PART 6000

INDIVIDUAL ORGANIC COMPOUNDS

Part Coordinators
John R. Gumpper, Ann E. Rosecrance and Edward M. Glick

Joint Task Group Chairs

6010 Introduction ..... Edward M. Glick
6020 Quality Assurance/Quality Control ..... Ann E. Rosecrance
6040 Constituent Concentration by Gas Extraction ..... Bart Koch
6200 Volatile Organic Compounds ..... Melissa S. Dale
6232 Trihalomethanes and Chorinated Organic Solvents ..... Nancy E. Grams
6251 Disinfection By-Products: Haloacetic Acids and Trichlorophenol ..... Sanwat N. Chaudhuri
6252 Disinfection By-Products: Aldehydes ..... Stephen D. Winslow
6610 Carbamate Pesticides ..... Andrew D. Eaton
6640 Acidic Herbicide Compounds ..... Russell Chinn

Summary of Major Changes since 1998

A solid-phase microextraction (SPME) method was added to Constituent Concentration by Gas Extraction (6040), the section covering closed-loop stripping.

Revisions in Disinfection By-Products: Haloacetic Acids (6251) include changes in holding times and new precision and bias data. Sampling and storage techniques were revised in Disinfection By-Products: Aldehydes (6252), and new data on method detection levels, new chromatograms, and improvements in calibration and calculation were added. Carbamate Pesticides (6610) contains new data on the stability of carbamates when different preservatives are used. New data on method detection levels, retention time, and precision and bias were added to Acidic Herbicide Compounds (6640), and the method was revised to include hydrolysis steps.

Also see methods in Supplement to the 21st Edition, pp. S-1 through S-16.

6010 INTRODUCTION

6010 A. General Discussion

The methods presented in Part 6000 are intended for the determination of individual organic compounds. Methods for determination of aggregate concentrations of groups of organic compounds are presented in Part 5000.

Most of the methods presented herein are highly sophisticated instrumental methods for determining very low concentrations of the organic constituents. Stringent quality control requirements are given with each method and require careful attention.

Many compounds are determinable by two or more of the methods presented in Part 6000. Table 6010:I shows the specific analytical methods applicable to each compound. Guidance on selection of method is provided in the introduction to each section.

Joint Task Group: Edward M. Glick (chair).

TABLE 6010:I. ANALYSIS METHODS FOR SPECIFIC ORGANIC COMPOUNDS*

Compound	Analysis Methods (section number)	Compound	Analysis Methods (section number)
Acenaphthene	6040B; 6410B; 6440B	Captan	6630B
Acenaphthylene	6410B; 6440B	Carbaryl	6610B
Acetaldehyde	6252B	Carbofuran	6610B
Aldicarb	6610B	Carbon tetrachloride	6200B, C
Aldicarb sulfone	6610B	Chlordane	6410B; 6630B, C
Aldicarb sulfoxide	6610B	Chlorobenzene	6040B; 6200B, C
Aldrin	6410B; 6630B, C	Chloroethane	6200B, C
Aminomethylphosphonic acid (AMPA)	6651B	Chloroethoxy methane	6040B; 6410B
Anthracene	6040B; 6410B; 6440B	Chloroethyl ether	6040B; 6410B
Baygon	6610B	Chloroethylvinyl ether	6200B, C
Bentazon	6640B	Chloroform	6200B, C; 6232B
Benzaldehyde	6252B	Chloroisopropyl ether	6410B
Benzene	6200B, C	Chloromethane	6200B, C
Benzidine	6410B	Chloromethyl benzene	6040B
Benzo(a)anthracene	6040B; 6410B; 6440B	Chloromethylphenol	6410B; 6420B
Benzo(a)pyrene	6410B; 6440B	Chloronaphthalene(s)	6040B; 6410B
Benzo(b)fluoranthene	6410B; 6440B	Chlorophenol(s)	6410B; 6420B
Benzo(ghi)perylene	6410B; 6440B	Chlorophenoxy benzene	6040B
Benzo(k)fluoranthene	6410B; 6440B	Chlorophenyl phenyl ether	6410B
BHC(s)	6410B; 6630C	Chlorotoluene	6200B, C
Bromobenzene	6040B; 6200B, C	Chrysene	6040B; 6410B; 6440B
Bromochloroacetic acid	6251B	Crotonaldehyde	6252B
Bromochloromethane	6200B, C	Cyclohexanone	6252B
Bromodichloromethane	6040B; 6200B, C; 6232B	2,4-D (dichlorophenoxyacetic acid)	6640B
Bromoform	6040B; 6200B, C; 6232B	Dalapon	6640B
Bromomethane	6200B, C	DDD	6410B; 6630B, C
Bromophenoxybenzene	6040B	DDE	6410B; 6630B, C
Bromophenyl phenyl ether	6410B	DDT	6410B; 6630B, C
Butanal	6252B	Decanal	6252B
Butyl benzyl phthalate	6410B	Dibenzo(a,h)anthracene	6410B; 6440B
Butylbenzene(s)	6200B, C	Dibromoacetic acid (DBAA)	6251B

TABLE 6010:I. CONT.

Compound	Analysis Methods (section number)	Compound	Analysis Methods (section number)
Dibromochloromethane	6040B; 6200B, C; 6232B	Methylene chloride	6200B, C
Dibromochloropropane	6200B, C; 6231B	Methyl glyoxal	6252B
Dibromoethane	6040B; 6200B, C; 6231B	Methylisoborneol	6040B, D
Dibromomethane	6200B, C	Methyl parathion	6630B
Dibutyl phthalate	6410B	Methomyl	6610B
Dicamba	6640B	Mirex	6630B
Dichloran	6630B	Monobromoacetic acid (MBAA)	6251B
Dichloroacetic acid (DCAA)	6251B	Monochloroacetic acid (MCAA)	6251B
Dichlorobenzene(s)	6040B; 6200B, C; 6410B	Naphthalene	6040B; 6200B, C; 6410B; 6440B
Dichlorobenzidine	6410B	Nitrobenzene	6410B
Dichlorodifluoromethane	6200B, C	Nitrophenol(s)	6410B; 6420B
Dichloroethane	6200B, C	Nitrosodi-*n*-propylamine	6410B
Dichloroethene(s)	6200B, C	Nitrosodimethylamine	6410B
Dichlorophenol(s)	6410B; 6420B	Nitrosodiphenylamine	6410B
Dichloropropane(s)	6200B, C	Nonanal	6252B
Dichloropropene	6040B; 6200B, C	Octanal	6252B
Dieldrin	6410B; 6630B, C	Oxamyl	6610B
Diethyl phthalate	6040B; 6410B	Parathion	6630B
Dimethyl phthalate	6410B	PCB-1016, 1221, 1232, 1242, 1248, 1254, 1260	6410B, 6630B
Dimethylphenol(s)	6410B; 6420B	Pentachloronitrobenzene	6630B
Dinitrophenol(s)	6410B; 6420B	Pentachlorophenol	6410B; 6420B; 6640B
Dinitrotoluene(s)	6410B	Pentanal	6252B
Dinoseb	6640B	Phenanthrene	6040B; 6410B; 6440B
Di-*n*-octyl phthalate	6410B	Phenol	6410B; 6420B
Diphenyl hydrazine	6040B	Phenylbenzamine	6040B
Endosulfan	6410B; 6630B, C	Picloram	6640B
Endosulfan sulfate	6410B; 6630C	Propanal	6252B
Endrin	6410B; 6630B, C	Propylbenzene	6040B; 6200B, C
Endrin aldehyde	6410B; 6630C	Pyrene	6040B; 6410B; 6440B
Ethenyl benzene (styrene)	6040B	Silvex (trichlorophenoxy propionic acid)	6640B
Ethylbenzene	6040B; 6200B, C	Strobane	6630B
Ethylhexyl phthalate	6410B	Styrene (ethenyl benzene)	6200B, C
Fluoranthene	6040B; 6410B; 6440B	2,4,5-T (trichlorophenoxy acetic acid)	6640B
Fluorene	6040B; 6410B; 6440B	2,4,5-TP	6640B
Formaldehyde	6252B	Tetrachloroethane(s)	6040B; 6200B, C
Geosmin	6040B, D	Tetrachloroethene	6040B; 6200B, C
Glyoxal	6252B	Toluene	6200B, C
Glyphosate	6651B	Toxaphene	6410B; 6630B, C
Heptachlor	6410B; 6630B, C	Trichloroanisole	6040B
Heptachlor epoxide	6410B; 6630B, C	Trichloroacetic acid (TCAA)	6251B
Heptaldehyde	6252B	Trichlorobenzene(s)	6040B; 6200B, C; 6410B
Heptanal	6252B	Trichloroethane(s)	6040B; 6200B, C
Hexachlorobenzene	6040B; 6410B	Trichloroethene	6040B; 6200B, C
Hexachlorobutadiene	6040B; 6200B, C; 6410B	Trichlorofluoromethane	6200B, C
Hexachlorocyclopentadiene	6410B	Trichlorophenol	6251B; 6410B; 6420B
Hexachloroethane	6040B; 6410B	Trichloropropane	6200B, C
Hexanal	6252B	Trifluralin	6630B
3-Hydroxycarbofuran	6610B	Trimethylbenzene(s)	6200B, C
Indeno(1,2,3-cd))pyrene	6410B; 6440B	Vinyl chloride	6200B, C
Isobutylmethoxy pyrazine	6040B	Xylene(s)	6040B; 6200B, C
Isophorone	6410B		
Isopropylbenzene	6200B, C		
Isopropyl methoxy pyrazine	6040B		
Isopropyltoluene	6200B, C		
Lindane (γ-BHC)	6630B		
Malathion	6630B		
Methane	6211		
Methiocarb	6610B		
Methoxychlor	6630B		
Methyldinitrophenol(s)	6410B; 6420B		

* Compounds are listed under the names by which they are most commonly known and called in specific methods.

6010 B. Sample Collection and Preservation

1. Volatile Organic Compounds

Use 25- or 40-mL vial equipped with a screw cap with a hole in the center* and TFE-faced silicone septum.† Wash vials, caps, and septa with detergent, rinse with tap and distilled water, and dry at 105°C for 1 h before use in an area free of organic vapors. NOTE — Do not heat seals for extended periods of time (> 1 h) because the silicone layer slowly degrades at 105°C. When bottles are cool, seal with TFE seals. Alternatively purchase precleaned vials free from volatile organic compounds.

Collect all samples in duplicate and prepare replicate field reagent blanks with each sample set. A sample set is all samples collected from the same general sampling site at approximately the same time. Prepare field reagent blanks in the laboratory by filling a minimum of two sample bottles with reagent water, sealing, and shipping to the sampling site along with empty sample bottles.

Fill sample bottle just to overflowing without passing air bubbles through sample or trapping air bubbles in sealed bottle. When sampling from a water tap, open tap and flush until water temperature has stabilized (usually about 10 min). Adjust flow rate to about 500 mL/min and collect duplicate samples from flowing stream. When sampling from an open body of water, fill a 1-L, wide-mouth bottle or beaker with a representative sample and carefully fill duplicate sample bottles from the container.

Preservation of samples is highly dependent on target constituents and sample matrix. Ongoing research indicates the following areas of concern: rapid biodegradation of aromatic compounds, even at low temperatures;[1] dehydrohalogenation reactions such as conversion of pentachloroethane to tetrachloroethane;[2] reactions of alkylbenzenes in chlorinated samples, even after acidification; and possible interactions among preservatives and reductants when dechlorination is used to prevent artifact formation, especially in samples potentially containing many target compounds.

There is as yet no single preservative that can be recommended. Ideally, maintain samples chilled (preferably at 4°C) and analyze immediately. In practice, delays between sampling and analysis often necessitate preservation. The recommended preservation techniques are summarized in Table 6010:II.

1) For samples and field blanks that contain volatile constituents but do not contain residual chlorine, add HCl (4 drops 6*N* HCl/40 mL) to prevent biodegradation and dehydrohalogenation. NOTE: HCl may contain traces of organic solvents. Verify freedom from contamination before using a specific lot for preservation.

2) For samples that contain residual chlorine and for field blanks, also add a reducing agent. In general, ascorbic acid (25 mg/40 mL) is the agent of choice for GC/MS determinations, while sodium thiosulfate (3 mg/40 mL) is more appropriate with conventional GC detectors. Alternative reducing agents may be specified or permitted in particular methods.

In all cases run reagent blanks with reducing agent and acid to ensure absence of interferences. Always add the reducing agent before adding the HCl to the sample. The reducing agent can be added to the sample bottle as a powder before the bottle is shipped into the field. Do not add HCl to a chlorinated sample because formation of chlorinated volatiles can occur. When both preservatives are being added in the field, add reducing agent, fill bottle, and then, after at least 1 min to reduce residual chlorine, add 4 drops 1:1 HCl (or other amount sufficient to lower sample pH to 2.0).

Tightly seal sample bottle, TFE face down. After sampling and preservation invert several times to mix. Chill samples to 4°C immediately after collection and hold chilled in an atmosphere free of organic solvent vapors until analysis. Normally analyze all samples within 14 d of collection. Shorter or longer holding times may be appropriate, depending on constituents and sample matrix. Develop data to show that alternate holding times are appropriate.

TABLE 6010:II. RECOMMENDED PRESERVATION FOR VOLATILE ORGANIC COMPOUNDS

Constituents	Chlorinated Matrix	Non-Chlorinated Matrix
Halocarbons	Reducing agent, then HCl	HCl
Aromatics	Reducing agent, then HCl	HCl
THMs	Reducing agent (HCl optional)*	None required
EDB/DBCP	None required	None required

* See 6232B.2.

2. Other Organic Compounds

See individual methods for sampling and preservation requirements.

3. References

1. BELLAR, T. & J. LICHTENBERG. 1978. Semi-automated headspace analysis of drinking waters and industrial waters for purgeable volatile organic compounds. *In* C. E. Van Hall, ed. Measurement of Organic Pollutants in Water and Wastewater. STP 686, American Soc. Testing & Materials. Philadelphia, Pa.
2. BELLAR, T. & J. LICHTENBERG. 1985. The Determination of Synthetic Organic Compounds in Water by Purge and Sequential Trapping Capillary Column Gas Chromatography. U.S. Environmental Protection Agency, Cincinnati, Ohio.

4. Bibliography

KEITH, L. H., ed. 1988. Principles of Environmental Testing. American Chemical Soc., Washington, D.C.

* Pierce 13075 or equivalent.
† Pierce 12722 or equivalent.

6010 C. Analytical Methods

1. General Discussion

The methods presented in Part 6000 for identification and quantitation of trace organic constituents in water generally involve isolation and concentration of the organic compounds from a sample by solvent or gas extraction (see Section 6040 and individual methods), separation of the components, and identification and quantitation of the compounds with a detector.

2. Gas Chromatographic Methods

Gas chromatographic (GC) methods are highly sophisticated microanalytical procedures. They should be used only by analysts experienced in the techniques required and competent to evaluate and interpret the data.

a. Gas chromatograph:

1) Principle—In gas chromatography a mobile phase (a carrier gas) and a stationary phase (column packing or capillary column coating) are used to separate individual compounds. The carrier gas is nitrogen, argon-methane, helium, or hydrogen. For packed columns, the stationary phase is a liquid that has been coated on an inert granular solid, called the column packing, that is held in borosilicate glass tubing. The column is installed in an oven with the inlet attached to a heated injector block and the outlet attached to a detector. Precise and constant temperature control of the injector block, oven, and detector is maintained. Stationary-phase material and concentration, column length and diameter, oven temperature, carrier-gas flow, and detector type are the controlled variables.

When the sample solution is introduced into the column, the organic compounds are vaporized and moved through the column by the carrier gas. They travel through the column at different rates, depending on differences in partition coefficients between the mobile and stationary phases.

2) Interferences—Some interferences in GC analyses occur as a result of sample, solvent, or carrier gas contamination, or because large amounts of a compound may be injected into the GC and linger in the detector. Methylene chloride, chloroform, and other halocarbon and hydrocarbon solvents are ubiquitous contaminants in environmental laboratories. Make strenuous efforts to isolate the analytical system from laboratory areas where these or other solvents are in use. An important sample contaminant is sulfur, which is encountered generally only in base/neutral extracts of water, although anaerobic groundwaters and certain wastewaters and sediment/sludge extracts may contain reduced sulfur compounds, elemental sulfur, or polymeric sulfur. Eliminate this interference by adding a small amount of mercury or copper filings to precipitate the sulfur as metallic sulfide. Sources of interference originating in the chromatograph, and countermeasures, are as follows:

- Septum bleed—This occurs when compounds used to make the septum on the injection port of the GC bleed from the heated septum. These high-molecular-weight silicon compounds can be distinguished readily from compounds normally encountered in environmental samples by their peak shape or baseline rise. Nevertheless, minimize septum bleed by using septum sweep, in which clean carrier gas passes over the septum to flush out the "bleed" compounds.
- Column bleed—This term refers to loss of column coating or breakdown products when the column is heated. This interference is more prevalent in packed columns, but also occurs to a much lesser extent in capillary columns. It occurs when the column temperature is high or when water or oxygen is introduced into the system. Solvent injection can damage the stationary phase by displacing it. Certain organic compounds acting as powerful solvents, acids, or bases can degrade the column coating. Injection of large amounts of certain surface-active agents may destroy GC columns. Signs of column bleed are a rising baseline or a large peak at end of run.
- Ghost peaks—These peaks occur when an injected sample contains either a large amount of a given compound, or a compound that adsorbs to the column coating or injector parts (e.g., septum). When a subsequent sample is injected, peaks can appear as a result of the previous injection. Eliminate ghost peaks by injecting a more dilute sample, by producing less reactive derivatives of a compound that may interact strongly with the column material, by selecting a column coating that precludes these interactions, by injecting solvent blanks between samples, or by heating the column for a longer time or to a higher temperature at the end of the run.

b. Detectors: Various detectors are available for use with gas chromatographic systems. See individual methods for recommendations on appropriate detectors.

1) Electrolytic conductivity detector—The electrolytic conductivity detector is a sensitive and element-specific detector that has gained considerable attention because of its applicability to the gas chromatographic analysis of environmentally significant compounds. It is utilized in the analysis of purgeable halocarbons, pesticides, herbicides, pharmaceuticals, and nitrosamines. This detector is capable of operation in each of four specific modes: halogen (X), nitrogen (N), sulfur (S), and nitrosamine (NO). Only organic compounds containing these elements will be detected.

Compounds eluting from a gas chromatographic column enter a reactor tube heated to 800°C. They are mixed with a reaction gas, hydrogen for X, N, or NO modes, and air for the S mode. The hydrogen catalytically reduces the compounds while the air oxidizes them. The gaseous products are transferred to the detector through a conditioned ion exchange resin or scrubber. In the halogen mode, only HX is detected, while NH_3 or H_2S are eliminated on the resin. In the nitrogen or nitrosamine mode, the NH_3 formed is ionized while HX and H_2S, if present, are eliminated with KOH/quality wool scrubber. The sulfur mode produces SO_2 or SO_3, which is ionized while HX is removed with a silver wire scrubber. All other products either are not ionizable or are produced in such low yield that they are not detectable.

The electrolytic conductivity detector contains reference and analytical electrodes, a gas-liquid contactor, and a gas-liquid separator. The conductivity solvent enters the cell and flows by the reference electrode. It combines with the gaseous reaction products in the gas-liquid phases in the gas-liquid contactor. This heterogeneous mixture is separated into gas and liquid phases in the gas-liquid separator, with the liquid phase flowing past the analytical electrode. The electrometer monitors the difference in

conductivity at the reference electrode (solvent) and the analytical electrode (solvent + carrier + reaction products).

2) Electron capture detector—The electron capture detector (ECD) usually is used for the analysis of compounds that have high electron affinities, such as chlorinated pesticides, drugs, and their metabolites. This detector is somewhat selective in its response, being highly sensitive toward molecules containing electronegative groups: halogens, peroxides, quinones, and nitrogen groups. It is insensitive toward functional groups, such as amines, alcohols, and hydrocarbons.

The detector is operated by passing the effluent from the gas chromatographic column over a radioactive beta particle emitter, usually nickel-63 or tritium adsorbed on platinum or titanium foil. An electron from the emitter ionizes the carrier gas, preferably nitrogen, and produces a burst of electrons. About 100 secondary electrons are produced for each initial beta particle. After further collisions, the energy of these electrons is reduced to the thermal level and they can be captured by electrophilic sample molecules.

The electron population in the ECD cell is collected periodically by applying a short voltage pulse to the cell electrodes and the resulting current is compared with a reference current. The pulse interval is adjusted automatically to keep the cell current constant, even when some of the electrons are being captured by the sample. The change in the pulse rate when a sample enters the ECD is then related to the sample concentration. The ECD offers linearity in the range of 10^4 and subpicogram detection limits for compounds with high electron affinities.

3) Flame ionization detector—The flame ionization detector (FID) is widely used because of its high sensitivity to organic carbon-containing compounds. The detector consists of a small hydrogen/air diffusion flame burning at the end of a jet. When organic compounds enter the flame from the column, electrically charged intermediates are formed. These are collected by applying a voltage across the flame. The resulting current is amplified by an electrometer and measured. The response of the detector is directly proportional to the total mass entering the detector per unit time and is independent of the concentration in the carrier gas.

The FID is perhaps the most widely used detector for gas chromatography because of several advantages: (*a*) it responds to virtually all organic carbon-containing compounds with high sensitivity (approximately 10^{-13} g/mL); (*b*) it does not respond to common carrier gas impurities such as water and carbon dioxide; (*c*) it has a large linear response range (approximately 10^7) and excellent baseline stability; (*d*) it is relatively insensitive to small column flow-rate changes during temperature programming; (*e*) it is highly reliable, rugged, and easy to use; and (*f*) it has low detector dead volume effects and fast response. Its limitations include: (*a*) it gives little or no response to noncombustible gases and all noble gases; and (*b*) it is a destructive detector that changes the physical and chemical properties of the sample irreversibly.

4) Photoionization detector—Photoionization occurs when a molecular species absorbs a photon of light energy and dissociates into a parent ion and an electron. The photoionization detector (PID) detects organic and some inorganic species in the effluent of a gas chromatograph with detection limits as low as the picogram range. The PID is equipped with a sealed ultraviolet light source that emits photons which pass through an optically transparent window (made of LiF, MgF_2, NaF, or sapphire) into an ionization chamber where photons are absorbed by the eluted species. Compounds having ionization potential less than the UV source energy are ionized. A positively biased high-voltage electrode accelerates the resulting ions to a collecting electrode and the resulting current is measured by an electrometer. This current is proportional to the concentration.

The PID has high sensitivity, low noise (approximately 10^{-14} A), and excellent linearity (10^7), is nondestructive, and can be used in series with a second detector for more selective detection. The PID can be operated as a universal detector or a selective detector by simply changing the photon energy of the ionization source. Tables of ionization potentials are used to select the appropriate UV source for a given measurement.

5) Mass spectrometer—The mass spectrometer (MS) has the ability to detect a wide variety of compounds, coupled with a capacity to deduce compound structures from fragmentation patterns. Among the different types of mass spectrometers, the quadrupole has become the most widely used in water and wastewater analysis.

The mass spectrometer detects compounds by ionizing molecules into charged species with a 70-eV beam. The ions are accelerated toward the quadrupole mass filter through a series of lenses held at 0 to 200 V. The differently sized, charged fragments are separated according to mass-to-charge ratio (related to molecular weight) by means of the quadrupole, which uses varying electric and radiofrequency (rf) fields. The quadrupole is connected to a computer, which varies these fields so that only fragments of one particular mass-to-charge ratio (± 0.5) can traverse the quadrupole at any one time. As the ions leave the quadrupole they are attracted to the electron multiplier through an electrical potential of several thousand volts. The charge fragments, in turn, are detected by the electron multiplier. Because the electric and the rf fields are cycled every few seconds, a fragmentation pattern is obtained. Each cycle is called a mass scan. Most chemicals have unique fragmentation patterns, called mass spectra. The computer contains, and can search, a library of known mass spectra to identify tentatively an unknown compound exhibiting a particular spectrum. Use authentic compounds for confirmation after tentative identifications are made.

Background mass interference can result from the ability of the mass spectrometer to detect any ions created in its ion volume (up to a specified mass). Any compounds continuously present in the source will be detected. Some mass ions always present are due to air components that leak into the system, such as oxygen (masses 16 and 32), nitrogen (masses 14 and 28), carbon dioxide (mass 44), argon (mass 40), and water (mass 18), or to helium carrier gas (masses 4 and 8), or to diffusion pump oil vapors.

3. High-Performance Liquid Chromatographic (HPLC) Methods

a. Principle: HPLC is an analytical technique in which a liquid mobile phase transports a sample through a column containing a liquid stationary phase. The interaction of the sample with the stationary phase selectively retains individual compounds and permits separation of sample components. Detection of the separated sample compounds is achieved mainly through the use of absorbance detectors for organic compounds and through conductivity and electrochemical detectors for metal and inorganic components.

b. Detectors:

1) Photodiode array detector (PDAD)—The PDAD measures the absorbance of a sample from an incident light source (UV-VIS). After passing through the sample cell, the light is directed through a holographic grating that separates the beam into its component wavelengths reflected on a linear array of photodiodes. This permits the complete absorbance spectrum to be obtained in 1 s or less and simultaneous multiwavelength analysis.

The PDAD is subject to the interference encountered with all absorbance detectors. Of special concern for HPLC is the masking of the absorbance region of the HPLC mobile phase and its additives. This may reduce the range and sensitivity of the detector to the sample components. Most interferences occur in monitoring the shorter wavelengths (200–230 nm). In this region, many organic compounds absorb light energy and can be sources of interference.

2) Post column reactor (PCR)—The PCR consists of in-line sample derivatizing/reacting equipment that permits chemical alteration of certain organic compounds. This equipment is used to enhance detection by attaching a chromophore to the compound(s) of interest. Sensitivity and selectivity of compounds that were initially undetectable are altered to make them detectable.

Interferences from this technique usually arise from the impurities in the reagents used in the reaction. When this technique is coupled with a selective detector such as fluorescence, these interferences are minimized. Generally, only compounds of the same class as the compounds of interest will cause interference.

3) Fluorescence detector—The fluorescence detector is an absorbance detector in which the sample is energized by a monochromatic light source. Compounds capable of absorbing the light energy do so and release it as fluorescence emission. Filters permit the detector to respond only to the fluorescent energy. The fluorescence detector is the most sensitive of the current HPLC detectors available and often is used in conjunction with a post column reactor.

Because of instrument sensitivity, minute quantities of contaminants can cause interferences to fluorescence detectors. Contamination can happen from glassware, mobile phase solvents, postcolumn reagents, etc. These sources will raise the background signal and thus narrow the range of the detector. Interference from individual compounds is minimal because of detector specificity (i.e., all interferences must fluoresce).

6020 QUALITY ASSURANCE/QUALITY CONTROL

6020 A. Introduction

Quality assurance (QA) and quality control (QC) for organic compound analysis include the operating principles stated in Sections 1020 Quality Assurance, 1030 Data Quality, and 1040 Method Development and Evaluation. This section consolidates the additional requirements common to the methods in Part 6000. The requirements are recommended minimum QA/QC activities; they should be followed unless the individual method gives different, but comparable, specifications. Some methods may have additional QA/QC requirements. Others may have broader acceptance criteria because of the unique difficulties associated with the determination of a constituent, e.g., the extraction efficiency for phenols.

Joint Task Group: 20th Edition—Ann Rosecrance (chair).

6020 B. Quality Control Practices

This section describes the elements of a quality control program deemed necessary to maintain proper control for organic analyses. The related data quality objectives (DQOs), i.e., the rationale for sampling and analyses, should define which elements are necessary for each individual analytical design.

1. Calibration

a. Initial calibration: Perform initial calibration with a minimum of five concentrations of analytical standards for the analyte(s) of interest (for example: 1, 5, 10, 20, 40). The lowest concentration should be at the minimum reporting level; for example, if the minimum reporting level is 1, the lowest concentration should be 1. The highest concentration should be at or near the upper end of the calibration range; for example, if the upper end of the calibration range is 40, the high standard should be 40. Concentration ranges should reflect concentrations in actual samples. Choose calibration concentrations with no more than one order of magnitude between concentrations; for example, for a calibration range of 1 to 1000, choose concentrations of 1, 10, 100, and 1000.

Use any of the following calibration functions, as appropriate: response factor for internal standard calibration, calibration factor for external standard calibration, or calibration curve. Calibration curves may be linear or quadratic, and may or may not pass through the origin. Use the following recommended acceptance criteria for the various calibration functions.

If response factors or calibration factors are used, relative standard deviation (RSD) for each analyte should be $\leq 20\%$. If RSD is $> 20\%$ for any analyte, identify and correct source of lack of linearity before sample quantitation. When using response factors (i.e., for GC/MS analysis), check performance or sensitivity of instrument for each analyte against minimum ac-

ceptance values for response factors. Refer to specific analytical method for the acceptance criteria for response factors.

If a linear regression is used, the correlation coefficient should be >0.995. If a calibration curve has been constructed, recalculate each calibration point compared to curve. Values should be within ±20%. If any of the recalculated values are not within ±20%, identify and correct source of outlier(s) before sample quantitation.

Use initial calibration with any of the above functions (response factor, calibration factor, or calibration curve) for quantitation of analytes in samples. Use continuing calibration, ¶ *b* below, only for checks on initial calibration and not for sample quantitation. Perform initial calibration when instrument is set up and whenever continuing calibration criteria are not met.

b. Continuing calibration: Continuing calibration is the periodic verification, by analysis of a calibration standard, that instrument performance has not changed significantly from initial calibration. Perform continuing calibration every 10 samples for GC analysis, every 20 samples for GC/MS analysis, or every 12 h, whichever is more frequent. Perform continuing calibration with one or more of the concentrations of analytical standards in the initial calibration, varying actual concentration of continuing calibration standard over the calibration range. The acceptance criteria for continuing calibration should be within ±20% (80 to 120% recovery) compared to the known or expected value of the calibration standard. If the acceptance criteria are not met, reanalyze continuing calibration standard or repeat initial calibration. When using response factors (i.e., GC/MS analysis), check performance or sensitivity of instrument for analytes against minimum acceptance values for the response factors. Refer to the specific analytical method for acceptance criteria for response factors for each analyte.

c. Closing standard: Finish all runs with a laboratory-fortified blank (LFB) for VOC analyses or closing standard (for methods with procedural standards) to demonstrate that performance was still acceptable for last sample analyzed. A LFB is a reagent blank to which a known concentration of analytes has been added. See ¶ 3*b* below. All samples must be bracketed by acceptable continuing calibrations.

2. Initial Quality Control

a. Initial demonstration of capability: Before analysis of any sample, require each analyst to demonstrate proficiency with the method of choice. Include at least the analysis of a laboratory reagent blank (LRB) and a minimum of four laboratory-fortified blanks (LFBs) that have added concentrations between 5 times the minimum reporting level and the midpoint of the calibration curve. The blank should not contain any analyte at a concentration greater than one-fourth the minimum quantitation level. The precision and percent recovery calculated from the four LFBs should be at least as good as the values listed in the method of choice.

b. Method detection level (MDL): Determine MDL before any samples are analyzed, using procedure described in Section 1030C or other specified procedure[1] required for the type of samples the laboratory is intending to analyze. As a starting point for determining concentration to use in performing the laboratory's MDL calculation, try about five times the estimated instrument detection level or refer to the selected method. Determine MDL as an iterative process. Repeat determinations if calculated MDLs are not within a factor of 10 of the fortified value. Determine MDL at least annually. Preferably analyze samples for MDL determination over a 3- to 5-d period to generate a more realistic value. Include all applicable sample preparatory techniques in MDL determinations.

c. Minimum quantitation level (MQL): The MQL is the lowest level that can be quantitated accurately. MQL is defined as four times the MDL. Report samples containing compounds of interest at a level less than the MQL as <MQL. Report samples containing compounds of interest at a level less than the MDL as ND (not detected).

e. Sample (batch) set: A sample set or batch is defined as those samples extracted in a single day, not to exceed 20 samples.

f. Analytical day: An analytical day is defined as a 12-h analytical period.

3. Batch Quality Control

a. Reagent blank: A reagent blank, also known as a method blank, consists of all reagents and preservatives that normally contact a sample when it is carried through the entire analytical procedure. Use a reagent blank to determine contribution of reagents and preparative analytical steps to error in the observed value. No analyte of interest should be present in a reagent blank at a level greater than one fourth the MQL. Include a minimum of one reagent blank with each sample set or batch.

b. Laboratory-fortified blank (LFB): An LFB, also known as spiked blank, is a reagent blank containing all the same reagents and preservatives as samples and to which a known concentration of analyte(s) has been added. Use LFB to evaluate laboratory performance and analyte recovery in a blank matrix. Make addition concentration at least 5 times the MQL or the midpoint of the calibration curve, and use to calculate recovery limits and to plot control charts as in Section 1020B. Prepare known-addition solution for blanks and samples from a different primary mix than that used to develop the calibration standard mix. Include a minimum of one LFB with each sample set or batch. Ensure that LFB meets performance criteria in the method of choice.

c. Internal standard (IS): An internal standard is a compound of known concentration added to each standard and sample/sample extract just before sample analysis. This compound should have chromatographic characteristics similar to those of the analytes of interest. Use IS to monitor retention time, relative response, and quantity of analytes in each sample. When quantifying by the internal standard method, measure all analyte responses relative to this standard. Internal standard response should be in the range of ±30% compared to calibration curve response. The retention time of this compound should separate from all analytes of interest and elute in a representative area of the chromatogram. If a single compound cannot be found to meet these criteria, use additional compounds.

d. Surrogate standard: A surrogate standard is a compound of a known concentration added to each environmental and blank sample before extraction (before analysis for VOCs or direct injection methods). Use compound(s) that have characteristics similar to those of the analytes of interest and that are unlikely to be found in environmental samples. Carry surrogate standard

through entire sample extraction and analytical process to monitor extraction efficiency of the method for each sample. Refer to method of choice for specific surrogates and acceptance criteria.

e. Quality control sample: Analyze an externally generated quality control sample of known quantity as a laboratory-fortified blank at least quarterly or whenever new stock solutions are prepared. This sample is used to validate the laboratory's standards both qualitatively and quantitatively.

f. Laboratory-fortified sample (LFS): A laboratory-fortified sample, also known as laboratory-fortified matrix or matrix spike, is another portion of a sample fortified with the analytes of interest at a concentration at least 5 times the MQL or around the midpoint of the calibration range. Include a minimum of one LFS with each sample set or batch. Make LFSs of sufficient concentrations that sample background levels do not adversely affect the recovery calculations. (Adjust addition concentrations if this is a known sample to be about five times background level.) Base sample batch acceptance on results of LFBs rather than on LFSs, because the sample matrix may interfere with method performance. Prepare addition solution for blanks and samples from a different primary mix than that used to develop calibration standard mix.

g. Laboratory-fortified sample duplicates (LFSDs): A laboratory-fortified sample duplicate, also known as laboratory-fortified matrix duplicate, spiked sample duplicate, or matrix spike duplicate, is a second portion of the sample to which a known amount of analyte is added. If sufficient sample volume is collected, add the analytes of interest to a second portion of sample and compare to first. If sufficient sample volume is not collected, use a second sample to obtain results on two separate LFSs rather than LFSDs. Include a minimum of one LFSD with each sample set or batch. Compare precision and bias to those listed in the method. Base sample batch acceptance on results of reagent blank additions rather than laboratory-fortified sample duplicates.

4. Reference

1. U.S. ENVIRONMENTAL PROTECTION AGENCY. 1995. Definition and procedure for the determination of the method detection limit, revision 1.11. 40 CFR Part 136, Appendix B. *Federal Register* 51:23703.

6040 CONSTITUENT CONCENTRATION BY GAS EXTRACTION*

6040 A. Introduction

The ability to analyze ultratrace levels of organic pollutants in water has been limited, in part, by the concentration technique. With the development of closed-loop stripping analysis (CLSA) (Method B), organic compounds of intermediate volatility and molecular weight, i.e., from the heavier volatiles to the lighter polynuclear aromatic hydrocarbons, can be extracted from water and concentrated to allow quantitative and semiquantitative analysis (depending on the compound) at nanograms-per-liter levels. This extract can be analyzed on a gas chromatograph (GC) connected to one of several detectors. A CLSA technique coupled with gas chromatographic/mass spectrometric (GC/MS) analysis for the determination of trace organic compounds is presented here. It is applicable to both treated and natural waters.

The purge and trap technique (Method C) is a valuable concentration method applicable to volatile organic compounds. The compounds are concentrated by bubbling an inert gas through the sample followed by collection in, and desorption from, a sorbent trap. This extract may be analyzed by GC or GC/MS methods. The technique is applicable to both water and wastewater.

The solid-phase microextraction method (D) for taste- and odor-causing compounds is more rapid than Method B and uses much smaller sample sizes, while achieving similar sensitivity. However, it has not been validated for as many compounds, or in as many matrices, as Method B.

* Approved by Standard Methods Committee, 1999.
Joint Task Group: (6040D)–Bart Koch (chair), Thomas S. Gittelman, Bruce A. Hale, John C. Nanci, Dat T. Nguyen, Francois Rodigari, Robert Shirey, Mark R. Simpson, Lely Suhady, Connie C. Young; 20th Edition—Bart Koch (chair), Tom E. Collins, Thomas S. Gittelman, Bruce A. Hale, Wolfgang Korth, Francois Rodigari, Mark R. Simpson.

6040 B. Closed-Loop Stripping, Gas Chromatographic/Mass Spectrometric Analysis

1. General Discussion

a. Principle: This CLSA-GC/MS procedure is suitable for the analysis of a broad spectrum of organic compounds in water. It can be used for the identification and quantitation of specific compounds, such as earthy-musty-smelling compounds [e.g., 2-methylisoborneol (MIB) and geosmin][1–3] or U.S. Environmental Protection Agency (EPA) priority pollutants.[4,5] The method is suitable for other taste- and odor-causing compounds (e.g., geranyl acetone 6-methylhept-5-en-2-one, β-ionone, and β-cyclocitral).[6]

TABLE 6040:I. METHOD DETECTION LEVELS FOR EARTHY-MUSTY SMELLING COMPOUNDS BY CLSA-GC/MS

Compound	Detection Level *ng/L** Unsalted Method[1]	Salting-Out Technique[3]
Geosmin	2	0.8
2-Methylisoborneol	2	0.8
2-Isopropyl-3-methoxy pyrazine	2	0.8
2-Isobutyl-3-methoxy pyrazine	2	0.8
2,3,6-Trichloroanisole	5	0.8

* Stripping at 25°C. Selective ion monitoring.

In closed-loop stripping, volatile organic compounds of intermediate molecular weight are stripped from water by a recirculating stream of air. The organics are removed from the gas phase by an activated carbon filter. They are extracted from the filter with carbon disulfide (CS_2) or methylene chloride. A portion of the extract is injected into a capillary-column GC/MS for identification of the organic compounds by retention time and spectrum matching; quantification is done by single-ion current integration. Alternatively, analysis may be made by capillary-column GC equipped with a flame ionization detector with identification by retention times on primary and secondary capillary columns.

b. Interference: Organic compounds that are stripped during this procedure may coelute with the compounds of interest. The uniqueness of the mass spectrum of each target compound makes it possible to confirm compound identity with a high probability when coeluting components are present. Problems may arise if several isomers of a compound are present that are not resolvable chromatographically.

c. Detection levels: Trace organics can be detected at low nanogram-per-liter levels. The CLSA-GC/MS detection levels are affected by many factors; especially important are the stripping efficiency and the condition of the GC/MS. Stripping efficiencies can be improved by using an elevated stripping temperature and/or the salting-out technique. The stripping and extraction portion of the method can be evaluated independently of the instrumentation portion. As an option, add internal standard after stripping and extraction, and transfer extract quantitatively.

