	ND
n-Nonane (C_9)	1967 ± 184	22 (12)	ND	ND
Phenol	3066 ± 573	81 (6)	44 (1)	18 (12)
n-Decane (C_{10})	1906 ± 231	49 (11)	ND	ND
m-, p-Cresol	5242 ± 743	81 (4)	63 (1)	39 (11)
n-Undecane (C_{11})	2125 ± 287	68 (9)	ND	ND
C_2-alkylphenol	1321 ± 183	89 (2)	109 (9)	65 (11)
n-Dodecane (C_{12})	1880 ± 257	75 (11)	17 (3)	ND
n-Tridecane (C_{13})	1598 ± 181	81 (8)	34 (8)	ND
n-Tetradecane (C_{14})	1618 ± 132	70 (7)	51 (9)	ND
n-Pentadecane (C_{15})	908 ± 117	86 (7)	82 (10)	ND
n-Hexadecane (C_{16})	536 ± 35	83 (7)	92 (10)	ND
n-Heptadecane (C_{17})	304 ± 14	81 (4)	88 (9)	12 (3)
n-Octadecane (C_{18})	138 ± 6	85 (4)	90 (9)	37 (2)
n-Nonadecane (C_{19})	115 ± 3	95 (3)	93 (8)	72 (2)
n-Pentacosane (C_{25})	31 ± 2	90 (2)	93 (2)	106 (6)
n-Hexacosane (C_{26})	41 ± 2	90 (5)	95 (2)	105 (5)
n-Heptacosane (C_{27})	44 ± 4	86 (5)	98 (7)	104 (6)
n-Octacosane (C_{28})	37 ± 5	92 (6)	111 (5)	110 (7)
n-Nonacosane (C_{29})	36 ± 4	89 (6)	103 (8)	105 (5)
n-Triacontane (C_{30})	26 ± 3	92 (8)	108 (6)	108 (4)

[a] Analyte concentration was determined from triplicate extractions with CO_2 at 400 atm and 60°C for 30 min (average value ± standard deviation).

[b] Percentage recovery relative to values obtained when bed of drying agent was used. Values in parentheses are the relative standard deviations of triplicate 30-min extractions with CO_2 at 400 atm and 60°C.

[c] Percentage recovery was normalized to wet mass.

[d] ND = Not detected. FID detection limit ≈4 μg/g as concentration in the original sample.

extraction temperatures, with modified fluids or with larger amounts of the "successful" drying agents.] Half of the drying agents investigated retained >80% of the spiked water at 400 atm CO_2 and 60°C, and a continuous extraction flow was attainable for the entire 10-min extraction. The "successful" drying agents stopped the initial removal of a large percentage of the water from the extraction cell, so that water no longer blocked the restrictor within seconds of commencing the extraction (as occurred without the drying agents). The rest of the drying agents investigated were either unable to stop water plugging the restrictor or were extracted into the

collection solvent, resulting in a plugged extraction cell outlet frit and/or restrictor (Table II).

None of the reagents investigated efficiently retained all the water in the extraction cell (Table II). The poor water retention of the "unsuccessful" drying agents may be related to the temperature (60°C) of the supercritical fluid extraction. Heating the drying agent to 60°C may partially dehydrate the reagent (e.g., $CaCl_2 \cdot 6H_2O$ was dehydrated to $CaCl_2 \cdot 2H_2O$ [18]) and decrease the water capacity of the drying agent. Even the "successful" drying agents which prevented restrictor plugging only retained ca. 80% (w/w) of the water as ca. 20% (w/w) of the

added water was recovered in the collection solvent. A similar amount of water was generally recovered from all the "successful" drying agents regardless of whether the water was retained by adsorption (molecular sieves, alumina and Florisil), absorption (water-soluble polymer), physical entrapment (molecular sieves) or the formation of a hydrate (metal sulfates).

