

METHOD 3540C

SOXHLET EXTRACTION

1.0 SCOPE AND APPLICATION

1.1 Method 3540 is a procedure for extracting nonvolatile and semivolatile organic compounds from solids such as soils, sludges, and wastes. The Soxhlet extraction process ensures intimate contact of the sample matrix with the extraction solvent.

1.2 This method is applicable to the isolation and concentration of water-insoluble and slightly water soluble organics in preparation for a variety of chromatographic procedures.

1.3 This method is restricted to use by or under the supervision of trained analysts. Each analyst must demonstrate the ability to generate acceptable results with this method.

2.0 SUMMARY OF METHOD

2.1 The solid sample is mixed with anhydrous sodium sulfate, placed in an extraction thimble or between two plugs of glass wool, and extracted using an appropriate solvent in a Soxhlet extractor.

2.2 The extract is then dried, concentrated (if necessary), and, as necessary, exchanged into a solvent compatible with the cleanup or determinative step being employed.

3.0 INTERFERENCES

Refer to Method 3500.

4.0 APPARATUS AND MATERIALS

4.1 Soxhlet extractor - 40 mm ID, with 500-mL round bottom flask.

4.2 Drying column - 20 mm ID Pyrex® chromatographic column with Pyrex® glass wool at bottom.

NOTE: Fritted glass discs are difficult to decontaminate after highly contaminated extracts have been passed through. Columns without frits may be purchased. Use a small pad of Pyrex® glass wool to retain the adsorbent. Prewash the glass wool pad with 50 mL of acetone followed by 50 mL of elution solvent prior to packing the column with adsorbent.

4.3 Kuderna-Danish (K-D) apparatus

4.3.1 Concentrator tube - 10-mL, graduated (Kontes K-570050-1025 or equivalent). A ground-glass stopper is used to prevent evaporation of extracts.

4.3.2 Evaporation flask - 500-mL (Kontes K-570001-500 or equivalent). Attach to concentrator tube with springs, clamps, or equivalent.

4.3.3 Snyder column - Three-ball macro (Kontes K-503000-0121 or equivalent).

4.3.4 Snyder column - Two-ball micro (Kontes K-569001-0219 or equivalent).

4.3.5 Springs - 1/2 inch (Kontes K-662750 or equivalent).

NOTE: The following glassware is recommended for the purpose of solvent recovery during the concentration procedures requiring the use of Kuderna-Danish evaporative concentrators. Incorporation of this apparatus may be required by State or local municipality regulations that govern air emissions of volatile organics. EPA recommends the incorporation of this type of reclamation system as a method to implement an emissions reduction program. Solvent recovery is a means to conform with waste minimization and pollution prevention initiatives.

4.4 Solvent vapor recovery system (Kontes K-545000-1006 or K-547300-0000, Ace Glass 6614-30, or equivalent).

4.5 Boiling chips - Solvent-extracted, approximately 10/40 mesh (silicon carbide or equivalent).

4.6 Water bath - Heated, with concentric ring cover, capable of temperature control ($\pm 5^{\circ}\text{C}$). The bath should be used in a hood.

4.7 Vials - Glass, 2-mL capacity, with polytetrafluoroethylene (PTFE)-lined screw or crimp top.

4.8 Glass or paper thimble or glass wool - Contaminant-free.

4.9 Heating mantle - Rheostat controlled.

4.10 Disposable glass pasteur pipet and bulb.

4.11 Apparatus for determining percent dry weight.

4.11.1 Drying oven - capable of maintaining 105°C .

4.11.2 Desiccator.

4.11.3 Crucibles - Porcelain or disposable aluminum.

4.12 Apparatus for grinding

4.13 Analytical balance - capable of weighing to 0.0001 g.

5.0 REAGENTS

5.1 Reagent grade inorganic chemicals shall be used in all tests. Unless otherwise indicated, it is intended that all reagents shall conform to the specifications of the Committee on Analytical Reagents of the American Chemical Society, where such specifications are available. Other grades may be used, provided it is first ascertained that the reagent is of sufficiently high purity to permit its use without lessening the accuracy of the determination.

5.2 Organic-free reagent water. All references to water in this method refer to organic-free reagent water, as defined in Chapter One.

5.3 Sodium sulfate (granular, anhydrous), Na_2SO_4 . Purify by heating at 400°C for 4 hours in a shallow tray, or by precleaning the sodium sulfate with methylene chloride. If the sodium sulfate is precleaned with methylene chloride, a method blank must be analyzed, demonstrating that there is no interference from the sodium sulfate.

5.4 Extraction solvents - All solvents must be pesticide quality or equivalent.

5.4.1 Soil/sediment and aqueous sludge samples shall be extracted using either of the following solvent systems:

5.4.1.1 Acetone/Hexane (1:1) (v/v), $\text{CH}_3\text{COCH}_3/\text{C}_6\text{H}_{14}$.

NOTE: This solvent system has lower disposal cost and lower toxicity.