The method detection levels for five earthy-musty-smelling compounds are shown in Table 6040:I. Detection levels for the salted CLSA method are less than half those for the unsalted method for each compound. Using the elevated stripping temperature rather than the salting-out technique produces comparable recoveries[7] and similar detection levels. Detection levels for various organic compounds of interest, obtained with an elevated stripping temperature/salting-out technique, ranged from 0.1 to 100 ng/L (see Table 6040:II).[8]

TABLE 6040:II. METHOD DETECTION LEVELS FOR SELECTED ORGANIC COMPOUNDS BY CLSA-GC/MS[8]

Compound	Detection Level *ng/L*†*	Compound	Detection Level *ng/L*†*
1,1,1-Trichloroethane	2.0	1,3,5-Trichlorobenzene	0.1
Trichloroethene	100	1,2,4-Trichlorobenzene	10
Dichlorobromomethane	5.0	1,2,3-Trichlorobenzene	2.0
1,3-Dichloropropene	2.0	*bis*(2-Chloro-ethoxy)methane	10
1,1,2-Trichloroethane	2.0	Methylisoborneol (MIB)	0.5
Chlorodibromomethane	1.0	Geosmin	0.2
1,2-Dibromoethane	2.0	Naphthalene	100
Tetrachloroethene	100	1,1,2,3,4,4-Hexachloro-1,3-butadiene	2.0
Chlorobenzene	10	1-Chloronaphthalene	0.5
Ethylbenzene	50	2-Chloronaphthalene	0.5
m,p-Xylene	100	Acenaphthene	0.5
Bromoform	1.0	Fluorene	2.0
Ethylbenzene	5.0	Diethylphthalate	100
o-Xylene	50	1-Chloro-4-phenoxybenzene	0.5
1,1,2,2-Tetrachloroethane	50	*N*-Phenylbenzamine	20
Bromobenzene	0.5	1,2-Diphenylhydrazine (as azobenzene)	1.0
Propylbenzene	0.5	1-Bromo-4-phenoxybenzene	0.5
1-Chloro-3-methylbenzene	0.5	Hexachlorobenzene	1.0
bis (2-Chloroethyl)ether	1.0	Phenanthrene	10
o-Dichlorobenzene	0.1	Anthracene	50
m-Dichlorobenzene	10	Fluoranthene	20
p-Dichlorobenzene	10	Pyrene	20
Hexachloroethane	20	Chrysene	50
N-Nitrosodi-*n*-propylamine	5.0	Benzo(a)anthracene	50

* Elevated stripping temperature and salting-out both utilized.

† Instrument detection level based on a 2:1 signal:noise ratio (where a background interference existed, the target compound was required to be at least twice background.)

2. Apparatus

Use clean glassware in sample collection and calibration standard preparation. Wash with soapy water, rinse with tap water, with demineralized water, and finally with reagent-grade acetone. As an alternative to acetone rinse, bake glassware in an annealing oven for 1 h at 400°C. Air-dry and bake at 180°C for 6 to 12 h. Do not bake sample bottle caps or volumetric ware. After drying and baking, store inverted or cover mouths with aluminum foil to prevent accumulation of dust or other contaminants.

a. Sample bottles, 1-L capacity or larger, glass, with TFE-lined screw caps.

b. CLSA apparatus, equipped with the following components (Figure 6040:1) or their equivalents.*

1) *Stripping bottle,* with mark at 1-L level and stainless-steel quick-connect stems (Figure 6040:2) or unpolished spherical glass joints sealed with TFE-covered silicone rubber O-rings and secured with metal clamps.† Immediately after use, clean stripping bottle by rinsing twice with demineralized water and once with organic-free water. For particularly adherent impurities, clean with acetone and bake at 180°C for at least 2 h. Turbid samples may cause a film to deposit on the stripping bottle and frit and may require washing with acid detergent.

2) *Gas heater,* with aluminum heating cylinder and soldering iron (25 W) controlled by a variable transformer (Figure 6040:3). Alternatively, use a temperature-controlled heater block to maintain a fixed temperature at the filter that is 10 to 20°C above temperature of the thermostatic water bath.

3) *Filter holder,* stainless steel or glass.‡ If glass is used, also use an auxiliary heating device, e.g., an infrared light, to maintain proper filter temperature.

4) *Pump,* with stainless-steel bellows,§ providing air flow in the range of 1 to 1.5 L/min. When using the salting-out technique, periodically disassemble the pump and clean. Salt will leave deposits on the pump bellows. If there is a noticeable drop in pump performance, clean valve assembly with acetone or replace.

5) *Automatic timer* (optional), connected to pump.

6) *Circuit,* with stainless-steel parts: 1/8-in. (0.3-cm)-OD tubing, 4-in. (10.2-cm) × 1/4-in. (0.6-cm)-OD flexible tubing, tube fittings, and quick-connect bodies;‖ or glass joints described in ¶ 1) above. Glass sample lines can be used except where circuit enters and exits pump. Use TFE ferrules in making connections to glass and flexible metal tubing. Whenever sample carryover is observed, clean circuit and pump as follows: Connect fittings to the quick-connect bodies on both ends of the circuit to open system. Turn on pump and flush with approximately 100 mL each organic-free water, acetone, and methanol. After last rinse, dry with a heat gun with pump still running (or flush with nitrogen) until there is no residual methanol. (NOTE: Overheating quick-connect units can cause deterioration of internal O-rings.)

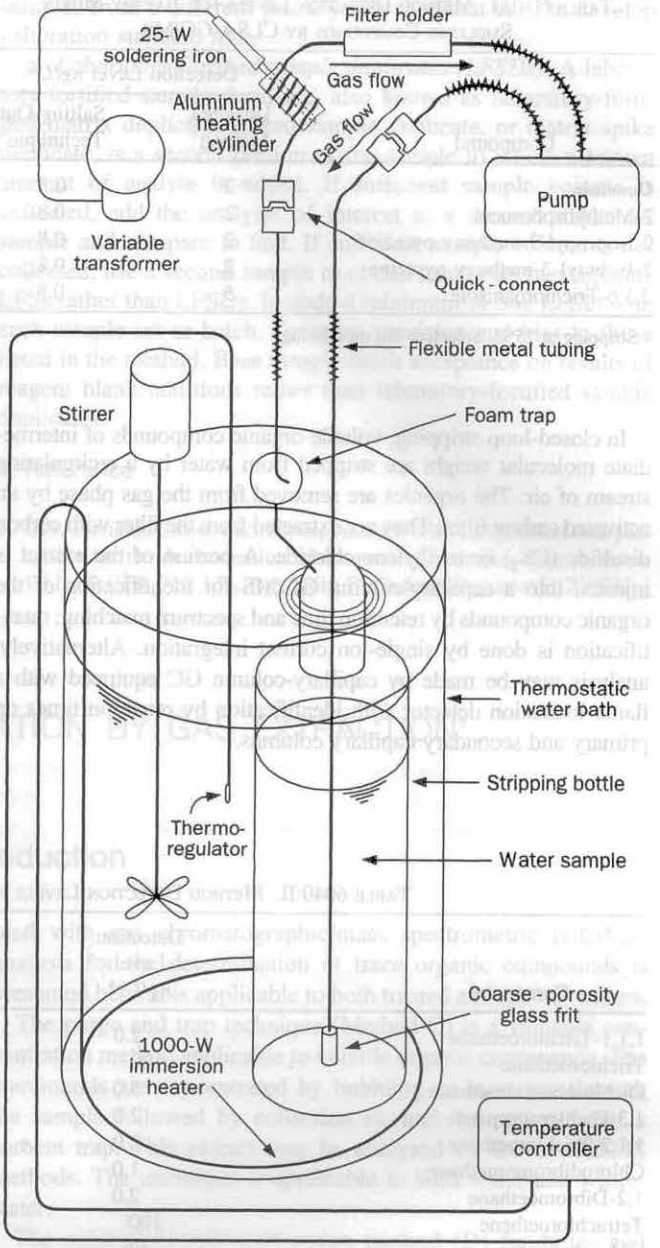

Figure 6040:1. Schematic of closed-loop stripping apparatus (not to scale). Source: KRASNER, S.W., C.J. HWANG & M.J. MCGUIRE. 1983. A standard method for quantification of earthy-musty odorants in water. *Water Sci. Technol.* 15(6/7):127.

7) *Thermostatic water bath,* with 222-mm-OD × 457-mm chromatography jar and thermoregulating system accurate to at least ±0.5°C. When the ambient temperature of the laboratory is greater than 25°C, maintain water bath at 25°C by inserting a coil of copper tubing connected to a cold water tap to recirculate cold water.

8) *Filters,* with 1.5 mg activated carbon# (Figure 6040:4). Use a set of filters matched in solvent flow resistance and

* Model CLS 1, Tekmar, Cincinnati, OH; Brechbühler AG, 8952 Schlieren ZH, Switzerland, available from Chromapon, Whittier, CA; or equivalent.
† Rotulex Sovirel, Brechbühler AG or equivalent.
‡ Brechbühler AG or equivalent.
§ Metal Bellows Model MB-21, Sharon, MA, or equivalent.
‖ Swagelok fittings or equivalent.

Brechbühler AG, Chromapon, Inc., or equivalent.

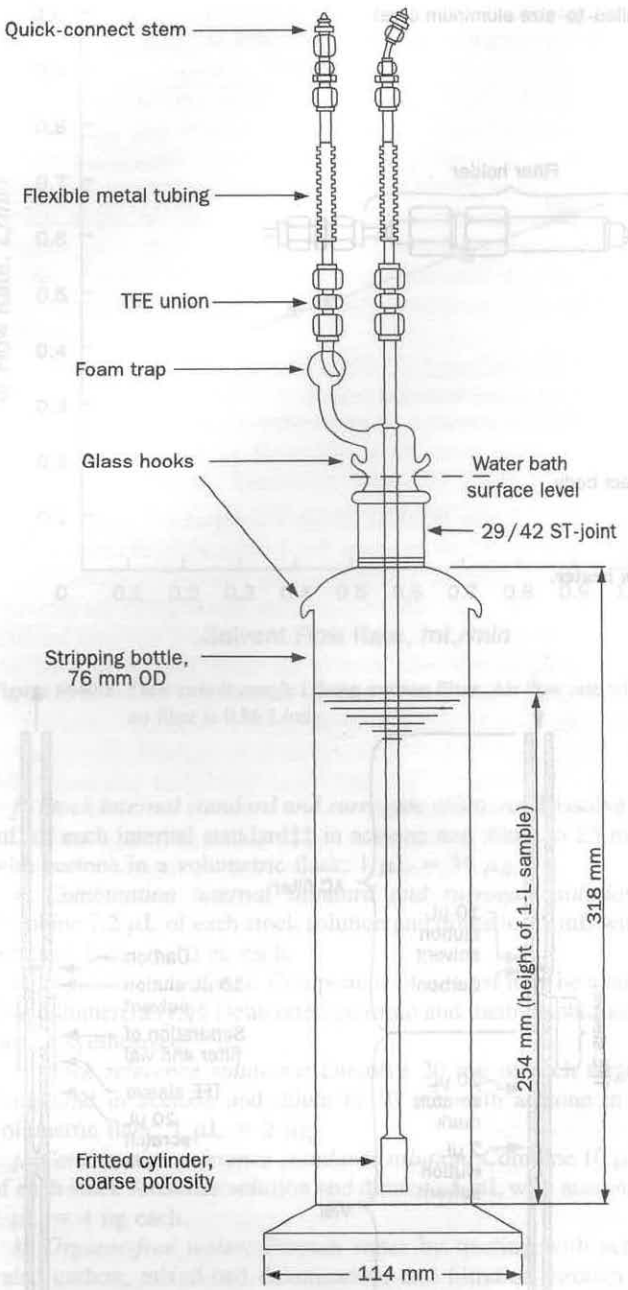

Figure 6040:2. One-liter "tall form" stripping bottle. Source: KRASNER, S.W., C.J. HWANG & M.J. MCGUIRE. 1983. A standard method for quantification of earthy-musty odorants in water. *Water Sci. Technol.* 15(6/7):127. (To obtain dimensions in inches, divide dimensions in millimeters by 25.4.)

performance for each group of samples and calibration standards. Determine filter resistance by measuring solvent flow rate through a cleaned, solvent-wetted filter. First fill the longer glass tube above the charcoal with organic-free water and let flow by gravity through filter disk. Rinse once with acetone, rinse twice with elution solvent, rinse twice with acetone, but on the final rinse, measure time necessary to empty the solvent (0.3-mL volume) from top of filter tube to surface of carbon. Rates for new, commercially prepared filters vary significantly, and decrease with use. Flow rates also depend on the solvent used. Determine optimal flow rate range from analyte recoveries. Preferably, verify filter performance by preparing check standards. Figure 6040:5 shows the reduction in air flow caused by using a "slow" filter. Figure 6040:6 shows the effect of filter resistance on recovery of earthy-musty odorants and one of the internal standards. Clean filter as soon as possible after use. Fill glass tube with organic-free water and let flow through filter. Repeat once with acetone, twice with elution solvent, and twice with acetone. Measure flow on final acetone rinse. If solvent flow is slow because of salt deposits, pull 1*N* HNO_3 through filter, using a vacuum connection. After acid washing, rinse with distilled water and acetone and continue with cleaning as above. After final rinse, remove residual solvent by connecting filter to a vacuum for approximately 5 min. Clean auxiliary filter after 40 uses or 2 weeks, whichever comes first.

If the salting-out technique is used, Na_2SO_4 may be carried over and ultimately may clog the filter. The initial water rinse is necessary to remove deposited salts and may be avoided if salt is not used.

c. Stirrer (optional), with 5-cm-long TFE stirring bar.

d. Microsyringes, 5-, 10-, 25-, and 50-μL capacity. Use a 25-μL gastight syringe with electrotapered (blunt-end) tip** for transferring extract.

e. Receivers, 50-μL capacity (Figure 6040:4). Receivers can be produced by a custom glass-blowing company: use a 1.6-mm-ID precision-bore capillary glass and grind to 5 mm OD, then heat-constrict to close off bore at approximately 29 mm from top. Mark at 20-μL level with glass scribe. To clean receivers and extract storage vials (¶ *g*, below), rinse seven times with elution solvent and bake at 180°C overnight or rinse with acetone and bake in annealing oven at 400°C for 1 h. Rinse receivers several times with elution solvent before using.

f. TFE sleeve, 5-mm-ID TFE flexible tubing approximately 19 mm long. If a 5-mm-OD receiver is not prepared as described above, then connect filter and receiver with a piece of heat-shrink TFE tubing that is custom-shrunk to the dimensions of the filter and receiver. Rinse sleeve with acetone after each use and store in acetone until ready to use.

g. Extract storage vials, 100-μL capacity conical-shaped vials with TFE-lined septum screw cap or crimp cap.†† Transfer sample extracts from receiver to storage vial for extract storage. The storage vials are compatible with various autosamplers.

h. Gas chromatograph (GC)/mass spectrometer (MS)/data system, equipped with:

1) *Capillary injector,* Grob-designed split-splitless injector or equivalent with 2.5-mm-ID glass insert or nonvaporizing, septumless, cold on-column injector.

2) *Capillary column,* 30-m or 60-m × 0.25-mm-ID DB-1 or DB-5 fused silica or other capillary column capable of producing adequate and reproducible resolution. If using a Grob on-column injector, use a 0.32-mm-ID column for injection when a stainless-steel needle is used, or use a 0.25-mm-ID column with a fused-silica needle.

** Hamilton Model 1805N electrotapered tip, Reno, NV.
†† Pierce Chemical Company #13100 or equivalent.

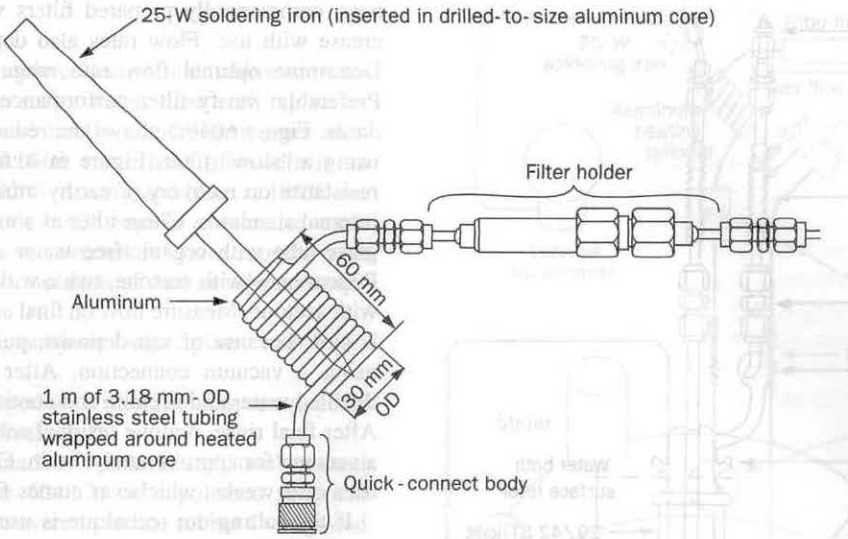

Figure 6040:3. Gas heater.

For other injector types, a 1-m precolumn of uncoated, deactivated 0.53-mm-ID fused silica is recommended. Connect precolumn to analytical column with a zero-dead-volume union.

3) *Microsyringes,* 5- and 10-µL capacity, with 75-mm-long needles. Use 0.23-mm-OD stainless-steel or 0.17-mm-OD fused-silica needle for on-column injection.

4) *Mass spectrometer analyzer:* See Section 6200B.2 for suggested specifications.

5) *Data system,* with software capable of performing reverse-library searches (optional).

6) *Autosampler injector* (recommended for improved precision of analysis). When methylene chloride is the elution solvent, manual injection is recommended.

3. Reagents

Use reagent-grade solvents or better and obtain purest standards available.

a. Carbon disulfide, CS_2: Use only after gas chromatographic verification of purity to ensure that the solvent does not contain components that coelute with the compounds of interest. CAUTION: *Use proper safety procedures; explosive, toxic, and occasionally allergenic.*

b. Acetone.

c. Methylene chloride, high-resolution grade. Preferably use for analyses conducted at high altitudes. MIB has been shown to be unstable in methylene chloride; extract with CS_2 whenever possible.[9]

d. Carrier gas: Helium gas, ultrapurified grade, moisture- and oxygen-free.

e. Internal standards and surrogates:

1) *1-Chlorooctane.*

2) *1-Chlorodecane.*

3) *1-Chlorododecane.*

4) *1-Chlorohexane, 1-chlorohexadecane,* and *1-chlorooctadecane* can be added for broad-spectrum analysis.

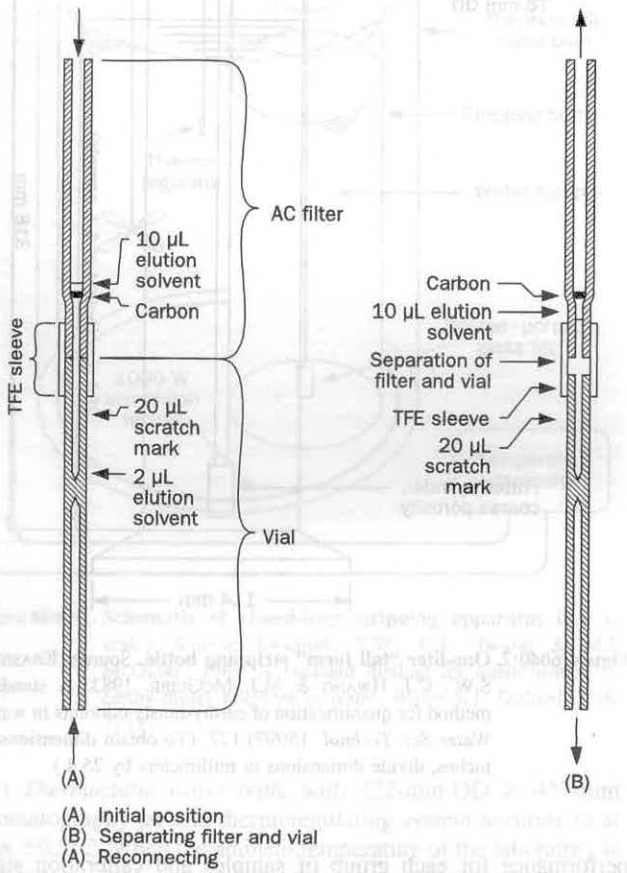

Figure 6040:4. Extraction of filter. Source: KRASNER, S.W., C.J. HWANG & M.J. MCGUIRE. 1981. Development of a closed-loop stripping technique for the analysis of taste- and odor-causing substances in drinking water. *In* L.H. Keith, ed. Advances in the Identification and Analysis of Organic Pollutants in Water, Vol. 2. Ann Arbor Science Publishers, Ann Arbor, Mich.

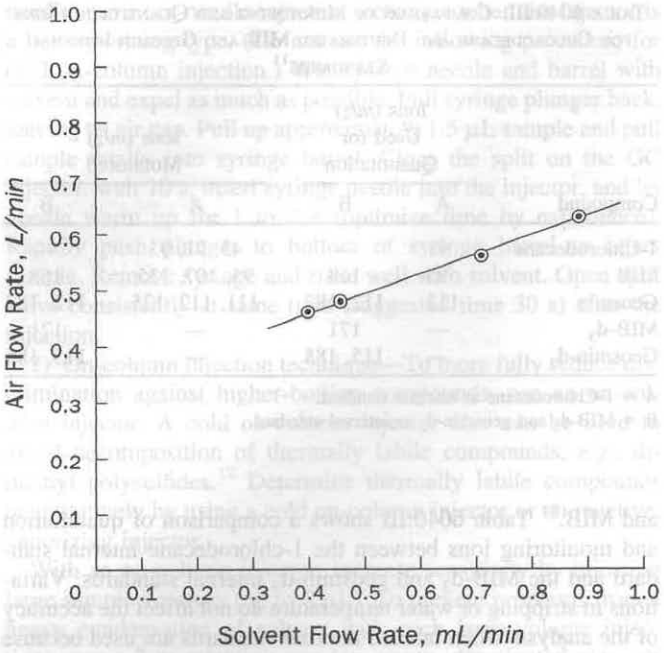

Figure 6040:5. Flow rate through 1.5-mg carbon filter. Air flow rate with no filter is 0.86 L/min.

f. Stock internal standard and surrogate solutions: Dissolve 1 mL of each internal standard‡‡ in acetone and dilute to 25 mL with acetone in a volumetric flask; 1 μL = 35 μg.

g. Combination internal standard and surrogate solution: Combine 7.2 μL of each stock solution and dilute to 25 mL with acetone; 1 μL = 10 ng each.

h. Reference standards: Compounds of interest may be available commercially.§§ Deuterated geosmin and methylisoborneol can be synthesized.[10]

i. Stock reference solutions: Dissolve 20 mg of each target compound in acetone and dilute to 10 mL with acetone in a volumetric flask; 1 μL = 2 μg.

j. Combination reference standards solution: Combine 10 μL of each stock reference solution and dilute to 5 mL with acetone; 1 μL = 4 ng each.

k. Organic-free water: Prepare water by treating with activated carbon, mixed-bed deionization, and filtration through a membrane filter.‖ ‖ Irradiate under ultraviolet light (185 and 254 nm) for 1 h (optional) and prestrip in the CLSA apparatus for 1 h (optional if laboratory blanks are consistently free of interferences), using a clean activated carbon filter or, alternatively, prestrip large quantities of water with nitrogen (ultra-high-purity grade) just before use. Store in a closed bottle tightly capped with TFE-lined screw cap, under nitrogen (optional), in a refrigerator and away from solvent contamination for not longer than 1 week.

l. Sodium sulfate, Na_2SO_4 (optional), granular, anhydrous. Bake at 400°C for 2 h before use; store at room temperature in desiccator.

‡‡ 1-Chlorohexadecane and 1-chlorooctadecane solidify upon refrigeration. Warm before removing a portion.

§§ Geosmin and 2-methylisoborneol are available from Wako Bioproducts, Wako Chemicals USA, Inc., 1600 Bellwood Rd., Richmond, VA 23237, or Dalton Chemical Lab, Inc., 4700 Keele St., Suite 119, FARQ, North York, Ontario, Canada M3J 1P3. NOTE: This synthetic geosmin is racemic and includes (+)-geosmin, which has an odor intensity different from that of the natural (−) compound. This precludes its use in quantitative sensory analysis; however, its GC/MS characteristics (i.e., retention time and spectrum) are the same as those of natural geosmin.

‖ ‖ Millipore Milli-QUV Plus or equivalent.

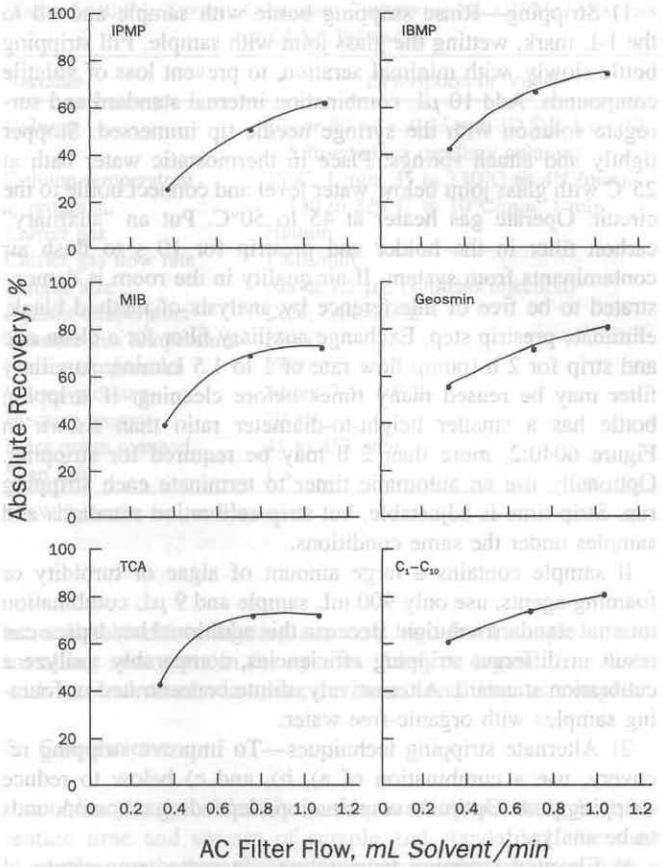

Figure 6040:6. Effect of filter resistance, measured as flow, on recovery of earthy-musty odorants and C_1–C_{10} internal standard. Reprinted with permission from: HWANG, C.J., S.W. KRASNER, M.J. MCGUIRE, M.S. MOYLAN & M.S. DALE. 1984. Determination of subnanogram per liter levels of earthy-musty odorants in water by the salted closed-loop stripping method. *Environ. Sci. Technol.* 18:535. Copyright 1984, American Chemical Society.

4. Procedure

a. Sample collection and storage: Rinse sample bottle with sample, fill to overflowing (with no air bubbles), and cap tightly. Collect duplicate samples and in the field keep in an insulated container stocked with ice. In the laboratory store at 4°C, but analyze as soon as possible, preferably within 3 d. For holding longer than 3 d, add 40 mg $HgCl_2$/L to inhibit biological activity. Adding a dechlorinating agent is optional, because disinfection by-products may be affected. CAUTION: *$HgCl_2$-containing samples must be disposed of as hazardous waste. See Section 1090 for precautions.*

b. Treatment of samples:

1) Stripping—Rinse stripping bottle with sample and fill to the 1-L mark, wetting the glass joint with sample. Fill stripping bottle slowly, with minimal aeration, to prevent loss of volatile compounds. Add 10 μL combination internal standard and surrogate solution with the syringe needle tip immersed. Stopper tightly and attach springs. Place in thermostatic water bath at 25°C with glass joint below water level and connect bottle to the circuit. Operate gas heater at 45 to 50°C. Put an "auxiliary" carbon filter in the holder and prestrip for 10 s to flush air contaminants from system. If air quality in the room is demonstrated to be free of interference by analysis of method blank, eliminate prestrip step. Exchange auxiliary filter for a clean one and strip for 2 h (pump flow rate of 1 to 1.5 L/min). Auxiliary filter may be reused many times before cleaning. If stripping bottle has a smaller height-to-diameter ratio than shown in Figure 6040:2, more than 2 h may be required for stripping. Optionally use an automatic timer to terminate each stripping run. Strip time is adjustable, but strip calibration standards and samples under the same conditions.

If sample contains a large amount of algae or turbidity or foaming agents, use only 900 mL sample and 9 μL combination internal standard solution. Because this additional headspace can result in different stripping efficiencies, comparably analyze a calibration standard. Alternatively, dilute concentrated or foaming samples with organic-free water.

2) Alternate stripping techniques—To improve stripping recovery, use a combination of a), b), and c) below to reduce stripping time. Optimize combination, depending on compounds to be analyzed.

a) Elevated stripping temperature—Increase temperature of thermostatic water bath to 45°C to increase recovery of many organic compounds.[5,7] Raise temperature of gas heater to at least 55°C (for a 45°C stripping temperature) to avoid condensation of water vapor on the activated carbon filter. Further increases in stripping temperature reduce recovery.[5]

b) Salting-out technique—Raising the sample ionic strength with Na_2SO_4 before stripping increases the stripping rate of many organic compounds.[3] Bring sample to room temperature before analysis by immersing it in a water bath at 25°C for approximately 15 min. Transfer 800 mL to the stripping bottle and add stirring bar. Using a glass funnel and with the stirring bar (at intermediate setting) in motion, add 72 g Na_2SO_4. Remove funnel and replace with a glass stopper. Continue stirring until Na_2SO_4 has dissolved (not more than 1 min). Remove stopper and stirring bar, then add remaining 100 mL of sample,## rinsing and wetting inside neck of bottle. Add 9 μL combination internal standard solution and strip at 25°C as described in standard stripping procedure above. If analysis is only for MIB and geosmin, a 1-h strip time is adequate. If additional target compounds are being analyzed, verify strip time needed for adequate recoveries of each target compound. Alternatively, combine salt and sample by pouring salt directly into 900 mL sample, stopper tightly, shake vigorously, let stand for several minutes, and add internal standards.

c) Alternative analysis—Use deuterium-labeled geosmin and MIB as internal standards for the determination of geosmin and MIB.[11] Table 6040:III shows a comparison of quantitation and monitoring ions between the 1-chlorodecane internal standard and the MIB-d_3 and geosmin-d_3 internal standards. Variations in stripping or water temperature do not affect the accuracy of the analysis when labeled internal standards are used because MIB and geosmin will strip at the same rate as MIB-d_3 and geosmin-d_3.[11] In addition, labeled standards, if added in the field, will document degradation of the target compound within a 3- to 4-week period. The labeled standards compensate for losses of analyte by physical, chemical, and biological processes during*** sample storage.[11]

TABLE 6040:III. COMPARISON OF MONITORING AND QUANTITATION IONS FOR CHLORODECANE AND DEUTERATED MIB AND GEOSMIN INTERNAL STANDARDS[11]

	Ions (*m/z*) Used for Quantitation		Ions (*m/z*) Monitored	
Compound	A	B	A	B
1-Chlorodecane	91	—	43, 91, 93	—
MIB	95	168	93, 107, 135	168
Geosmin	112	112, 182	111, 112, 125	112, 182
MIB-d_3	—	171	—	171
Geosmin-d_3	—	115, 185	—	115, 185

A = 1-Chlorodecane as internal standard.
B = MIB-d_3 and geosmin-d_3 as internal standard.

3) Extracting the filter—Remove activated carbon filter from holder. In a fume hood, extract with CS_2 as indicated in Figure 6040:4. Keep solvents well within the hood to avoid inhalation by analyst or contamination of stripping apparatus. Add 2 μL elution solvent to a clean receiver and connect filter and receiver with a TFE sleeve so as to leave no dead space between glass parts. Place 10 μL elution solvent above the carbon, taking care not to touch carbon with the needle. Warm receiver with hands and alternately pass elution solvent across carbon 10 times. Cool receiver with ice, taking care not to freeze the elution solvent, in order to draw the elution solvent below the carbon. Tap the filter/vial assembly gently on a hard surface to complete transfer of elution solvent to bottom of vial.

Repeat filter extraction with a 10-μL and a 5-μL portion of elution solvent. Separate vial from filter, adjust volume precisely to the 20-μL mark,††† and, using a gastight electrotapered tip syringe, transfer extract to a clean, conical-shaped storage vial. Label and store at −20°C until analysis.

Filter may be extracted while maintaining tight seal between filter and vial during the procedure. Using ice chips, cool closed volume in the vial; solvent accumulates on lower side of the filter disk. Push solvent back to upper side by warming the closed vial between two fingers. Repeat and then extract with more solvent as above.

c. Gas chromatography/mass spectrometry:

1) "Hot-needle" injection technique—To reduce discrimination against higher-boiling compounds by distillation from the

A total sample volume of 900 mL is preferred to minimize foaming-over due to salt addition.

*** NOTE: Because synthetic labeled geosmin is racemic, best results for the compensation of prolonged biological processes are achieved by monitoring the degradation of natural (−)-geosmin using (−)-geosmin-d_3.

††† Calibration mark needs to be verified periodically.

needle, use a hot-needle injection technique when the injector is a hot vaporizing type. (Do not use the following procedure for cold on-column injection.) Wet syringe needle and barrel with solvent and expel as much as possible. Pull syringe plunger back, leaving an air gap. Pull up approximately 1.5 μL sample and pull sample totally into syringe barrel. Close the split on the GC injector, wait 10 s, insert syringe needle into the injector, and let needle warm up for 1 to 2 s (optimize time by experience). Rapidly push plunger to bottom of syringe barrel to inject sample. Remove syringe and rinse well with solvent. Open split valve consistently at same time (suggested time 30 s) after the injection.

2) On-column injection technique—To more fully reduce discrimination against higher-boiling compounds, use an on-column injector. A cold on-column injector also can be used to avoid decomposition of thermally labile compounds, e.g., dimethyl polysulfides.[12] Determine thermally labile compounds quantitatively by using a cold on-column injector or an inactive, vaporizing injector.

With an on-column injector, increase sensitivity by injecting large sample volumes (up to 8 μL). To prevent problems from a heavy condensation of solvent with such large-volume injections, use a 2-m retention gap (an empty, deactivated piece of 0.53-mm ID fused silica tubing connected to the head of the column with a zero-dead-volume connector).[13] To preclude backpressure from large-volume injections, inject slowly at about 1 μL/5 s. Keep initial column temperature at 10°C above boiling point of solvent for a full solvent effect and to produce sharp peaks (narrow peak widths).[13] Because the entire injection is deposited directly into the head of the column, the column can develop active sites after as few as 50 to 80 injections. Check activity by injecting a polarity test mixture at least weekly. Breaking approximately 30 cm off the head of the column can restore inertness.

3) Operating conditions for GC/MS—After initial installation of the capillary column, condition it according to the manufacturer's instruction. Daily, make a conditioning run with a CS_2 injection or method blank extraction injection before injecting any samples (optional). Typical instrument conditions are given in Table 6040:IV.

d. Calibration standard: The method is semiquantitative for a large number of compounds, but has been shown to be quantitative for many of the compounds listed in this section. Prepare a 20-ng/L target-compound calibration standard by dosing 1 L organic-free water in the stripping bottle with 10 μL combination internal standard solution plus 5 μL combination reference standards solution. (Internal standards concentration is 100 ng/L each.) If the salting-out technique is used, add 72 g Na_2SO_4 to a total volume of 900 mL organic-free water before dosing with 9 μL combination internal standard solution plus 4.5 μL combination reference standards solution. Analyze as directed above. Inject the calibration standard extract, preferably daily, to determine GC/MS response factors and verify spectra.

Verify working linear range by analyzing standards and representative samples with added organics at different concentrations. Calibrate at least every two weeks. Use calibration levels that bracket the levels found in samples. At minimum, use a three-level calibration curve.

e. Blanks: Run a procedural blank daily to assess contamination from reagents, apparatus, and other sources. Run a blank immediately after analyzing any very high-level sample or after installing new parts in the system. Analyze organic-free water with internal standards under the same conditions as samples.

Table 6040:IV. Typical Operating Conditions for GC/MS Analysis of CLSA Extracts

Variable	Description or Value
Column	30- or 60-m × 0.25-mm-ID DB-1 or DB-5 fused silica capillary column*
Column temperature program	35°C, 1 min; 35 to 130°C @ 4°C/min; 130 to 220°C @ 10°C/min; 1 min
Carrier gas	Helium
Carrier gas flow rate	1 mL/min
Sample size	About 1.5 μL (splitless injection)
Injector temperature	Cold, on-column
Transfer line temperature	280°C
Ionizer temperature	280°C
Source pressure	About 7×10^4 Pa
Electron energy	70 eV
Mass range scanned	41 to 453 amu
Scan time	1 s

*J&W Scientific, Inc., or equivalent.

5. Calculations

a. Identification: Identify a compound by matching both retention time and spectra of sample and standard. If they are available, use both a reverse-search computer program with a target-compound library and a forward-search program with the National Institute of Standards and Technology library for tentative identification of other compounds present.

1) Retention times—Use each internal standard to calculate relative retention times for all compounds in the same part of the chromatogram (Table 6040:V). For compounds eluting on the solvent tail, use an early-eluting internal standard (e.g., 1-chlorohexane). Sample retention times should match predicted retention times within ±15 s.

$$\text{Predicted } T_{z,x} = \frac{T_{z,s}}{T_{I,s}} \times T_{I,x}$$

where:

$T_{z,x}$ = retention time of target compound in sample analysis,
$T_{z,s}$ = retention time of target compound in calibration standard analysis,
$T_{I,s}$ = retention time of internal standard in calibration standard analysis, and
$T_{I,x}$ = retention time of internal standard in sample analysis.

2) Spectra—Peaks of at least three characteristic ions should all maximize at the same retention time and have standard intensity ratios (spectra) within ±20% of those of the calibration-standard compounds. Characteristic ions and their typical relative intensities for three of the internal standards and two earthy-musty-smelling compounds are given in Table 6040:V. Preferably, use reference spectra of 10 to 14 key masses. Determine reference spectra by analysis of standards; verify these frequently. The spectra of MIB are particularly dependent on

TABLE 6040:V. GC/MS DATA FOR THREE INTERNAL STANDARDS AND TWO EARTHY-MUSTY SMELLING COMPOUNDS

Compound	Retention Time* *min*	Quantification Mass *amu*	Characteristic Ions (with relative intensities)
1-Chlorooctane	30.8	91	43 (100), 91 (86), 93 (27)
2-Methylisoborneol	36.4	95, 107†	95 (100), 107 (26), 135 (9)
1-Chlorodecane	39.8	91	43 (100), 91 (87), 93 (28)
Geosmin	45.1	112	112 (100), 111 (28), 125 (18)
1-Chlorododecane	47.2	91	43 (100), 91 (61), 93 (19), 85 (12)

*See Table 6040:IV for GC conditions. Data accumulated using 30-m DB-5 capillary column.
†Quantify using two different masses and obtain an average value.

instrument condition; both 107 and 95 amu have been reported as base peaks (Figure 6040:7). Figure 6040:8 shows the mass spectrum for geosmin.

b. Quantitation: Determine concentrations by comparison of peak areas of specific quantitation ions. A quantitation ion should be relatively intense in the mass spectrum, yet be free from interference problems caused by closely eluting compounds (see Table 6040:V). Calculate a response factor for each compound from CLSA of a calibration standard as follows:

$$R_z = \frac{A_z \times C_I}{C_z \times A_I}$$

where:

R_z = response factor for target compound z,
A_z = peak area of target compound z,
A_I = peak area of internal standard,
C_I = concentration of internal standard, and
C_z = concentration of target compound z.