The drying agents which were "successful" at maintaining an extraction flow at 60°C were also "successful" at eliminating restrictor plugging at 150°C (Table II). However, at the higher extraction temperature of 150°C all the "successful" drying agents were dehydrated [ca. 75% (w/w) water removed] owing to the temperature limitation of the reagents (e.g., $MgSO_4 \cdot 7H_2O$ is dehydrated to $MgSO_4 \cdot H_2O$ [18]) and the order of magnitude increase in the solubility of water in CO_2 (i.e., 0.008 mole fraction of water in CO_2 at 400 atm and 50°C compared with 0.06 mole fraction at 150°C [19]), which increased the extraction efficiency of water from the drying agents (Table II). The Joule–Thomson cooling effect at the restrictor tip was also decreased at the high extraction temperature (Fig. 2), so that
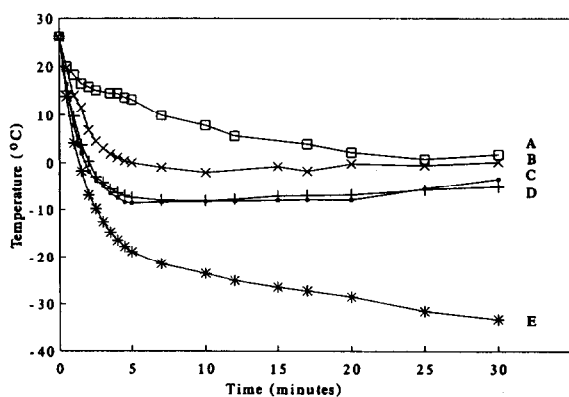

Fig. 2. Collection solvent temperature using various extraction and collection conditions. Temperature profiles are shown for the collection solvent (5 ml of acetone) in a 7.4-ml vial with a 10 cm × 32 μm I.D. restrictor using: (A) CO_2 at 400 atm and 60°C with the collection vial in a beaker of water; (B) CO_2 at 400 atm and 150°C with the collection vial free standing in air; (C) CO_2–10% (v/v) methanol at 400 atm and 60°C with the collection vial free standing in air; (D) CO_2–10% (v/v) toluene at 400 atm and 80°C with the collection vial free standing in air; and (E) CO_2 at 400 atm and 60°C with the collection vial free standing in air.

water no longer froze in the restrictor and the collection vial did not require a beaker of water to obtain a continuous extraction flow. However, the drying agents were still needed because, in their absence, liquid water quickly plugged the restrictor on commencing the extraction (Table II). Even though several of the drying agents successfully avoided restrictor plugging, only Florisil and molecular sieve 3A were able to retain a moderate amount [ca. 40% (w/w)] of the water at the 150°C extraction temperature.

Examination of the extraction cell and collection solvent after extracting the wet drying agents at 400 atm CO_2 and 60°C revealed that several of the reagents had precipitated in the extraction cell outlet frit, restrictor and/or collection solvent (Table II). The plugging problem associated with the extraction of the drying agents was partially related to the 60°C extraction temperature, which melted one of the hydrated reagents ($Na_2SO_4 \cdot 10H_2O$, which has a melting point of 32°C [18]), blocking the outlet frit of the extraction cell and the restrictor. Fines or small fragments removed from the drying agents during SFE also caused frit and restrictor plugging problems. These fines are probably formed as a dust during the manufacture of the reagents or from agitation of the drying agent container. However, the fines were successfully removed and the plugging eliminated from several of the reagents (e.g., magnesium sulfate and Hydromatrix) by sieving the reagents prior to extraction. Unfortunately, five of the drying agents (calcium sulfate, copper sulfate, calcium oxide, potassium carbonate and calcium chloride) still plugged the frit and restrictor even after they were sieved, and small particles of drying agents were observed in the collection solvent (Table II). Impurities from some of the drying agents (e.g., carboxymethylcellulose and guar gum) were also found in the supercritical fluid extracts by GC–FID (Table II), which made these reagents unsuitable for SFE.