5.4.1.2 Methylene chloride/Acetone (1:1 v/v), $\text{CH}_2\text{Cl}_2/\text{CH}_3\text{COCH}_3$.

5.4.2 Other samples shall be extracted using the following:

5.4.2.1 Methylene chloride, CH_2Cl_2 .

5.4.2.2 Toluene/Methanol (10:1) (v/v), $\text{C}_6\text{H}_5\text{CH}_3/\text{CH}_3\text{OH}$.

5.5 Exchange solvents - All solvents must be pesticide quality or equivalent.

5.5.1 Hexane, C_6H_{14} .

5.5.2 2-Propanol, $(\text{CH}_3)_2\text{CHOH}$.

5.5.3 Cyclohexane, C_6H_{12} .

5.5.4 Acetonitrile, CH_3CN .

6.0 SAMPLE COLLECTION, PRESERVATION, AND HANDLING

See the introductory material to this chapter, Organic Analytes, Section 4.1.

7.0 PROCEDURE

7.1 Sample Handling

7.1.1 Sediment/soil samples - Decant and discard any water layer on a sediment sample. Mix sample thoroughly, especially composited samples. Discard any foreign objects such as sticks, leaves, and rocks.

7.1.2 Waste samples - Samples consisting of multiple phases must be prepared by the phase separation method in Chapter Two before extraction. This extraction procedure is for solids only.

7.1.3 Dry waste samples amenable to grinding - Grind or otherwise subdivide the waste so that it either passes through a 1-mm sieve or can be extruded through a 1-mm hole. Introduce sufficient sample into the grinding apparatus to yield at least 10 g after grinding.

7.1.4 Gummy, fibrous, or oily materials not amenable to grinding should be cut, shredded, or otherwise reduced in size to allow mixing and maximum exposure of the sample surfaces for the extraction. The addition of anhydrous sodium sulfate to the sample (1:1) may make the mixture amenable to grinding.

7.2 Determination of percent dry weight - When sample results are to be calculated on a dry weight basis, a second portion of sample should be weighed at the same time as the portion used for analytical determination.

WARNING: The drying oven should be contained in a hood or be vented. Significant laboratory contamination may result from drying a heavily contaminated sample.

Immediately after weighing the sample for extraction, weigh 5 - 10 g of the sample into a tared crucible. Dry this aliquot overnight at 105°C. Allow to cool in a desiccator before weighing. Calculate the % dry weight as follows:

$$\% \text{ dry weight} = \frac{\text{g of dry sample}}{\text{g of sample}} \times 100$$

This oven-dried aliquot is not used for the extraction and should be disposed of appropriately once the dry weight has been determined.

7.3 Blend 10 g of the solid sample with 10 g of anhydrous sodium sulfate and place in an extraction thimble. The extraction thimble must drain freely for the duration of the extraction period. A glass wool plug above and below the sample in the Soxhlet extractor is an acceptable alternative for the thimble.

7.3.1 Add 1.0 mL of the surrogate standard spiking solution onto the sample (see Method 3500 for details on the surrogate standard and matrix spiking solutions).

7.3.2 For the sample in each analytical batch selected for spiking, add 1.0 mL of the matrix spiking standard.

7.3.3 Consult Secs. 5.5 and 8.3 of Method 3500 for the appropriate choice of matrix spiking compounds and concentrations.

7.4 Place approximately 300 mL of the extraction solvent (Sec. 5.4) into a 500-mL round bottom flask containing one or two clean boiling chips. Attach the flask to the extractor and extract the sample for 16 - 24 hours at 4 - 6 cycles/hour.

7.5 Allow the extract to cool after the extraction is complete.

7.6 Assemble a Kuderna-Danish (K-D) concentrator (Sec. 4.3), if necessary, by attaching a 10-mL concentrator tube to a 500-mL evaporation flask.

7.7 Attach the solvent vapor recovery glassware (condenser and collection device) (Sec. 4.4) to the Snyder column of the K-D apparatus following manufacturer's instructions.

7.8 Dry the extract by passing it through a drying column containing about 10 cm of anhydrous sodium sulfate. Collect the dried extract in a K-D concentrator. Wash the extractor flask and sodium sulfate column with 100 to 125 mL of extraction solvent to complete the quantitative transfer.

7.9 Add one or two clean boiling chips to the flask and attach a three-ball Snyder column. Prewet the Snyder column by adding about 1 mL of methylene chloride to the top of the column. Place the K-D apparatus on a hot water bath (15 - 20°C above the boiling point of the solvent) so that the concentrator tube is partially immersed in the hot water and the entire lower rounded surface of the flask is bathed with hot vapor. Adjust the vertical position of the apparatus and the water temperature, as required, to complete the concentration in 10 - 20 minutes. At the proper rate of distillation the balls of the column will actively chatter, but the chambers will not flood. When the apparent volume of liquid reaches 1 - 2 mL, remove the K-D apparatus from the water bath and allow it to drain and cool for at least 10 minutes.