Alternatively, a calibration fit can be used.

Compound concentration in the sample (x) is:

$$C_{z,x},\ \text{ng/L} = \frac{A_{z,x} \times C_{I,x}}{R_z \times A_{I,x}}$$

where:

$C_{z,x}$ = concentration of target compound in sample,
$A_{z,x}$ = peak area of target compound in sample,
$C_{I,x}$ = concentration of internal standard in sample, and
$A_{I,x}$ = peak area of internal standard in sample.

Use the internal standard 1-chlorodecane for determining response factors. Use other internal standards as a check on the system; calculated values should be to ±20%. Computerized reverse-search spectral matching and automatic quantitation are recommended to improve identification in complex matrices and to facilitate data processing.

Where calibration standards are unavailable, estimate concentrations by comparing the total ion current of the compounds to that of the internal standard 1-chlorodecane.

6. Quality Assurance/Quality Control

The CLSA method is semiquantitative for some compounds because of the variability of stripping efficiencies. However,

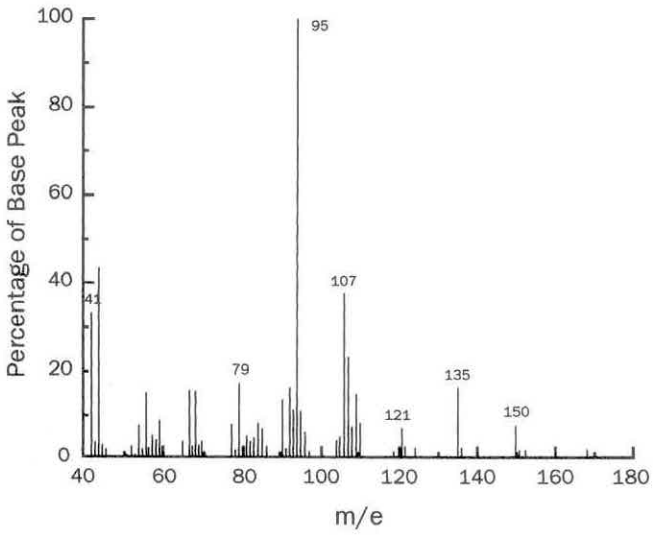

Figure 6040:7. Mass spectrum of 2-methylisoborneol.

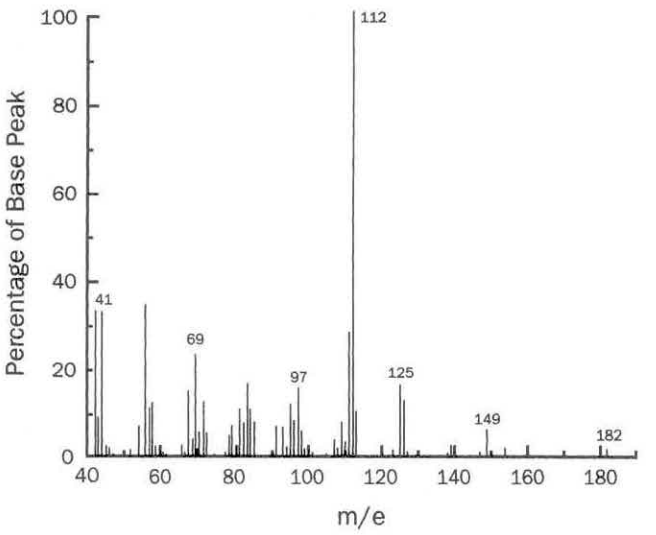

Figure 6040:8. Mass spectrum of geosmin.

TABLE 6040:VI. SINGLE-LABORATORY BIAS FOR SELECTED ORGANIC COMPOUNDS CAUSING TASTE AND ODOR

Stripping Technique Compound	Dose Level *ng/L*	Number of Samples*	Mean Recovery† %	Standard Deviation %
Unsalted method‡				
2-Isopropyl-3-methoxy pyrazine	4–20	23	89	21
2-Isobutyl-3-methoxy pyrazine	4–20	22	101	28
2-Methylisoborneol	4–120	30	101	21
2,3,6-Trichloroanisole	4–20	22	88	27
Geosmin	4–120	28	109	20
Salting-out method‡				
2-Isopropyl-3-methoxy pyrazine	4–20	44	120	24
2-Isobutyl-3-methoxy pyrazine	4–20	48	106	18
2-Methylisoborneol	4–20	48	106	15
2,3,6-Trichloroanisole	4–20	45	99	22
Geosmin	4–20	48	105	15

*Finished and natural surface waters.
†Standard-adjusted recovery.
‡Stripping at 25°C.
Reprinted with permission from *Environmental Science and Technology*, 18:535.[3] Copyright 1984. American Chemical Society.

quantitative data are obtainable for compounds that are reproducibly stripped (e.g., MIB and geosmin).[1–3,7] Follow general quality assurance/quality control requirements (e.g., calibration, initial quality control, and batch quality control) described in Section 6020.

Analyze a replicate sample at least once per 10 samples to check precision. If unusual or unexpected results are obtained, analyze a replicate to confirm. Typically, single-analyst determinations for a relatively simple matrix have a coefficient of variation less than or equal to 10%. Otherwise, precision is usually within 20%. For compounds that are poorly stripped, a higher coefficient of variation may be obtained.

Analyze a sample with a known addition at least once per 10 samples to check accuracy and recovery. If matrix problems exist, this will confirm the accuracy of results. Adjust these recoveries against the calibration standards results. Even when absolute recoveries are less than 50%, standard adjusted recoveries, which correct for stripping efficiencies, are usually between 80 and 120%.

Internal standard response should equal ± 40% of the daily standard. An unacceptable internal standard response requires extract reinjection. If the reinjection is still unacceptable, investigate cause, restrip sample, and reanalyze. If consecutive samples fail the internal standard acceptance criterion, immediately analyze a calibration standard. If the calibration standard internal standard response also is unacceptable, recalibrate the instrument.

Ideally prepare and analyze an intralaboratory check sample monthly. Prepare from an independent stock solution of the standards.

TABLE 6040:VII. PRECISION DATA FOR SELECTED ORGANIC COMPOUNDS CAUSING TASTE AND ODOR*

		Multiple Laboratories†			Single Laboratory‡		
Compound	Dose Level *ng/L*	Mean *ng/L*	Standard Deviation *ng/L*	Coefficient of Variation %	Mean *ng/L*	Standard Deviation *ng/L*	Coefficient of Variation %
Sample A§							
2-Isopropyl-3-methoxy pyrazine	5.9	5.6	1.6	28	6.6	0.6	9
2-Isobutyl-3-methoxy pyrazine	3.0	3.0	0.7	24	3.2	0.3	11
2-Methylisoborneol	4.3	4.8	1.0	20	5.1	0.1	2
2,3,6-Trichloroanisole	8.7	7.3	3.1	43	9.3	2.0	22
Geosmin	2.9	3.3	0.9	27	3.6	0.5	14
Sample B§							
2-Isopropyl-3-methoxy pyrazine	25	22	8.0	36	27	2.8	10
2-Isobutyl-3-methoxy pyrazine	15	14	4.2	30	17	2.5	14
2-Methylisoborneol	20	18	6.2	34	23	2.0	9
2,3,6-Trichloroanisole	35	32	9.7	30	37	3.3	9
Geosmin	16	16	5.9	37	20	2.8	14

*Stripping at 25°C, unsalted method.
†Five analysts at three laboratories.
‡Three analysts at one laboratory.
§Organic-free water dosed with taste and odor compounds.

TABLE 6040:VIII. RECOVERY AND PRECISION DATA FOR SELECTED PRIORITY POLLUTANTS*

Compound	Amount ng	Mean Recovered Amount† ng	Range	Recovery Efficiency %	RSD
Thiophene	25	9	7–11	35	16
Dibromochloromethane	29	17	13–21	57	13
Styrene	22	17	16–20	80	7
Isopropylbenzene	24	26	24–29	107	8
2-Chlorotoluene	26	23	22–27	90	8
bis(2-Chloroethyl)ether	24	3	3–4	12	11
α-Methylstyrene	22	19	18–22	90	6
1,4-Dichlorobenzene	20	18	16–21	93	8
2-Ethyl-1,3-dimethylbenzene	24	22	21–25	92	6
4-Chloro-*o*-xylene	26	22	20–26	85	9
1,1-Dimethylindan	22	24	22–26	110	7
p-Methylphenol	27	ND‡			
Tetrahydronaphthalene	23	23	20–26	99	10
1,2,4-Trichlorobenzene	23	19	18–21	83	7
Hexachloro-1,3-butadiene	27	31	28–34	114	9
2-Methylbiphenyl	24	25	22–27	101	8
1,6-Dimethylnaphthalene	24	10	8–11	39	12
2-Isopropylnaphthalene	23	21	19–22	91	6
Pentachlorobenzene	26	12	11–14	48	11
Hexachlorobenzene	20	6	5–7	31	13
2,2′,4,4′,6,6′-Hexachlorobiphenyl	27	28	26–32	104	7
2,2′,4,5,5′-Pentachlorobiphenyl	28	23	18–28	82	14

* Stripping at 40°C, unsalted method.
† Based on six purging analyses using single ion quantification.
‡ ND = not detected.
Reprinted with permission from *Environmental Science and Technology*, 17:571.[4] Copyright 1983. American Chemical Society.

7. Precision and Bias

Precision and bias data are given in Tables 6040:VI and VII for the analysis of earthy-musty-smelling compounds. Table 6040:VIII shows recovery and precision data for selected pollutants.

8. References

1. KRASNER, S.W., C.J. HWANG & M.J. MCGUIRE. 1981. Development of a closed-loop stripping technique for the analysis of taste- and odor-causing substances in drinking water. *In* L.H. Keith, ed. Advances in the Identification and Analysis of Organic Pollutants in Water, Vol. 2. Ann Arbor Science Publishers, Ann Arbor, Mich.
2. KRASNER, S.W., C.J. HWANG & M.J. MCGUIRE. 1983. A standard method for quantification of earthy-musty odorants in water. *Water Sci. Technol.* 15(6/7):127.
3. HWANG, C.J., S.W. KRASNER, M.J. MCGUIRE, M.S. MOYLAN & M.S. DALE. 1984. Determination of subnanogram per liter levels of earthy-musty odorants in water by the salted closed-loop stripping method. *Environ. Sci Technol.* 18:535.
4. COLEMAN, W.E., J.W. MUNCH, R.W. SLATER, R.G. MELTON & F.C. KOPFLER. 1983. Optimization of purging efficiency and quantification of organic contaminants from water using a 1-L closed-loop-stripping apparatus and computerized capillary column GC/MS. *Environ. Sci. Technol.* 17:571.
5. THOMASON, M.M. & W. BERTSCH. 1983. Evaluation of sampling methods for the determination of trace organics in water. *J. Chromatogr.* 279:383.
6. JONES, G.J. & W. KORTH. 1995. In situ production of volatile odor compounds by river and reservoir phytoplankton populations in Australia. *Water Sci. Technol.* 31(11):145.
7. MALLEVIALLE, J. & I.H. SUFFET, eds. 1987. Identification and Treatment of Tastes and Odors in Drinking Water. American Water Works Association & American Water Works Association Research Foundation, Denver, Colo.
8. Report of Analysis for Semivolatile Organics by Closed-Loop Stripping GC/MS. 1982. Environmental Research Lab., James M. Montgomery, Consulting Engineers, Inc., Pasadena, Calif.
9. KORTH, W., J. ELLIS & K. BOWMER. 1992. The stability of geosmin and MIB and their deuterated analogues in surface waters and organic solvents. *Water Sci. Technol.* 25(2):115.
10. KORTH, W., J. ELLIS & K. BOWMER. 1991. Synthesis of deuterium labelled geosmin and methylisoborneol. *J. Labelled Compounds Radiopharm.* 29:823.
11. KORTH, W., K. BOWMER & J. ELLIS. 1991. New standards for the determination of geosmin and methylisoborneol in water by gas chromatography/mass spectrometry. *Water Res.* 25:319.
12. WAJON, J.E., R. ALEXANDER & R.I. KAGI. 1985. Determination of trace levels of dimethyl polysulphides by capillary gas chromatography. *J. Chromatogr.* 319:187.
13. GROB, K. 1978. On-column injection onto capillary columns. Part 2: Study of sampling conditions; practical recommendations. *J. High Resolution Chromatogr. Chromatogr. Commun.* 1:263.

9. Bibliography

BUDDE, W.L. & J.W. EICHELBERGER. 1979. Organic Analysis Using Gas Chromatography- Mass Spectrometry. Ann Arbor Science Publishers, Ann Arbor, Mich.

GROB, K. & K. GROB, JR. 1978. Splitless injection and the solvent effect. *J. High Resolution Chromatogr. Chromatogr. Commun.* 1:57.

GROB, K. & G. GROB. 1979. Practical gas chromatography—a systematic approach. *J. High Resolution Chromatogr. Chromatogr. Commun.* 2:109.

REINHARD, M., J.E. SCHREINER, T. EVERHART & J. GRAYDON. 1987. Specific analysis of trace organics in water using gas chromatography and mass spectroscopy. *J. Environ. Pathol., Toxicol. Oncol.* 7:417.

6040 C. Purge and Trap Technique

For applications of this technique to analyses for volatile organics, volatile aromatic organics, and volatile halocarbons, see Section 6200.

6040 D. Solid-Phase Microextraction (SPME)

1. General Discussion

a. Principle: Solid-phase microextraction (SPME) of taste- and odor-causing compounds relies on the adsorption of target compounds on a SPME fiber, which consists of a fiber coated with divinylbenzene-carboxen-polydimethylsiloxane cross-link. The fiber is placed in the headspace above the sample in a closed vessel and allowed to equilibrate with the aqueous sample. After a predetermined equilibration time the fiber is removed and inserted directly into the injection port of a GC or GC/MS system. The port is heated and the analytes desorbed into the instrument. Preferably perform analysis using GC/MS in the selected ion monitoring mode, monitoring the multiple major masses (see Table 6040:II) for each of the target analytes, if possible, to ensure more accurate identification and quantitation. The method has been validated for at least three of the taste and odor compounds, methyl isoborneol (MIB), geosmin, and isopropyl methoxy pyrazine (IPMP), and may be applicable to others. SPME is much faster than closed-loop stripping and uses a much smaller sample size. However, it is critical that samples and standards be processed in *exactly* the same manner because extraction conditions determine the response factor.

b. Interference: Any organic compounds present in the sample may be adsorbed onto the fiber and desorb in the GC/MS. The mass spectrum serves to identify the compound of interest and eliminate possible interferences. For matrices containing intracellular geosmin or MIB, demonstrate recovery by using matrices with known additions of these compounds and preferably comparing to Method B.

c. Sampling and storage: Collect samples in either a single amber glass bottle of at least 250 mL or in two amber glass vials (minimum size 60 mL), preferably with no headspace. Minimize aeration of the sample. Preserve sample with 10 mg mercuric chloride/L sample if it will not be analyzed within 3 d of sampling. Pack samples in ice and ship to laboratory for next-day delivery if they are not analyzed immediately. Store samples under refrigeration.

d. Minimum detectable concentration: With a 30-min equilibration time and 45 mL of sample in a 60-mL vial, concentrations as low as 1 to 5 ng/L of most of the target compounds can be determined. Table 6040:IX shows results of an MDL determination performed over an 8-d period with 8 replicates and a known addition level of 3 ng/L at one laboratory and a similar determination in a single day at 5 ng/L at another laboratory. Under these conditions, the response for MIB, geosmin, and IPMP is linear from 10 ng/L to at least 100 ng/L (<20% RSD) with internal standard calibration (with IBMP used as an internal standard) (Table 6040:X) and can be measured with a calibration curve to higher levels. Some laboratories have demonstrated linearity to 100 ng/L or higher without the use of an internal standard (Table 6040:XI).

2. Apparatus

a. Gas-chromatograph/mass spectrometer (GC/MS): Any GC/MS system capable of meeting the specifications described in Section 6200 is acceptable.* Several GC columns have been tested;† others may be acceptable but have not yet been tested.

b. Solid-phase microextraction holder.‡

c. Water-bath capable of maintaining at least 50°C. Higher temperatures (65°C) typically produce more precise results. A covered water bath is preferable. Optional use of sonication or stirring shortens equilibration times but is not necessary.

d. Graduated cylinder, glass, 50 mL (Class A).

e. Block heater or hot plate (if using aluminum block).

f. Vial with septum, 60-mL.§

* Data reported herein were obtained with Finnigan-Mat 4000 MS, Varian 3400 GC, and HP 5890/5972 MSD.

† Supelcowax-10 (Supelco) and DB5-MS (Restek).

‡ Supelco catalog no. 5-7330 or equivalent.

§ I-Chem RC346-0060 or equivalent.

TABLE 6040:IX. METHOD DETECTION LEVEL (MDL) IN REAGENT WATER FOR MIB, GEOSMIN, AND IPMP

	MDL For Given Compound and Laboratory *ng/L*				
	MIB		Geosmin		IPMP
Replicate	Laboratory A*	Laboratory B	Laboratory A*	Laboratory B	Laboratory A
1	3.4	5.3	4.2	5.4	5.3
2	2.5	5.1	3.3	4.7	6.1
3	1.8	4.9	3.3	4.7	5.0
4	1.7	5.7	3.8	4.6	4.9
5	2.2	4.9	3.0	5.5	4.2
6	2.3	5.0	2.9	5.0	5.3
7	2.6	5.0	2.2	4.7	5.0
8	2.0	4.4	1.9	5.0	5.3
9	–	5.2	–	5.3	4.7
10	–	5.4	–	5.5	4.5
Average	2.3	5.1	3.1	5.0	5.0
Std deviation	0.5	0.4	0.8	0.4	0.52
MDL	1.6	1.0	2.3	1.0	1.6

* 3 ng/L known-addition level on 8 separate days.

3. Reagents

a. SPME fiber (divinylbenzene-carboxen-polydimethylsiloxane cross-link coating).‖

Other fiber chemistries may be applicable, but must be validated by the user.

b. Stock standard solutions, 100 mg/L MIB, geosmin, IBMP, and IPMP. Prepare stock standards from neat compounds weighed gravimetrically and dissolved in acetone or methanol using procedures described in 6040B. Standards may be prepared as a mixture or as single solutions.

c. Working standard solutions, 10 mg/L MIB and geosmin and/or IPMP. Prepare working standard by diluting stock standard 1:10 in acetone or methanol. Standard is stable for at least 30 d.

d. Calibration standard, 45 µg/L MIB and geosmin and/or IPMP. From 10 mg/L prepare a 45-µg/L calibration standard by diluting 45 µL 10-mg/L solution in 10 mL acetone or methanol. Standard is good for 3 d if properly refrigerated. Prepare calibration curve by adding appropriate amounts of calibration standard to 45 mL reagent water (10 µL = 10 ng/L, 20 µL = 20 ng/L, etc.).

e. Laboratory control standard and laboratory-fortified matrix standard, 15 to 20 ng/L MIB and geosmin. Prepare laboratory control standard (LCS) and laboratory-fortified matrix standard (LFM) from the working standard of a source different from that of the calibration standard, or from the same source but by a different chemist if a different source is not available.

f. Internal standard, 45 µg IBMP/L: Prepare internal standards by dilution from 100-mg/L stock standards and dissolving in acetone or methanol. IBMP has a response factor similar to those of MIB and geosmin and is not generally found in samples. Final concentration in samples will be 10 ng/L.

g. Surrogate, 45 µg/L IPMP: IPMP is an effective surrogate to allow monitoring of fiber performance for each sample. Prepare surrogate by dilution from calibration standard as in ¶ *f.* Final concentration in samples will be 10 ng/L. Higher levels (20 to 50 ng/L final concentration) also may be used if desired. Surrogate can be prepared as a mix with the internal standard if desired.

h. Sodium chloride, NaCl, reagent-grade: Check salt for contamination. Bake if necessary to eliminate contaminants. If baking does not work, use a different lot of salt.

‖ Supelco catalog no. 5-7348U.

TABLE 6040:X. INTERNAL STANDARD CORRECTED RESPONSE FACTOR FOR 5–100 NG/L TASTE- AND ODOR-CAUSING COMPOUNDS IN REAGENT WATER

Temperature	Concentration	Response Factor for Given Compound		
°C	*ng/L*	MIB	Geosmin	IPMP
50	5	1.87	1.06	0.77
	20	1.27	0.92	0.87
	25	1.32	0.93	0.94
	40	1.13	0.96	0.87
	50	1.11	0.95	0.86
	75	1.05	1.10	0.81
	100	1.04	1.25	0.75
	Avg.	1.25	1.02	0.84
	SD	0.29	0.12	0.07
	% RSD	23.2	11.8	7.8
65	5	1.02	0.98	0.75
	10	0.71	1.31	0.86
	20	0.83	1.31	0.92
	50	0.61	1.35	0.75
	70	0.66	1.37	0.86
	100	0.75	1.85	0.75
	Avg.	0.71	1.44	0.83
	SD	0.06	0.23	0.07
	% RSD	11.6	16.1	9.2

TABLE 6040:XI. CALIBRATION FOR 1–100 NG/L TASTE- AND ODOR-CAUSING COMPOUNDS WITHOUT AN INTERNAL STANDARD AT 65°C

Concentration ng/L	Primary Ion Count		
	MIB	Geosmin	IPMP
1	612	1 483	513
5	3 994	7 363	3 076
10	6 944	13 979	6 126
20	11 134	29 225	11 254
50	29 834	75 165	25 251
70	36 935	99 763	34 456
100	58 540	138 987	48 652
R^2	0.995	0.999	0.999

4. Procedure

Load SPME fiber in SPME holder. Condition fiber at 270°C overnight before first use. To each 60-mL vial, add at least 13.5 g NaCl. Use same amount of salt in samples and standards.

Pour 45 mL sample into a NaCl-prepared vial. If using internal standard method, add to each sample 10 μL 45-μg/L internal standard and surrogate. Otherwise add only surrogate. Swirl gently for 15 s to mix well. Secure vial in a water-bath set at 65°C. Lower temperatures (50°C) are acceptable but may lead to nonlinear calibration and poorer precision. Alternatively, use aluminum heating block to extract multiple samples at once, being very careful to control temperatures. Adjust SPME holder to a depth setting of 1. Insert holder into vial and secure holder in place. Depress plunger to expose the fiber into vial's headspace at the premarked level.

Extract for 30 to 35 min. Longer times are acceptable, as long as all samples and standards are extracted for exactly the same period of time. Shorter times do not give consistent recoveries. Fully retract fiber into holder before its removal. Check holder's needle for water; if present, blot water with clean tissue.

Adjust holder depth setting such that when the fiber is in the extended position, it is several millimeters above the injector.**

Insert holder's needle into GC injector port. Depress plunger to expose fiber in injector and immediately start the run. Allow fiber to remain in injector port for 10 min to clean up fiber for the next extraction.

Analyze samples using conditions recommended for the apparatus used.††

Monitor primary masses for quantitation. Optionally also monitor secondary masses for confirmation.

MIB — 95 (primary), 93, 107, 108, 135 (secondary)
Geosmin — 112 (primary), 126 (secondary)
IPMP — 137 (primary), 152, 124 (secondary)
IBMP — 124 (primary), 151, 94 (secondary)

Inconsistent recoveries may be due to variations in exposure time, temperature, or condition of fiber. Check to ensure the fiber is in good condition and that both time and temperature are the same as those for the calibration curve. Low recovery usually is due to water present at the needle tip after extraction. Be sure to blot out any residual water that may be present before injection. Use of a surrogate in each sample monitors this problem. High, low, or variable water-bath temperatures may lead to lower recoveries and greater variability. Ensure that the bath or heating block temperature is no greater than 65°C and no less than 50°C. If a heating block is used, temperatures up to 60°C can be used without greatly affecting the headspace temperature. Be sure to maintain exactly the same temperature for standards and samples.

If it is suspected that the sample contains IPMP or IBMP, or if IS or surrogate recoveries are >130% (indication of possible interference), reanalyze sample without addition of IS and surrogate to verify absence of interference.

5. Calculations

$$C = \frac{A}{R \times I \times D}$$

where:

C = concentration, ng/L,
A = area of primary peak observed from chromatogram,
R = response factor from calibration,
I = internal standard (IS) factor (IS injected/IS recovered), and
D = dilution factor.

Conventional external standard calibration techniques using an initial five-point curve can be used if linearity is demonstrated. Alternatively, if response is nonlinear, quantify against a calibration curve of at least six points bracketing the expected concentration and demonstrate that calculated values for the curve are within 10% of prepared values.

6. Quality Control

For each new fiber used, prepare a calibration curve with it to show that the fiber can achieve linearity with % RSD<20% over the desired calibration range.

Each day or when a new fiber is first used, whichever is more frequent, validate method by analyzing a midrange LCS. LCS recovery must be between 80 and 120% of nominal value. Recalibrate if QC criteria are not met. There is no limit to the number of samples that can be analyzed by a fiber.

For each fiber, analyze a check sample at the desired minimum reporting level (MRL) with each 20 samples or weekly, whichever is more frequent, at regular intervals (preferably daily) to demonstrate that the fiber is maintaining extraction efficiency. If the fiber is no longer able to extract samples reliably down to the MRL level, discard. The acceptance criterion for the MRL check sample is ±2 ng/L.

Analyze a laboratory-fortified matrix (LFM) with each 20 samples and with each new matrix or weekly, whichever is more frequent. The LFM should be near the midpoint of the calibration range or near the level of interest. The acceptance criterion for the LFM is 70 to 130%. Flag values outside these limits as indicating a matrix effect, as long as LFM addition at least doubles the ambient concentration.

** For Varian 3400 and Hewlett Packard GCs, preferably use a depth setting of 3.

†† For example:
Finnigan 4000 MS; Restek DB-5 column; detector: quadrupole, SIM mode; injector: splitless @ 250°C, split vent at 0.70 min @40 mL/min; temperature program: 50°C for 1 min, ramp 12°C/min, 250°C for 6 min.
HP 5972; Supelcowax 10 column; detector: quadrupole, SIM mode; injector: splitless@ 250°C, split vent at 2.0 min @ 33 mL/min, 0.75 mm liner; temperature program: 90°C for 8 min, ramp 15°C/min, 180°C for 6 min.

TABLE 6040:XII. COMPARISON OF METHODS 6040B AND D IN A SINGLE LABORATORY

	Concentration† ng/L			
	MIB		Geosmin	
Water Analyzed*	6040B	6040D	6040B	6040D
RWS	13	9	8.9	9
LFM	41	41	43	43
RWS	6.5	5.8	6.5	3.3
RWS	32	40	22	28
RWS	19	16	4.1	2.5
LFM	39	39	24	24
RWS	77	91	2.8	2.9
RWS	3.8	3.5	ND(<3)	0.3
RWS	ND(<3)	1.3	5.6	5.2
RWS	ND	ND	ND	ND
RWS	ND(<3)	1.7	8.3	7.7
RWS	ND	ND	12.5	11.3
RWS	16/10	11	13.1	13.1
RWS	11	8	5.2/3.9	2.7
RWS	3.7	1.3	3.7	3.1
RWS	10.1	8.7	9	7.1
RWS	11	5.7/5.2	4.4	3.2
RWS	22	24	4.3	1.4
			4.8	4.7

* RWS = raw water sample; LFM = laboratory-fortified matrix.
† ND = not determined.
NOTE: Closed-loop stripping (B) performed within 3 d of collection; solid-phase microextraction (D) performed after storage of up to 2 months. Extraction performed at 50°C. Results with value/value represent duplicate analyses.

Analyze a blank daily. Analytes should not be detected in the blank.

Preferably include surrogate with each sample. Acceptance criterion for surrogate is 80 to 120%.

7. Precision and Bias

The MDL for this method was determined to be <5 ng/L for MIB, geosmin, and IPMP (Table 6040:IX) under the conditions described above.

A number of raw water samples were analyzed for MIB and geosmin by both Method 6040B and 6040D in a single laboratory, and two laboratories analyzed a series of samples by 6040D

TABLE 6040:XIII. COMPARISON OF RESULTS FOR MIB AND GEOSMIN IN TWO DIFFERENT LABORATORIES

Analyte	Laboratory A ng/L	Laboratory B ng/L
MIB	ND*	ND
	ND*	Interference
	Interference*	ND
	13*	16
	9	11
	5.8	3.2
	40†	77†
	16 (19 by CLS)	23
Geosmin	ND(<3)*	5.3
	ND*	5.3
	3.5*	2.3
	48*	64
	9	10
	3.3	3
	28 (22 by CLS)	20
	2.5	2.2

* Analyzed by Method B.
† Reanalysis by CLS in Laboratory A gave a result of 32 and did not resolve the difference between the laboratories. 6040D extraction at 50°C in Laboratory A and 65°C in Laboratory B.

using the different extraction and analysis options included in the method. Comparison results are shown in Tables 6040:XII and XIII. Recoveries for LCS at 20 ng/L in a single laboratory are 95 ± 10% for MIB and 97± 12% for geosmin for a set of 30 points collected over a 2-month period.

8. Bibliography

BRAND, G. 1995. Evaluation of SPME technology for applicability of drinking water analysis. Proc. Water Qual. Technol. Conf., San Francisco, Calif., Nov. 6–10, 1994. Pt. 1, p. 273. American Water Works Assoc., Denver, Colo.

EISERT, R. & J. PAWLISZYN. 1997. New trends in solid-phase microextraction. *Crit. Rev. Anal. Chem.* 27:103.

PAWLISZYN, J. 1997. Solid Phase Microextraction Theory and Practice. Wiley-VCH, New York, N.Y.

MCCALLUM, R., P. PENDLETON, R. SCHUMANN & M.U. TRINH. 1998. Determination of geosmin and 2-methylisoborneol in water using solid-phase microextraction and gas chromatography - chemical ionization/electron impact ionization - ion-trap mass spectrometry. *Analyst* 123:2155.

6200 VOLATILE ORGANIC COMPOUNDS*

6200 A. Introduction

1. Source and Significance

Many organic compounds have been detected in ground and surface waters. While most groundwater contamination episodes are traceable to leaking underground fuel or solvent storage vessels, landfills, agriculture practices, and wastewater disposal, the most probable cause for contamination of some aquifers and surface waters has never been firmly established. Contamination may be due to past practices of on-site (leach field) disposal of domestic and industrial wastes or to illegal discharges. Organohalides, particularly the trihalomethanes, are present in most chlorinated water systems, especially those using surface waters as a source of supply. Toxicological studies on animal models have shown that some of these organics have the potential for teratogenesis or carcinogenesis in human beings. To minimize these health risks, sensitive detection and accurate and reproducible quantitation of organics is of paramount importance.

2. Selection of Method

Two capillary gas chromatographic methods for purgeable organic compounds are presented. The scope of analytes is detector-dependent. Method B is a gas chromatographic/ mass spectrometric (GC/MS) technique. Method C combines GC with photoionization detection (PID)/electrolytic conductivity detection (ELCD) in series. Methods B and C are applicable to a wide range of purgeable organics. Both methods can be applied to finished drinking water, drinking water in any stage of treatment, source water, or wastewater.

The methods presented are highly sophisticated micro-analytical procedures that should be used only by analysts experienced in chromatography and data evaluation and interpretation. While the methods are similar, they are not interchangeable from a regulatory point of view.[1,2]

3. Scope

Table 6200:I lists the compounds that can be determined by these methods. All are determinable by both Method B and Method C. Other compounds may be amenable to these methods.

4. Sampling and Storage

See Section 6010B.1

5. Method-Required Quality Control Criteria

a. Initial quality control:

1) Initial demonstration of capability—Conduct initial demonstration of capability study at least once, before analysis of any sample, by each analyst, to demonstrate proficiency with the method of choice. Include at least analysis of a reagent blank and four reagent blank samples fortified at a concentration between 10 times the minimum reporting level and the midpoint of the calibration curve. The blank should not contain any compound of interest at a concentration greater than minimum reporting level. Mean percent recovery for each compound calculated from the four fortified samples should be 80% to 120%, and the relative standard deviation (RSD) should be <20%.

2) Method detection level (MDL)—The MDL is a statistical determination of the minimum concentration that can be measured by the method with a confidence level of 99% that the analyte concentration is greater than zero. Determine MDL before any samples are analyzed, using the procedure described in Section 1030 or other appropriate procedure[3] as required for each matrix to be analyzed. For MDL calculation, start with a concentration about five times the estimated instrument detection level. Perform MDL determination as an iterative process. The values listed in Table 6200:III were generated using a concentration of 0.5 µg/L. Conduct MDL determination at least annually. Analyze samples for MDL determination over a 3- to 5-d period to generate a more realistic value.

3) Quality-control sample—Analyze an externally generated quality-control sample as a laboratory fortified blank at least quarterly or whenever new stock solutions are generated. Obtain this sample from sources external to the immediate laboratory, and use it to validate the laboratory's standards both qualitatively and quantitatively. Acceptance criteria are supplied by the manufacturer. If all criteria are not met, determine cause of error, and correct it before continuing.

4) Minimum quantitation level (MQL)—The MQL is the lowest level that can be quantified accurately. The MQL is defined as four times the MDL.

b. Calibration:

1) Initial calibration—Perform initial calibration with a minimum of five concentrations of analytical calibration standards (CALs) for the compound(s) of interest. The lowest concentration should be at the working reporting level; the highest concentration should be at the upper end of the calibration range. Do not report values that are outside of the defined calibration range. For the calibration concentrations, there should be no more than one order of magnitude between concentrations.

Use any of the following calibration functions, as appropriate: response factor for internal standard calibration, calibration factor for external standard calibration, or calibration curve. Calibration curves may be linear through the origin, linear not through the origin, or quadratic through or not through the origin. Use the following recommended acceptance criteria for the various calibration functions.

If using response factors or calibration factors, relative standard deviation (RSD) for each compound of interest should be

* Approved by Standard Methods Committee, 1997.
Joint Task Group: 20th Edition—Melissa S. Dale (chair), Anthony Bucciferro, Melly L. Fabro, T.J. Richards, William A. Saner, Robert Slater.

TABLE 6200:I. COMPOUNDS DETERMINABLE BY GAS CHROMATOGRAPHIC METHODS FOR PURGEABLE ORGANIC COMPOUNDS

Analyte	Chemical Abstract Services Registry Number	Analyte	Chemical Abstract Services Registry Number
Benzene	71-43-2	2,2-Dichloropropane	590-20-7
Bromobenzene*	108-86-1	1,1-Dichloropropene*	563-58-6
Bromochloromethane	74-97-5	*cis*-1,3-Dichloropropene*	10061-01-5
Bromodichloromethane	75-27-4	*trans*-1,3-Dichloropropene*	10061-02-6
Bromoform	75-25-2	Ethylbenzene*	100-41-4
Bromomethane	74-83-9	Hexachlorobutadiene*	87-68-3
n-Butylbenzene*	104-51-8	Isopropylbenzene*	98-82-8
sec-Butylbenzene*	135-98-8	*p*-Isopropyltoluene*	99-87-6
tert-Butylbenzene*	98-06-6	Methyl *t*-butyl ether*	1634-04-4
Carbon tetrachloride	56-23-5	Methylene chloride	75-09-2
Chlorobenzene*	108-90-7	Naphthalene*	91-20-3
Chloroethane	75-00-3	*n*-Propylbenzene*	103-65-1
Chloroform	67-66-3	Styrene*	100-42-5
Chloromethane	74-87-3	1,1,1,2-Tetrachloroethane	630-20-6
2-Chlorotoluene*	95-49-8	1,1,2,2-Tetrachloroethane	79-34-5
4-Chlorotoluene*	106-43-4	Tetrachloroethene*	127-18-4
Dibromochloromethane	124-48-1	Toluene*	108-88-3
1,2-Dibromo-3-chloropropane	96-12-8	1,2,3-Trichlorobenzene*	87-61-6
1,2-Dibromoethane	106-93-4	1,2,4-Trichlorobenzene*	120-82-1
Dibromomethane	74-95-3	1,1,1-Trichloroethane	71-55-6
1,2-Dichlorobenzene*	95-50-1	1,1,2-Trichloroethane	79-00-5
1,3-Dichlorobenzene*	541-73-1	Trichloroethene*	79-01-6
1,4-Dichlorobenzene*	106-46-7	Trichlorofluoromethane	75-69-4
Dichlorodifluoromethane	75-71-8	1,2,3-Trichloropropane	96-18-4
1,1-Dichloroethane	75-34-3	1,2,4-Trimethylbenzene*	95-63-6
1,2-Dichloroethane	107-06-2	1,3,5-Trimethylbenzene*	108-67-8
1,1-Dichloroethene*	75-35-4	Vinyl chloride*	75-01-4
cis-1,2-Dichloroethene*	156-59-4	*o*-Xylene*	95-47-6
trans-1,2-Dichloroethene*	156-60-5	*m*-Xylene*	108-38-3
1,2-Dichloropropane	78-87-5	*p*-Xylene*	106-42-3
1,3-Dichloropropane	142-28-9		

* Compound can be determined using Method 6200C with PID only.

less than 20%. If the RSD is not less than 20% for any compound of interest, then identify and correct source of lack of linearity before sample quantitation. When using response factors (i.e., for GC/MS analysis), check performance or sensitivity of the instrument for the compound of interest against minimum acceptance values for the response factors. See specific analytical method for the acceptance criteria for the response factors for each compound.

For a linear regression, the correlation coefficient should be >0.994. Recalculate each calibration point compared to curve. Resulting values should be within ±20%. If any of the recalculated values are not within ±20%, identify and correct source of outlier(s) before sample quantitation.

Use initial calibration, with any of the above functions (response factor, calibration factor, or calibration curve) for quantitation of the analytes of interest in samples. Use continuing calibration, described in ¶ 2) below, only for checks on initial calibration and not for sample quantitation. Perform initial calibration when instrument is set up and whenever continuing calibration criteria are not met.

2) Continuing calibration—Continuing calibration (CCAL) is the periodic analysis of a calibration standard used to verify that the instrument response has not changed significantly from the initial calibration. Perform continuing calibration every 10 samples for GC analysis, every 20 samples for GC/MS analysis, or every 12 h, whichever is more frequent. Perform continuing calibration with one or more of the concentrations of analytical standards in the initial calibration. Vary actual concentration of continuing calibration standard over calibration range, with a minimum concentration greater than two times the reporting limit. The acceptance criterion for continuing calibration is 70% to 130% recovery compared to the known or expected value of the calibration standard (at the analyst's discretion, the acceptance criterion for the gases may be extended to 60% to 140% recovery). If the acceptance criteria are not met, re-analyze continuing calibration standard or repeat initial calibration. When using response factors, check performance or sensitivity of instrument for analytes of interest against minimum acceptance values for response factors.

3) Closing standard—Finish all sample sets with a closing standard to demonstrate that performance was still acceptable for the last sample analyzed. Use acceptance criteria as for the CCAL.

c. Batch quality control:

1) Analytical day—An analytical day is defined as a 12-h analytical period.