Examination of the drying agent after SFE indicated that a number of the reagents had been compressed into a hard plug which may block the extraction cell outlet frit (Table II). For example, the water-soluble polymers (carboxymethylcellulose, guar gum and polyacrylamide

TABLE II

FLOW CHARACTERISTICS OF WET DRYING AGENTS USING CO_2 AT 400 ATM AND 60 OR 150°C

Reagent	Qualitative flow-rate[a] (CO_2, 400 atm, 60°C)	Water in collection solvent (%)[b] (CO_2, 400 atm, 60°C)	Qualitative flow-rate[a] (CO_2, 400 atm, 150°C)	Water in collection solvent (%)[b] (CO_2, 400 atm, 150°C)	Extractable components from reagents[c] (CO_2, 400 atm, 60°C)	Physical description of reagents after SFE (CO_2, 400 atm, 60°C)	Temperature of reagent on hydration[d] (°C)
Glass beads	Blocked	–	Blocked	–	–	Wet particles	21
Carboxymethylcellulose	Continuous	13	Continuous	–	Yes	Hard plug[f]	22
Xanthan gum	Continuous	13	Continuous	79	No	Rubbery strand	23
Guar gum	Intermittent	–	–	–	Yes	Rubbery strand	20
Polyacrylamide	Blocked	–	–	–	–	Hard plug[f]	23
Hydromatrix	Continuous	19	Continuous	88	No	Wet particles	21
Molecular sieve 3A	Continuous	18	Continuous	56	No	Wet particles	40
Molecular sieve 4A	Continuous	15	Continuous	74	No	Wet particles	60
Molecular sieve 5A	Continuous	14	Continuous	79	No	Wet particles	35
Molecular sieve 13X	Continuous	18	Continuous	74	No	Wet particles	85
Alumina	Continuous	17	Continuous	83	No	Wet particles	21
Silica	Intermittent	–	–	–	No	Wet particles	23
Florisil	Continuous	14	Continuous	60	No	Wet particles	21
Sodium sulfate	Blocked	–	–	–	–	White paste	25
Magnesium sulfate ($MgSO_4$)	Continuous	12	Continuous	73	No	Hard plug[f]	94
Magnesium sulfate ($MgSO_4 \cdot H_2O$)	Continuous	43	Continuous	71	No	Semi-hard plug	61
Calcium sulfate	Blocked	–	–	–	–[e]	Wet particles	26
Copper sulfate	Blocked	–	–	–	–[e]	Hard plug[f]	69
Calcium oxide	Blocked	–	–	–	–[e]	Hard plug[f]	93
Boron trioxide	Intermittent	–	–	–	–[e]	Semi-hard plug	103
Potassium carbonate	Blocked	–	–	–	–[e]	White paste	68
Calcium chloride	Blocked	–	–	–	–[e]	White paste	91

[a] The flow was assessed as continuous (constant flow-rate ± 0.2 ml/min), intermittent (flow initially constant but within a few minutes of extraction plugging starts to occur) or blocked (no flow). Collection vial was placed in a beaker of water (initially at room temperature).

[b] Percentage of spiked water recovered in collection solvent as determined by Karl Fischer titration. Water content was only determined for agents showing a continuous flow.

[c] Extractable components from drying agent were detected in collection solvent by GC–FID.

[d] The drying agent (1 g) was placed inside a polystyrene cup and hydrated with water (3 ml). The change in temperature with the addition of water was measured with a J-thermocouple.

[e] Particles of drying agent were seen in collection solvent.

[f] Drying agent formed hard plug during extraction which proved difficult to remove from the extraction cell.

gel) form sticky hydrated gels at atmospheric pressure, but at the 400 atm extraction pressure the hydrated gels become hard or rubbery strands. On inspection of a polymer plug or strand removed from the extraction cell after depressurization, it was observed that the exterior portion of the plug had formed into a hard, clear solid, whereas the interior of the plug was dry or sticky in texture. These physical differences suggest that water preferentially flowed along the interior extraction cell wall so that "plug" flow within the cell was not achieved. Mixing a dispersant (100 μm glass beads) with the polymers did not improve the flow characteristics. However, magnesium sulfate also forms a hard plug, but mixing glass beads with the reagent made it easier to unpack the extraction cell after SFE. (Note that without glass beads a hammer and chisel were required to remove the drying agent.) Monohydrated magnesium sulfate formed a semi-hard plug which was easier to remove from the extraction cell, but the reagent retained *ca.* 30% less water than anhydrous magnesium sulfate (Table II).

Several of the drying agents also produced a large amount of heat on hydration (Table II).