7.10 If a solvent exchange is required (as indicated in Table 1), momentarily remove the Snyder column, add approximately 50 mL of the exchange solvent and a new boiling chip, and reattach the Snyder column. Concentrate the extract as described in Sec. 7.9, raising the temperature of the water bath, if necessary, to maintain proper distillation. When the apparent volume again reaches 1 - 2 mL, remove the K-D apparatus from the water bath and allow it to drain and cool for at least 10 minutes.

7.11 Remove the Snyder column and rinse the flask and its lower joints into the concentrator tube with 1 - 2 mL of methylene chloride or exchange solvent. If sulfur crystals are a problem, proceed to Method 3660 for cleanup. The extract may be further concentrated by using the techniques described in Sec. 7.12 or adjusted to 10.0 mL with the solvent last used.

7.12 If further concentration is indicated in Table 1, either micro Snyder column technique (Sec. 7.12.1) or nitrogen blowdown technique (Sec. 7.12.2) is used to adjust the extract to the final volume required.

7.12.1 Micro Snyder column technique

7.12.1.1 Add another one or two clean boiling chips to the concentrator tube and attach a two-ball micro Snyder column. Prewet the column by adding about 0.5 mL of methylene chloride or exchange solvent to the top of the column. Place the K-D apparatus in a hot water bath so that the concentrator tube is partially immersed in the hot water. Adjust the vertical position of the apparatus and the water temperature, as required, to complete the concentration in 5 - 10 minutes. At the proper rate of distillation the balls of the column will actively chatter, but the chambers will not flood.

7.12.1.2 When the apparent volume of liquid reaches 0.5 mL, remove the K-D apparatus from the water bath and allow it to drain and cool for at least 10 minutes. Remove the Snyder column and rinse the flask and its lower joints with about 0.2 mL of solvent and add to the concentrator tube. Adjust the final volume to 1.0 - 2.0 mL, as indicated in Table 1, with solvent.

7.12.2 Nitrogen blowdown technique

7.12.2.1 Place the concentrator tube in a warm water bath (approximately 35°C) and evaporate the solvent volume to the required level using a gentle stream of clean, dry nitrogen (filtered through a column of activated carbon).

CAUTION: Do not use plasticized tubing between the carbon trap and the sample, since it may introduce contaminants.

7.12.2.2 The internal wall of the tube must be rinsed several times with the appropriate solvent during the operation. During evaporation, the solvent level in the tube must be positioned to prevent water from condensing into the sample (i.e., the solvent level should be below the level of the water bath). Under normal operating conditions, the extract should not be allowed to become dry.

CAUTION: When the volume of solvent is reduced below 1 mL, semivolatile analytes may be lost.

7.13 The extracts obtained may now be analyzed for the target analytes using the appropriate organic technique(s) (see Sec. 4.3 of this Chapter). If analysis of the extract will not be performed immediately, stopper the concentrator tube and refrigerate. If the extract will be stored longer than 2 days, it should be transferred to a vial with a PTFE-lined screw cap or crimp top, and labeled appropriately.

8.0 QUALITY CONTROL

8.1 Any reagent blanks, matrix spikes, or replicate samples should be subjected to exactly the same analytical procedures as those used on actual samples.

8.2 Refer to Chapter One for specific quality control procedures and Method 3500 for extraction and sample preparation procedures.

9.0 METHOD PERFORMANCE

Refer to the determinative methods for performance data.

10.0 REFERENCES

None.

TABLE 1
SPECIFIC EXTRACTION CONDITIONS FOR VARIOUS DETERMINATIVE METHODS

Determinative method	Extraction pH	Exchange solvent for analysis	Exchange solvent for cleanup	Volume of extract for cleanup (mL)	Final extract volume for analysis (mL) ^a
8041	as received	2-propanol	hexane	1.0	1.0, 0.5 ^b
8061	as received	hexane	hexane	2.0	10.0
8070	as received	methanol	methylene chloride	2.0	10.0
8081	as received	hexane	hexane	10.0	10.0
8082	as received	hexane	hexane	10.0	10.0
8091	as received	hexane	hexane	2.0	1.0
8100	as received	none	cyclohexane	2.0	1.0
8111	as received	hexane	hexane	2.0	10.0
8121	as received	hexane	hexane	2.0	1.0
8141	as received	hexane	hexane	10.0	10.0
8270 ^c	as received	none	-	-	1.0
8310	as received	acetonitrile	-	-	1.0
8321	as received	methanol	-	-	1.0
8325	as received	methanol	-	-	1.0
8410	as received	methylene chloride	methylene chloride	10.0	0.0 (dry)

^a For methods where the suggested final extract volume is 10.0 mL, the volume may be reduced to as low as 1.0 mL to achieve lower detection limits.

^b Phenols may be analyzed by Method 8041, using a 1.0-mL 2-propanol extract by GC/FID. Method 8041 also contains an optional derivatization procedure for phenols which results in a 0.5-mL hexane extract to be analyzed by GC/ECD.

^c The specificity of GC/MS may make cleanup of the extracts unnecessary. Refer to Method 3600 for guidance on the cleanup procedures available if required.

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