2) Sample set (batch)—A sample set (batch) is defined as those samples extracted in an analytical day, not to exceed 20 samples.

3) Laboratory reagent blank (LRB)—A LRB is a blank sample consisting of all reagents that normally contact a sample when carried through the entire analytical procedure. Use reagent blank to determine contribution of reagents and preparative analytical steps to observed value. No compound of interest should be present in reagent blank at a level greater than the MQL. Include a minimum of one reagent blank with each sample set (batch).

4) Laboratory-fortified blank (LFB)—See ¶ *b*2) above. NOTE: For this method the LFB and CCAL are the same.

5) Internal standard (IS)—An internal standard is a compound of known concentration added to each standard and sample just before sample analysis. Because of the nature of purge and trap analysis, the IS is taken through the entire analytical process, just as is the surrogate standard [see ¶ 6) below]. However, the IS is used for quantitation, whereas the surrogate standard is used to monitor ongoing purge recovery. Use IS to monitor retention time, relative response, and concentration of analytes in each sample. When quantifying by the internal standard method, measure all compound responses relative to this standard. Internal standard area response should be in the range of ±30% compared to the mean calibration curve area response. The IS compound should mimic the chromatographic conditions of the analytes of interest. The retention time of this compound should separate from all analytes of interest and elute in a representative area of the chromatogram. If a specific compound cannot be found to meet these criteria, use additional compounds to satisfy analytical needs.

6) Surrogate standard (SS)—A surrogate standard is a compound added to each standard and sample at a known concentration before extraction. Choose a compound(s) that is chemically similar to the analytes and that is unlikely to be found in environmental samples. Carry surrogate standard through entire sample extraction and analytical process to monitor extraction recovery for each sample. Surrogate recovery should remain reasonably constant over time. Recovery should not vary more than 30% from the known value. Refer to method of choice for specific surrogates.

7) Laboratory-fortified sample (LFS)—A LFS is an additional portion of a sample to which the analytes of interest have been added at a concentration at least two times the MRL or around the middle of the calibration range. Include a minimum of one LFS with each sample set (batch). Make LFSs at sufficient concentrations that sample background levels do not adversely affect recovery calculations. (If this is a known sample, adjust addition concentrations to be about five times background level). Base sample batch acceptance on results of CCALs and LFBs rather than on LFSs, because the matrix of the sample may interfere with method performance. Prepare fortifying solution for blanks and samples from a different primary mix than that used to develop working standard mix.

8) LFS duplicates—A LFS duplicate is a second LFS used to evaluate the precision of the method in a matrix sample. If sufficient sample volume is collected, fortify a large enough volume to yield two sample portions for analysis. If sufficient sample volume is not collected, use a second bottle of the same sample fortified to the same concentration as the first. Include a minimum of one LFS duplicate with each sample set (batch). Compare precision and bias to those listed in the method. Base sample batch acceptance on results of CCAL and LFB additions rather than LFS duplicates.

6. References

1. U.S. ENVIRONMENTAL PROTECTION AGENCY. 1987. National Primary Drinking Water Regulations—synthetic organic chemicals; monitoring for unregulated contaminants; final rule. 40 CFR 141 & 142; *Federal Register* 52, No. 130.
2. U.S. ENVIRONMENTAL PROTECTION AGENCY. 1986. Guidelines establishing test procedures for the analysis of pollutants under the Clean Water Act. 40 CFR Part 136; *Federal Register* 51, No. 125.
3. U.S. ENVIRONMENTAL PROTECTION AGENCY. 1995. Definition and procedure for the determination of the method detection limit, revision 1.11. 40 CFR Part 136, Appendix B.

6200 B. Purge and Trap Capillary-Column Gas Chromatographic/Mass Spectrometric Method

This method[1] is applicable to the determination of a wide range of purgeable organic compounds (see Table 6200:I). The method can be extended to include other volatile organic compounds, provided that all performance criteria are met. It should be used only by analysts experienced in the operation of GC/MS systems and in evaluation and interpretation of mass spectra.

1. General Discussion

a. Principle: Volatile organic compounds are transferred efficiently from the aqueous to the gaseous phase by bubbling an inert gas (e.g., helium) through a water sample contained in a specially designed purging chamber at ambient temperature. The vapor is swept through a sorbent trap that adsorbs the analytes of interest. After purging is complete, the trap is heated and back-flushed with the same inert gas to desorb the compounds onto a gas chromatographic column. The gas chromatograph is temperature-programmed to separate the compounds. The detector is a mass spectrometer. See Section 6010C for discussion of gas chromatographic and mass spectrometric principles.

b. Interferences: Impurities in the purge gas and organic compounds outgassing from the plumbing upstream of the

trap account for most contamination problems. Demonstrate that the system is free from contamination under operational conditions by analyzing laboratory reagent blanks daily. (NOTE: Use blanks for monitoring only; corrections for blank values are unacceptable.) Avoid using non-TFE plastic tubing, non-TFE thread sealants, or flow controllers with rubber components in the purge and trap system. Ensure that the analytical area is not subject to contamination from laboratory solvents, particularly methylene chloride and methyl *tert*-butyl ether (MtBE).

Samples can be contaminated by diffusion of volatile organics (particularly fluorocarbons and methylene chloride) through the septum seal during shipment and storage. Use a field reagent blank prepared from reagent water and carried through the sampling, handling, and shipping procedures as a check on such contamination.

Contamination by carryover can occur whenever high-level and low-level concentration samples are analyzed sequentially. To reduce carryover, rinse purging device and sample syringe with reagent water between samples. Follow analysis of an unusually high concentration sample with a LRB to check for carryover contamination. For samples containing large amounts of water-soluble materials, suspended solids, high boiling compounds, or high levels of volatile compounds, wash purging device with a detergent solution, rinse it with distilled water, and dry it in an oven at 105°C between analyses. The trap and other parts of the system also are subject to contamination; therefore, frequently bake and purge entire system.

c. Detection levels: Method detection levels (MDLs) are compound-dependent and vary with purging efficiency and instrument response. In a single laboratory using reagent water and known-addition concentrations of 0.5 μg/L, observed MDLs were in the range of 0.025 to 0.450 μg/L. The applicable calibration range of this method is compound- and instrument-dependent, but is approximately 0.2 to 200 μg/L. Compounds that are inefficiently purged from water will not be detected when present at low concentrations, but they can be measured with acceptable bias and precision when present at sufficient concentration. Determination of some geometrical isomers (e.g., xylenes) may be hampered by co-elution.

d. Safety: The toxicity or carcinogenicity of each analyte has not been precisely defined. Benzene, carbon tetrachloride, bis(1-chloroisopropyl)ether, 1,4-dichlorobenzene, 1,2-dichloroethane, hexachlorobutadiene, 1,1,2,2-tetrachloroethane, 1,1,2-trichloroethane, chloroform, 1,2-dibromoethane, tetrachloroethene, trichloroethene, and vinyl chloride have been classified tentatively as known or suspect carcinogens. Handle pure standard materials and stock standard solutions of these compounds in a hood and wear a NIOSH/MESA-approved toxic gas respirator when handling high concentrations.

2. Apparatus

a. Purge and trap system: The purge and trap system consists of purging device, trap, and desorber. Several complete systems are available commercially.

1) *Purging device,* designed to accept 25-mL samples with a water column at least 5 cm deep. A smaller 5-mL purging device is acceptable if required method detection levels and performance criteria are met. Keep gaseous headspace between water column and trap to a total volume of less than 15 mL. Pass purge gas through water column as finely divided bubbles with a diameter of less than 3 mm at the origin. Introduce purge gas no more than 5 mm from base of water column. The purging device illustrated in Figure 6200:1 meets these criteria.

Needle spargers may be used instead of the glass frit shown in Figure 6200:1; however, in either case, introduce purge gas at a point <5 mm from base of water column.

2) *Trap,* at least 25 cm long and with an inside diameter of at least 3 mm, packed with the following minimum lengths of adsorbents: 1.0 cm methyl silicone coated packing, 7.7 cm 2,6-diphenylene oxide polymer, 7.7 cm silica gel, and 7.7 cm coconut charcoal. If analysis is not to be made for dichlorodifluoromethane, the charcoal may be eliminated and the polymer section lengthened to 15 cm. Alternative sorbents may be used provided that all quality control criteria are met. Various sorbent traps are available commercially*; ensure that trap keeps total purge gas volume and purge time constant (i.e., 40 mL/min for 11 min) and that performance will meet all quality control criteria. The minimum specifications for the trap are illustrated in Figure 6200:2.

Methyl silicone coated packing is recommended, but not mandatory. The packing protects the diphenylene oxide polymer adsorbent from aerosols, re-coating any active site that may develop during the heating process, and ensures that the polymer is fully enclosed within the heated zone of the trap, thus eliminating potential cold spots. Alternatively, silanized glass wool may be used as a spacer in the trap inlet.

Before initial use, condition trap overnight following manufacturer's instructions. Vent trap effluent to the room, not to analytical column. Before daily use, condition trap for 10 min with back-flushing. Optimally, vent trap to analytical column during daily conditioning; however, run column through temperature program before sample analysis.

b. Gas chromatograph (GC)†: Use a temperature-programmable GC, suitable for on-column injection. Deactivate all glass components (e.g., injector liners) with a silanizing agent.

c. Capillary GC columns: Use any capillary GC column that meets all performance criteria. Ensure that desorb flow rate is compatible with the column of choice. Four examples of acceptable columns are listed below.

1) *Column 1:* 60-m-long × 0.75-mm-ID VOCOL‡ wide-bore capillary column with 1.5-μm film thickness.

2) *Column 2:* 30-m-long × 0.53-mm-ID DB-624§ mega-bore capillary column with 3-μm film thickness.

3) *Column 3:* 30-m-long × 0.32-mm-ID DB-5§ capillary column with 1-μm film thickness.

4) *Column 4:* 30-m-long × 0.25-mm-ID DB-624§ capillary column with 1.4-μm film thickness.

---

* Tekmar VOCARB 4000 or equivalent.

† Gas chromatographic methods are extremely sensitive to the materials used. Mention of trade names by *Standard Methods* does not preclude the use of other existing or as-yet-undeveloped products that give *demonstrably* equivalent results.

‡ Supelco, Inc. or equivalent.

§ J&W or equivalent.

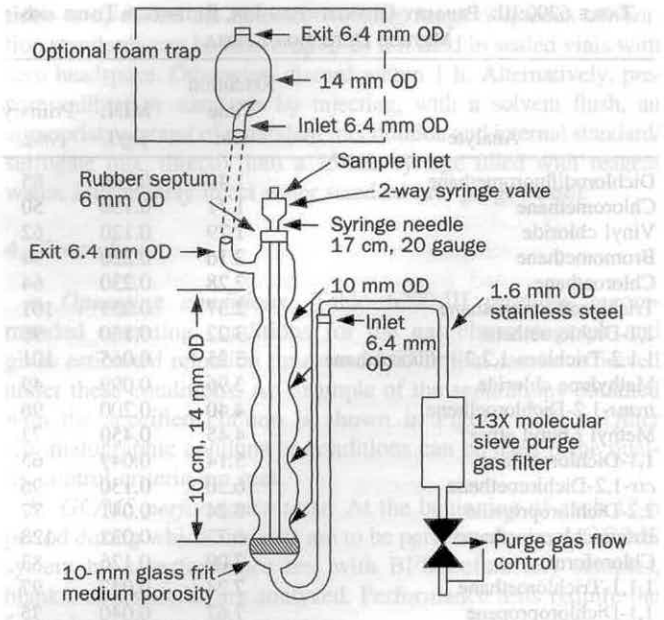

Figure 6200:1. Purging device.

d. Mass spectrometer, capable of scanning from 35 to 300 amu every 2 s or less, utilizing 70 eV (nominal) electron energy in the electron impact ionization mode, and producing a mass spectrum that meets all criteria in Table 6200:II when 25 ng or less of 4-bromofluorobenzene is introduced into GC. To ensure sufficient precision, the desired scan rate permits acquisition of at least five spectra while a sample component elutes from the GC.

e. Purge and trap – GC/MS interface: Use an open-split or direct-split interface, depending on which column is used. Alternatively, if the narrow-bore column (4) is used, a capillary concentrator preceding the GC may be necessary. This interface condenses desorbed materials onto an uncoated fused silica pre-column and when flash-heated transfers compounds onto the capillary column. The uncoated section of column is cooled to −150°C during desorption and heated to 250°C to transfer condensed materials.

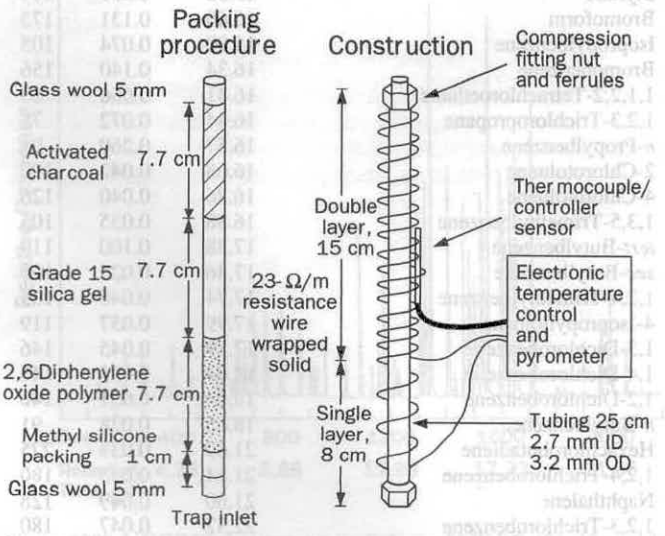

Figure 6200:2. Trap packings and construction to include desorb capability.

TABLE 6200:II. BFB KEY *m/z* ABUNDANCE CRITERIA

Mass	*m/z* Abundance Criteria
50	15 to 40% of mass 95
75	30 to 60% of mass 95
95	Base peak, 100% relative abundance
96	5 to 9% of mass 95
173	<2% of mass 174
174	>50% of mass 95
175	5 to 9% of mass 174
176	95 to 101% of mass 174
177	5 to 9% of mass 176

f. Data system: To the mass spectrometer attach a computer that allows continuous acquisition and storage of all mass spectra obtained throughout the chromatographic program. Computer software should allow for a search of all acquired spectra for specific *m/z* (masses) and the plot of such *m/z* abundances versus time or scan number. This type of plot is an extracted ion current profile (EICP). Software also should allow the integration of the abundances in any EICP over a specified time or scan limit.

g. Syringes, 0.5-, 1.0-, 5-, and 25-mL glass hypodermic with detachable tip.‖

h. Syringe valves, two-way, with detachable tip.‖

i. Microsyringes, 10-, 25-, and 100-μL with a 5-cm × 0.15-mm-ID and 220 bevel needle.#

j. Bottles, 40-mL with TFE-lined screw cap.

3. Reagents

a. Reagent water, in which no interferent is observed at or above the MDL of the constituents of interest. Prepare by passing tap water through a carbon filter bed containing about 0.5 kg activated carbon, by distillation, or by using a water purification system.**

b. Trap packing materials:

1) *2,6-Diphenylene oxide polymer,* 60/80 mesh, chromatographic grade.

2) *Methyl silicone packing,* 3 OV-1.

3) *Silica gel,* 35/60 mesh.

c. Methanol, purge-and-trap grade.

d. Hydrochloric acid: HCl, 1 + 1.

e. Vinyl chloride, 99.9% pure.

f. Ascorbic acid.

g. Stock standard solutions: Prepare from pure standard materials or purchase as certified solutions. Prepare stock standard solutions in methanol using assayed liquids or gases as appropriate. CAUTION: *Toxic substances.* See ¶ 1*d*.

Place about 9.8 mL methanol in a 10-mL ground-glass-stoppered volumetric flask. Let stand unstoppered for about 10 min or until all alcohol-wetted surfaces have dried. Weigh flask to nearest 0.1 mg.

‖ Luerlok or equivalent.
Hamilton # 702 or equivalent.
** Millipore Super Q or equivalent.

Add assayed reference materials as follows: For liquids, using a 100-μL syringe or disposable capillary-tip glass pipet, immediately add two or more drops of assayed reference material to flask, then reweigh. Ensure that the drops fall directly into the alcohol without contacting flask neck. For halocarbon gases that boil below 30°C (bromomethane, chloroethane, chloromethane, dichlorofluoromethane, trichlorofluoromethane, vinyl chloride), attach a vinyl plastic†† tube to port of gas bottle containing reference material, with open end bubbling into a beaker of methanol showing flow through the tubing; insert needle of 5-mL valved gastight syringe into tube and pull gas into syringe slowly to 5.0-mL mark. Lower syringe needle to within 5 mm of methanol surface and slowly force gas onto surface. The gas will dissolve into the methanol and will be seen as a vortex as it dissolves into the solvent. Reweigh flask (difference is amount of gas dissolved into methanol), dilute to volume, stopper, and mix by inverting several times. Calculate concentration in micrograms per microliter from net gain in weight. When compound purity is assayed to be 96% or greater, calculate concentration of stock standard from uncorrected weight. Preferably use commercially prepared stock standards at any concentration if they are certified by the manufacturer or an independent source. Transfer stock standard solution into a TFE-sealed screw-cap bottle. Store with minimum headspace at −10 to −20°C away from light.

h. Secondary dilution standards: Using stock standard solutions, prepare in methanol secondary dilution standards that contain the compounds of interest, either singly or mixed together. Prepare secondary dilution standards at concentrations that will permit aqueous calibration standards (¶ *j* below) to bracket working range of the analytical system. Store secondary dilution standards with minimal headspace in a freezer and check frequently for signs of evaporation (which would indicate need for regeneration). Always bring to room temperature before preparing calibration standards. Prepare standards fresh weekly for gases. Replace all other standards monthly, or sooner if comparison with check standards indicates a problem.

i. Internal standard/surrogate standard known addition: Prepare a solution containing fluorobenzene (internal standard) and 1,2-dichlorobenzene-d_4 (surrogate) in methanol. Alternate internal standard and surrogate compounds may be used, provided that they meet method criteria and do not interfere with any method analyte(s). Prepare secondary dilution standard at a concentration of 5 μg/mL of each compound. Adding 5.0 μL standard to 25.0 mL sample or calibration standard yields a concentration equivalent to 1.0 μg/L. Alternate secondary standard concentrations can be used if addition volume is adjusted accordingly and all internal standard criteria are met. Add this mixture to each sample, standard, and blank.

j. Calibration standards: Prepare at least five concentration levels for each compound by adding appropriate amounts of secondary standard solution to reagent water and inverting water sample twice. Prepare one standard at a concentration near, but above, the MDL (i.e., 4 × MDL for potable-water-type samples) or a level that defines the low end of the working range and the others to correspond to the expected range of sample concentra-

†† Tygon or equivalent.

TABLE 6200:III. PRIMARY QUANTITATION ION, RETENTION TIMES AND METHOD DETECTION LEVELS

Analyte	Retention Time *min*	MDL *μg/L*	Primary *m/z*
Dichlorodifluoromethane	1.49	0.190	85
Chloromethane	1.71	0.150	50
Vinyl chloride	1.79	0.120	62
Bromomethane	2.16	0.220	94
Chloroethane	2.28	0.230	64
Trichlorofluoromethane	2.57	0.059	101
1,1-Dichloroethene	3.22	0.130	96
1,1,2-Trichloro-1,2,2-trifluoroethane	3.25	0.065	101
Methylene chloride	3.96	0.099	49
trans-1,2-Dichloroethene	4.40	0.200	96
Methyl *t*-butyl ether	4.45	0.450	73
1,1-Dichloroethane	5.14	0.047	63
cis-1,2-Dichloroethene	6.30	0.130	96
2,2-Dichloropropane	6.24	0.041	77
Bromochloromethane	6.77	0.032	128
Chloroform	7.00	0.126	83
1,1,1-Trichloroethane	7.24	0.043	97
1,1-Dichloropropene	7.67	0.040	75
Carbon tetrachloride	7.65	0.042	117
Benzene	8.07	0.036	78
1,2-Dichloroethane	8.14	0.055	62
Trichlorethene	9.44	0.045	95
1,2-Dichloropropane	9.85	0.053	63
Dibromomethane	10.07	0.035	93
Bromodichloromethane	10.47	0.112	83
cis-1,3-Dichloropropene	11.29	0.048	75
Toluene	11.81	0.047	91
trans-1,3-Dichloropropene	12.27	0.051	75
1,1,2-Trichloroethane	12.56	0.043	83
1,3-Dichloropropane	12.83	0.090	76
Tetrachloroethene	12.77	0.047	166
Dibromochloromethane	13.24	0.133	129
1,2-Dibromoethane	13.35	0.133	107
Chlorobenzene	14.21	0.052	112
1,1,1,2-Tetrachloroethane	14.37	0.048	131
Ethylbenzene	14.42	0.032	91
m,p-Xylene	14.63	0.038	91
o-Xylene	15.27	0.038	91
Styrene	15.30	0.031	104
Bromoform	15.60	0.131	173
Isopropylbenzene	15.90	0.074	105
Bromobenzene	16.34	0.140	156
1,1,2,2-Tetrachloroethane	16.41	0.066	83
1,2,3-Trichloropropane	16.44	0.072	75
n-Propylbenzene	16.57	0.260	91
2-Chlorotoluene	16.68	0.042	126
4-Chlorotoluene	16.86	0.040	126
1,3,5-Trimethylbenzene	16.88	0.035	105
tert-Butylbenzene	17.38	0.100	119
sec-Butylbenzene	17.46	0.025	105
1,2,4-Trimethylbenzene	17.74	0.046	105
4-Isopropyltoluene	17.99	0.037	119
1,3-Dichlorobenzene	17.89	0.045	146
1,4-Dichlorobenzene	18.04	0.033	146
1,2-Dichlorobenzene	18.64	0.031	146
n-Butylbenzene	18.65	0.028	91
Hexachlorobutadiene	21.69	0.033	225
1,2,4-Trichlorobenzene	21.34	0.043	180
Naphthalene	21.80	0.049	128
1,2,3-Trichlorobenzene	22.32	0.047	180

GC conditions: Column: J&W DB-624, 30 m, 0.25-mm ID, 1.4-μm film; Temperature program: 35°C, 4 min; 4°C/min; 50°C, 0 min; 10°C/min; 175°C, 4 min.

tions or to define the detector working range. Aqueous calibration standards can be stored up to 24 h if held in sealed vials with zero headspace. Otherwise, discard within 1 h. Alternatively, prepare calibration standards by injecting, with a solvent flush, an appropriate amount of a standard mix dilution and internal standard/surrogate mix, directly into a 25-mL syringe filled with reagent water; immediately inject water standard into purge vessel.

4. Procedure

a. Operating conditions: Table 6200:III provides recommended operating conditions for the gas chromatograph and gives estimated retention times and MDLs that can be achieved under these conditions. An example of the separations obtained with the specified column is shown in Figure 6200:3. Other chromatographic columns or conditions can be used if the quality control criteria are met.

b. GC/MS performance tests: At the beginning of each 12-h period during which analyses are to be performed, check GC/MS system by a performance test with BFB before any samples, blanks, or standards are analyzed. Performance tests require the following instrument parameters:

Electron energy: 70 eV (nominal)
Mass range: 35 to 300 amu
Scan time: at least 5 scans/peak but not more than 2 s/scan

Inject 25 ng BFB directly on GC column. If direct injection is not easily performed, add 1 μL 25-μg/mL BFB solution to 25 mL reagent water in syringe used for sample transfer to purge device and analyze as a sample. Obtain a background-corrected mass spectrum of BFB and confirm that all key *m/z* criteria in Table 6200:II are achieved. If all criteria are not achieved, re-tune mass spectrometer and repeat test until all criteria are met.

c. Calibration: Calibrate system as follows:

1) System setup—Condition trap initially overnight at 180°C by back-flushing with inert gas at 20 mL/min. Condition trap daily for 10 min at manufacturer's suggested temperature. Connect purge and trap system to GC using recommended temperature program and flow-rate conditions. Calibrate system by either the internal or the external standard technique.

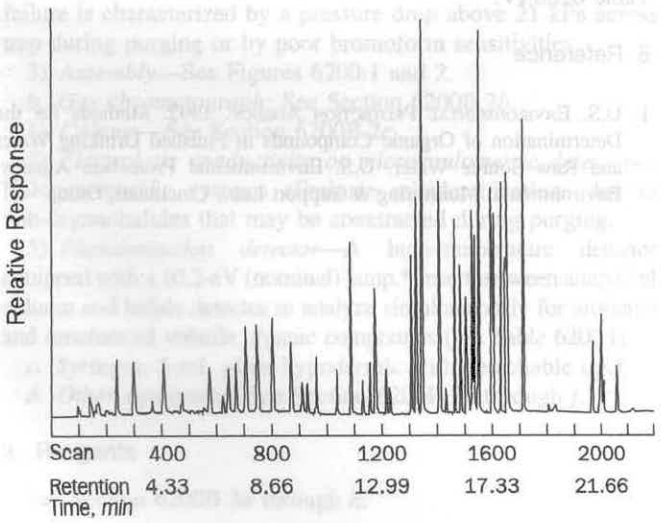

Figure 6200:3. GC/MS chromatogram. Column: J&W DB-624, 30 m, 0.25-mm ID, 1.4-μm film; temperature program: 35°C, 4 min; 4°C/min; 50°C, 0 min; 10°C/min; 175°C, 4 min.

2) Internal standard calibration technique—Select one or more internal standards similar in analytical behavior to the compounds of interest. Fluorobenzene is a recommended internal standard compound. Demonstrate that measurement of internal standard is not affected by method or matrix interference. Because of such limitations, no one internal standard may be applicable to all samples. The compounds used as surrogates (e.g., 1,2-dichlorobenzene-d_4) for quality control also can be used successfully as internal standards. Prepare calibration standards at a minimum of five concentration levels for each compound as described in ¶ 3*j* above. Prepare a secondary dilution standard containing each of the internal standards (¶ 3*i* above). Analyze each calibration standard according to procedure for samples, adding internal standard solution directly to syringe. Tabulate peak height or area responses against concentration for each compound and internal standard, and calculate response factors (RF) for each compound in each calibration standard as follows:

$$RF = \frac{(A_s)(C_{is})}{(A_{is})(C_s)}$$

where:

A_s = response for compound to be measured,
A_{is} = response for internal standard,
C_{is} = concentration for internal standard, and
C_s = concentration of compound to be measured.

Calculate % RSD for the average RFs for each compound. Average RF can be used if RSD is less than 20%.

3) External standard calibration technique—Prepare standards as directed in ¶ 3*j*. Analyze each calibration standard and tabulate peak area responses versus concentration. Prepare calibration curve for each compound. Alternatively, if ratio of response to concentration (calibration factor) is a constant over the working range (<20% RSD), assume linearity through the origin and use average calibration factor in place of a calibration curve.

4) Calibration check—See ¶ A.5*b*2).

d. Sample analysis: Bring sample to ambient temperature. Remove plunger from 25-mL syringe and close attached valve. Open sample bottle and carefully pour sample into syringe barrel to just short of overflowing. Replace syringe plunger, invert syringe, and open valve. Vent any air and adjust sample volume to 25.0 mL, in duplicate if sufficient sample is available (once sample cap has been removed, sample cannot be stored, because of headspace). Add an appropriate amount of surrogate/internal standard through valve bore, and close valve. Attach to purge device, open valves, and inject sample into purge vessel. Close valves and purge sample for 11.0 min at ambient temperature at a flow rate of 40 mL/min (helium or nitrogen). If water vapor causes problems in the mass spectrometer, use a 3-min dry purge and/or a moisture control module.

Desorb trapped materials onto head of chromatographic column at 180°C while back-flushing trap for 4 min with inert gas at a flow rate compatible with the column of choice, and begin GC temperature program.

TABLE 6200:IV. SINGLE-LABORATORY BIAS AND PRECISION DATA IN REAGENT WATER*

Analyte	Recovery %	Standard Deviation	Relative Standard Deviation %
Benzene	107	0.046	9
Bromobenzene	111	0.034	6
Bromochloromethane	88	0.052	12
Bromodichloromethane	104	0.036	7
Bromoform	107	0.042	8
Bromomethane	89	0.049	11
n-Butylbenzene	115	0.048	8
sec-Butylbenzene	113	0.043	8
tert-Butylbenzene	116	0.057	10
Carbon tetrachloride	119	0.048	8
Chlorobenzene	108	0.033	6
Chloroethane	115	0.073	13
Chloroform	108	0.043	8
Chloromethane	74	0.036	10
2-Chlorotoluene	111	0.045	8
4-Chlorotoluene	112	0.049	9
Dibromochloromethane	108	0.042	8
1,2-Dibromoethane	102	0.042	8
Dibromomethane	132	0.113	17
1,2-Dichlorobenzene	106	0.043	8
1,3-Dichlorobenzene	108	0.052	10
1,4-Dichlorobenzene	106	0.045	8
Dichlorodifluoromethane	80	0.058	15
1,1-Dichloroethane	109	0.049	9
1,2-Dichloroethane	102	0.031	6
1,1-Dichloroethene	99	0.059	12
cis-1,2-Dichloroethene	103	0.062	12
trans-1,2-Dichloroethene	113	0.045	8
1,2-Dichloropropane	129	0.064	10
1,3-Dichloropropane	107	0.046	9
2,2-Dichloropropane	106	0.049	9
1,1-Dichloropropene	110	0.044	8
cis-1,3-Dichloropropene	99	0.044	9
trans-1,3-Dichloropropene	101	0.038	7
Ethylbenzene	109	0.049	9
Hexachlorobutadiene	112	0.053	9
Isopropylbenzene	112	0.044	8
4-Isopropyltoluene	117	0.046	8
Methylene chloride	85	0.050	12
Methyl *t*-butyl ether	81	0.017	11
Naphthalene	121	0.068	11
n-Propylbenzene	107	0.048	9
Styrene	101	0.039	8
1,1,1,2-Tetrachloroethane	113	0.037	7
1,1,2,2-Tetrachloroethane	104	0.053	10
Tetrachloroethene	106	0.046	9
Toluene	106	0.045	8
1,2,3-Trichlorobenzene	118	0.054	9
1,2,4-Trichlorobenzene	109	0.049	9
1,1,1-Trichloroethane	106	0.040	8
1,1,2-Trichloroethane	97	0.041	9
Trichloroethene	105	0.041	8
Trichlorofluoromethane	105	0.045	9
1,2,3-Trichloropropane	104	0.034	6
1,1,2-Trichloro-1,2,2-trifluoroethane	113	0.042	7
1,2,4-Trimethylbenzene	116	0.044	8
1,3,5-Trimethylbenzene	110	0.051	9
Vinyl chloride	85	0.037	9
m,p-Xylene	110	0.057	10
o-Xylene	106	0.044	8

*For all analytes, seven samples, each of 0.5 µg/L concentration, were analyzed.

Set system auto-drain to empty purge chamber while trap is being desorbed into GC, or alternatively, use sample syringe to empty vessel. Washing chamber with two 25-mL flushes of reagent water is useful if highly contaminated samples are being analyzed. Be sure all areas wetted during purging are also wetted during rinsing to maximize flushing.

Recondition trap by baking at conditioning temperature for 5 to 7 min. Let trap cool to ambient before introduction of next sample into purge vessel. When all sample compounds have been eluted from chromatographic column, end data acquisition and store data files. Use data system software to display full range mass spectra and appropriate extracted ion current profiles (EICP). If any ion abundances exceed system working range, dilute sample in second syringe with reagent water and analyze. NOTE: *Take care with sample because compounds can be very volatile and can be lost if sample is reopened.* Estimate amount of dilution needed and expel excess sample from second syringe, inject that portion into purge vessel, and with a second syringe, add necessary reagent water to a total of 25.0 mL in purge vessel.

5. Calculation

When compounds have been identified, base quantitation on integrated area abundance from the EICP of the primary characteristic *m/z* given in Table 6200:III. If sample produces an interference for the primary *m/z*, calculate a response factor or calibration curve using a secondary characteristic *m/z*, and use secondary *m/z* to quantitate. Report results in micrograms per liter. Report all quality control data with sample results.

6. Quality Control

See Section 6200A.5.

7. Precision and Bias

Typical single-laboratory precision and bias data are shown in Table 6200:IV.

8. Reference

1. U.S. ENVIRONMENTAL PROTECTION AGENCY. 1992. Methods for the Determination of Organic Compounds in Finished Drinking Water and Raw Source Water. U.S. Environmental Protection Agency, Environmental Monitoring & Support Lab., Cincinnati, Ohio.

6200 C. Purge and Trap Capillary-Column Gas Chromatographic Method

This method[1] is applicable to the determination of purgeable halocarbons and aromatic organic compounds (Table 6200:I) in finished drinking water, raw source water, drinking water in any treatment stage, and wastewater.

1. General Discussion

a. Principle: See Section 6200B.1*a*.

b. Interferences: See Section 6200B.1*b*.

c. Detection levels: In a single laboratory using reagent water and known additions of 0.5 μg/L, calculated method detection levels (MDLs) for these compounds were in the range of 0.01 to 0.05 μg/L, depending on the compound. Some laboratories may not be able to achieve these detection levels because results depend on instrument sensitivity and matrix effects. Analysis of complex mixtures containing partially resolved compounds may be hampered by concentration differences larger than a factor of 10. This problem commonly occurs in analyses of finished drinking waters because of the relatively high trihalomethane content.

d. Safety: The toxicity or carcinogenicity of each reagent has not been defined precisely. Carbon tetrachloride, 1,2-dichloroethane, 1,1,2,2-tetrachloroethane, 1,1,2-trichloroethane, chloroform, 1,2-dibromoethane, tetrachloroethene, trichloroethene, and vinyl chloride have been classified tentatively as known or suspected human or mammalian carcinogens. Prepare primary standards of these compounds in a hood and wear a NIOSH/MESA-approved toxic gas respirator when handling high concentrations.

2. Apparatus

a. Purge and trap system: The purge and trap system consists of three separate pieces of equipment: purging device, trap, and desorber. Several complete systems are commercially available.

1) *Purging device*—See Section 6200B.2*a*1).

2) *Trap*—See Section 6200B.2*a*2). If only compounds boiling above 35°C are to be analyzed, both silica gel and charcoal can be eliminated and polymer increased to fill entire trap. Trap failure is characterized by a pressure drop above 21 kPa across trap during purging or by poor bromoform sensitivities.

3) *Assembly*—See Figures 6200:1 and 2.

b. Gas chromatograph: See Section 6200B.2*b*.

1) *Column*—See Section 6200B.2*c*.

2) *Electrolytic conductivity or microcoulometric detector*—Halogen-specific systems eliminate misidentifications due to non-organohalides that may be coextracted during purging.

3) *Photoionization detector*—A high-temperature detector equipped with a 10.2-eV (nominal) lamp.* Insert between analytical column and halide detector to analyze simultaneously for aromatic and unsaturated volatile organic compounds (see Table 6200:I).

c. Syringes, 5-mL glass hypodermic with detachable tip.†

d. Other equipment: See Section 6200B.2*g* through *j*.

3. Reagents

See Section 6200B.3*a* through *h*.

* Tracor Model 703 or equivalent.
† Luerlok or equivalent.

4. Procedure

a. Operating conditions: Table 6200:V summarizes recommended operating conditions for the gas chromatograph, estimated retention times, and method detection levels. Examples of separations obtained with the specified column are shown in Figures 6200:4 and 5.

b. Calibration: See Section 6200A.5*b*. Use either internal or external calibration technique. If using internal standard technique, prepare a dilution standard as described in Section 6200B.3*i*.

c. Instrument performance: See Section 6200A.5. Ensure that all peaks in standard chromatograms are sharp and symmetrical. Correct any peak tailing significantly in excess of that shown in method chromatograms. Tailing problems generally are traceable to active sites on the GC column or to detector operation. If only compounds eluting before chloroform give random responses or unusually wide peak widths, are poorly resolved, or are missing, the problem usually is traceable to the trap/desorber. If only brominated compounds show poor peak geometry or do not respond properly at low concentrations, replace trap. Excessive detector reactor temperatures also can cause low bromoform response. If negative peaks appear in the chromatogram, replace both ion-exchange column and electrolyte in detector. Check precision between replicate analyses. A properly operating system shows an average relative standard deviation of less than 10%. Poor precision generally is traceable to pneumatic leaks, especially around sample purger and detector reactor inlet and exit, electronic problems, or sampling and storage problems. Monitor retention times for each compound using calibration standards and laboratory control standard. If individual retention times vary by more than 10% over an 8-h period or do not fall within 10% of an established norm, locate and correct source of retention data variance.

d. Sample analysis: See Section 6200B.5.

5. Calculation

Identify each organohalide in sample chromatogram by comparing retention time of suspect peak to retention times generated by calibration standards and laboratory control standard. Determine concentrations of individual compounds. If external standard calibration procedure is used, calculate concentration of compound being measured from peak response using calibration curve or calibration factor previously determined.

If internal standard calibration procedure is used, calculate concentration using response factor [*RF*, ¶ B.4*c*2)] by the following equation:

$$\text{Concentration, } \mu\text{g/L} = \frac{A_s \times C_{is}}{A_{is} \times RF}$$

where:

A_s = response for compound to be measured,
A_{is} = response for internal standard, and
C_{is} = concentration of internal standard.

Report results in micrograms per liter without correction for recovery. Report quality control data with sample results.

TABLE 6200:V. RETENTION TIMES AND METHOD DETECTION LEVELS

Analyte	Retention Time *min*	Method Detection Level: Electrolytic Conductivity Detector *µg/L*	Method Detection Level: Photoionization Detector *µg/L*
Dichlorodifluoromethane	6.22	0.037	—
Chloromethane	7.09	0.041	—
Vinyl chloride	7.68	0.025	0.088
Bromomethane	9.45	0.103	—
Chloroethane	9.76	0.025	—
Trichlorofluoromethane	11.04	0.042	—
1,1-Dichloroethene	13.59	0.018	0.035
1,1,2-Trichloro-1,2,2-trifluoroethane	13.07	0.047	—
Methylene chloride	15.83	0.068	—
Methyl *t*-butyl ether	16.49	—	0.411
trans-1,2-Dichloroethene	16.78	0.015	0.015
1,1-Dichloroethane	18.49	0.015	—
2,2-Dichloropropane	20.27	0.220	—
cis-1,2-Dichloroethene	20.54	0.012	0.032
Chloroform	21.04	0.017	—
Bromochloromethane	21.53	0.025	—
1,1,1-Trichloroethane	22.14	0.014	—
1,1-Dichloropropene	22.57	0.019	0.008
Carbon tetrachloride	22.80	0.022	—
Benzene	23.38	—	0.017
1,2-Dichloroethane	23.62	0.074	—
Trichloroethene	25.30	0.012	0.014
1,2-Dichloropropane	25.92	0.021	—
Bromodichloromethane	26.63	0.040	—
cis-1,3-Dichloropropene	28.38	0.067	0.041
Dibromomethane	28.40	0.057	—
Toluene	29.16	—	0.023
trans-1,3-Dichloropropene	30.00	0.029	0.046
1,1,2-Trichloroethane	30.39	0.042	—
Tetrachloroethene	31.04	0.013	0.014
1,3-Dichloropropane	31.18	0.020	—
Dibromochloromethane	31.86	0.039	—
1,2-Dibromoethane	32.36	0.070	—
Chlorobenzene	33.67	0.029	0.027
Ethylbenzene	33.89	—	0.028
1,1,1,2-Tetrachloroethane	33.91	0.020	—
m,p-Xylene	34.08	—	0.021
o-Xylene	35.44	—	0.024
Styrene	35.67	—	0.027
Isopropylbenzene	36.64	—	0.018
Bromoform	36.72	0.023	—
1,1,2,2-Tetrachloroethane	37.43	0.034	—
1,2,3-Trichloropropane	37.88	0.048	—
n-Propylbenzene	37.94	—	0.023
Bromobenzene	37.98	0.026	0.026
1,3,5-Trimethylbenzene	38.44	—	0.019
2-Chlorotoluene	38.48	0.017	0.017
4-Chlorotoluene	38.63	0.026	0.028
1,2,4-Trimethylbenzene	39.61	—	0.030
tert-Butylbenzene	39.76	—	0.018
sec-Butylbenzene	40.34	—	0.018
4-Isopropyltoluene	40.80	—	0.019
1,3-Dichlorobenzene	41.01	0.017	0.028
1,4-Dichlorobenzene	41.40	0.059	0.061
n-Butylbenzene	42.21	—	0.028
1,2-Dichlorobenzene	42.62	0.023	0.031
1,2,4-Trichlorobenzene	48.21	0.019	0.028
Hexachlorobutadiene	48.75	0.026	0.019
Naphthalene	49.05	—	0.043
1,2,3-Trichlorobenzene	49.92	0.018	0.032

GC conditions: Column—Supelco VOCOL, 60 m, 0.75-mm ID, 1.5-µm film; Temperature program–0°C, 8 min; 4°C/min; 185°C, 1.5 min.