For example, adding water to boron trioxide increased the temperature of the mixture sufficiently to boil the water. The significant amount of heat generated from several of the drying agents may make them undesirable for use with samples containing volatile analytes (as discussed below).

Water retention of drying agents with extraction by modified CO_2

Of the 21 drying agents initially investigated, eleven "successfully" prevented restrictor plugging by water during supercritical fluid extraction with pure CO_2. The "successful" drying agents were further evaluated with a water-insoluble (toluene) and water-soluble (methanol) modified CO_2 supercritical fluid (Table III), as organic modifiers are frequently used to increase the supercritical fluid extraction efficiency of many organic pollutants from environmental matrices [20]. Hence the ideal drying agent would be expected to retain water efficiently when used with these modified fluids.

Most of the drying agents proved suitable for use with the CO_2–toluene extraction fluid, the reagents retaining >75% of the water in the

TABLE III

FLOW CHARACTERISTICS OF WET DRYING AGENTS USING CO_2–10% (v/v) TOLUENE AT 400 ATM AND 80°C AND CO_2–10% (v/v) METHANOL AT 400 ATM AND 60°C

Drying agent	Qualitative flow-rate[a]		Water in collection solvent (%)[b]		Particles of reagent in collection solvent	
	CO_2–10% (v/v) toluene	CO_2–10% (v/v) methanol	CO_2–10% (v/v) toluene	CO_2–10% (v/v) methanol	CO_2–10% (v/v) toluene	CO_2–10% (v/v) methanol
Glass beads	Blocked	Continuous	–	73	No	No
Xanthan gum	Continuous	Continuous	12	59	No	No
Hydromatrix	Continuous	Blocked	11	–	No	Yes
Molecular sieve 3A	Continuous	Continuous	20	92	No	Yes
Molecular sieve 4A	Continuous	Continuous	10	88	No	Yes
Molecular sieve 5A	Continuous	Blocked	23	–	No	Yes
Molecular sieve 13X	Continuous	Continuous	15	87	No	Yes
Alumina	Continuous	Continuous	17	76	No	Yes
Florisil	Continuous	Blocked	11	–	No	Yes
Magnesium sulfate (MgSO$_4$)	Intermittent	Blocked	22	–	Yes	Yes
Magnesium sulfate (MgSO$_4$ · H$_2$O)	Intermittent	Blocked	24	–	Yes	Yes

[a] The flow-rates are assessed as continuous (constant flow-rate ± 0.2 ml/min), intermittent (flow initially constant but within a few minutes of extraction plugging starts to occur) or blocked (no flow). Collection vial was placed in a beaker of water (initially at room temperature).

[b] Percentage of spiked water recovered in collection solvent as determined by Karl Fischer titration. Water content was only determined for agents showing a continuous flow.