6. Quality Control

See Section 6200A.5.

7. Precision and Bias

See Table 6200:VI.

8. Reference

1. U.S. ENVIRONMENTAL PROTECTION AGENCY. 1991. Volatile organic compounds in water by purge and trap capillary column gas chromatography with photoionization and electrolytic conductivity detectors in series. Method 502.2 *in* Methods for the Determination of Organic Compounds in Finished Drinking Water and Raw Source Water. U.S. Environmental Protection Agency, Environmental Monitoring & Support Lab., Cincinnati, Ohio.

TABLE 6200:VI. SINGLE-LABORATORY BIAS AND PRECISION DATA IN REAGENT WATER*

Analyte	Photoionization Detector			Electrolytic Conductivity Detector		
	Recovery %	Standard Deviation	Relative Standard Deviation %	Recovery %	Standard Deviation	Relative Standard Deviation %
Benzene	70	0.006	2	—	—	—
Bromobenzene	—	—	—	89	0.008	2
Bromochloromethane	—	—	—	83	0.008	2
Bromodichloromethane	—	—	—	135	0.021	3
Bromoform	—	—	—	81	0.007	2
Bromomethane	—	—	—	73	0.033	9
n-Butylbenzene	63	0.009	3	—	—	—
sec-Butylbenzene	65	0.009	3	—	—	—
tert-Butylbenzene	72	0.006	2	—	—	—
Carbon tetrachloride	—	—	—	79	0.007	2
Chlorobenzene	70	0.009	2	97	0.009	2
Chloroethane	—	—	—	64	0.008	2
Chloroform	—	—	—	83	0.006	1
Chloromethane	—	—	—	96	0.063	13
2-Chlorotoluene	—	—	—	91	0.005	1
4-Chlorotoluene	73	0.009	2	81	0.008	2
Dibromochloromethane	—	—	—	88	0.013	3
1,2-Dibromoethane	—	—	—	139	0.022	3
Dibromomethane	—	—	—	79	0.018	5
1,2-Dichlorobenzene	67	0.010	3	93	0.007	2
1,3-Dichlorobenzene	70	0.009	3	95	0.005	1
1,4-Dichlorobenzene	70	0.019	6	91	0.019	4
Dichlorodifluoromethane	—	—	—	71	0.027	8
1,1-Dichloroethane	—	—	—	82	0.005	1
1,2-Dichloroethane	—	—	—	78	0.024	6
1,1-Dichloroethene	61	0.011	4	81	0.006	1
cis-1,2-Dichloroethene	61	0.010	3	76	0.004	1
trans-1,2-Dichloroethene	79	0.005	1	77	0.005	1
1,2-Dichloropropane	—	—	—	85	0.007	2
1,3-Dichloropropane	—	—	—	148	0.018	2
2,2-Dichloropropane	—	—	—	74	0.045	12
1,1-Dichloropropene	54	0.003	1	74	0.006	2
cis-1,3-Dichloropropene	57	0.013	5	78	0.021	5
trans-1,3-Dichloropropene	63	0.015	5	78	0.009	2
Ethylbenzene	70	0.009	3	—	—	—
Hexachlorobutadiene	55	0.006	2	76	0.008	2
Isopropylbenzene	67	0.006	2	—	—	—
4-Isopropyltoluene	65	0.006	2	—	—	—
Methylene chloride	—	—	—	83	0.022	5
Methyl *t*-butyl ether†	75	0.130	3	—	—	—
Naphthalene	73	0.014	4	—	—	—
n-Propylbenzene	70	0.007	2	—	—	—
Styrene	70	0.009	3	—	—	—
1,1,1,2-Tetrachloroethane	—	—	—	83	0.001	0
1,1,2,2-Tetrachloroethane	—	—	—	88	0.011	2
Tetrachloroethene	54	0.005	2	79	0.004	1
Toluene	69	0.007	2	—	—	—
1,2,3-Trichlorobenzene	72	0.010	3	84	0.006	1
1,2,4-Trichlorobenzene	70	0.009	3	94	0.006	1
1,1,1-Trichloroethane	—	—	—	79	0.005	1
1,1,2-Trichloroethane	—	—	—	118	0.014	2
Trichloroethene	57	0.004	2	80	0.004	1
Trichlorofluoromethane	—	—	—	70	0.013	4
1,2,3-Trichloropropane	—	—	—	87	0.015	3
1,1,2-Trichloro-1,2,2-trifluoroethane	—	—	—	79	0.029	7
1,2,4-Trimethylbenzene	65	0.010	3	—	—	—
1,3,5-Trimethylbenzene	70	0.006	2	—	—	—
Vinyl chloride	73	0.022	6	67	0.008	2
m,p-Xylene	73	0.007	2	—	—	—
o-Xylene	68	0.008	2	—	—	—

*For all analytes, seven samples, each at a concentration of 0.5 µg/L (unless otherwise noted), were analyzed.
†Sample concentration 5 µg/L.

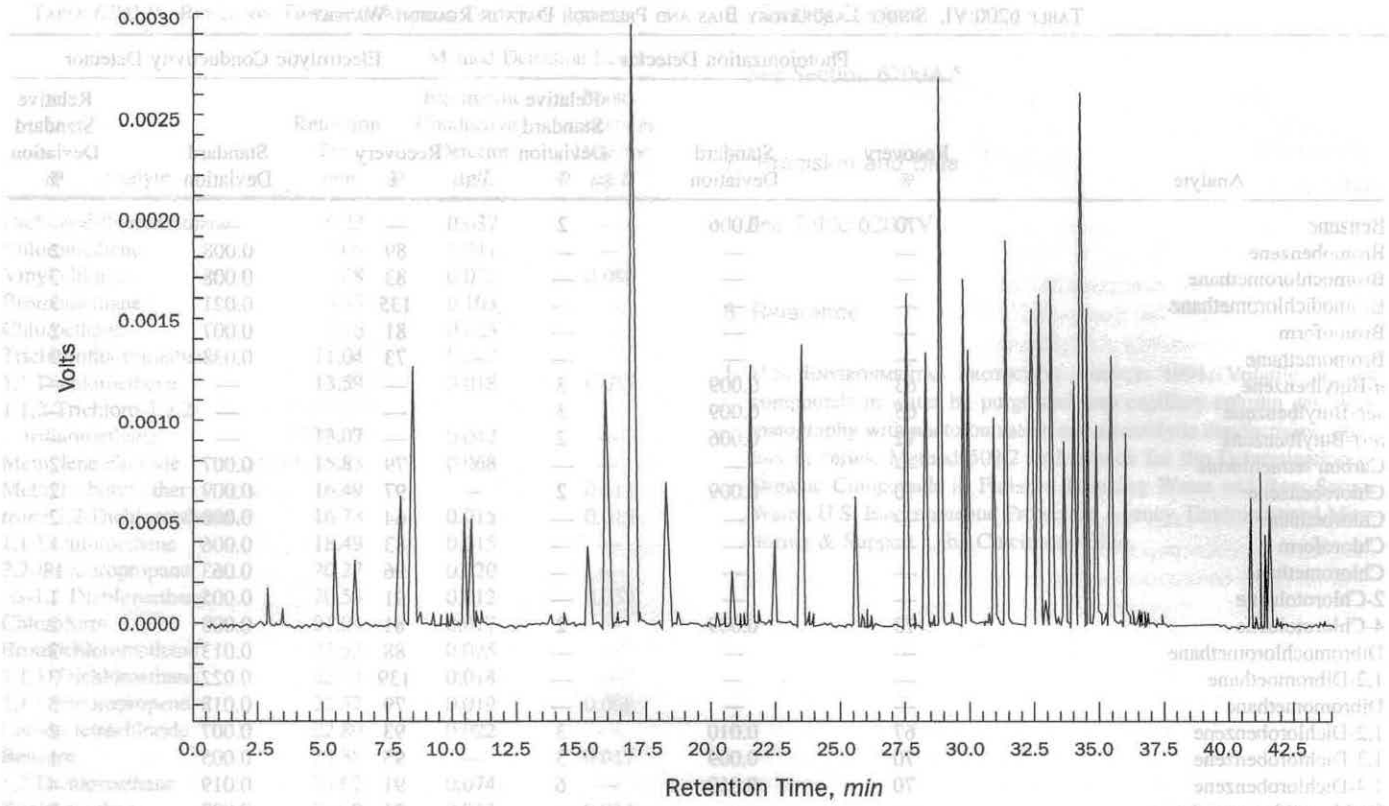

Figure 6200:4. PID chromatogram. GC conditions: Column: Supelco VOCOL, 60 m, 0.75-mm ID, 1.5-μm film; temperature program: 0° C, 8 min: 4° C/min; 185° C, 1.5 min.

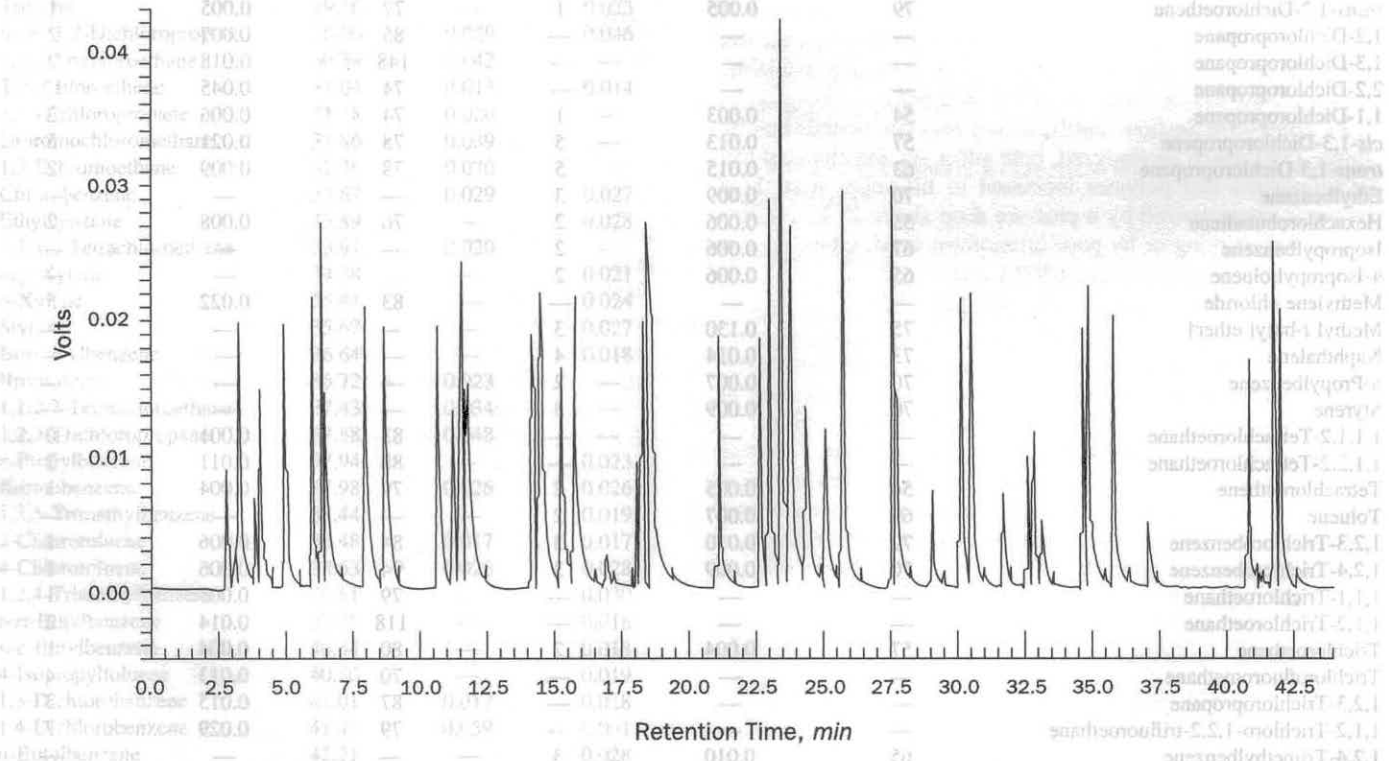

Figure 6200:5. ELCD chromatogram. GC conditions: Column: Supelco VOCOL, 60 m, 0.75-mm ID, 1.5-μm film; temperature program: 0° C, 8 min; 4° C/min; 185° C, 1.5 min.

6211 METHANE*

6211 A. Introduction

1. Occurrence and Significance

Methane (CH_4) is a colorless, odorless, tasteless combustible gas occasionally found in groundwaters. Escape of this gas from water may cause an explosive atmosphere not only in a utility's tanks, pumphouses, and other facilities, but also on the consumer's property, particularly where water is sprayed through poorly ventilated spaces such as public showers.

The explosive limits of CH_4 in air are 5 to 15% by volume. At sea level, a 3.95% CH_4 concentration in air theoretically could be reached in a poorly ventilated space sprayed with hot (68°C) water having a CH_4 concentration of only 0.7 mg/L. At higher water temperatures, the vapor pressure of water is so great that no explosive mixture can form. At lower barometric pressures, the theoretical hazardous concentration of methane in water will be reduced proportionately. In an atmosphere of N_2 or other inert gas, at least 12.8% O_2 must be present for there to be an explosion hazard.

Methane also is produced from wastewater and may be present in sewers and wastewater treatment plants (see Section 2720).

2. Selection of Method

The combustible-gas indicator method (B) offers the advantages of simplicity, speed, and great sensitivity. The volumetric method (C) can be made more accurate for concentrations of 4 to 5 mg/L and higher, but will not be satisfactory for very low concentrations. The volumetric method also can be applied to differentiate between CH_4 and other gases, as when a water supply is contaminated by liquid petroleum gas or other volatile combustible materials.

Methane also may be determined with the gas chromatograph as described in Sludge Digester Gas, Section 2720. This method permits differentiation between H_2 and CH_4, and/or its higher homologs.

* Approved by Standard Methods Committee, 2000.

6211 B. Combustible-Gas Indicator Method

1. General Discussion

a. Principle: An equilibrium according to Henry's law is established between CH_4 in solution and the partial pressure of CH_4 in the gas phase above the solution. The partial pressure of CH_4 can be determined with a combustible-gas indicator. The operation of the instrument is based on the catalytic oxidation of a combustible gas on a heated platinum filament that is made a part of a Wheatstone bridge. The heat generated by the oxidation of the gas increases the electrical resistance of the filament. The resulting imbalance of the electrical circuit causes deflection of a milliammeter that may be calibrated in terms of percentage of CH_4 or percentage of the lower explosive limit of the gas sampled.

b. Interference: Small amounts of ethane usually are associated with CH_4 in natural gas and presumably would be present in water that contains methane. Hydrogen gas has been observed in well waters and would behave similarly to CH_4 in this procedure. Hydrogen sulfide may interfere if the pH of the water is low enough for an appreciable fraction of the total sulfide to exist in the un-ionized form. The vapors of combustible oils also may interfere. In general, these interferences are of no practical importance because primary interest is in calculating the explosion hazard to which all combustible gases and vapors contribute.

Interference due to H_2S can be reduced by the addition of solid NaOH to the container before sampling.

c. Minimum detectable concentration: The limit of sensitivity of the test is approximately 0.2 mg/L.

d. Sampling: If the water is supersaturated with CH_4, a representative sample cannot be obtained unless the water is under sufficient pressure to keep all of the gas dissolved. Operate wells long enough to insure sampling water coming directly from the aquifer. Representative samples can be expected only when the well is equipped with a pump operating at sufficient submergence to assure that no gas escapes from the water.

2. Apparatus

*a. Combustible-gas indicator:** Connect a three-way stopcock to the inlet to zero instrument on atmospheric air immediately before obtaining sample reading. For laboratory use, replace the suction bulb with a filter pump throttled to draw gas through the instrument at a rate of approximately 600 mL/min. See Figure 6211:1.

b. Laboratory filter pump.

c. Glass bottle, 4-L, fitted with a two-hole rubber stopper. Extend inlet tube to within 1 cm of bottom. End outlet tube approximately 1 cm below stopper. Use metal or glass tubes, each fitted with stopcocks or with short (approximately 5-cm) lengths of rubber tubing and pinchcocks. The entire assembly should be capable of holding a low vacuum for several hours. Determine volume of assembly by filling with water and measuring volume, or weight, of water contained.

3. Reagent

Sodium hydroxide, NaOH, pellets.

* Marketed under the following trade names: "Explosimeter," "Methane Gas Detector," and "Methane Tester," all manufactured by Mine Safety Appliance Co., Pittsburgh, PA 15235, and "J-W Combustible Gas Indicator," manufactured by Bacharach Instrument Co., Mountain View, CA 94043, or equivalent.

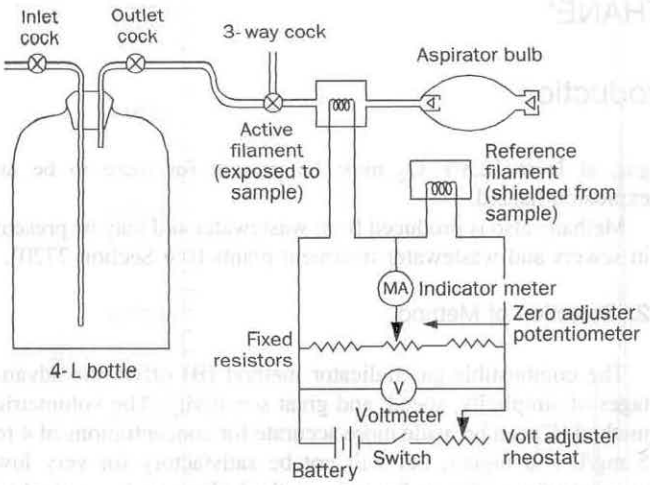

Figure 6211:1. Combustible gas indicator circuit and flow diagram.

4. Procedure

a. Rough estimation of CH_4 concentration: Fill bottle about half full of water, using a rubber tube connecting sampling tap and inlet tube, with outlet tube open. With both inlet and outlet tubes closed, shake bottle vigorously for approximately 15 s and let stand for approximately 1 min. Sample gas phase by withdrawing gas from the outlet, leaving inlet open to admit air. If the needle swings rapidly to a high level on the meter and then drops to zero, the CH_4-air mixture is too rich to burn; take a smaller sample for the final test. If needle deflection is too small to be read accurately, take a larger volume of water.

b. Accurate determination: If the water contains H_2S, add approximately 0.5 g NaOH pellets to empty bottle to suppress interference. Evacuate bottle, using filter pump. Fill bottle not more than three-quarters full by connecting inlet tube to sampling cock, with outlet tube closed. After collecting desired volume of water, let bottle fill with air through inlet tube. Close inlet cock, shake bottle vigorously for 60 s, and let stand for at least 2 h. Sample gas phase through outlet tube with inlet cock open. Take reading as rapidly as possible before the entering air has diluted sample appreciably. Measure volume of water sampled.

5. Calculation

The weight of CH_4 (w), in mg, in the sample is given by the equation:

$$w = P\left(\frac{1.928\ V_g}{T + 273} + \frac{890\ V_l}{H}\right)$$

where:

P = partial pressure of CH_4, kPa,
T = temperature, °C,
V_g = volume of gas phase, mL,
V_l = volume of liquid phase, mL, and
H = Henry's law constant, kPa/mole CH_4/mole of water.

Values for Henry's constant are as follows:

Temperature °C	Henry's Constant H*	Temperature °C	Henry's Constant H*
0	2.265	40	5.261
5	2.625	45	5.577
10	3.010	50	5.846
15	3.413	60	6.342
20	3.804	70	6.749
25	4.181	80	6.911
30	4.544	90	7.013
35	4.926	100	7.106

* Multiply given values by 10^6.

For most determinations, it may be assumed that atmospheric pressure is 100 kPa, and that the temperature is 20°C. The concentration of CH_4 in the sample is then given by:

$$\text{mg } CH_4/\text{L} = Rf\left(6.7\frac{V_0 - V_1}{V_1} + 0.24\right)$$

where:

R = scale reading,
V_0 = total volume of sample bottle, mL,
V_1 = volume of water sampled, mL, and
f = factor depending on instrument used.

If the instrument reads directly in percentage of methane, f = 1.00. If the instrument reads in percentage of the lower explosive limit of CH_4, f = 0.05. For instruments that require additional factors, consult the manufacturer. For example, one commercial instrument with a scale that reads in percentage of the lower explosive limit of combustible gases requires an additional factor of 0.77 for CH_4. Hence, the value of f in the above equation would be 0.77 × 0.05, or 0.0385.

For more accurate work, or in locations where normal barometric pressure is significantly lower than 100 kPa, use the equation:

$$\text{mg } CH_4/\text{L} = RBf\left(19.277\frac{V_0 - V_1}{TV_1} + \frac{8900}{H}\right)$$

where:

B = barometric pressure, kPa,

and other symbols are as above.

6. Accuracy

The accuracy of the determination is limited by the accuracy of the instrument used. Errors of approximately 10% may be expected. Calibration of instrument on known CH_4-air mixtures will improve accuracy.

7. Bibliography

ROSSUM, J.R., P.A. VILLARRUZ & J.A. WADE. 1950. A new method for determining methane in water. *J. Amer. Water Works Assoc.* 42:413.

6211 C. Volumetric Method

1. General Discussion

a. Principle: If CH_4 is slowly mixed with an excess of O_2 in the presence of a platinum coil heated to yellow incandescence, most of the CH_4 will be converted to CO_2 and H_2O in a smooth reaction. Several passes of the mixed gases may be needed to burn substantially all the CH_4. An excess of O_2 is mixed with the sample before passage through the assembly. By differential absorption and volumetric changes the product CO_2 is measured.

b. Interference: Low-boiling hydrocarbons other than ethane and vapors from combustible oils interfere. These substances, however, are not likely to be present in water in sufficiently high concentration to affect the results significantly.

c. Minimum detectable concentration: This method is not satisfactory for determining CH_4 in water where the concentration is less than 2 mg/L.

d. Sampling: Collect sample as directed in Method B and observe the same precautions to obtain representative samples (Section 6211B.1*d*). Omit NaOH pellets and fill sample bottle with water up to 90% of capacity.

2. Methane Determination

See Section 2720B for a description of apparatus, reagents, procedure, calculation, and precision and bias.

Use percentage of CH_4 found by this method with Henry's law to obtain the CH_4 concentration in original sample. Substitute CH_4 percentage for R (scale reading) and $f = 1$ in the calculation given under Section 6211B.5 preceding.

3. Bibliography

DENNIS, L.M. & M.L. NICHOLS. 1929. Gas Analysis. Macmillan Co., New York, N.Y.

HALDANE, J.S. & J.I. GRAHAM. 1935. Methods of Air Analysis. Charles Griffin & Co., London.

BUSWELL, A.M. & T.E. LARSON. 1937. Methane in ground waters. *J. Amer. Water Works Assoc.* 29:1978.

BERGER, L.B. & H.H. SCHRENK. 1938. Bureau of Mines Haldane gas analysis apparatus. U.S. Bur. Mines Information Circ. No. 7017.

LARSON, T.E. 1938. Properties and determination of methane in ground waters. *J. Amer. Water Works Assoc.* 30:1828.

6231 1,2-DIBROMOETHANE (EDB) AND 1,2-DIBROMO-3-CHLOROPROPANE (DBCP)*

6231 A. Introduction

1. Sources and Significance

Dibromoethane and dibromochloropropane have been found in groundwater supplies in many areas of the United States; typically they are found in agricultural areas where these compounds have been applied in the past as fumigants. Toxicological studies suggest that they may have detrimental effects on human health, and therefore many states have established maximum contaminant levels for them.

2. Selection of Method

Method 6231B is a liquid-liquid extraction gas chromatographic (GC) method that uses microextraction and two capillary columns (one for primary quantitation and the second for confirmation). Another detector, such as MS, also may be used for confirmation (see 6231C). Method B is the preferred method. In addition, these compounds can be detected by the purge and trap gas chromatographic/mass spectrometric (GC/MS) and GC methods (6200B and C), and dibromoethane by closed-loop stripping analysis (see Section 6040). For additional information on applicability, sensitivity, precision, and bias, see specific methods.

* Approved by Standard Methods Committee, 2000.

6231 B. Liquid-Liquid Extraction Gas Chromatographic Method

This method[1–3] is applicable to the determination of 1,2-dibromoethane (EDB) and 1,2-dibromo-3-chloropropane (DBCP) in drinking water and untreated groundwater.

1. General Discussion

a. Principle: The sample is extracted with hexane and injected into a gas chromatograph equipped with a linearized electron capture detector for separation and analysis. Identification is confirmed by analyzing the sample with a dissimilar column. See Section 6010C for discussion of gas chromatographic principles.

b. Interferences: Impurities in the extracting solvent usually account for most analytical problems. Analyze solvent blanks on each new bottle of solvent before use. Obtain indirect daily checks on the extracting solvent by monitoring sample blanks; whenever an interference is noted, reanalyze the extracting sol-

vent. If necessary, remove interference by distillation or column chromatography[3] or, more simply, obtain a new source solvent. Interference-free solvent contains less than 0.1 μg/L individual compound interference. Store solvents in an area free of organochlorine solvents.

Accidental sample contamination can occur by diffusion of volatile organics through the septum seal into the sample bottle during shipment and storage. Trip blanks monitor this type of contamination.

EDB at low concentrations may be masked by very high levels of dibromochloromethane (DBCM) when the confirmation column is used.

For further information on interferences in gas chromatographic methods, see Section 6010C.

c. Detection levels: The method detection levels (MDL)[4] for EDB and DBCP are 0.01 μg/L. The method is useful over a concentration range from approximately 0.03 to 200 μg/L. Actual detection levels are highly dependent on the characteristics of the gas chromatographic system used.

d. Safety: The toxicity or carcinogenicity of each reagent has not been defined precisely. EDB and DBCP have been classified tentatively as known or suspected human or mammalian carcinogens. Handle pure standard materials and stock standard solutions in a hood or glovebox and wear a NIOSH/MESA-approved toxic gas respirator when handling high concentrations.

2. Sampling and Storage

Collect all samples in duplicate and prepare replicate trip blanks with each sample set. A sample set is all of the samples collected from the same general sampling site at approximately the same time. Prepare the trip reagent blanks in the laboratory by filling a minimum of two sample bottles with reagent water, sealing, and shipping to the sampling site along with sample bottles.

Fill sample bottle to overflowing without air bubbles. When sampling from a water tap, open tap and flush until water temperature has stabilized (usually about 10 min). Adjust flow rate to about 500 mL/min and collect duplicate samples from the flowing stream. When sampling from a well, fill a wide-mouth bottle or beaker with sample, and carefully fill duplicate 40-mL sample bottles.

Keep samples chilled in an atmosphere free of organic solvent vapors, from day of collection until analysis. Do not acidify. Thiosulfate has no effect on EDB and DBCP. For regulatory compliance monitoring, thiosulfate is required to prevent interferences from other disinfection by-products.

Analyze all samples within 28 d of collection.

3. Apparatus

a. Sample containers, 40-mL screw-cap vials* each with a TFE-faced silicone septum.† Wash vials and septa with detergent and rinse with tap and distilled water before using. Let vials and septa air dry at room temperature, place in a 105°C oven for 1 h, then remove and let cool in an area free of organics.

* Pierce #13075 or equivalent.
† Pierce #12722 or equivalent.

Table 6231:I. Chromatographic Conditions for 1,2-Dibromoethane (EDB) and 1,2 Dibromo-3-Chloropropane (DBCP)

Compound	Retention Time, min	
	Column 1	Column 2
EDB	9.5	8.9
DBCP	17.3	15.0

Column 1 conditions: Durawax-DX 3 (0.25-μm film thickness) in a 30 m long × 0.32-mm ID fused silica capillary column with helium carrier gas at linear velocity of 25 cm/s. Column temperature held isothermal at 40°C for 4 min, then programmed at 8°C/min to 180°C for final hold.
Column 2 conditions: DB-1 (0.25-μm film thickness) in a 30 m long × 0.32-mm ID fused silica capillary column with helium carrier gas at linear velocity of 25 cm/s. Column temperature held isothermal at 40°C for 4 min, then programmed at 10°C/min to 270°C for final hold.

b. Vials, auto sampler, screw cap with septa, 1.8 mL.‡

c. Microsyringes, 10- and 100-μL.

d. Microsyringe, 25-μL with a 51- by 0.15-mm needle.§

e. Pipets, 2.0- and 5.0-mL transfer.

f. Volumetric flasks, 10- and 100-mL, glass stoppered.

g. Standard solution storage containers, 15-mL bottles with TFE-lined screw caps.

h. Gas chromatograph:‖ See Section 6200B.2*b*. The system is equipped with a linearized electron capture detector and a capillary column splitless injector.

Two GC columns are recommended. Column 1 is a highly efficient column that provides separations for EDB and DBCP without interferences from trihalomethanes. Use Column 1 as the primary analytical column unless routinely occurring compounds are not adequately resolved. Use Column 2 as a confirmatory column when GC/MS confirmation is not available.

1) *Column 1,* 30 m long × 0.32-mm ID fused silica capillary with dimethyl silicone mixed phase.# See Table 6231:I. Injector temperature: 200°C; detector temperature: 290°C. See Figure 6231:1 for a sample chromatogram.

2) *Column 2* (confirmation column), 30 m long × 0.32-mm ID fused silica capillary with methyl polysiloxane phase.** See Table 6231:I. Injector temperature: 200°C; detector temperature: 290°C.

4. Reagents

a. Reagent water: See Section 6200B.3*a*.

b. Hexane extraction solvent, UV grade.††

c. Methanol, pesticide quality or equivalent.

d. Sodium chloride, NaCl: Before using, pulverize and place in a muffle furnace at room temperature. Increase temperature to 400°C for 30 min. Store in capped bottle.

e. 1,2-Dibromoethane, 99%.‡‡

‡ Varian #96-000099-00 or equivalent.
§ Hamilton 702N or equivalent.
‖ Gas chromatographic methods are extremely sensitive to the materials used. Mention of trade names by *Standard Methods* does not preclude the use of other existing or as-yet-undeveloped products that give *demonstrably* equivalent results.
Durawax-DX3, 0.25-μm film, or equivalent.
** DB-1, 0.25-μm film, or equivalent.
†† Burdick and Jackson #216 or equivalent.
‡‡ Such as that available from Aldrich Chemical Company.

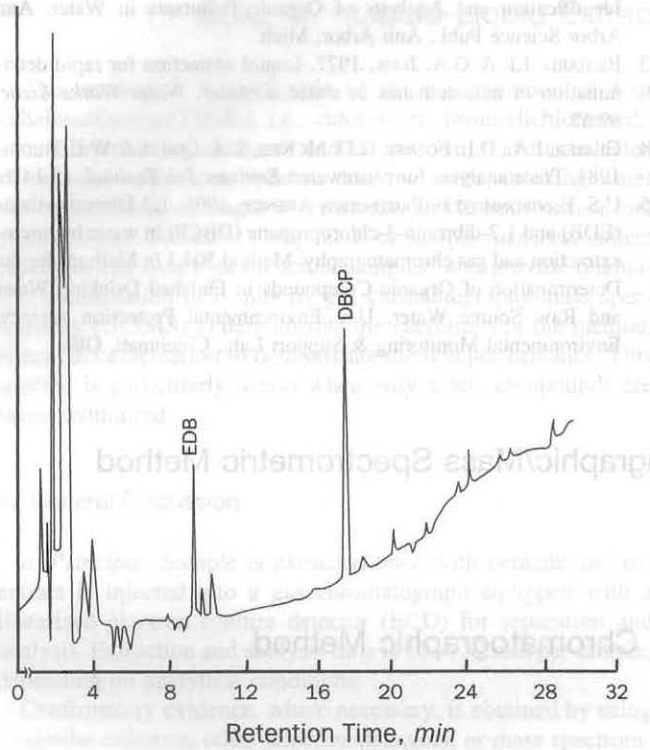

Figure 6231:1. Extract of reagent water with 0.114 μg/L added EDB and DBCP. Column: fused silica capillary; liquid phase: Durawax-DX3; film thickness: 0.25 μm; column dimensions: 30 m × 0.317 mm ID.

f. 1,2-Dibromo-3-chloropropane, 99.4%.§§

g. Standard stock solutions: See Section 6200B.3*g*. Store in 15-mL bottles with TFE-lined screw caps. Methanol solutions prepared from liquid standard materials are stable for at least 4 weeks when stored at 4°C.

h. Secondary dilution standards: See Section 6200B.3*h*. Dilution standards are as stable as stock solutions.

5. Procedure

a. Operating conditions: Table 6231:I summarizes recommended operating conditions for the gas chromatograph and provides estimated retention times.

b. Calibration: Prepare calibration standards as directed in Section 6200B.3*j* and analyze according to ¶ 5*d* below. Follow rest of calibration procedure in Section 6200A.5*b*), but limit variations from predicted response to ± 15% rather than ± 20%.

c. Instrument performance: See Section 6200C.4*c*.

d. Sample analysis: Let samples and standards come to room temperature. For samples and trip blanks, open bottle, discard 5 mL using a 5-mL transfer pipet, and replace container cap. Weigh to nearest 0.1 g; record weight for subsequent volume determination. For calibration standards, QC check standards, and reagent blank, measure 35 mL using a 50-mL graduated cylinder and transfer to a 40-mL sample container.

§§ Such as that available from AMVAC Chemical Corporation, Los Angeles, CA.

TABLE 6231:II. SINGLE-LABORATORY PRECISION AND BIAS FOR EDB AND DBCP IN TAP WATER

Compound	Number of Samples	Addition μg/L	Average Bias %	Relative Standard Deviation %
1,2-Dibromoethane	7	0.03	114	9.5
	7	0.24	98	11.8
	7	50.0	95	4.7
1,2-Dibromo-3-chloropropane	7	0.03	90	11.4
	7	0.24	102	8.3
	7	50.0	94	4.8

Remove container cap and add 7 g NaCl. Add 2.0 mL hexane with a transfer pipet. Recap and shake vigorously by hand for 1 min. Let water and hexane phases separate. (If sample is stored at this stage, keep container upside down). Carefully transfer 0.5 mL of hexane layer into an autosampler vial using a disposable glass pipet. Transfer remaining hexane phase, but not any of the water phase, into a second autosampler vial. Hold second vial at 4°C for reanalysis if necessary. Transfer first sample vial to an autosampler set up to inject 2.0-μL portions into the gas chromatograph. Alternatively, manually inject 2-μL portions.

To determine sample volume for samples and field blanks, remove cap and discard remaining sample/hexane mixture. Shake off remaining drops using short, brisk wrist movements. Reweigh empty container with original cap and calculate net weight of sample by difference to the nearest 0.1 g. This net weight is equivalent to the volume of water (in mL) extracted. Alternatively, weigh vial before collection and reweigh full vial. Sample volume then equals gross weight (g) − [tare weight (g) + 5 g].

6. Calculation

Identify EDB and DBCP in sample chromatogram by comparing retention time of suspect peak to retention times generated by calibration and laboratory control standards. Retention times of samples should be within ± 0.1 min of standard for positive identification.

Use calibration curve or calibration factor to calculate uncorrected concentration (C_i) of each compound (e.g., calibration factor × response). Calculate sample volume (V_s) as equal to the net sample weight:

$$V_s = \text{gross weight} - \text{bottle tare}$$

The corrected sample concentration is:

$$\text{Concentration, } \mu g/L = C_i \times \frac{35}{V_s}$$

Round off results to the nearest 0.1 μg/L or two significant figures.

7. Quality Control

Follow procedures given in Section 6200A.5.

8. Precision and Bias

Single-laboratory precision and bias at several concentrations in tap water are presented in Table 6231:II.[5]

9. References

1. GLAZE, W.H. & C.C. LIN. 1984. Optimization of Liquid-Liquid Extraction Methods for Analysis of Organics in Water. EPA-600/S4-83-052, U.S. Environmental Protection Agency.
2. HENDERSON, J.E., G.R. PEYTON & W.H. GLAZE. 1976. A convenient liquid-liquid extraction method for the determination of halomethanes in water at the parts-per-billion level. *In* L. H. Keith, ed. Identification and Analysis of Organic Pollutants in Water. Ann Arbor Science Publ., Ann Arbor, Mich.
3. RICHARD, J.J. & G.A. JUNK. 1977. Liquid extraction for rapid determination of halomethanes in water. *J. Amer. Water Works Assoc.* 69:62.
4. GLASER, J.A., D.L. FOERST, G.D. MCKEE, S. A. QUAVE & W.L. BUDDE. 1981. Trace analyses for wastewater. *Environ. Sci. Technol.* 15:1426.
5. U.S. ENVIRONMENTAL PROTECTION AGENCY. 1995. 1,2-Dibromoethane (EDB) and 1,2-dibromo-3-chloropropane (DBCP) in water by microextraction and gas chromatography. Method 504.1 *in* Methods for the Determination of Organic Compounds in Finished Drinking Water and Raw Source Water. U.S. Environmental Protection Agency, Environmental Monitoring & Support Lab., Cincinnati, Ohio.

6231 C. Purge and Trap Gas Chromatographic/Mass Spectrometric Method

See Section 6200B for capillary-column method.

6231 D. Purge and Trap Gas Chromatographic Method

See Section 6200C for capillary-column method.

6232 TRIHALOMETHANES AND CHLORINATED ORGANIC SOLVENTS*

6232 A. Introduction

1. Sources and Significance

The trihalomethane (THM) compounds have been found in most chlorinated water supplies in the United States; typically they are produced in the treatment process as a result of chlorination. The formation of these compounds is a function of precursor concentration, contact time, chlorine dose, and pH. Toxicological studies suggest that chloroform is a potential human carcinogen. Consequently, total trihalomethanes are being regulated in potable waters. Chlorinated organic solvents are found in many raw waters because of industrial contamination.

2. Selection of Method

Several methods are available for measurement of the trihalomethanes and chlorinated organic solvents. Some of these are specific for these compounds and others have a much broader spectrum. Method 6232B is a simple liquid-liquid extraction gas chromatographic (GC) method that is highly sensitive and very precise for these compounds and certain other chlorinated solvents. Method C refers to purge and trap gas chromatographic/mass spectrometric (GC/MS) methods that can detect not only THMs but also a wide variety of other compounds. Method D refers to purge and trap GC methods with similar target compounds. All of these methods have approximately the same sensitivity for the trihalomethanes; method choice depends on availability of equipment, operator choice, and the list of desired target compounds. In addition, closed-loop stripping analysis can be used for several of these compounds (see Section 6040).