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TABLE IV

RECOVERY OF SEMI-VOLATILE POLLUTANTS FROM DRYING AGENTS USING CO_2 AT 400 ATM AND 60°C

Form of drying agent	Compound	Recovery (%)[a]									
		Xanthan[b]	Hydromatrix	M.S. 3A	M.S. 4A	M.S. 5A	M.S. 13X	Alumina	Florisil	MgSO₄	MgSO₄·H₂O
Wet	2-Chlorophenol	66 (51)	92 (7)	96 (6)	97 (3)	95 (13)	99 (3)	99 (3)	95 (5)	93 (3)	105 (3)
	o-Cresol	65 (52)	93 (7)	93 (6)	95 (2)	99 (5)	96 (5)	97 (5)	95 (5)	93 (4)	102 (1)
	Nitrobenzene	72 (56)	94 (7)	94 (8)	92 (8)	98 (4)	98 (2)	97 (3)	95 (6)	94 (4)	100 (1)
	2-Chloroethoxymethane	68 (53)	95 (6)	95 (2)	98 (1)	99 (5)	98 (2)	99 (3)	96 (6)	95 (4)	100 (1)
	1,2,4-Trichlorobenzene	66 (54)	95 (8)	93 (7)	99 (1)	99 (4)	100 (1)	99 (13)	98 (5)	95 (6)	100 (2)
	4-Chloroaniline	64 (55)	98 (6)	94 (8)	99 (6)	101 (6)	100 (4)	99 (3)	96 (6)	97 (3)	96 (2)
	Tetradecane	66 (55)	95 (6)	92 (8)	98 (2)	98 (4)	98 (2)	99 (4)	95 (7)	94 (4)	96 (1)
	Dibenzofuran	65 (55)	98 (4)	96 (6)	96 (3)	96 (8)	98 (5)	98 (4)	94 (7)	93 (8)	97 (3)
	Diethyl phthalate	67 (55)	95 (4)	102 (3)	96 (1)	96 (4)	98 (2)	98 (3)	94 (6)	95 (7)	99 (1)
	Phenanthrene	66 (56)	100 (2)	103 (3)	96 (1)	98 (4)	98 (3)	98 (3)	99 (3)	96 (6)	100 (1)
	Chrysene	65 (54)	104 (5)	101 (2)	102 (2)	104 (5)	100 (4)	100 (4)	99 (3)	96 (11)	94 (6)
Dry	2-Chlorophenol		101 (8)	97 (2)	19 (57)	93 (5)	0 (0)	0 (0)	97 (7)	95 (5)	98 (5)
	o-Cresol		97 (9)	89 (3)	60 (20)	89 (4)	0 (0)	0 (0)	88 (7)	103 (1)	98 (5)
	Nitrobenzene		99 (9)	96 (3)	98 (2)	91 (4)	0 (0)	87 (3)	96 (9)	98 (9)	100 (8)
	2-Chloroethoxymethane		98 (7)	90 (2)	96 (2)	90 (5)	0 (0)	90 (1)	92 (7)	98 (2)	94 (6)
	1,2,4-Trichlorobenzene		98 (8)	94 (3)	98 (3)	91 (5)	87 (21)	93 (3)	93 (7)	102 (2)	97 (7)
	4-Chloroaniline		94 (11)	92 (2)	0 (0)	80 (5)	0 (0)	0 (0)	11 (15)	0 (0)	0 (0)
	Tetradecane		97 (8)	94 (3)	98 (1)	90 (3)	88 (21)	94 (3)	92 (7)	101 (1)	97 (6)
	Dibenzofuran		97 (8)	94 (2)	99 (2)	90 (1)	37 (48)	89 (2)	93 (7)	102 (1)	97 (6)
	Diethyl phthalate		96 (10)	12 (48)	0 (0)	89 (4)	0 (0)	2 (91)	0 (0)	99 (2)	94 (8)
	Phenanthrene		98 (9)	96 (3)	100 (0)	90 (3)	53 (37)	92 (2)	92 (7)	102 (1)	97 (6)
	Chrysene		100 (9)	96 (3)	101 (3)	90 (3)	50 (33)	71 (13)	94 (8)	102 (0)	94 (7)

[a] Values in parentheses are relative standard deviations (%) of triplicate 10-min extractions. Collection vial was in a beaker of water (initially at room temperature).
[b] Values in parentheses are relative standard deviations (%) of six 10-min extractions from xanthan gum only.

extraction cell to achieve a continuous extraction flow (Table III). However, the drying agents were unsuitable for use with the CO_2-methanol extraction fluid, as the reagents were generally co-extracted with the majority of the water, and the reagents precipitated in the extraction cell outlet frit, restrictor and collection solvent (Table III). The most severe plugging problems were with anhydrous and monohydrated magnesium sulfate, which were extracted by both the CO_2-toluene and CO_2-methanol, and irreversibly blocked the frit and restrictor (Table III). It is interesting that only xanthan gum was not extracted by either modified extraction fluid, possibly because the reagent formed a gel on hydration.

The addition of an organic modifier to CO_2 reduced the rate at which the collection solvent was cooled (Fig. 2), and a continuous extraction flow was achieved with both modified fluids without placing the collection vial in a beaker of water. The polar CO_2-methanol extracted the majority (ca. 80%, w/w) of the water from the drying agents, whereas the aromatic CO_2-toluene (Table III), with a low affinity for water, extracted about the same amount of water from the reagents as pure CO_2 at 60°C (Table II). Further, in contrast to the CO_2-toluene, the CO_2-methanol extraction fluid did not require a drying agent to maintain a continuous extraction flow (Table III). To confirm this observation, the 0.5-ml extraction cell was filled with water, extracted with CO_2-methanol and the extract collected in an organic solvent which was allowed to cool to below 0°C during the extraction. A liquid was observed leaving the restrictor for the first 50 s of the extraction, followed by bubbles of CO_2 typically seen during normal SFE. After 5 min of extraction all the water had been removed from the extraction cell, and no restrictor plugging had occurred during the entire extraction procedure.