---

* Approved by Standard Methods Committee, 2000.
Joint Task Group: 20th Edition — Nancy E. Grams (chair), Bradford R. Fisher.

6232 B. Liquid-Liquid Extraction Gas Chromatographic Method

This method[1–3] is applicable to the determination of four trihalomethanes (THMs), i.e., chloroform, bromodichloromethane, dibromochloromethane, and bromoform, and the selected chlorinated solvents in finished drinking water, drinking water during intermediate stages of treatment, and in both surface and ground water. For other compounds or sample matrices, collect precision and bias data on actual samples[4] and provide qualitative confirmation of results by gas chromatography/mass spectrometry (GC/MS) to demonstrate the usefulness of the method. Retain documentation to demonstrate method performance. This method is particularly useful when only a few compounds are being monitored.

1. General Discussion

a. Principle: Sample is extracted once with pentane and the extract is injected into a gas chromatograph equipped with a linearized electron capture detector (ECD) for separation and analysis. Extraction and analysis time is 10 to 30 min per sample, depending on analytical conditions.

Confirmatory evidence, where necessary, is obtained by using dissimilar columns, other selective detectors, or mass spectrometry. Component concentrations must be sufficiently high (i.e., >50 μg/L) for confirmatory analyses using a mass spectrometer. See other methods in this section for alternative means of confirming positive results.

Standards added to organic-free water and samples are extracted and analyzed in the same manner, under identical conditions. This step is essential to adjust for the less-than-100% extraction efficiency of the simplified extraction technique. Extreme differences in ionic strength or organic content between standards and samples can result in different equilibria of sample constituents with the extracting solvent and a method bias may result. Monitor known additions recoveries on various matrices for bias.

Where required, sum the concentrations of the four trihalomethanes and report as total trihalomethanes in micrograms per liter.

See Section 6010 for discussion of gas chromatographic principles.

b. Interferences: Impurities contained in the extracting solvent account for many analytical problems. Maintain records of the reagent's manufacturer, lot number, purity, date bottle was opened, and expiration date. Analyze solvent blanks before using a new bottle of solvent. Make indirect daily checks on the extracting solvent by monitoring the method blanks. Whenever an interference is noted in the method blank, analyze a solvent blank. Discard (or use for another purpose) extraction solvent if a high level of interference is traced to it. Low-level interferences can be removed by distillation or column chromatography;[5] however, it usually is more economical to obtain new solvent or select an approved alternative solvent. Interference-free solvent is defined as a solvent containing less than the laboratory determined detection limit of interference for each constituent. Protect interference-free solvents by storing in an area known to be free of organohalogen solvents. *Do not subtract blank values from sample analysis results as a correction for contamination.*

Sample contamination has been attributed to diffusion of volatile organics through the septum seal on a sample bottle during shipment and storage. Use the trip blank to monitor for this problem.

Contamination also may occur whenever equipment and materials used to store, extract, or analyze samples are inadequately cleaned, prepared, tested, or stored. There are many sources of contamination including contamination of reagents during storage and contamination of equipment reused in the sequential extraction of samples and standards. Maintain records of dates of preparation and cleaning and inclusive dates of use of reagents, standards, bottles, and equipment. Test all reagents and standards before initial use. To reduce possibility of carryover contamination, always clean equipment thoroughly after each use. Where equipment contamination is a concern, processing and analysis of additional method blanks beyond the minimum requirements of this method may be useful. Start by placing reagent water in a sample vial of the same lot that was used for samples and add preservative as was done for samples. Process this method blank in conjunction with samples using the same reagents, materials, and equipment. Where analysis of a method blank indicates contamination, investigate possible sources and isolate the cause. Take and document corrective action. Following analysis of a sample containing late-eluting interferences, or containing over-range concentrations of constituents of interest, analyze a solvent blank or method blank to demonstrate freedom from carry-over.

This liquid-liquid extraction technique efficiently extracts a wide boiling range of nonpolar organic compounds and also extracts polar organic components with varying efficiencies. To analyze rapidly for trihalomethanes and chlorinated solvents with sensitivities in the low microgram-per-liter range in the presence of these other organic compounds, use the semi-specific electron capture detector. Trihalomethanes are primarily products of the chlorination process and seldom appear in raw unchlorinated source water. The absence of peaks with retention times similar to the trihalomethanes in raw source water analysis is supporting evidence of an interference-free finished drinking water analysis. Because of possible interferences, analysis of a representative raw source water when analyzing finished drinking water provides evidence of freedom from this interference source. When potential interferences are noted in the raw source water, use the alternate chromatographic columns to reanalyze the sample set. If interferences still are noted, make confirmatory qualitative identifications as directed in ¶ 1*a*. If the peaks are determined to be other than the constituents of interest and they add significantly to the constituents' value in the finished drinking water, analyze sample set by the purge and trap method.[6]

Where chlorinated solvents are present in finished drinking water the most likely source is the raw water. Analyze individual raw water samples to isolate the source of contamination. Always consider the possibility of coeluting interferences. Analysis using capillary chromatography minimizes this possibility. Analysis using dissimilar columns may confirm the presence of interferences through differences in retention time between the constituent of interest and the unknown compound in the sample;

however, the most definitive confirmation routinely available is GC/MS.[7]

Because the simplified extraction technique depends on equilibria between solvent and water, because extraction is not 100% efficient, and because efficiency is dependent on concentration, it is important to: extract samples and standards in the same manner; monitor matrix recoveries to assess differences in equilibria; and, where the concentration is found to be above the linear range of the method, to either dilute samples carefully before extraction, or prepare standards in water at the estimated sample concentration and carefully dilute *both* sample and standard extracts. Always process standards and samples together and hold constant such variables as water temperature, solvent temperature, room temperature, extraction time, and separation times.

For further information on interferences in gas chromatographic methods, see Section 6010C.

c. Detection levels: The method is useful for trihalomethane and selected chlorinated solvents at concentrations from approximately 0.1 to 200 μg/L. Actual detection levels are highly dependent on the characteristics of the gas chromatographic system used, the ratio of solvent to water, and interferences present in the solvent. See Section 1030.

2. Sampling and Storage

See Sections 6010B.1 and 5710.

If trihalomethane formation potential is to be measured, do not add any preservatives at the time of sample collection. If chemical stabilization is not used at time of sampling, add the reducing agent just before extracting the sample or add it at the time specified in the formation potential method for quenching the sample.[8,9]

The raw source water sample history should resemble that of the finished drinking water. Take into account the average retention time of the finished drinking water within the water plant when sampling the raw source water.

Store blanks and samples, collected at a given site (sample set), together in a protected area known to be free from contamination. At a water treatment plant, duplicate raw source water, duplicate finished water, and duplicate sample blanks comprise the minimum sample set. When samples are collected and stored under conditions specified in 6010B.1, no measurable loss has been detected over extended periods of time.[8] Analyze samples within 14 d of collection.

For samples collected soon after chlorination, quenching with reducing agent may not be sufficient to prevent further formation of THMs completely, because of hydrolysis of intermediates. In that case, acidification is necessary and consistent with the recommended preservation techniques.

3. Apparatus

a. Sample storage vials: Clean, baked 40-mL glass open screw-top TFE-faced septum VOA vials or equivalent. See Section 6010B for additional information on cleaning, storage, and preparation.

b. Microsyringes, 10, 25, and 100-μL. Microsyringes with extended barrels are suggested for proper injection of methanolic standards when preparing aqueous standards in volumetric flasks.

c. Volumetric flasks, glass-stoppered, 10, 25, 50, 100, 250, 500, and 1000 mL, Class A. Choose size according to final volume of aqueous standard required and concentration of methanolic standards.

d. Extraction vessels: Use sample storage vials. Optionally, where samples are transferred to smaller extraction vessels, select an extraction container on the basis of specific requirements for final extract volume, solvent-to-water volume ratio, and availability. If a separate vessel is used for extraction, place standards along with samples in sample storage vials and follow identical procedures for preservation and transfer to the extraction vessel. Use the same lots of vessels for sample and standard extraction. Use of clean, oven-baked glass vessels and TFE-faced septa is critical.

e. Extract storage vials: 1.8-mL autosampler vials with open screw-top caps and TFE septa, or equivalent.

f. Gas chromatograph, preferably temperature-programmable with linearized electron-capture detector.

*g. Chromatographic columns:**

1) 0.32-mm ID × 30 m fused silica capillary, 1 μm DB-5,† or equivalent, at linear velocity of 20 cm/s. Temperature program: 35°C for 5 min, ramp 10°C/min to 70°C, then 20°C/min to 200°C. See Figure 6232:1 for a typical standard chromatogram.

2) 0.53-mm ID × 30 m, 1.5 μm DB-5† or equivalent, at 25 cm/s. Starting at 30°C for 1 min, ramp 6°C/min to 150°C.

3) 2-mm ID × 2 m long glass packed with 1% SP-1000‡ on Carbopack B (60/80) operated at 50°C with 60 mL/min flow, or, if temperature-programmable GC is available, 45°C for 1 min, ramp 8°C/min to 240°C.

4) 2-mm ID × 2 m long glass packed with 10% squalene on Chromosorb WAW (80/100 mesh) operated at 67°C with 25 mL/min flow.

5) 2-mm ID × 3 m long glass packed with 6% OV-11/4% SP-2100 on Supelcoport (100/120 mesh); temperature program 45°C for 12 min, then 1°C to 70°C with 25-mL/min flow rate.

h. Mechanical shaker: Optionally, a rotary platform shaker.

i. Solvent pipettor: 2-mL transfer pipet, pipettor, or all-glass and TFE repipettor that attaches to the pentane storage bottle.

j. Transfer pipets, 5 mL. Preferably use a pipettor with disposable tips, cleaned and dried as recommended for TFE septa.

k. Analytical balance, capable of measuring to ± 0.01 g.

4. Reagents

a. Extraction solvent: See ¶ 1*b*. For capillary column split injection technique, preferably use only pentane. For other techniques, recommended solvent is pentane; alternatively, use hexane, methylcyclohexane, methyl-*tert*-butyl ether (MtBE) or 2,2,4-trimethylpentane. For alternative solvents, collect and document precision and bias data, evaluate extraction efficiency and effect of constituent concentration on efficiency, and maintain documents demonstrating

* Chromatographic methods are extremely sensitive to the materials used. Mention of trade names by *Standard Methods* does not preclude the use of other existing or as-yet-undeveloped products that give *demonstrably* equivalent results.
† J&W Scientific.
‡ Supelco.

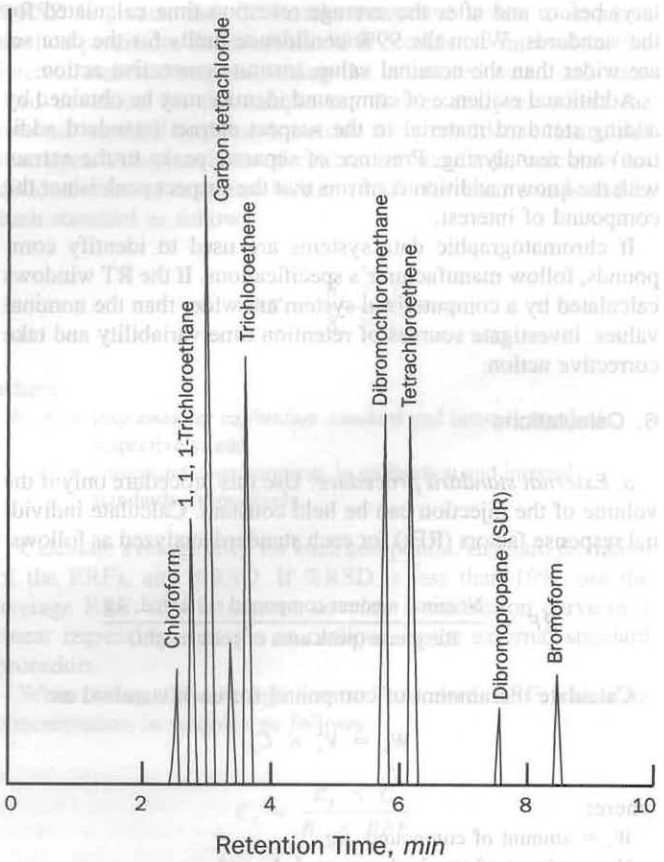

Figure 6232:1. Chromatogram for THMs and chlorinated organic solvents. Concentration was 50 μg/L for each compound; primary column DB-5.

applicability. Demonstrate that solvent is free of compounds for which the analysis is being performed.

b. Methyl alcohol, demonstrated to be free of interferences.

c. Neat standard materials: Use materials of 96% purity or greater for:

1) *Calibration standards:* bromoform ($CHBr_3$), bromodichloromethane (BDCM), dibromochloromethane (DBCM), chloroform ($CHCl_3$), 1,1,1-trichloroethane (TCA), tetrachloroethene (PCE), trichloroethene (TCE), and carbon tetrachloride (CCl_4).

2) *Internal standard:* 1,2 dibromopropane. A compound selected as an internal standard should have baseline resolution to separate it from constituents of interest and any interferences. Because this requirement is highly dependent on the samples and the analytical conditions and equipment used, no single internal standard is universally applicable. 1,2-dibromopropane has been found to be generally useful.

d. Reagent water: Generate VOC-free water, defined as water free of interference when used in the procedure described herein, by passing tap water through a carbon filter. Alternatively, prepare VOC-free water as follows: boil water for 15 min, then maintain at 90°C while bubbling a contaminant-free inert gas through water at 100 mL/min for 1 h. While water is still hot, transfer to a narrow-mouth screw-cap bottle with a TFE seal. Test VOC-free water each day before use by analyzing a method blank for constituents of interest. If any chlorine residual remains after such treatment, destroy it. See Section 1080 for additional information and general discussion of reagent-grade water.

e. Stock standard solutions: See Section 6200B.3*g*. Alternatively, purchase prepared standard solutions in methanol.

CAUTION: *Trihalomethanes and chlorinated solvents are toxic and may be carcinogenic: prepare primary stock solutions in a hood and wear appropriate personal protective equipment.*

f. Secondary dilution standards: From standard stock solutions, prepare multi-component secondary standards in methyl alcohol so that standards over the working range of the instrument can be prepared using no more than 20 μL methanolic standard solution /100 mL reagent water. See Method 6200B.*3h*.

g. Internal standard solution: Prepare stock solution from neat material in hexane. Make secondary dilution directly into storage container of pentane extracting solvent to produce a concentration of 30 μg/L internal standard in pentane.

h. Aqueous calibration standards: Construct a calibration curve for each constituent using a minimum of three different concentrations, but preferably use five to seven concentrations. Bracket each sample with two of the concentrations. Use one concentration near, but above, the laboratory-determined detection limit. Where a sample component exceeds the range bracketed by standards, dilute a fresh volume of sample and re-extract, or prepare new standards in reagent water to bracket the concentration and dilute sample and standard extracts to bring them into the linear range of the detector. To prepare calibration standards, rapidly inject the required volume of alcoholic standard into the expanded area of a reagent-water-filled volumetric flask. Using an extended barrel syringe, inject the methanol *well below* the reagent water surface. Preferably incline the volumetric flask at an approximately 45-deg angle while injecting the standard. Remove syringe and stopper flask. Mix aqueous standards by gently inverting flask three times only. Discard to waste the contents in the neck of flask before transferring standards to sample vials. Add any preservatives to both samples and standards before extraction. Process standards through extraction in conjunction with sample sets. Aqueous standards, when stored with a headspace, are not stable; discard after 1 h. When stored in headspace-free sample storage vials, aqueous standards may be used for 24 h.

Avoid standard preparation procedures that require delivery of less than 10 μL of methanolic standards into volumetric flasks. Instead, use a larger volumetric flask and a larger volume of methanolic standard.

i. Quality control (QC) check standards: Obtain concentrate in methanol from a vendor of proficiency-testing materials or NIST for each compound, or if not available, from a second source vendor. If no second source is available, prepare stock standards separately from neat materials used for calibration standards. Prepare a mixed secondary dilution standard containing each compound and then an aqueous QC check standard at a concentration approximating the midlevel calibration standard.

5. Procedure

a. Extraction: Let samples and standards come to room temperature. Open each sample vial and remove 5 mL of sample and discard to waste, preferably using a transfer pipettor with disposable tips. Replace cap, weigh vial to nearest 0.1 g, and record

weight. Alternatively, use a 25-mL gastight syringe to measure sample volume (6200B.4*d*) and perform subsequent extraction.

Using a clean, dedicated volumetric measurement device (¶ 3*i*) carefully measure 2.00 mL pentane and add to sample vial. *Vigorously* shake by hand for 1 min or use a rotary platform shaker set at 60 to 100 rpm.

Let phases separate for at least 2 min. Where emulsions do not separate on standing, centrifuge or transfer entire emulsion to a separate vial and cool extract below 4°C to promote separation. Using a disposable glass pipet, transfer at least 1 mL of upper pentane extract to extract storage vials. Optionally, transfer half of the pentane extract to each of two vials to provide for reanalysis where necessary. Protect pentane extracts from warm temperatures and minimize extract holding time at room temperature. Store extracts at 4°C.

Empty sample/extraction vial to waste, rinse, and shake dry. Reweigh empty container with original cap to nearest 0.1 g and record weight. Calculate weight of sample extracted to the nearest 0.1 g by subtraction of vial-only weight from sample-plus-vial weight. For an assumed density of 1 g/mL, weight of sample extracted is equal to volume of sample extracted, in milliliters. Convert volume in milliliters to liters and record.

b. Sample and standard analysis: Before extraction of samples or standards, prepare and analyze a method blank to verify freedom from interferences. Once extracts have been prepared, analyze standards and calculate a calibration curve or calibration factor as outlined in ¶ 6 below. Inject 1 to 5 μL of standard extract depending on the configuration of the instrument and the required sensitivity. Inject exactly the same volume of extract each time, preferably using an autosampler. To test that injection volumes are repeatable, inject replicates of a single standard extract, and determine the standard deviation. Percent relative standard deviation (%RSD) should not be more than 5%. If this precision is not routinely achievable, use the internal standard calibration procedure.

After calibration, analyze the method blank, samples, and quality control samples. Extract and analyze a quality-control check standard each twentieth analysis (5%) and at the end of the analytical sequence. The percent recovery for the QC check standard should be between 80 and 120%. Develop historical mean control charts of QC check standard recovery for each compound and use the 99% confidence about historical data as the control criteria for rejection of QC check standards validity. Where criteria are failed, repeat analysis of any samples analyzed since the last QC check standard was in control.

c. Internal standard analysis procedure: Add the internal standard to the pentane solvent in the storage container at the concentration specified, and proceed with extraction and analysis of samples and standards as outlined above.

d. Compound identification: Identification of compounds in samples is based on comparison of retention times (RT) of suspect peaks to the confidence limits for RT of the authentic compounds in standards. Using the retention times of the standards analyzed, determine the average retention time for each compound and the standard deviation of the retention time. Tentatively identify peaks in sample chromatograms as compounds on basis of the 99% confidence interval around the calculated mean value using the calculated standard deviation. Nominally, the retention time window would be expected to be no wider than 0.25 min (packed column) and 0.05 min (capillary) before and after the average retention time calculated for the standards. When the 99% confidence limits for the data set are wider than the nominal value, institute corrective action.

Additional evidence of compound identity may be obtained by adding standard material to the suspect extract (standard addition) and reanalyzing. Presence of separate peaks in the extract with the known addition confirms that the suspect peak is not the compound of interest.

If chromatographic data systems are used to identify compounds, follow manufacturer's specifications. If the RT windows calculated by a computerized system are wider than the nominal values, investigate sources of retention time variability and take corrective action.

6. Calculations

a. External standard procedure: Use this procedure only if the volume of the injection can be held constant. Calculate individual response factors (RFs) for each standard analyzed as follows:

$$RF = \frac{\text{Nominal amount compound extracted, } \mu g}{\text{Response (peak area or peak height)}}$$

Calculate the amount of compound for each standard as:

$$W_s = V_s \times C_s$$

where:

W_s = amount of compound, μg,
V_s = volume of standard extracted, L, and
C_s = concentration of prepared standard, μg/L.

For each compound determine average RF and standard deviation of the RFs using all calibration standards analyzed. If the percent relative standard deviation [%RSD =(SD/mean RF) × 100] is less than 10%, use average RF to calculate sample concentration.

If the %RSD is greater than 10%, plot a calibration curve of amount injected versus response. Use the graph to determine the amount of compound present in each sample. Then determine the concentration by dividing amount, μg, by the volume, L, of sample extracted. Optionally use a data system to prepare a linear regression and use the linear regression equation to calculate compound amounts in samples from response values.

Where average RF is used, determine sample concentration as follows:

$$C_x = \frac{RF \times R_x}{V_x}$$

where:

C_x = compound concentration, μg/L,
R_x = sample response (mm, area, etc.), and
V_x = volume of sample extracted, L.

Round all final sample results to two significant figures.

b. Internal standard procedure: For all analyses made in a given analytical sequence, determine average internal standard response and standard deviation of the internal standard response. Calculate percent relative standard deviation. If the

%RSD is greater than 25%, take corrective action to improve method precision. Establish the 99% confidence interval for the internal standard response using the calculated mean and standard deviation for the sample set. Reject analyses where the internal standard response is outside these confidence limits, and reanalyze. After analysis of calibration standards, calculate individual relative response factors (RRF) for each compound in each standard as follows:

$$RRF = \frac{R_s \times C_i}{R_i \times C_s}$$

where:

R_s, R_i = responses for calibration standard and internal standards, respectively, and
C_s, C_i = compound concentrations in calibration and internal standards, respectively.

Calculate average RRF for each compound, standard deviation of the RRFs, and %RSD. If %RSD is less than 10%, use the average RRF; if it is greater, develop a calibration curve or a linear regression equation as outlined in the external standard procedure.

When using the average internal standard RRF, calculate concentration in samples as follows:

$$C_x = \frac{R_x \times C_i}{R_i \times RRF}$$

where:

C_x = compound concentration in sample, µg/L, and
R_x = sample response.

c. Total trihalomethane concentration: Calculate total trihalomethane concentration by summing the concentration of the four individual trihalomethanes in each sample. This is required for USEPA reporting purposes but it is preferable to report only individual THMs.

7. Quality Control

A minimum program of quality control consists of an initial demonstration of proficiency for each analyst and each instrument system and an ongoing program of quality control analysis. Record initial quality by documenting initial performance relative to published performance criteria. Maintain records of performance by comparing ongoing quality control checks to performance criteria and objectives for data quality. Document this performance as outlined in Sections 1010, 1030, and 6020.

a. Analyst proficiency: The analyst should be experienced in the operation of a GC/ECD and produce an initial demonstration of proficiency in accordance with the procedure outlined in 6200A.5*a*1).

b. Method blanks: Prepare and analyze method blanks on each shift after calibration and before first sample analysis. Concentrations of compounds in the method blank should not exceed the experimentally determined method detection level. If the method blank is out of control isolate the source of contamination, apply corrective action, and process a new method blank. Under no circumstances subtract method blank values from the sample result.

c. Quality control (QC) check standards: Preferably obtain QC standards from a separate source and prepare independently from calibration standards. Analyze QC check standards as though they were samples at a frequency of 5% (every 20 samples) and at the end of the analytical sequence. Compare results to known concentration of the check standard and calculate percent recovery. Percent recovery nominally should be between 80 and 120%. Develop mean recovery control charts of QC check standards results and use historical 99% confidence limits to accept or reject the ongoing calibration. Where historical confidence limits are wider than the nominal limits, investigate standard materials, preparation and storage procedures, and other potential sources of error. Take and document corrective actions.

d. Detector sensitivity: Maintain a log of detector response, in area counts or peak height, using one standard that is analyzed each day, to monitor changes in detector sensitivity. Optionally, plot these data to observe trends in detector sensitivity. Note the sensitivity at which method detection level studies were performed and replace or repair detectors where minimum detectable quantities are significantly affected by declining detector sensitivity.

e. Laboratory-fortified samples with known additions: In a laboratory analyzing more than 10 samples daily, extract and analyze a known addition on each tenth sample. Be sure this is representative of different sample types because there is some evidence of matrix effects with liquid-liquid extraction methods. See Section 1020B.6. In a laboratory analyzing fewer than 10 samples daily, each time sample extractions are performed, extract and analyze at least one laboratory-generated known-addition sample. Chart percent recovery as outlined in Section 1020B.12 using a means chart. To evaluate method bias, see Section 1030.

f. Duplicate analysis: Randomly select, then extract and analyze in duplicate, 10% of all samples. Maintain an up-to-date log on bias and precision data collected on known-addition samples and duplicate samples. Evaluate results as outlined in Section 1030. If results are significantly different from those cited in ¶ 8 below, check entire analytical scheme to determine why the laboratory's precision and bias limits are excessive.

g. Laboratory control standards (performance evaluation standards): Quarterly, add an external reference laboratory evaluation standard to organic-free water, extract, and analyze. Obtain this standard from an authorized proficiency-testing provider. The results from this sample should agree within 20% of the true value for each compound. If not, check each step in preparation and analysis to isolate the problem. Document external reference standard results and any corrective action taken.

8. Precision and Bias

The single-laboratory precision and bias data in Table 6232:I were generated by adding known amounts of trihalomethanes and chlorinated organic solvents to organic-free water. The mixtures were analyzed as true unknowns.

TABLE 6232:I. PRECISION AND BIAS DATA FOR THM-CHLORINATED ORGANIC SOLVENT METHOD, DB-5 COLUMN

Compound	Added Amount	Amount Recovered µg/L A	B	C	D	E	F	G	H	Bias % Recovery	Precision % RSD
Chloroform ($CHCl_3$)	20.0	18.7	18.6	19.4	19.5	19.2	18.5	19.5	19.7	95.6	2.4
Bromodichloromethane (BDCM)	20.0	19.8	20.2	20.7	20.8	20.3	19.7	20.6	20.5	101.6	2.1
Dibromochloromethane (DBCM)	20.0	18.7	19.4	20.0	20.2	19.7	18.7	20.1	20.6	98.3	3.6
Bromoform ($CHBr_3$)	20.0	17.4	18.5	18.7	19.2	19.3	17.9	18.8	19.8	93.5	4.1
Trichloroethane (TCA)	20.0	18.5	18.8	19.7	19.9	19.8	18.5	20.1	20.4	97.3	3.8
Carbon tetrachloride (CCl_4)	20.0	20.1	20.4	20.0	20.2	20.6	20.0	20.1	20.0	100.8	1.2
Trichloroethene (TCE)	20.0	17.9	18.3	18.9	19.2	19.1	17.9	19.2	19.6	93.8	3.4
Tetrachloroethene (PCE)	20.0	19.8	20.4	20.6	20.9	20.7	19.7	20.7	20.7	102.2	2.2
Internal standard	100.0	99.0	95.0	100.0	102.0	101.0	99.0	105.0	105.0	100.8	3.3

9. References

1. U.S. ENVIRONMENTAL PROTECTION AGENCY. 1980. Analysis of trihalomethanes in drinking water by liquid/liquid extraction. 40 CFR Part 141, Appendix C. Part II.
2. MIEURE, J.P. 1977. A rapid and sensitive method for determining volatile organohalides in water. *J. Amer. Water Works Assoc.* 69:60.
3. REDING, R., W. KOLLMAN, M. WEISNER & H. BRASS. 1978. THM's in drinking water: Analysis by LLE and comparison to purge and trap. *In* C.E. Van Hall, ed. Measurement of Organic Pollutants in Water and Wastewater. STP 686, American Soc. Testing & Materials, Philadelphia, Pa.
4. Handbook for Analytical Quality Control in Water and Waste Water Laboratories. 1972. Analytical Quality Control Lab., National Environmental Research Center, Cincinnati, Ohio.
5. RICHARD, J.J. & G.A. JUNK. 1977. Liquid extraction for rapid determination of halomethanes in water. *J. Amer. Water Works Assoc.* 69:62.
6. U.S. ENVIRONMENTAL PROTECTION AGENCY. 1979. The Analysis of Trihalomethanes in Finished Water by the Purge and Trap Method. Environmental Monitoring & Support Lab., Environmental Research Center, Cincinnati, Ohio.
7. BUDDE, W.L. & J.W. EICHELBERGER. 1979. Organic Analysis Using Gas Chromatography-Mass Spectrometry. Ann Arbor Science Publ., Ann Arbor, Mich.
8. BRASS, H.J., et al. 1977. *In* R.B. Pojasek, ed. National Organic Monitoring Survey: Sampling and Purgeable Organic Compounds. Drinking Water Quality Through Source Protection, p. 398. Ann Arbor Science Publ., Ann Arbor, Mich.
9. KOPFLER, F.C., et al. 1976. GC/MS determination of volatiles for the National Organics Reconnaissance Survey (NORS) of Drinking Water. *In* L.H. Keith, ed. Identification and Analysis of Organic Pollutants in Water. Ann Arbor Science Publ., Ann Arbor, Mich.
10. WHITE, L.D., et al. 1970. Convenient optimized method for the analysis of selected solvent vapors in industrial atmosphere. *Amer. Ind. Hyg. Assoc. J.* 31:225.

6232 C. Purge and Trap Gas Chromatographic/Mass Spectrometric Method

See Section 6200B for capillary-column method.

6232 D. Purge and Trap Gas Chromatographic Method

See Section 6200C for capillary column method.

6251 DISINFECTION BY-PRODUCTS: HALOACETIC ACIDS AND TRICHLOROPHENOL*

6251 A. Introduction

1. Sources and Significance

The haloacetic acids (HAAs) are formed by the chlorination of natural organic (humic and fulvic) matter. Utilities using chlorine as a water disinfectant generate haloacetic acids, usually as the second most prevalent group of known disinfection by-products[1]; the primary group formed is usually the trihalomethanes. Toxicological studies indicate that dichloroacetic acid and trichloroacetic acid are animal carcinogens.[2] The USEPA has promulgated a maximum contaminant level for the sum of five haloacetic acids.[3]

* Approved by Standard Methods Committee, 1994.
Joint Task Group: Sanwat N. Chaudhuri (chair), Russell Chinn, Patricia Snyder Fair, Yuefeng Xie.

This section is currently undergoing revision. It is reprinted from the 20th Edition with editorial changes recommended by the Joint Task Group.

2. References

1. U.S. Environmental Protection Agency & Association of Metropolitan Water Agencies. 1989. Disinfection By-Products in U.S. Drinking Water. Vol. 1 Report, James M. Montgomery Consulting Engineers, Pasadena, Calif.
2. Alceon Corp. 1993. An Overview of Available Information on the Toxicity of Drinking Water Disinfectants and Their By-Products. Cambridge, Mass.
3. U.S. Environmental Protection Agency. 1998. Disinfectants and Disinfection Byproducts Final Rule. *Federal Register* 63(241): 69390.

6251 B. Micro Liquid-Liquid Extraction Gas Chromatographic Method

This method[1] was developed to analyze simultaneously for monochloroacetic acid (MCAA), monobromoacetic acid (MBAA), dichloroacetic acid (DCAA), trichloroacetic acid (TCAA), bromochloroacetic acid (BCAA), dibromoacetic acid (DBAA), and 2,4,6-trichlorophenol (TCPh) in treated and untreated drinking water. Additional haloacetic acids may be present and analyzed by this method; however, standards for these compounds are not readily available and are less stable.

1. General Discussion

a. Principle: The sample is extracted with methyl *tertiary*-butyl ether (MtBE) at an acidic pH to extract the nondissociated acidic compounds to be determined. A salting agent is added to increase extraction efficiency. The extracted compounds are methylated with diazomethane solution to produce methyl ester or ether derivatives that can be separated chromatographically. A temperature-programmable gas chromatograph using a fused silica capillary column and an electron capture detector (ECD) is used for analysis. Simultaneous analysis and confirmation using a single injection can be effected by setting up both the analytical column and the confirmation column to share a common injection port. Alternatively, use separate analytical and confirmation columns. Alternative detectors may be used if QC criteria can be met. Aqueous calibration standards are extracted, methylated, and analyzed in the same manner to compensate for less than 100% extraction and derivatization efficiencies.

b. Interferences: Impurities in extraction solvent and on glassware and other equipment can interfere. Follow specifications and cleaning procedures carefully to minimize interference. As shown in Figure 6251:1, the analysis separates peaks of haloacetic acids from those of other common disinfection by-products. Use of two columns is recommended because for waters with high carbonate contents, false-positive MCAA peaks have been observed on the column described in ¶ 3*h*4).

c. Detection levels: Method detection levels (MDL) are given in Table 6251:I.[2] The method has been shown to be useful for haloacetic acids over a working range of 0.5 to 30 µg/L (1.0 to 30 µg/L for MCAA) and 0.25 to 15 µg/L for TCPh. The calibration range can be extended, depending on the compound and detector characteristics.

d. Safety: The toxicity and carcinogenicity of each reagent has not been defined precisely. Minimize exposure to these chemicals and use them only in a properly operating ventilation hood.

Avoid exposure to DCAA and TCAA because they are carcinogens.[3] Avoid contact with the other haloacetic acids and their solutions.

MNNG (1-methyl-3-nitro-1-nitrosoguanidine) is carcinogenic. Keep in properly labelled plastic containers, containing activated carbon, with tight-fitting lids and store in a refrigerator used only for chemical storage. Store spatulas and glassware for the handling of MNNG in specially labelled plastic containers and use only for MNNG.

Diazomethane is toxic, carcinogenic, and an explosion hazard.[4] Follow special precautions whenever handling this material. Use only in a properly operating fume hood; Caution: *Do not breathe vapors.* To avoid explosions, do not heat above 90°C and do not use glassware with ground-glass surfaces (e.g., ground-glass joints, sleeve bearings) or glass stirrers. Special glassware for diazomethane generation and handling, as well as screw-cap volumetric flasks, are available commercially. Always use a safety shield when generating diazomethane. Always quench excess diazomethane with silica gel. Do not store diazomethane/ether solutions; they are extremely hazardous and tend to become contaminated.

Store ether in tightly-closed amber bottles in an explosion-safe or -proof refrigerator. Store only with compatible chemicals. Eliminate all sources of ignition; keep away from heat, sparks, and flames. Handle ether only in a hood and avoid direct physical contact. Do not breathe vapors. If ether is spilled or leaks, evacuate area, ventilate, and absorb on vermiculite or similar material. Wear appropriate OSHA equipment before entering spill area. Also see Section 1090.

2. Sampling and Storage

See Section 6010B.1.

Preferably collect grab samples in quadruplicate to allow sufficient volume for replicates and known additions. Flush sampling tap until water temperature stabilizes and stagnant lines are cleared. Collect samples in nominal 40- or 60-mL vials containing approximately 65 mg crystalline NH_4Cl (bake overnight at >100°C to eliminate contaminants), which converts free chlorine to a combined chlorine residual, and sealed with TFE-

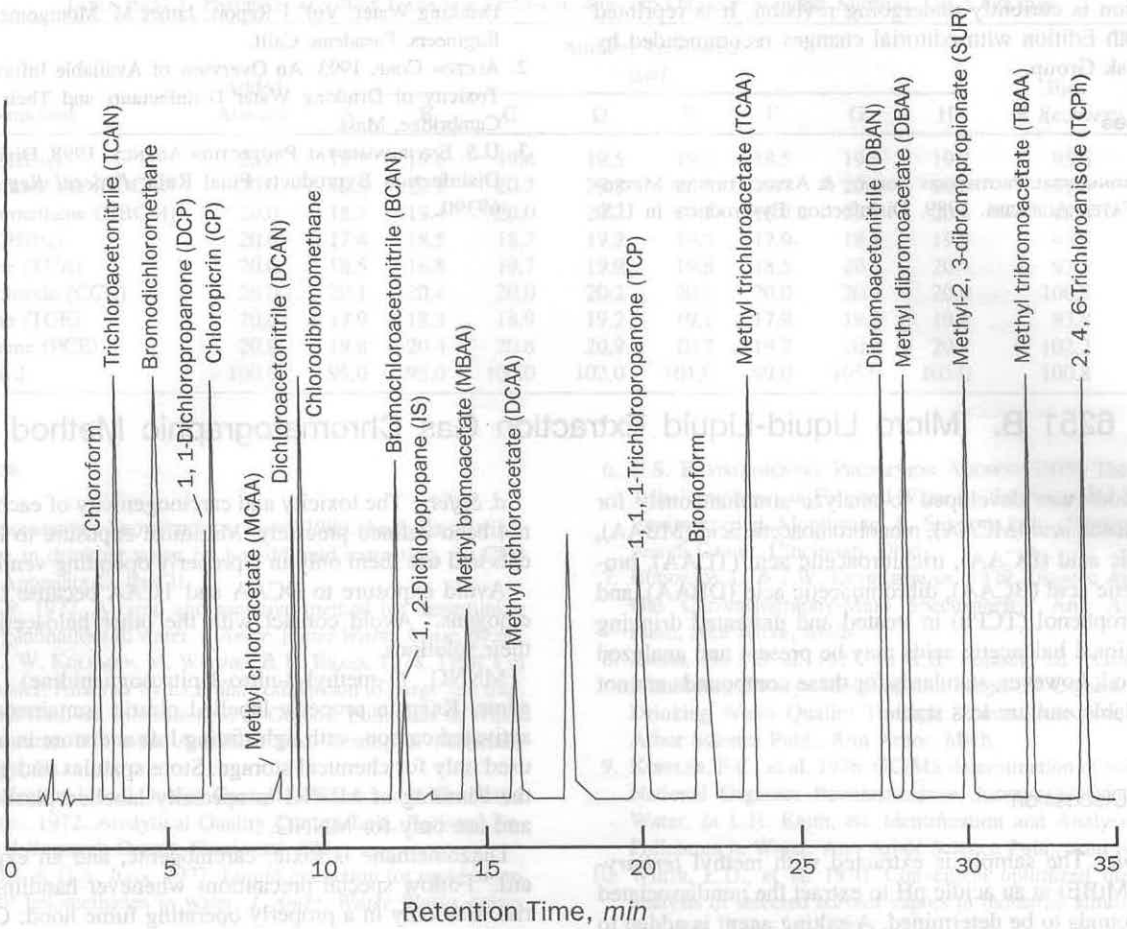

Figure 6251:1. Haloacetic acids separation from other commonly produced disinfection by-products on a DB-1701 column. Chromatogram produced by mixing the methyl esters of each haloacetic acid with other disinfection by-products in MtBE (disinfection by-products tentatively identified).

faced septa and screw caps. To minimize aeration, fill vials so that no air bubbles pass through the sample. Do not rinse with sample and do not let vial overfill. Seal sample vials with no headspace.

Analyze samples as soon as possible after collection. Store dechlorinated samples at 4°C, but for no more than 14 d;[5,6] check compound stability in any unknown sample matrix. Sample extracts can be held in a freezer at −11°C for 21 d.