Retention of semi-volatile pollutants on drying agents

Ideally, the "successful" drying agents should be able to retain selectively the extracted water from real samples but not retain the extracted analytes of interest. Therefore, the affinity of the "successful" drying agents for semi-volatile pollutants of various volatility, polarity and functional groups was investigated (Table IV). As the drying agents were intended to be used with wet samples, the semi-volatile pollutant mixture was spiked directly on to wet (150 μl of added water) drying agents. Supercritical CO_2 at 400 atm and 60°C quantitatively recovered all the analytes from the majority of the wet drying agents (Table IV). The quantitative recoveries were attributed to the deactivation of the active sites of the drying agents with water. For example, the deactivation of alumina with 5% (w/w) water prior to extraction enabled organochlorine pesticides to be recovered quantitatively from the sorbent using supercritical CO_2 [21]. However, poor analyte recoveries with high relative standard deviations were obtained with xanthan gum. The low average recoveries and poor reproducibilities obtained for all the analytes regardless of polarity were due to the "non-plug" flow or channeling of the CO_2 through the polymer, resulting in only partial hydration of the drying agent (i.e., dry interior) which, as discussed below, had a significant effect on analyte retention.

The recovery of the analytes from "dry" drying agents was also evaluated (Table IV) as all the drying agents were dehydrated to some extent under SFE conditions, and a routine procedure for the extraction of environmental samples could include drying agents regardless of whether the sample is wet or dry. Without water present, a number of the drying agents proved highly retentive, completely retaining at least one of the test analytes (most often 4-chloroaniline and/or diethyl phthalate; Table IV). Poor analyte recoveries were seen for the porous molecular sieves (4A and 13X), particularly molecular sieve 13X, which has the largest pore diameter (10 Å) and adsorbed the majority of analytes regardless of polarity. As might be expected, the polar basic alumina adsorbent retained the weakly acidic phenols in addition to 4-chloroaniline and diethyl phthalate. The acidic Florisil adsorbent selectively retained diethyl phthalate and the basic 4-chloroaniline, whereas the weakly acidic magnesium sulfate retained only the 4-chloroaniline. Only Hydromatrix gave

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quantitative recoveries of all the analytes investigated. As most of the drying agents retained at least one of the analytes, a spike recovery of the target analytes should always be performed to determine the appropriate drying agent for a particular set of analytes.

Thus, of the eleven drying agents investigated only five (i.e., those displaying the least retention of the target analytes, namely Hydromatrix, molecular sieves 3A and 5A and anhydrous and monohydrated magnesium sulfate) proved suitable for use with environmental samples. As shown in Table II, these drying agents did not cause detectable contamination of the extracts when analyzed by GC–FID. To evaluate further the potential for contamination, extracts from each of the five drying agents were also analyzed by GC–ECD. None of the drying agents added detectable GC–ECD contaminants (compared with ca. 10 pg injected of 1,2,4-trichlorobenzene) to the SFE (CO_2 at 400 atm and 60°C) extracts. However, a few trace contaminants from the acetone solvent and/or CO_2 were found in the collection solvent blanks when no drying agent was present.

Evaluation of drying agents with real samples

Three real samples with different water contents and SFE-extractable components were chosen to test the ability of the drying agents to avoid restrictor plugging when using CO_2 at 400 atm and 60°C. The samples were (i) a lake sediment soil [ca. 20% (w/w) water] containing a very low concentration (<0.05%, w/w) of extractable hydrocarbons; (ii) a biosludge [90% (w/w) water] with ca. 0.5% (w/w) extractable hydrocarbons; and (iii) a petroleum waste sludge [ca. 6% (w/w) water] with a consistency and color similar to those of molasses and with a high concentration (ca. 20%, w/w) of extractable hydrocarbons. In the absence of drying agents, all three samples caused restrictor and/or frit plugging problems.