TABLE 6251:I. METHOD DETECTION LEVELS AND PRECISION DATA*

Compound	Added Conc. µg/L	Found Conc. µg/L	Standard Deviation µg/L	Relative Standard Deviation %	Method Detection Level µg/L
Monochloroacetic acid	0.50	0.54	0.026	4.8	0.082
Monobromoacetic acid	0.50	0.80	0.028	3.4	0.087
Dichloroacetic acid	0.50	0.5	0.017	3.5	0.054
Trichloroacetic acid	0.50	0.5	0.017	3.4	0.054
Bromochloroacetic acid	0.50	0.49	0.015	3.1	0.04
Dibromoacetic acid	0.50	0.47	0.021	4.4	0.065
2,4,6-Trichlorophenol	0.25	0.27	0.011	4.1	0.034

*Based on the analysis of seven portions of reagent water with known additions.[2]

3. Apparatus

Preferably dedicate all analytical glassware used in this method to this procedure.

*a. Sample containers and extraction vials,** 40- or 60-mL screw-cap vials with TFE-faced silicone septa. Clean vials by washing with detergent, rinsing thoroughly with tap water, rinsing with 1:10 HCl, rinsing again with tap water, and finally rinsing with reagent water. Heat in an oven at 180° for at least 1 h. Clean caps and septa by rinsing with acetone, then hexane. Heat at 80°C for not more than 1 h in a clean, forced-air convection oven.

b. Microsyringes, 5, 10, 25, 50, 100, 250, 500, and 1000 µL.

c. Syringe, 30-mL glass hypodermic, metal luer-lok tip with 8.9-cm- (3.5-in.-) long × 17 gauge stainless steel pipetting needle (alternatively use a 30-mL volumetric pipet). See ¶ *a* above for glassware cleaning procedure.

* Wheaton: Industrial Glassware, Millville, NJ; or equivalent.

d. Micro volumetric flasks,† TFE-lined screw-cap: 2-mL, 5-mL, and 10-mL. Immediately after use, rinse three times with methanol. Invert to drain. Let air-dry completely in a ventilation hood.

e. Mechanical shaker,‡ to automate MtBE extraction. Insert vials into a wooden holding block (20 vial capacity) made from laminated plywood with drilled holes to accept vials, dimensioned to fit snugly onto the shaker table.

f. Extract and standard solution storage container, 1.8-mL clear glass, 7- and 14-mL amber glass screw-cap vials with TFE-lined silicone septa. For cleaning procedures, see ¶ *a* above.

g. Transfer pipets, 14.6- and 23-cm (5.75- and 9-in.) disposable glass pasteur pipets. See ¶ *a* above for glassware cleaning procedure.

h. Gas chromatograph, temperature-programmable (preferably with multiple ramp capability) with injector. Optimally use an autosampler for sample injection and a computer data system for peak integration and quantitation. (A detector base that can mount two electron capture detectors is ideal.)

1) *Gas handling equipment:* Use carrier (helium) and makeup (nitrogen) gases of high purity (99.999%) grade that pass through indicating calcium sulfate, molecular sieve 5A, activated charcoal, and an oxygen purifying cartridge. Use two-stage metal diaphragm high-purity regulators at the compressed gas sources. Use flow controllers to regulate carrier gas flow. Make all gas lines 0.3-cm (1/8-in.) copper (or stainless steel) tubing; rinse with acetone and bake before use.

2) *Injector*, split/splitless (using straight open bore insert).

3) *Analytical column*,§ 30 m long × 0.25 mm ID, fused silica capillary column with a 0.25-μm film thickness or equivalent.

4) *Confirmation column*, 30 m long × 0.25 mm ID, fused silica capillary column‖ with a 0.25-μm film thickness or a 30 m long × 0.25 mm ID, fused silica capillary column# with a 0.5-μm film thickness.

5) *Detectors*, a constant-current pulse-modulated ^{63}Ni ECD with standard size cell (use two ECDs for simultaneous confirmation analysis).

i. Salt scoops for sodium sulfate, made from stainless steel 1.3-cm- (0.5-in.-) diam bar stock drilled out to a volume of 1.73 mL so that each level scoopful contains 3 g. Alternatively, weigh the salt.

j. Pipetting dispensers, adjustable 5- and 2-mL sizes with TFE transfer lines, that can be mounted on the supplier's reagent bottles. Use for dispensing H_2SO_4 and MtBE. Alternatively use a 3-mL volumetric pipet and a 5-mL graduated pipet with manual pipet bulbs.

k. Diazomethane generator: Use millimole-size generator with "o"-ring joint (Figure 6251:2).** Immediately after use, rinse inner tube twice with 20% NaOH, then rinse twice with tap water. Immediately add 1 g silica gel to the outside tube to quench any residual diazomethane solution, rinse twice with methanol, and twice with tap water. Rinse both inner and outer tubes with reagent water three times. Bake at 180°C until dry in a clean, forced-air convection oven.

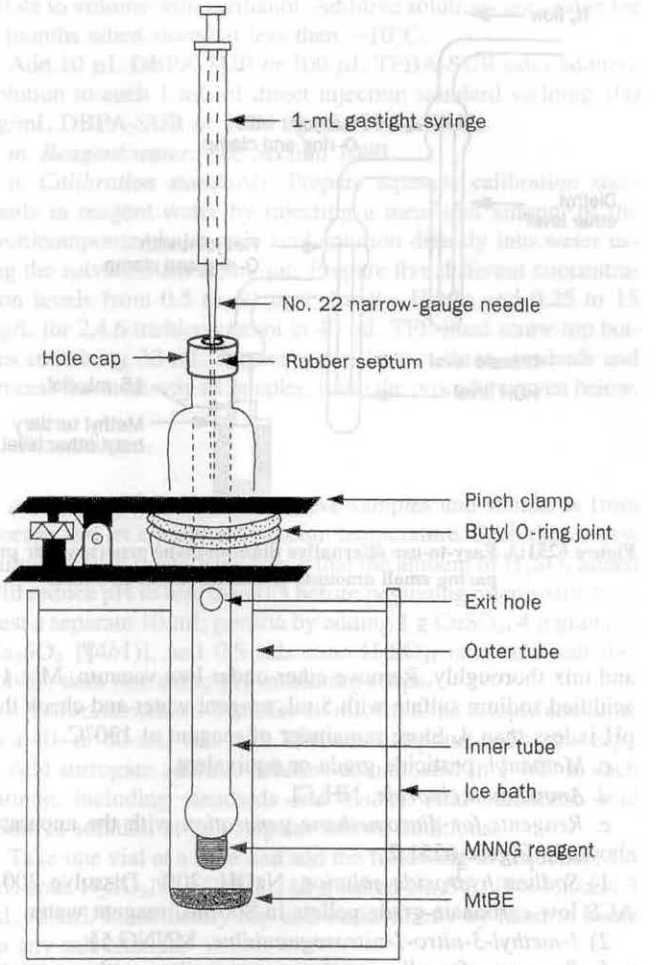

Figure 6251:2. Easy-to-use diazomethane generator apparatus for preparing small amounts of diazomethane in methyl *tertiary*-butyl ether (MtBE).

Alternatively use the millimole-size diazomethane generator shown in Figure 6251:3.†† To clean, rinse with reagent water and then with methanol, invert, and let air-dry.

l. pH-strips, pH indicating strips, 0 to 2.5 range.

4. Reagents

a. Extraction solvent, 99+ % MtBE, preservative-free.‡‡

b. Sodium sulfate reagents:

1) *Granular sodium sulfate*, Na_2SO_4, reagent grade, suitable for pesticide analysis. Heat at 400°C overnight in a shallow stainless steel pan covered with aluminum foil. Store in a 1-L glass bottle with TFE-lined polypropylene cap.

2) *Acidified sodium sulfate:* To 100 g anhydrous sodium sulfate, heated as above and cooled, add diethyl ether to just cover the solid; make a slurry. Add 0.1 mL conc sulfuric acid

† Kontes or equivalent.
‡ Eberbach or equivalent.
§ Durabond-1701, J&W Scientific, or equivalent.
‖ Durabond-5, J&W Scientific, or equivalent.
Durabond-210, J&W Scientific, or equivalent.
** Aldrich or equivalent.
†† Paxton Woods Glass, Cincinnati, OH, or equivalent.
‡‡ Omnisolv, manufactured by EM Science, Gibbstown, NJ, or equivalent.

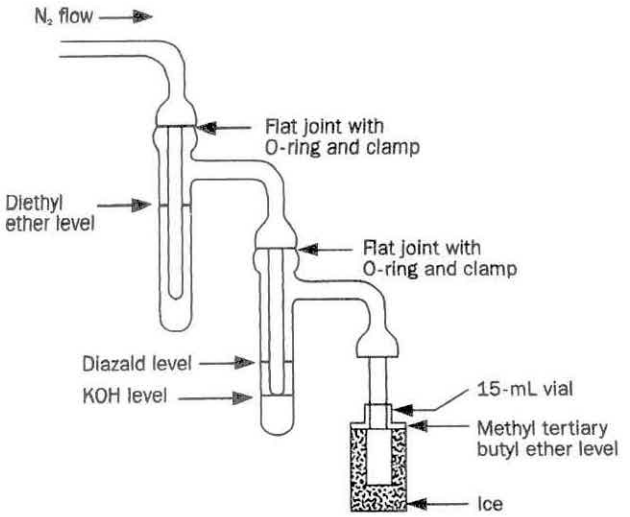

Figure 6251:3. Easy-to-use alternative diazomethane generator for preparing small amounts of diazomethane in MtBE.

and mix thoroughly. Remove ether under low vacuum. Mix 1 g acidified sodium sulfate with 5 mL reagent water and check that pH is less than 4. Store remainder of reagent at 130°C.

c. Methanol, pesticide grade or equivalent.

d. Ammonium chloride, NH_4Cl.

e. Reagents for diazomethane generation with the apparatus shown in Figure 6251:2:

1) *Sodium hydroxide solution,* NaOH, 20%: Dissolve 200 g ACS low-carbonate-grade pellets in 800 mL reagent water.

2) *1-methyl-3-nitro-1-nitrosoguanidine,* MNNG.§§

f. Reagents for diazomethane generation with apparatus shown in Figure 6251:3:

1) *Diethylene glycol monoethyl ether.*‖‖

2) N-*methyl*-N-*nitroso*-p-*toluene sulfonamide.*##

3) *Ethyl ether,* absolute.

4) N-*methyl*-N-*nitroso*-p-*toluene sulfonamide solution:* Mix 10 g reagent *f* 2) in 100 mL 1:1 (v:v) solution of ethyl ether and reagent *f*1). Solution is stable for 1 month when stored at 4°C in an amber-colored bottle with a TFE-lined screw cap.

5) *Potassium hydroxide solution,* KOH: Dissolve 37 g in 100 mL reagent water.

g. Silica gel, 35/60 mesh activated at 180°C and stored in a desiccator.

h. Sulfuric acid, H_2SO_4, conc.

i. Copper (II) sulfate pentahydrate, $CuSO_4 \cdot 5H_2O$.

j. Standard material, see Table 6251:II for source and physical information.

1) *Individual haloacetic acid standard stock solutions:* Prepare individual haloacetic acid and the trichlorophenol stock solutions as follows: Weigh 0.150 g of each acid. Dilute each standard in MtBE to 10 mL in a screw-top volumetric flask. (NOTE: Do not use methanol for dilution, because spontaneous methylation of the haloacetic acid may occur in methanolic solution.[8] Transfer each to a separate clean 14-mL amber vial and store in a freezer at −11°C. Stock standards are usable for 6 months.

2) *Multicomponent haloacetic acid additive solution:* Prepare a six-component additive solution using individual haloacetic acid stock solutions. Dilute 16.7 μL of each stock standard into a 10-mL volumetric flask containing 9 mL MtBE, but use 8.4 μL of 2,4,6-trichlorophenol solution. After adding all stock solutions, dilute to volume with MtBE. This gives 25 μg/mL of each HAA and 12.5 μg/mL for 2,4,6-trichlorophenol. The additive solution is usable for 3 months when stored at less than −10°C. Alternatively, prepare known-addition solution monthly in methanol and store at 4°C.

Measure microliter volumes with a gastight syringe using the solvent flush delivery technique. Do solvent flush with a 25-μL syringe by first drawing up 2.5 μL of solvent and then drawing the syringe plunger to the 5-μL mark with air. From the 5-μL mark measure amount of stock solution desired and then deliver the entire contents to the volumetric flask.

3) *Individual haloester standard stock solutions:* Prepare individual methyl ester stock solutions as follows: Weigh 0.1 × (molecular weight of ester/molecular weight of acid) g of each methyl ester in a 10-mL volumetric flask and dilute to mark with MtBE. Prepare methyl ester for dibromoacetic acid by derivatizing 1 mL of a 20 000-μg/mL acid solution with 100 μL methanol (follow derivatization steps in 5*e* below, but substitute dibromoacetic acid stock solution as the solution added to the outer tube for collection of the diazomethane). After derivatizing, transfer ester quantitatively to a 2-mL volumetric flask with

TABLE 6251:II. ANALYTICAL STANDARDS

Compound*	Purity %	Molecular Weight	Boiling Point[7] °C @ mm†
MCAA	99	94.5	183
MBAA	99+	138.95	208
DCAA	99+	128.94	194
TCAA	98	163.39	198
BCAA	97	173.39	215‡
DBAA	99	217.86	195 @ 250
TCPh	95	197.45	246
IS-DBP	95	201.9	140–142
IS-TCP§	99	147.43	156
SUR-DBPA	99	231.88	160 @ 20
SUR-TFBA	99	194.09	—
MeCA	99+	108.52	130 @ 740
MeBA	98	152.98	144
MeDCA	99+	142.97	143
MeTCA	99	177.42	152–153
MeBCAA	98	187.42	155
MeDBA	—	—	182
TCAn	99	211.48	132 @ 28

*Sources: BCAA, Radian Corp., Austin, TX; DBAA, Fluka Chemika-Biochemika, Switzerland; TCPh obtainable from Chem Service, Inc., Westchester, PA; MeDBA and SUR ester, derivatized acid at Metropolitan Water District of Southern California laboratory; other compounds from Aldrich Chemical Company, Inc., Milwaukee, WI.

†°C at reduced pressure in mm Hg.

‡Decomposes.

§Ensure that TCP is not a contaminant when it is used as IS.

§§ Aldrich or equivalent.

‖‖ Carbitol (Aldrich), or equivalent.

Diazald (Aldrich), or equivalent.

a TFE-lined screw cap and dilute to mark with MtBE. Stock standards are usable for 6 months when stored at less than −10°C.

4) *Multicomponent haloester additive solution:* Prepare a multicomponent additive solution by diluting 10 μL of each haloester stock standard, but use 5 μL of 2,4,6-trichloroanisole (methyl ether of the phenol), in a 10-mL volumetric flask and bring to volume with MtBE. This will yield a mixture containing approximately 10 μg/mL each, except for 2,4,6-trichloroanisole, which will be approximately 5 μg/mL. Additive solution is usable for 3 months when stored at less than −10°C.

5) *Direct injection haloester standards:* Prepare direct injection standards using 10 μg/mL multicomponent haloester additive solution, a 30-μg/mL internal standard additive solution [see ¶ 4*k*2)], and a 10-μg/mL methanol solution of methyl-2,3-dibromopropionate [surrogate ester, see ¶ 4*l*4)]. Prepare direct injection standards by diluting appropriate volumes of multicomponent haloester additive mix, internal standard additive solution, and surrogate ester solution with enough MtBE to give a final volume of 1.0 mL.

k. Internal standard, 1,2,3-trichloropropane (IS-TCP), 98% pure (alternatively use 1,2-dibromopropane (IS-DBP).

1) *Internal standard stock solutions:* Weigh 50 mg into a 10-mL volumetric flask and bring to volume with methanol. This will yield a 5000-μg/mL stock solution. Stock standards are usable for 6 months when stored at less than −10°C.

2) *Internal standard additive solution,* 30 μg/mL: Deliver 60 μL internal standard stock solution into a 10-mL volumetric flask and dilute to volume with methanol. Divide evenly among six 1.8-mL vials and store at −11°C. Additive solution is usable for 3 months.

Add 20 μL of internal standard additive solution to each 2 mL extract, yielding internal standard of 300 ng/mL.

l. Surrogate, (DBPA-SUR) 2,3-dibromopropionic acid, 99% pure or 2,3,5,6-tetrafluorobenzoic acid (TFBA-SUR).

1) *Surrogate stock solution,* 20 000 μg/mL: Weigh 0.2000 g SUR acid into a 10-mL screw-cap volumetric flask and dilute to mark with MtBE. Stock solutions are usable for 6 months when stored at less than −10°C.

2) *Surrogate additive solution:*

a) *DBPA-SUR,* 10 μg/mL: Deliver 5 μL DBPA-SUR stock solution into a 10-mL volumetric flask and dilute to volume with methanol. Divide evenly among six 1.8-mL vials and store at −11°C. Additive solutions are usable for 3 months.

Add 30 μL DBPA-SUR additive solution to each 30-mL sample portion, yielding DBPA-SUR 10 μg/L.

b) *TFBA-SUR,* 20 μg/mL: Deliver 300 μL TFBA-SUR stock solution into 1-L volumetric flask and dilute to volume with reagent water. Add 0.5 mL TFBA-SUR additive solution to each 30-mL sample, yielding 100 μg/L.

3) *Esterified surrogate stock solution,* 10 000 μg/mL: Derivatize 1 mL SUR stock solution and 100 μL methanol, according to derivatization steps in ¶ 5*e*, but substitute SUR stock solution and 100 μL methanol for MtBE. After derivatizing, transfer quantitatively to a 2-mL volumetric flask with a TFE-lined screw cap and dilute to mark with MtBE. Stock solutions are usable for 6 months when stored at less than −10°C.

4) *Surrogate ester additive solution,* 10 μg/mL: Deliver 10 μL SUR ester stock solution into a 10-mL volumetric flask and dilute to volume with methanol. Additive solutions are usable for 3 months when stored at less than −10°C.

Add 10 μL DBPA-SUR or 100 μL TFBA-SUR ester additive solution to each 1 mL of direct injection standard yielding 100 ng/mL DBPA-SUR or 1000 ng/mL TFBA-SUR.

m. Reagent water: See Section 1080.

n. Calibration standards: Prepare aqueous calibration standards in reagent water by injecting a measured amount of the multicomponent haloacetic acid solution directly into water using the solvent flush technique. Prepare five different concentration levels from 0.5 to 30 μg/L for the HAAs and 0.25 to 15 μg/L for 2,4,6-trichlorophenol in 40-mL TFE-lined screw-top bottles containing 30 mL reagent water. Extract these standards and process the same way as samples, using the procedure given below.

5. Procedure

a. Sample preparation: Remove samples and standards from storage and let equilibrate to room temperature. Each time a new sample matrix is analyzed check that the amount of H_2SO_4 added will reduce pH to less than 0.5 before beginning microextraction. Test a separate 10-mL portion by adding 1 g $CuSO_4$, 4 g granular Na_2SO_4 [¶4*b*1)], and 0.5 mL conc H_2SO_4, mix until salt dissolves, then test using pH indicating strips.

b. Microextraction: Transfer 30 mL from the sample container to a 40- or 60-mL vial with TFE-faced septum and screw cap.

Add surrogate additive solution as indicated in ¶ 4*l*2) to each sample, including standards and blanks. Add haloacetic acid additive solution at this step for known additions.

Take one vial at a time and add the following in sequence: 1.5 mL conc H_2SO_4, 3 g $CuSO_4$, 12 g baked Na_2SO_4 [¶4*b*1)], and 3 mL MtBE. Immediately cap and shake briefly by hand to break up any salt clumps.

When using automated extraction, place vials in mechanical shaker wooden holding block. Shake vials at fast speed (approximately 300 cycles/min) for 9 min; alternatively shake manually for 2 min until salt is dissolved.

Remove vials, place upright, and let stand for at least 3 min until the phases separate.

c. Preparation of diazomethane[9]: Using the apparatus in Figure 6251:2, add approximately 130 mg MNNG to the inside tube of the generator. Add 0.5 mL reagent water to the MNNG and secure cap and septum. Add 2 mL MtBE to the outside tube. Place butyl o-ring in glass joint, place inside tube firmly on top of o-ring, and clamp securely with a screw-type pinch clamp.

Place generator in an ice water bath containing enough ice to keep diazomethane MtBE solution at 0°C until used.

Add 600 μL 20% NaOH (1 drop/5 s) using a 1-mL gastight syringe (22-gauge needle) through the generator septum (check that the syringe needle is on the opposite side of the vapor exit hole). Let derivatization continue for 30 min after adding NaOH; use product as soon as possible. Add more ice if necessary to maintain temperature.

If more diazomethane is needed, prepare two or more batches and combine before use.

Alternatively use the apparatus in Figure 6251:3 to prepare diazomethane. Add enough ethyl ether to tube 1 to cover the first impinger. Add 10 mL MtBE to 15-mL collection vial. Set nitrogen flow at 5 to 10 mL/min. Add 4 mL sulfonamide solution and 3 mL 37% KOH solution to the second impinger. Connect

tubing as shown and let nitrogen flow purge diazomethane from the reaction vessel into the collection vial for 30 min. Cap vial when collection is complete and hold at 0°C. When stored at 0°C diazomethane solution may be used over a period of 48 h.

d. Separation and concentration: NOTE: Ensure that all items that come into contact with the sample prior to methylation have been washed with a dilute solution of sulfuric acid.

The drying step included here may be used if excess diazomethane is required to maintain the persistent yellow color of the sample (5*e*). It is not necessary in every case and may be used at the discretion of the analyst. Plug a small disposable pipet with a small amount of acid-washed glass wool. Add approximately 1 g acidified Na_2SO_4 [¶4*b*2)] to the pipet and pass exactly 2 mL of the top MtBE sample extract through the salt, being careful not to transfer any of the aqueous phase. Rinse the salt in the pipet with two 250-μL volumes of solvent and collect together in a receiver ampule (a 2-mL volumetric flask with TFE-lined screw cap) for subsequent concentration and methylation.

Concentrate MtBE extract to approximately 1.7 mL using a moderate stream of nitrogen blowing on the surface of the extract.

e. Derivatization: Add 20 μL of 30 μg/mL internal standard additive solution to each concentrated extract. (The internal standard is added at this time to minimize manipulation in the presence of diazomethane.) Cool in an explosion-safe freezer or in an ice bath for 7 min and add diazomethane (cooling extracts is unnecessary if diazomethane is generated by apparatus shown in Figure 6251:3).

Uncap one volumetric flask and add 250 μL of cold diazomethane/MtBE solution. Cap immediately with TFE-lined screw cap; mix gently by inverting once. Repeat for remaining extracts. A persistent yellow color after addition of diazomethane indicates that an excess is available for esterification. If necessary add more diazomethane solution.

Hold for 15 min at 4°C in an explosion-safe or explosion-proof refrigerator. Alternatively, keep extracts in an ice bath (cooling is unnecessary if diazomethane was generated by the apparatus shown in Figure 6251:3).

After holding 15 min, place extracts in a hood and let stand another 15 min until they reach room temperature. Dilute to mark with MtBE and invert flask to mix. If using an autosampler, transfer each extract evenly between two labeled autosampler vials containing approximately 0.01 g silica gel with a 23-cm (9-in.) disposable pasteur pipet to quench excess diazomethane. Keep each extract in contact with diazomethane for approximately the same amount of time before quenching. Store extra autosampler vial in freezer at −10°C as a backup extract. Alternatively, add silica gel to volumetric flask after derivatization is complete.

f. Gas chromatography: Typical operating conditions for the chromatograph are as follows:

Injector temperature 160°C; split valve opened at 0.5 min.

Temperature program: 37°C for 21 min, rising 11°C/min to 136°C, holding 3 min at 136°C, rising 20°C/min to 236°C, holding 3 min at 236°C.

Detector temperature: 300°C.

Carrier gas flow: 30 cm/s at 37°C.

Makeup gas flow: 23 mL/min.

At the beginning of each analytical run, inject two MtBE solvent blanks to condition the GC and to verify that interferences are absent. A 2-μL extract is injected in splitless mode. Always inject same sample volume and use sample dilution to obtain response in the calibration range. If levels greater than the highest standard are obtained, reanalyze diluted sample extract and readjust internal standard concentration. Calculate concentration only for those compounds that were at levels higher than the calibration curve; for other compounds use values obtained from the undiluted sample extract. See Figure 6251:4 for a chromatogram of an extracted and derivatized 30-μg/L standard on the analytical column. See Table 6251:III for retention times. A direct standard may be injected after the MtBE solvent blanks to verify continued system performance.

g. Calibration: Use five levels of calibration standards to define the quantitation range. The lowest standard should be near the limit of quantitation (LOQ) (see Table 6251:IV) for each compound. Use other standards to bracket the expected range of sample concentrations; do not exceed linear range of detector. Prepare standards by adding haloacetic acids and trichlorophenol to reagent water and then extract with the same solvent and derivatize with the same batch of diazomethane as used for the samples. Use the same extraction/esterification procedure for both standards and samples to correct for recovery characteristics. Analyze calibration standards under the same chromatographic conditions as samples.

Analyze calibration standard at one or more levels with each sample set to verify the working calibration curve. If the calibration standard is within ± 15% of the expected value, a new five-level calibration curve is not needed.

6. Calculation

A 2-μL injection of each calibration level will provide peak area (A_a) data for each compound and an internal standard peak area (A_i) for each level; use these peak areas to calculate relative response for each compound.

$$\text{Relative response} = A_a/A_i$$

A calibration curve passing through zero is generated from the plotted points for each compound using the relative response versus standard concentration. Use the internal standard quantitation method to determine unknown concentrations by a linear, quadratic, or point-to-point curve fit.

7. Quality Control

a. Quality control program: Because sample preparation requires many manipulations, chances for errors are increased. Consequently, at least follow minimum quality control requirements to monitor and maintain method performance. Include method blanks, an initial demonstration of laboratory capability and detection levels, assessment of the internal standard recovery, determination of surrogate compound recoveries, evaluation of calibration data and curves, sample matrix additions, and precision of replicate sample analysis. Additional quality control measures may be used.

b. Method blanks: Process a method blank (30 mL reagent water) with each set of samples. If the blank produces any peak within the retention time window of a compound that would prevent its determination, seek out and eliminate the source of contamination and reanalyze samples.

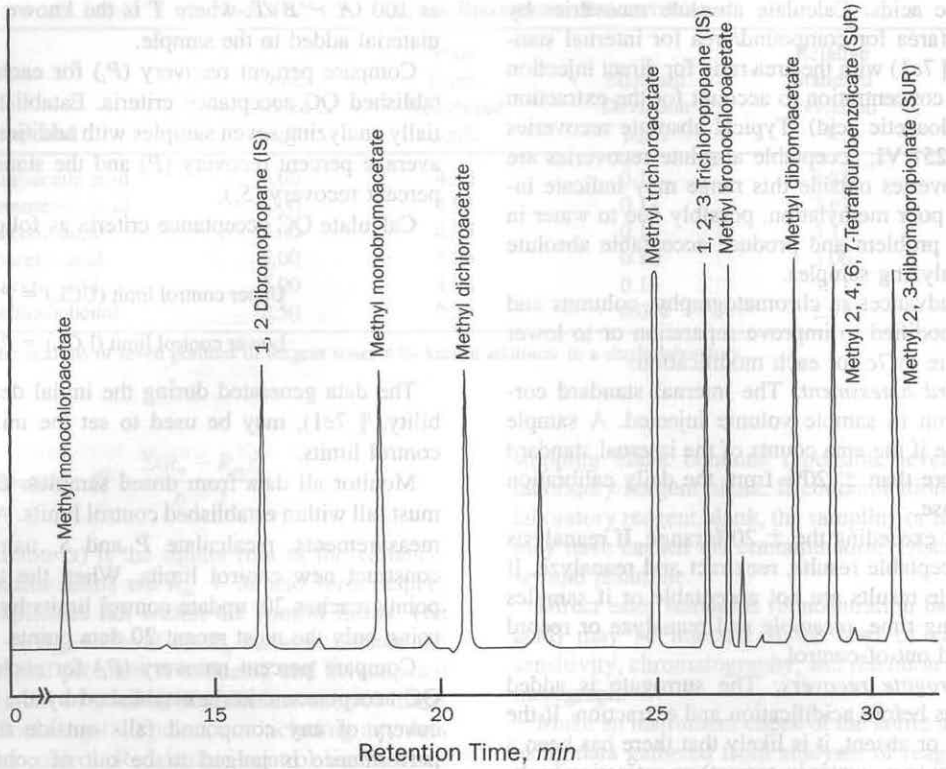

Figure 6251:4. Chromatogram produced by reagent water with known additions: 30 μg/L extracted standard on DB-1701 column.

c. Initial demonstration of capability: To demonstrate an adequate level of performance, conduct the following operations before analyzing samples and whenever any major analytical change, such as new analyst or switch in type of column, is made.

TABLE 6251:III. RETENTION TIMES

Compound	Retention Time *min*		
	DB-1701 Column	DB-5 Column	DB-210 Column
Methyl chloroacetate (MeCA)	11.11	5.80	10.97
Methyl bromoacetate (MeBA)	18.80	9.23	13.03
Methyl dichloroacetate (MeDCA)	20.53	10.11	12.72
Methyl trichloroacetate (MeTCA)	24.78	18.94	14.37
Methyl bromochloroacetate	26.23	19.19	15.11
Methyl dibromoacetate (MeDBA)	28.64	25.51	16.83
2, 4, 6-Trichloroanisole (TCAn)	~ 34.7	33.74	22.08
1,2-Dibromopropane (internal standard)	15.97	10.78	—
1,2,3-Trichloropropane (internal standard)	25.87	18.43	13.87
Methyl-2,3-dibromopropionate (surrogate ester)	30.74	28.60	—
Methyl-2,3,6,7- tetrafluoro-benzoicate (surrogate)	29.19	26.84	—

1) Accuracy as percent recovery—Establish a calibration curve as in ¶ 5*g* and select a representative additive concentration (5 μg/L is convenient) for each target compound. Using a syringe, add the appropriate amount of stock standard mix to each of a minimum of seven 30-mL portions of reagent water and analyze.

Calculate average percent recovery (P) and standard deviation of the recovery (S_r). Compare results to the single-laboratory recovery and precision data in Table 6251:V. Compare precision at similar concentrations, that is $P \pm 30\%$ of the additive level. Acceptable mean recovery values are within the interval $P \pm 30\%$. For compounds not meeting this criterion, repeat with another seven samples until satisfactory performance has been demonstrated.

2) Absolute recovery—Use direct injection haloester standards, ¶ 4*j*5), to check absolute recoveries of extracted and

TABLE 6251:IV. RECOMMENDED QUANTITATION LIMITS

Compound	Recommended Quantitation Limit μg/L
Monochloroacetic acid (MCAA)	1.0
Monobromoacetic acid (MBAA)	0.5
Dichloroacetic acid (DCAA)	0.6
Trichloroacetic acid (TCAA)	0.6
Bromochloroacetic acid (BCAA)	0.8
Dibromoacetic acid (DBAA)	0.6
2,4,6-Trichlorophenol (TCPh)	0.4

derivatized haloacetic acids. Calculate absolute recoveries by comparing the ratio (area for compound/area for internal standard) for samples in ¶ 7*c*1) with the area ratio for direct injection standard at a tenfold concentration to account for the extraction (expressed as the haloacetic acid). Typical absolute recoveries are given in Table 6251:VI; acceptable absolute recoveries are within ± 30%. Recoveries outside this range may indicate insufficient shaking or poor methylation, possibly due to water in the extracts. Correct problem and produce acceptable absolute recoveries before analyzing samples.

Because of rapid advances in chromatography, columns and conditions may be modified to improve separation or to lower cost. Repeat procedure in 7*c* for each modification.

d. Internal standard assessment: The internal standard corrects for any deviation in sample volume injected. A sample injection is acceptable if the area counts of the internal standard peak do not vary more than ± 20% from the daily calibration standard(s) IS response.

Reinject an extract exceeding the ± 20% range. If reanalysis does not produce acceptable results, reextract and reanalyze. If the reextracted sample results are not acceptable or if samples have exceeded holding time, resample and reanalyze or record results as suspect and out-of-control.

e. Evaluating surrogate recovery: The surrogate is added directly to all samples before acidification and extraction. If the surrogate area is low or absent, it is likely that there has been a derivatization problem (e.g., water in extract) or extraction problem (e.g., water insufficiently acidified).

An extract is acceptable if the area counts of the surrogate standard recovery are ± 30% from the surrogate standard recovery for the daily calibration standard(s).

When surrogate recovery is not acceptable, check the following: locate possible errors in calculations or procedure, degradation of standard solution, contamination sources, and instrument performance. If these steps do not reveal the problem, reanalyze the extract. If reanalysis does not produce acceptable results, reextract and reanalyze samples. If the reextracted sample results are not acceptable or if samples have exceeded holding time, record results as suspect and out-of-control.

f. Extracted standard calibration: Quantitation is done by internal standard referencing with relative areas. Produce a minimum five-level extracted standard calibration curve for sample quantitation.

If the response for any compound falls outside the predicted response by more than 15% from a previous calibration, make a new calibration standard and analyze it until an acceptable curve is obtained.

Analyze calibration standards with each sample set after an acceptable five-level calibration curve is generated. If the continuing calibration standards are not within ± 15%, check for errors or degradation of standards and construct a new calibration curve.

g. Matrix additions: Add each target compound into one sample per sample set (a sample set is all samples extracted within a 24-h period) or 10% of the samples, whichever is greater.

The added concentration should be near to or greater than the background. Take care, particularly with dichloroacetic acid, to ensure that the addition plus background concentration does not exceed calibration range (extract dilution may be needed).

Analyze one sample portion to determine the background concentration (B) of each compound. Add working standard mix to a second sample portion and analyze to determine the concentration of each compound (A). Calculate percent recovery (P_i) as 100 $(A - B)/T$, where T is the known concentration of the material added to the sample.

Compare percent recovery (P_i) for each compound with established QC acceptance criteria. Establish QC criteria by initially analyzing seven samples with additions and calculating the average percent recovery (P) and the standard deviation of the percent recovery (S_r).

Calculate QC acceptance criteria as follows:

$$\text{Upper control limit (UCL)} = P + 3S_r$$

$$\text{Lower control limit (LCL)} = P - 3S_r$$

The data generated during the initial demonstration of capability, ¶ 7*c*1), may be used to set the initial upper and lower control limits.

Monitor all data from dosed samples. Compound recoveries must fall within established control limits. After 10 new recovery measurements, recalculate P and S_r using all the data, and construct new control limits. When the total number of data points reaches 20, update control limits by calculating P and S_r using only the most recent 20 data points.

Compare percent recovery (P_i) for each compound with the QC acceptance criteria established by the control limits. If recovery of any compound falls outside the designated range, performance is judged to be out of control. Seek source of problem immediately and resolve before continuing the analysis.

However, if recovery of a compound meets calibration, blank, internal standard, surrogate, and replicate quality control, laboratory performance is in control, and the recovery problem is matrix-related. Label result for that compound in the sample as suspect/matrix.

h. Replicate analysis: Analyze sample duplicates to monitor precision. Analyze duplicates on at least 10% of all samples randomly selected.

Determine control limits by calculating the range as a function of the relative standard deviation. The range, R, is the absolute difference of the duplicate values, X_1 and X_2, as follows:

$$R = |X_1 - X_2|$$

The normalized range (R_n) is calculated by dividing the range by the average of the duplicate values (X_m):

$$R_n = \frac{R}{X_m}$$

$$X_m = \frac{X_1 + X_2}{2}$$

Calculate mean normalized range (R_m) for 20 pairs of duplicate data points initially and 20 pairs of points quarterly:

$$R_m = \frac{\Sigma R_n}{n}$$

where:

n = number of duplicate pairs.

Table 6251:V. Additive Recovery in Reagent Water*

Compound	Conc. Added µg/L	Mean Conc. Recovered µg/L	Standard Deviation µg/L	Relative Standard Deviation %	Mean Recovery %
Monochloroacetic acid	5.00	4.90	0.19	3.88	98.0
Monobromoacetic acid	5.00	4.95	0.13	2.67	99.0
Dichloroacetic acid	5.00	4.95	0.15	3.11	99.0
Trichloroacetic acid	5.00	5.06	0.16	3.06	101
Dibromoacetic acid	5.00	4.98	0.16	3.11	99.6
2,4,6-Trichlorophenol	2.50	2.51	0.075	2.99	100

* Based on the analysis of seven portions of reagent water with known additions in a single laboratory.

$$\text{Variance} = S^2 = \frac{\Sigma(R_n - R_m)^2}{n - 1}$$

The standard deviation (s) is the square root of the variance. Upper and lower control limits are $R_m + 3s$ and zero, respectively. Acceptable duplicates fall within the control limits. The upper warning limit is $R_m + 2s$. If an R_n value is outside the warning limit, a potential problem is indicated and investigated before the analysis is out of control. Recalculate control limits quarterly using the most recent 20 points not including any data points that are out of control. Recalculate control limits when any major analytical changes are made and after at least 20 points have been collected.

Analyze any problem and correct it. If the duplicate is not acceptable, reextract only for those compounds out of control. If the duplicates are still unacceptable or the sample holding time has been exceeded, resample and reanalyze. If this is not possible record results as suspect and out of control. Do not use such data in range calculations.

i. Additional quality control: Each quarter, analyze QC check standards from an external source. Independent confirmation may include interlaboratory split sampling for comparison. Analyze performance evaluation samples, preferably from USEPA or appropriate state agency, at least once a year. Results for each target compound must be within established acceptance limits.

Shipping blanks are containers filled with reagent water containing appropriate amount of NH_4Cl (see ¶ 2), shipped to all sample locations with sample bottles, and returned with the samples. They are used to assess contamination during sampling and transit. Analyze a shipping blank with each sample set. If the shipping blank contains reportable levels, compare with the laboratory reagent blank. If contamination is not detected in the laboratory reagent blank, the sampling or transportation practices may have caused the contamination. Discard all samples in the set and resample.

Direct ester standards (concentration based on corresponding acid) may be injected at the start of each set to verify the sensitivity, chromatography, and retention times on the gas chromatograph.

Make an instrument check of the entire analytical system daily using data gathered from analyses of reagent blanks, standards, and replicate samples.

8. Precision and Bias

Single-laboratory method detection levels (MDL) and extracted recovery data from reagent water are presented in Table 6251:I. Data for absolute recoveries in reagent water are given in Table 6251:VI. Laboratory data from two different laboratories showing duplicate precision and matrix additions recoveries are presented in Tables 6251:VII and 6251:VIII.