Only the drying agents that had a low affinity for the semi-volatile pollutants (i.e., retained none or only one of the analytes in Table IV) were investigated with the real samples (Table V). Two extraction cell-loading procedures were

used. The drying agents were placed inside the extraction cell either as a "bed" of reagent with the environmental sample placed on top, or the reagents were mixed with the environmental sample and were then placed inside the extraction cell. With both procedures, no attempt to regulate the collection solvent temperature was made. However, if restrictor plugging was still encountered when using a drying agent, the collection vial was placed in a beaker of water at room temperature to maintain the collection solvent above 0°C.

Plugging problems were encountered with all three environmental samples if just dispersants (e.g., 100 μm glass beads) were used as a bed or mixed with the samples. Conversely, when a drying agent (bed or mixture) was used with the samples a good extraction flow was usually attainable, demonstrating that the reagents were not just dispersants but also sorbents which retained the water. The success of the drying agents at eliminating restrictor plugging generally depended on the nature of the sample (Table V). For the lake sediment soil, a 1:1 drying agent-to-sample ratio was sufficient to eliminate the restrictor plugging problems. Continuous extraction flows could be obtained without keeping the collection solvent above 0°C as was required when the soil sample was mixed with the glass beads. The soil contained virtually no extractable hydrocarbons but a high concentration of water (20%, w/w), hence the restrictor plugging was associated with the water. Therefore, the drying agents retained a sufficient amount of the water or slowed the rate of water removal from the extraction cell to reduce or eliminate freezing water at the restrictor tip. (Note that occasional plugging of the restrictor still occurred but that within a few seconds the flow would restart unaided.) The various cell-loading procedures using a bed or mixture of drying agent and sample (with the exception of Hydromatrix) had no observable effect on the extraction flow.

With very wet samples such as the biosludge [ca. 90% (w/w) water, which was equivalent to 180 μl of water], the 1:1 ratio of drying agent to sample was unable to eliminate restrictor plug-

TABLE V

FLOW CHARACTERISTICS OF REAL SAMPLES WITH DRYING AGENTS USING CO_2 AT 400 ATM AND 60°C

Drying agent	Extraction flow[a]				
	Soil (1:1)[b]	Biosludge (1:1)[b]	Biosludge (4:1)[b]	Petroleum sludge (1:1)[b]	Petroleum sludge (4:1)[b]
$MgSO_4$ (bed)	Continuous	Intermittent	Continuous	Blocked	Intermittent
$MgSO_4$ (mixed)	Continuous	Intermittent	Intermittent	Intermittent	Intermittent
$MgSO_4$ (bed, collection vial in water)	Continuous	Continuous	Continuous	Continuous	Continuous
$MgSO_4 \cdot H_2O$ (bed)	Continuous	Intermittent	Intermittent	Blocked	Intermittent
$MgSO_4 \cdot H_2O$ (mixed)	Continuous	Intermittent	Intermittent	Intermittent	Intermittent
$MgSO_4 \cdot H_2O$ (bed, collection vial in water)	Continuous	Continuous	Continuous	Continuous	Continuous
Molecular sieve 3A (bed)	Continuous	Intermittent	Continuous	Intermittent	Intermittent
Molecular sieve 3A (mixed)	Continuous	Intermittent	Intermittent	Intermittent	Intermittent
Molecular sieve 3A (bed, collection vial in water)	Continuous	Continuous	Continuous	Continuous	Continuous
Molecular sieve 5A (bed)	Continuous	Intermittent	Continuous	Intermittent	Intermittent
Molecular sieve 5A (mixed)	Continuous	Intermittent	Intermittent	Intermittent	Intermittent
Molecular sieve 5A (bed, collection vial in water)	Continuous	Continuous	Continuous	Continuous	Continuous
Hydromatrix (bed)	Intermittent	Intermittent	Intermittent	Intermittent	Intermittent
Hydromatrix (mixed)	Continuous	Intermittent	Intermittent	Intermittent	Intermittent
Hydromatrix (bed, collection vial in water)	Continuous	Continuous	Continuous	Continuous	Continuous

[a] The extraction flow was assessed as continuous (flow obtained without intervention, occasionally the flow would stop but would restart unaided), intermittent (flow required periodic gentle warming of the tip of the restrictor by placing the restrictor and ones thumb against the wall of the collection vial) or blocked (no flow).
[b] Ratio of drying agent to sample (w/w).

ging (Table V). Again, the plugging problem was associated with the high concentration of water, and a continuous extraction flow was only obtained when the collection solvent temperature was maintained above 0°C. By increasing the amount of anhydrous magnesium sulfate or molecular sieve 3A and 5A to a 4:1 drying agent-to-sample ratio, a sufficient amount of water was retained in the extraction cell to obtain a continuous extraction flow with a cooled collection solvent (i.e., the beaker of water was no longer required, Table V). The cell loading procedure did have an effect on the extraction flow of the biosludge, because mixing the drying agent with the sample matrix was not as effective as using a bed of drying agent to stop water from plugging the restrictor. However, King et al. [22] have shown that mixing the drying agent with the

sample can be advantageous for samples such as a spent bleaching clay which can become severely compacted without a reagent, so that the rate of extraction is reduced. Therefore, placing the mixture of drying agent and sample on a bed of drying agent may be more appropriate when extracting highly compressible samples.

Severe frit and restrictor plugging problems were encountered with the petroleum waste sludge (Table V). Employing a drying agent at a 1:1 or 4:1 reagent-to-sample ratio enabled a continuous extraction flow to be obtained with the sludge, although in both instances the collection vial had to be kept in a beaker of water to stop the restrictor from plugging. The need to regulate the collection solvent temperature was related to the nature of the sample matrix. The petroleum waste sludge contained a high concen-

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tration (20%, w/w) of extractable hydrocarbons and a relatively low concentration (6%, w/w) of water so that the majority of the restrictor plugging appeared to be associated with the extracted hydrocarbons and not the water.

The cell-loading procedure also had an effect on the recovery of volatile analytes from a sample when mixed with an exothermic drying agent. For example, mixing anhydrous magnesium sulfate with a wet [30% (w/w) water] petroleum waste sludge caused significant losses of C_8–C_{12} n-alkanes, although these losses were not as severe as those produced from air or oven drying the sample (Fig. 1, Table I). During the mixing, the temperature of anhydrous magnesium sulfate and the wet sludge noticeably increased and a strong fuel odor was produced. Conversely, if the wet sludge was placed on a "bed" of anhydrous magnesium sulfate so that the physical contact between the drying agent and the sample was greatly reduced, the recovery of the volatile n-alkanes was significantly increased (Table I).

CONCLUSIONS

Of the 21 drying agents tested, eleven (Hydromatrix, molecular sieves, xanthan gum, carboxymethylcellulose, magnesium sulfate, alumina and Florisil) successfully prevented water from plugging a capillary restrictor during extraction with CO_2 at 400 atm and 60°C. However, none of these "successful" drying agents irreversibly retained the water. Instead, the reagents decreased the rate at which the water was removed from the cell. Increasing the extraction temperature to 150°C or adding a polar modifier (methanol) to the CO_2 greatly decreased the amount of water retained by the drying agents, whereas the addition of an aromatic modifier (toluene) resulted in no such decrease. Even though none of the drying agents retained significant amounts of polar and non-polar test analytes in the presence of water, all the reagents except Hydromatrix caused specific losses of one or more polar analytes when no water was present in the extraction cell. Therefore, a spike recovery study of the target analytes should always be undertaken to determine the appropriate drying agent

for a particular set of analytes. Using the best five drying agents (i.e., those displaying the least retention of polar analytes, namely Hydromatrix, molecular sieves 3A and 5A and anhydrous and monohydrated magnesium sulfate) a 1:1 (w/w) drying agent-to-sample ratio was sufficient to prevent water from plugging the restrictor when used with moderately wet samples [20% (w/w) water], but a 4:1 reagent-to-sample ratio was required with very wet samples [90% (w/w) water]. For samples containing volatile analytes, mixing the sample with exothermic drying agents (e.g., magnesium sulfate) should be avoided to minimize losses of volatile analytes from the heat of hydration.

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