The precision and bias of analyses performed under the Information Collection Rule[10] (ICR) using this method are presented in Tables 6251:IX and 6251:X, respectively. Precision was determined as relative percent difference for duplicate analyses [(Rn)(100)] and was calculated only when both analyses in the duplicate pair showed concentrations at or greater than the ICR minimum reporting level (MRL). Bias was evaluated as percent recovery for cases in which the fortified amount was at least half the backgound concentration. The fortifying concentrations

Table 6251:VI. Absolute Recovery Data for Reagent Water with Known Additions

Compound	Conc. Added µg/L	Mean Conc. Recovered µg/L	Standard Deviation µg/L	Relative Standard Deviation %	Mean Recovery %
Monochloroacetic acid	1.00	0.789	0.047	5.92	78.9
Monobromoacetic acid	1.00	0.706	0.034	4.76	70.6
Dichloroacetic acid	1.00	1.10	0.048	4.38	110
Trichloroacetic acid	1.00	0.927	0.051	5.49	92.7
Bromochloroacetic acid	0.50	0.49	0.015	3.07	98
Dibromoacetic acid	1.00	1.16	0.032	2.75	116
2,4,6-Trichlorophenol	0.50	0.523	0.030	5.89	105

TABLE 6251:VII. SAMPLE DUPLICATE DATA FROM TWO LABORATORIES

Laboratory	Compound	Number of Pairs of Replicates	Average Difference Between Duplicates %	Standard Deviation of Difference Between Duplicates %
A	MCAA	5	7.6	10.6
	MBAA	3	1.9	1.5
	DCAA	7	1.5	0.8
	TCAA	6	1.4	1.0
	DBAA	5	6.0	6.0
	BCAA	11	1.8	1.3
B	MCAA	10	16.7	14.8
	MBAA	3	8.9	8.4
	DCAA	11	8.5	10.6
	TCAA	11	5.5	3.6
	DBAA	5	5.4	4.0
	BCAA	10	5.3	4.3

TABLE 6251:VIII. FIELD SAMPLE RECOVERY WITH KNOWN ADDITIONS TO DRINKING WATER, IN TWO LABORATORIES

Laboratory	Compound	Added Conc. µg/L	Number of Samples	Mean Recovery %	Relative Standard Deviation %
A	MCAA	5.0	7	99	4
	MBAA	5.0	7	101	4
	DCAA	5.0	7	96	4
	TCAA	5.0	7	100	3
	BCAA	10.0	14	96	5
	DBAA	5.0	7	102	5
	TCPh	2.5	7	100	6
B	MCAA	5.0	13	101	8
	DCAA	4.0	14	103	7
	TCAA	4.0	14	103	6
	MBAA	5.0	14	97	8
	BCAA	2.0	14	106	8
	DBAA	4.0	14	102	7
	TCPh	0.4	14	104	15

ranged from the ICR MRL (2 µg/L for MCAA, 1 µg/L for all other HAAs) to 40 µg/L.

9. References

1. CHINN, R. & S.W. KRASNER. 1989. A simplified technique for the measurement of halogenated organic acids in drinking water by electron capture gas chromatography. Presented at the American Chemical Soc. Pacific Conf. on Chemistry and Spectroscopy, Pasadena, Calif., Oct. 18–21, 1989.
2. U.S. ENVIRONMENTAL PROTECTION AGENCY. 1984. Definition and procedure for the determination of the method detection limit. 40 CFR Part 136, Appendix B. *Federal Register* 49, No. 209.
3. ALCEON CORP. 1993. An Overview of Available Information on the Toxicity of Drinking Water Disinfectants and Their By-Products. Cambridge, Mass.
4. DE BOER, T.J. & H.J. BACKER. 1963. Organic Synthesis, Collective, Vol. IV. John Wiley & Sons, New York, N.Y.
5. U.S. ENVIRONMENTAL PROTECTION AGENCY. 1996. ICR Sampling Manual. EPA 814-B-96-001, U.S. Environmental Protection Agency, Cincinnati, Ohio.
6. MUNCH, D.J., J.W. MUNCH & A.M. PAWLECKI. 1995. Method 552.2, Determination of Haloacetic Acids and Dalapon in Drinking Water by Liquid-Liquid Extraction, Derivatization and Gas Chromatography with Electron Capture Detection. U.S. Environmental Protection Agency, Cincinnati, Ohio.
7. LIDE, D.R., ed. 1991. CRC Handbook of Chemistry and Physics, 71st ed. CRC Press, Boca Raton, Fla.
8. XIE, Y., D.A. RECKOW & R.V. RAJAN. 1993. Spontaneous methylation of haloacetic acids in methanolic stock solutions. *Environ. Sci. Technol.* 27:1232.
9. FALES, H.M. T.M. JAOUNI & J.F. BABASHAK. 1973. Simple device for preparing ethereal diazomethane without resorting to codistillation. *Anal. Chem.* 45:13.
10. U.S. ENVIRONMENTAL PROTECTION AGENCY. 1996. Monitoring requirements for public drinking water supplies: *Cryptosporidium, Giardia,* viruses, disinfection byproducts, water treatment plant data and other information requirements. Final rule. *Federal Register* 61(94):24354.

TABLE 6251:IX. RELATIVE PERCENT DIFFERENCE (RPD) DETERMINATIONS FROM DUPLICATE SAMPLES

Haloacetic Acid*	MRL µg/L	N	RPD in Given Percentile					Median Sample Concentration µg/L
			10th	25th	Median	75th	90th	
HAA5		775	0.0	0.83	2.9	6.4	12	21
Monochloroacetic acid	2.0	232	0.0	1.6	4.9	12.	28	3.2
Dichloroacetic acid	1.0	774	0.0	0.0	2.9	6.3	11	10
Trichloroacetic acid	1.0	716	0.0	0.0	2.4	6.1	10	9.8
Monobromoacetic acid	1.0	98	0.0	0.0	6.9	15.	27	1.4
Dibromoacetic acid	1.0	294	0.0	0.0	3.6	7.4	11	2.4
HAA6		771	0.07	0.86	2.8	6.3	11	24
Bromochloroacetic acid	1.0	656	0.0	0.0	3.2	6.7	12	3.4

* HAA5 data are the sum of the individual concentrations of MCAA + DCAA + TCAA + MBAA + DBAA; HAA6 data are the sum of HAA5 + BCAA.

TABLE 6251:X. PERCENT RECOVERY DETERMINATIONS FROM FORTIFIED SAMPLES

Haloacetic Acid*	N	Percent Recovery in Given Percentile					Median Sample Concentration
		10th	25th	Median	75th	90th	μgL
HAA5	811	93	98	103	108	120	16.
Monochloroacetic acid	937	88	96	105	116	138	<2.0
Dichloroacetic acid	806	89	95	100	106	116	7.2
Trichloroacetic acid	793	90	97	102	110	120	4.9
Monobromoacetic acid	953	91	99	105	112	140	0.0
Dibromoacetic acid	925	92	98	103	114	127	0.0
HAA6	804	93	99	103	108	118	19
Bromochloroacetic acid	873	90	98	102	108	120	1.8

* HAA5 data are the sum of the individual concentrations of MCAA + DCAA + TCAA + MBAA + DBAA; HAA6 data are the sum of HAA5 + BCAA.

10. Bibliography

U.S. ENVIRONMENTAL PROTECTION AGENCY. 1990. Methods for the Determination of Organic Compounds in Drinking Water. Supplement 1. EPA-600/4-90-020.

BARTH, R.C. & P.S. FAIR. 1992. Comparison of the microextraction procedure and Method 552 for the analysis of HAAS and chlorophenols. *J. Amer. Water Works Assoc.* 84:94.

6252 DISINFECTION BY-PRODUCTS: ALDEHYDES (PROPOSED)*

6252 A. Introduction

1. Sources and Significance

Ozone reactions during water treatment are complex and often produce a wide range of unstable oxidation by-products, usually oxygenated and polar. Among the intermediate products formed, when ozone attacks the organic matter present in raw waters, are low-molecular-weight by-products such as aldehydes. If oxidized further, these aldehydes can produce aldo-acids and carboxylic acids. Formaldehyde, a ubiquitous component of the environment, may be introduced into drinking water by ozone treatment, natural metabolism, and commercial processes.

There are two postulated mechanisms for aldehyde formation during ozone treatment. The first involves a two-step Criegee attack at unsaturated C-C bonds by molecular ozone with ozonides or epoxides formed as intermediates.[1] The second involves an indirect reaction of OH radicals.[2] Although the levels of aldehyde formation are usually a function of ozone dose, their concentrations are often controlled in water treatment by increasing the pH and thus the alkalinity of the water.

Aldehydes are unlikely to pose a serious health hazard to the consumer at microgram-per-liter concentrations as usually encountered in drinking water treatment. However, they react with nucleophiles even at these low levels and can therefore be a potential threat.[3] Thus, for example, formaldehyde, acetaldehyde, and crotonaldehyde are known animal carcinogens. Formaldehyde is a known human carcinogen.[4] Aldehydes also may serve as important components of assimilable organic carbon in promoting undesirable bioactivity.

2. Selection of Method

The most effective method for the determination of aldehydes in aqueous solutions involves the use of *O*-(2,3,4,5,6-pentafluorobenzyl)hydroxylamine (PFBHA)† as a derivatizing agent. PFBHA reacts with low-molecular-weight carbonyl compounds, including aldehydes, to form the corresponding oximes. Unless the carbonyl compound is a symmetrical ketone or formaldehyde, two geometric isomers of the oxime derivatives are formed. These derivatives are extractable with organic solvents and are highly sensitive to analysis by gas chromatography with electron capture detection (GC/ECD) and gas chromatography with selective ion mass spectrometric detection (GC/SIM-MS).

3. References

1. GLAZE, W.H., M. KOGA & D. CANCILLA. 1989. Ozonation by-products. 2. Improvement of an aqueous-phase derivatization method for the detection of formaldehyde and other carbonyl compounds formed by the ozonation of drinking water. *Environ. Sci. Technol.* 23:838.
2. BAILEY, P.S. 1978. Ozonation in Organic Chemistry, Vol. I, Olefinic Compounds. Chap. 4. Academic Press, New York, N.Y.

* Approved by Standard Methods Committee, 1999.
Joint Task Group: Stephen D. Winslow (chair), Devon A. Cancilla, Patricia Snyder Fair, Shane S. Que Hee, Harumi Yamada.

† This reagent is known under various synonyms. The more common are *O*-(2,3,4,5,6-pentafluorophenyl) methylhydroxylamine hydrochloride with CAS RN 57981-02-9 and pentafluorobenzyloxylamine hydrochloride (PFBOA). It also has appeared with the acronym PFBHOX.

3. NATIONAL ACADEMY OF SCIENCES. 1987. Drinking Water and Health. National Acad. Sciences, Washington, D.C.
4. AMERICAN CONFERENCE OF GOVERNMENTAL INDUSTRIAL HYGIENISTS. 1993. Documentation of the Threshold Limit Values and Biological Exposure Indices, 6th ed. American Conf. Governmental Industrial Hygienists, Cincinnati, Ohio.

6252 B. PFBHA Liquid-Liquid Extraction Gas Chromatographic Method

This method measures straight-chain, low-molecular-weight aldehydes in raw and treated drinking water and simultaneously can analyze for C_1–C_{10} mono-carbonyl saturated aliphatic aldehydes, benzaldehyde, the dialdehyde glyoxal, and the keto-aldehyde methyl glyoxal.[1] The effectiveness of the derivatizing agent (PFBHA) in its reactions with these carbonyl compounds has been reviewed.[2]

1. General Discussion

a. Principle: Samples at room temperature are buffered to pH 4, PFBHA is added, and the samples are placed in a constant-temperature water bath. The carbonyl compounds are converted to their corresponding oximes during reaction with PFBHA. Sulfuric acid is used to protonate excess PFBHA and the oxime derivatives are extracted with hexane. After H_2SO_4 cleanup, the organic extract is analyzed by gas chromatography where the volatile derivatives are easily separated in a temperature-programmable gas chromatograph equipped with a fused-silica capillary column and either an electron-capture detector or selective ion mass spectrometer. Simultaneous analysis and confirmation with a single injection can be effected by setting up both the analytical column and the confirmation column to share a common injection port. Alternatively, use separate analytical and confirmation columns. Aqueous calibration standards similarly are derivatized, extracted, and analyzed. A surrogate recovery standard is added to the samples before derivatization to indicate any variation in derivatization and extraction efficiency.

With the exception of symmetrical ketones and formaldehyde, most carbonyl compounds form two geometrical isomers of oxime derivative. Methyl glyoxal, however, produces only one prominent isomer.

The method described may be used if appropriate quality control can be demonstrated for quantification of all C_1–C_{10} mono-carbonyl, saturated aliphatic aldehydes, benzaldehyde, glyoxal, and methyl glyoxal, but precision and quality control data are presented only for the most commonly found ozonation by-products, namely, formaldehyde, acetaldehyde, heptanal, benzaldehyde, glyoxal, and methyl glyoxal.

b. Interferences: Dissolved ozone, residual chlorine, and other oxidizing substances interfere with the PFBHA reaction. Quantitative addition of potassium iodide (KI) as a reducing agent before derivatization or the addition of ammonium chloride or sulfate at the time of sampling (KI if ozone is present) prevents this interference. Ketones and quinones or large quantities of aldehydes may deplete the PFBHA reagent excess necessary to ensure complete reaction. Waters with high sulfide content inhibit the derivatization of carbonyl compounds. The occurrence of artifacts by aldehyde formation from thermal decomposition of water components is a potential positive interference. Because formaldehyde is used as a preservative for membranes, purified water produced by reverse osmosis is also a potential positive interference. In addition, formaldehyde and acetaldehyde are air pollutants and some formaldehyde in the air can be traced to certain insulation materials. The use of reducing agents such as sodium thiosulfate, sodium sulfite, and ascorbic acid can cause the formation of aldehydes if the sample water contains the corresponding organic acids.

c. Detection level: The method detection level (MDL) and precision data for those aldehydes most commonly found in ozonated waters are given in Table 6252:I. These levels were evaluated for the extracted oximes in hexane from aldehyde-free water. The level of quantitation (LOQ) for these aldehydes usually is set at five times the MDL. In effect, the LOQ for all aldehydes analyzed by this method is 0.5 μg/L except for acetaldehyde and glyoxal, where the value is 1 μg/L. Background formaldehyde contamination may require elevating the LOQ to as high as 5 μg/L. The precision data presented may be matrix-sensitive. Use known standard additions to the matrix if oxime standards are not available.

This method is useful for detecting carbonyl compounds such as short-chain aldehydes ($C_1 - C_{10}$), benzaldehyde, glyoxal, and methyl glyoxal in the range of 1 to 100 μg/L. A clean laboratory reagent water blank, free of these contaminants, is essential.

d. Safety: The toxicity or carcinogenicity of each reagent used in this method has not been defined precisely. Acetaldehyde contains material that can cause cancer in animals, formaldehyde contains material that can cause cancer in humans, and crotonaldehyde causes respiratory tract and eye burns if inhaled or swallowed. Observe proper ventilation and handling procedures. Wear quantitatively-fitted negative-pressure respirators with charcoal air-purifying filter canisters, gloves (such as butyl but not natural rubber, latex, or nitrile[2]), and protective garments resistant to the degrading effects and permeation of these chemicals. Glyoxal and methyl glyoxal are mutagenic in in-vitro tests and the former has subchronic oral toxicity. Take care when handling high concentrations of aldehydes during preparation of primary standards. When handling hexane solutions of the oxime derivatives, wear nitrile gloves (not butyl or latex).

2. Sampling and Storage

See Section 6010B.1 and note the following additional requirements:

Seal sample vials with TFE-lined polypropylene screw caps.* Do not use bakelite black caps made from a formulation con-

* I-Chem Research, Hayward, CA, or equivalent.

TABLE 6252:I. METHOD DETECTION LEVELS AND PRECISION DATA*

Compound	Added Concentration µg/L	Found Concentration µg/L	Standard Deviation µg/L	Relative Standard Deviation %	Method Detection Level† µg/L
Formaldehyde	0.450	1.49‡	0.0485	3.25	0.15
Acetaldehyde†	0.450	0.757‡	0.0242	3.20	0.076
Propanal	0.150	0.200	0.0083	4.15	0.026
Butanal	0.450	0.585	0.0504	8.62	0.16
Crotonaldehyde	0.250	0.242	0.0148	6.12	0.047
Pentanal	0.250	0.326	0.0153	4.70	0.048
Hexanal	0.250	0.307	0.0154	5.02	0.048
Cyclohexanone	0.150	0.199	0.0090	4.52	0.028
Heptanal	0.250	0.323	0.0170	5.25	0.053
Octanal	0.150	0.151	0.0136	8.99	0.043
Benzaldehyde	0.450	0.385	0.0405	10.5	0.13
Nonanal	0.450	0.602	0.0403	6.70	0.13
Decanal	0.450	0.669	0.0561	8.38	0.18
Glyoxal	0.250	0.133	0.220	16.5	0.069
Methyl glyoxal	0.150	0.133	0.0096	7.22	0.030

* Based on the analysis of seven portions of organic-free water with known additions.
† Method detection level for $n = 7$ found by multiplying the standard deviation by 3.14.
‡ Level of quantitation (LOQ) should take into consideration the level of target compound contamination rather than MDL.

taining phenol and formaldehyde. Do not add HCl for this method. Reduce residual free chlorine according to the following method. Additional reagents are required for ozonated samples as described below. If free chlorine is present in the samples, add ammonium chloride or sulfate (0.1 mL of a 20% solution /40 mL sample) before sample collection. Monochloramine may form but will not change the aldehyde concentration of samples subsequently stored at 4°C. If residual ozone is present, the levels of aldehydes may change as the ozone-natural organic matter reaction continues. To prevent this, quench residual ozone by adding 50 µL of a 6.40-g KI/L solution to each 40-mL vial before shipment to the field.

In chlorinated waters, addition of KI will eliminate an interferant that coelutes with the surrogate. The reduction of oxidizing compounds with KI may cause iodine to be extracted into the hexane extract, resulting in a pink-colored extract. Iodine in the extract also may interfere, chromatographically, with the internal standard. The formation of iodine can be suppressed by the addition of 50 µL of 8.52-g mercuric chloride/L solution to each 40-mL vial before shipment to the field. In most situations an ozone residual is unstable and the use of KI and the toxic $HgCl_2$ is unnecessary.

Before sample collection, add 20 mg (for 40 mL sample) of copper sulfate pentahydrate to each bottle to prevent microbial decay of the method analytes. If mercuric chloride is added to suppress iodine formation, copper sulfate pentahydrate need not be added, because mercuric chloride is an effective biocide at the concentration given above. Without a biocide, aldehydes are extremely susceptible to microbial decay. Preservation of the sample is important for obtaining valid data. Ideally, derivatize aldehyde samples immediately after collection. If sample is preserved with a biocide and stored at 4°C, extract within 7 d.

Prepare field reagent blanks from organic-free reagent water (¶ 4*e*).

3. Apparatus

a. Sample containers and extraction vials: 40-mL screw-top, glass sample vials with aldehyde-free caps. (NOTE: Do not use thermoset, phenol-formaldehyde, or urea-formaldehyde caps.) Prepare these, together with the 14-mL amber vials for storing stock solutions, as follows: Wash with detergent, rinse with tap water, soak in 10% HNO_3 for at least 30 min, rinse with tap water, rinse with laboratory organic-free water (¶ 4*e* below), and oven dry at 180°C for at least 1 h.

Clean caps and septa by rinsing with methanol, then with hexane, and dry at 80°C for no more than 1 h in a clean, forced-air convection oven.

b. Microsyringes or micropipets with glass tips, to measure the following volumes: 5, 10, 25, 50, 100, 250, 500, and 1000 µL.

c. Volumetric flasks, 5, 10, and 25 mL, borosilicate glass. Prepare initially by the method of ¶ 3*a*, except that after rinsing with organic-free water, rinse with methanol and invert to drain. Air-dry only. *Do not dry in oven.*

d. Syringe: 20-mL glass hypodermic, metal luer lock tip with 8.9-cm- (3.5-in.-) long × 17 gauge stainless steel pipetting needle (alternatively, use a 20-mL volumetric pipet). Clean as above.

e. Automatic pipet dispensers: To simplify batch processing, add reagent by use of these dispensers. Preferably use adjustable 1-mL and 4-mL sizes with PTFE transfer lines that can be mounted on the suppliers' reagent bottles. If these are not available use 1-mL and 4-mL volumetric pipets.

f. Constant-temperature water bath or incubator, capable of holding multiple 40-mL sample vials and maintaining 35°C ± 0.5°C.

g. Pasteur pipets: Have a selection of short-tipped (14.6-cm or 5.75-in.) and long-tipped (23-cm or 9-in.) pipets.

TABLE 6252:II. ANALYTICAL STANDARDS OF CARBONYL COMPOUNDS USED IN THE PFBHA METHOD

Compound	Purity %	Molecular Weight mg/mmol	Boiling Point °C	Density g/mL
Formaldehyde*	†	33.03	96	1.083
Acetaldehyde‡	99.9	44.05	21	0.788
Propanal (propionaldehyde)*	97	58.08	46–50	0.805
Butanal (*n*-butyraldehyde)‡	99.9	72.11	75	0.800
Pentanal(*n*-valeraldehyde)*	99	86.13	103	0.810
Hexanal (caproaldehyde)*	98	100.16	131	0.834
Heptanal (heptaldehyde)*	95	114.19	153	0.818
Octanal (caprylic aldehyde)*	99	128.22	171	0.821
Nonanal (nonyl aldehyde)‡	99.9	142.24	93	0.827
Decanal (decyl aldehyde)‡	99.9	156.27	207–209	0.830
Benzaldehyde‡	99.9	106.12	178–179	1.044
Glyoxal (ethanedial)‡	§	58.04	50	1.14
Methyl glyoxal (pyruvic aldehyde or 2-oxopropionaldehyde)*	§	72.06	72	1.045

* Aldrich Chemical Company, Inc., Milwaukee, WI, or equivalent.
† Available in 37% solution (by weight) in water.
‡ ChemService, Inc., West Chester, PA, or equivalent.
§ Available in 40% solutions (by weight) in water.

h. Mechanical shaker,† to automate hexane extraction (see Section 6251B.3*e*). Alternatively, use a vortex mixer or manual shaking for 1 min.

i. Storage vials: 7-mL glass, screw-cap vials with PTFE-lined silicone septa cleaned as described above.

j. Gas chromatograph, with capillary columns, temperature programmable, and supplied with a temperature-controlled injector and electron-capture detector.

1) *Gas handling equipment:* Use carrier (helium) and make-up (nitrogen or 95% argon/5% methane) gases of high purity (99.999%) that pass through indicating calcium sulfate, molecular sieve 5A, activated charcoal, and an oxygen-purifying cartridge. Use two-stage metal diaphragm high-purity regulators at the compressed gas sources. Use flow controllers to regulate carrier gas flow. Ensure that all gas lines use 0.3-cm (0.125-in.) copper (or stainless steel) tubing; rinse with high-purity acetone, and bake before use.

2) *Injector*, split/splitless (using straight open-bore insert).

3) *Analytical column*,‡ 30 m long × 0.25-mm ID, fused silica capillary column with a 0.25-μm film thickness.

4) *Confirmation column*,§ 30 m long × 0.25-mm ID, fused silica capillary column with a 0.25-μm film thickness.

5) *Detectors*, a constant current pulse modulated ^{63}Ni ECD with standard size cell (use two ECDs for simultaneous confirmation analysis).

4. Reagents

a. Extraction solvent, UV-grade, glass-distilled hexane.‖

b. Solvent for standard preparation, reagent-grade acetonitrile, free of the target aldehydes.#

c. Preservation agents, ammonium chloride, NH_4Cl, or sulfate, $(NH_4)_2SO_4$, copper sulfate pentahydrate, $CuSO_4 \cdot 5H_2O$, and mercuric chloride, anhydrous, $HgCl_2$.

d. Sulfuric acid, H_2SO_4, conc and 0.2*N*.

e. Organic-free reagent water: Treat water prepared in commercially available water systems to remove all traces of aldehydes. Two methods have been demonstrated: Either expose reagent water produced by a laboratory purification system** to UV irradiation for 1 h or distill reagent water from acidified potassium permanganate (500 mL water with 64 mg potassium permanganate and 1 mL conc H_2SO_4).

Alternative purification techniques, such as addition of another granular activated carbon filtration step, may be used if they can be shown to effectively eliminate background levels of aldehydes. *Do not use a reagent water with formaldehyde contamination to quantify formaldehyde in aqueous samples.*

f. Buffer pH 4 reagent: Add 200 mg potassium hydrogen phthalate per 20 mL aqueous sample.

g. Derivatizing agent, PFBHA:†† Weigh *O*-(2,3,4,5,6-pentafluorobenzyl)hydroxylamine hydrochloride into organic-free water to give a solution concentration of 15 mg/mL. Prepare fresh daily. Prepare enough to add 1 mL/20 mL sample.

h. Standard materials: See Table 6252:II for source and physical characteristics of the standards. Obtain purity assay of each purchased standard before use.

1) *Individual aldehyde standard stock solutions:* Prepare by weighing aldehyde standards in acetonitrile. Weigh between 20 and 70 mg of each standard into a 10-mL volumetric flask. Weigh solid standards directly into the empty flask, then fill with acetonitrile. Add liquid standards to the flask, which has been filled to the neck previously with acetonitrile. Place on a weighing balance and stabilize the weight. Inject liquid standard with a microsyringe directly into the bulk of the acetonitrile and

† Eberbach Corp., or equivalent.
‡ Durabond-5, J&W Scientific, or equivalent.
§ Durabond-1701, J&W Scientific, or equivalent.
‖ Burdick & Jackson, Muskegon, MI, or equivalent.
Sigma Chemical Co., St. Louis, MO, or equivalent.

** Milli-Q, Millipore Corp., Bedford, MA, or equivalent.
†† Aldrich Chemical Co., or equivalent.

TABLE 6252:III. RECOVERY OF TRIPLICATE IN-SITU DERIVATIZED ALDEHYDES COMPARED TO THE RECOVERY OF PURE OXIME DERIVATIVES FROM ORGANIC-FREE WATER

Derivatized Aldehyde	Concentration of Oxime Added* μg/L	Mean Concentration Recovered† μg/L	Standard Deviation μg/L	Relative Standard Deviation %	Mean Recovery‡ %
Formaldehyde	19.2	21.2	0.118	0.56	90.4
Acetaldehyde	19.3	19.3	0.095	0.49	100
Heptaldehyde	19.9	23.6	0.104	0.44	84.3
Benzaldehyde	19.8	18.6	0.138	0.74	107
Glyoxal	19.4	23.7	0.235	0.99	82.0
Methyl glyoxal	20.5	22.1	0.194	0.88	92.5

* Amount of standard oxime added to OPW.
† Evaluated by comparing the relative response of the extracted standard oximes to the calibration curve using in-situ derivatized aldehydes.
‡ Calculated recovery of in-situ derivatized aldehydes compared to standard oximes.

determine exact weight after the addition. Because some aldehyde standards are supplied as aqueous solutions, evaluate weight of actual standard component and make an approximate determination of the required volume to be added from either the density (for pure liquids) or the percentage by weight (for solutions). Due to the high volatility of acetaldehyde, keep in the refrigerator at all times and place measurement syringe in a freezer for 10 min before preparing the stock solution. After diluting to 10 mL with acetonitrile, cap flask and invert three times to mix. Transfer stock solutions to separate 14-mL amber vials with screw caps and PTFE liners and store at 4°C bound with self-adhesive film.‡‡ Stock solutions (except formaldehyde), are usable for up to 3 months. Let them come to room temperature before pipetting. Overcome presence of turbidity or a precipitate by ultrasonication in warm water. If a precipitate persists, make a new stock solution. Prepare formaldehyde stock solutions each month.

Verify the aldehyde concentrations of the aqueous solutions after filtering through 0.45-μm PTFE filters by the sodium bisulfite-iodine titration method.[3]

2) *Multicomponent aldehyde additive standards:* Prepare additive standards solution using individual stock solutions of those aldehydes of interest. Make mixture weekly. The concentration of each component in this additive standard solution should be about 10 mg/L when added to 20-mL aqueous samples. When preparing a calibration curve in 100 mL of organic-free water, prepare two or more multicomponent additive standards from which a volume in the range 10 to 100 μL can be injected directly into the water. For example, if the stock solution concentration is 50 mg in 10 mL, add 20 μL in 10 mL acetonitrile to produce a 10-mg/L additive standard. This is best achieved by first filling the 10-mL volumetric flask to just above the neck with acetonitrile. Inject required volume of each of the stock solutions, using a clean microsyringe for each component, into the bulk of the acetonitrile. After adding all stock solutions, fill to the mark with acetonitrile. Cap and invert three times to mix.

i. Standards derivatives: To determine reaction and extraction efficiency of each aldehyde in different matrices compare the chromatographic response of the derivatized standard in the matrix to that of authentic standards of the oximes. The surrogate standard used to establish optimum conditions for derivatization and laboratory-synthesized oximes for six of the aldehydes have been used to verify recovery of derivatized aldehydes from organic-free water. See Table 6252:III. Unforeseen matrix effects can occur, and because PFBHA-derivatized aldehyde standards (oximes) are not available commercially, some representative syntheses of these derivatives are available.[4]

j. Internal standard, 1,2-dibromopropane and decafluorobiphenyl, 98% purity.§§

1) *Internal standard stock solution:* Weigh 50 mg into a 10-mL volumetric flask containing acetonitrile up to the neck. Fill to mark with acetonitrile. This 5-g/L stock solution can be used for up to 6 months when stored as described in ¶ *h*1) above.

2) *Internal standard working solvent*, 100 μg/L in hexane: Deliver 20 μL internal standard stock solution directly into 1 L hexane in the solvent bottle to be used in the extraction. Cap bottle and invert three times to ensure thorough mixing. This dilution can be used for 4 weeks. To ensure suitability for extraction, run a sample of this working solvent on the GC before extraction of aqueous samples. Before processing samples, provide enough working solvent to extract all calibration and aqueous samples to be analyzed. *Never make up fresh working solvent for use during sample processing.*

k. Surrogate (SUR), 2,3,5,6-tetrafluorobenzaldehyde, 98% pure. Alternative surrogate: 4′-(trifluoromethyl)acetophenone.

1) *Surrogate stock solution*, 20 g/L: Weigh 0.2 g SUR into a 10-mL volumetric flask containing acetonitrile up to the neck. After determining the weight difference, fill to mark with acetonitrile. Stock solutions can be used for up to 6 months if stored as described in ¶ *h*1) above.

2) *Surrogate additive solution*, 20 mg/L: Deliver 10 μL SUR stock solution into a 10-mL volumetric flask and dilute to volume with acetonitrile. This solution can be used for up to 3 months when stored at 4°C. At the beginning of sample processing, add 10-μL surrogate additive solution to each 20-mL sample portion, yielding a surrogate concentration of 10 μg/L.

l. Calibration standards: Prepare aqueous calibration standards in 100 mL organic-free water by injecting a measured amount of the multicomponent aldehyde additive standard solution directly into the water using the solvent flush technique. Prepare five different concentration levels within the expected

‡‡ Parafilm®, American Can Co., Greenwich, CT, or equivalent.

§§ Aldrich Chemical Company, Inc., Milwaukee, WI, or equivalent.

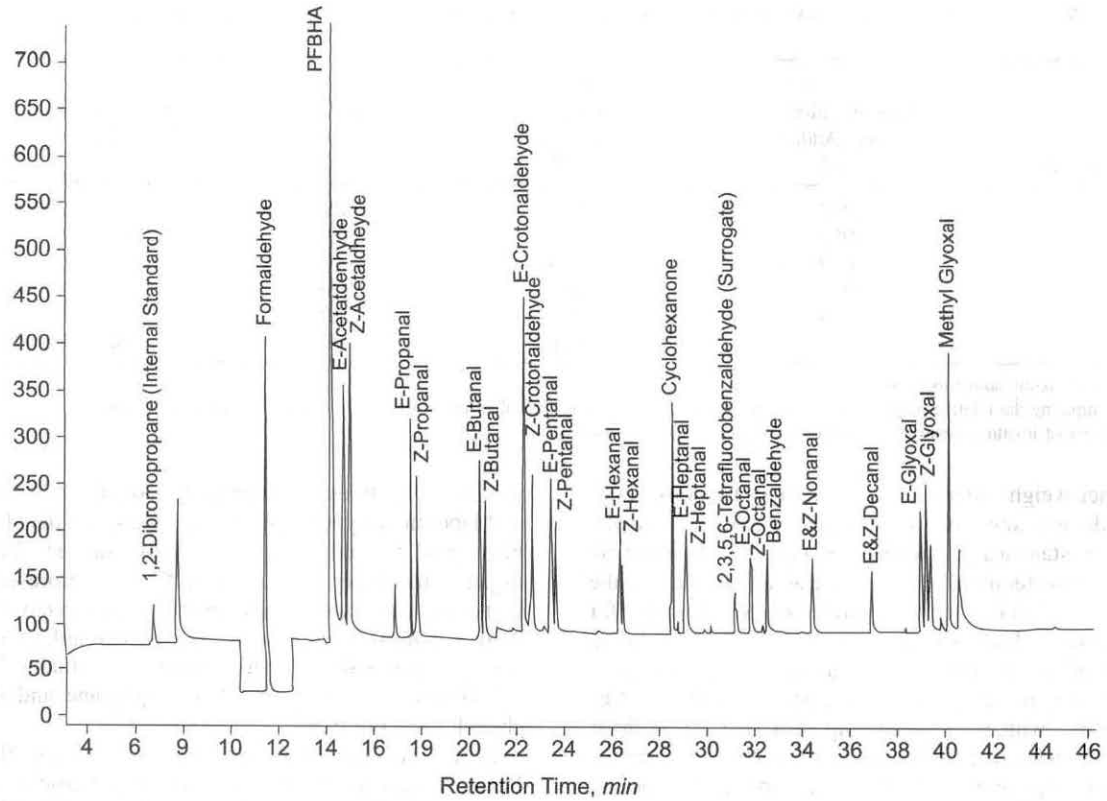

Figure 6252:1. Chromatogram for analytical (primary) column. The formaldehyde peak was attenuated by a factor of four.

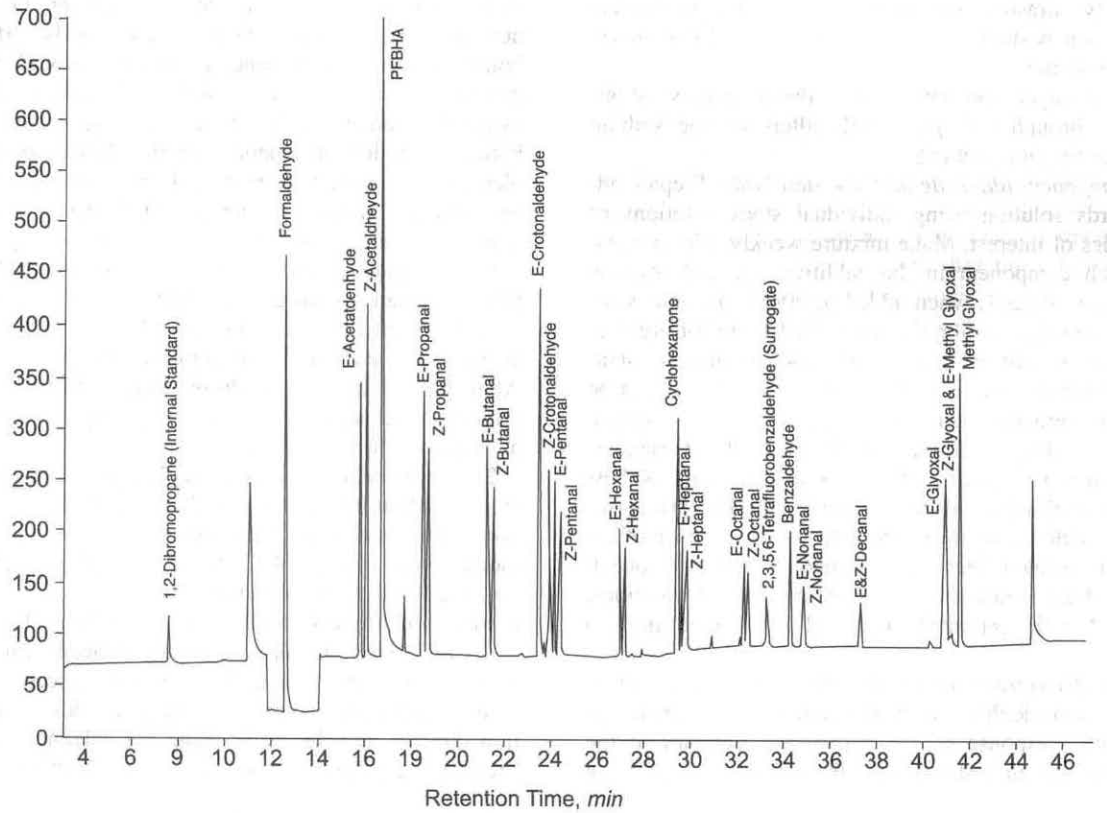

Figure 6252:2. Chromatogram for confirmation column. The formaldehyde peak was attenuated by a factor of four.

TABLE 6252:IV. RETENTION TIMES (RTs) FOR DERIVATIZED CARBONYLS, DERIVATIZED SURROGATE STANDARD, AND INTERNAL STANDARD ON ELECTRON-CAPTURE DETECTOR

Derivatized Carbonyls	Retention Time on Primary Column of ¶ 3*j*3) *min*	Retention on Confirmation Column of ¶ 3*j*4) *min*
1,2-Dibromopropane (internal standard)*	6.81	7.82
Formaldehyde	11.49	12.82
E- & Z-Acetaldehyde†	14.77 & 15.04	15.95 & 16.22
E- & Z-Propanal†	17.64 & 17.86	18.63 & 18.88
E- & Z-Butanal†	20.56 & 20.77	21.44 & 21.68
E- & Z-Crotonaldehyde†	22.41 & 22.73	23.56 & 24.08
E- & Z-Pentanal†	23.52 & 23.70	24.28 & 24.50
E- & Z-Hexanal†	26.44 & 26.59	27.10 & 27.28
Cyclohexanone	28.67	29.54
E- & Z-Heptanal†	29.25 & 29.34	29.78 & 29.92
2,3,5,6-Tetrafluorobenzaldehyde (surrogate)	31.34	33.34
E- & Z-Octanal†	31.96 & 32.04	32.40 & 32.51
Benzaldehyde	32.70	34.38
E- & Z-Nonanal†	34.61 (coelution)	34.93 & 35.01
E- & Z-Decanal†	37.12 (coelution)	37.36 (coelution)
E- & Z-Glyoxal†	39.16 & 39.40	40.92‡
Methyl glyoxal§	40.28	41.64

* 1,2-Dibromopropane, the internal standard, is not derivatized.
† These aldehydes form E- and Z- pentafluorobenzyloxime (PFBO) isomers in an assumed order of elution that has not been confirmed.
‡ On the confirmation column, only the E-isomer of glyoxal was used for quantitation, because Z-glyoxal coeluted with an isomer of derivatized methyl glyoxal.
§Derivatized methyl glyoxal had several conformational isomers, though only one peak was used for quantitation. The isomer peak used had significantly more peak area than two other isomer peaks and was free of coelutions.

sample range of approximately 2 to 40 μg/L. The lowest level of standard will depend on level of blank contamination for each analyte.

5. Procedure

a. Sample preparation: Remove samples and standard solutions from storage and let reach room temperature.

b. Derivatization: Withdraw 20 mL sample from sample vial using a 20-mL glass syringe or glass pipet. Discard remaining sample, shake the vial dry by hand, and return the syringe contents to the vial. Add 10 μL surrogate additive solution using either a microsyringe or automatic pipettor, to all samples and standards. Add 200 mg potassium hydrogen phthalate and dissolve. Add 1 mL freshly prepared 15-mg/mL PFBHA solution to each vial by automatic pipet, secure cap, and swirl to mix gently. Place all samples in a constant-temperature water bath set at 35 ± 0.5°C for 2 h. Remove vials and cool to room temperature for 10 min.

c. Microextraction: To each vial add 0.05 mL (approximately 2 drops) conc H_2SO_4 to quench the derivatization reaction and then add 4 mL hexane working solvent containing the internal standard. When using automated extraction, place vials in a mechanical wooden shaker box. Shake vials on fast setting (approximately 300 cycles) for 3.5 min. Alternatively, shake manually for approximately 3 min. Remove vials and place upright. Let stand for approximately 5 min to permit phases to separate.

d. Extraction cleanup: Draw off top hexane layer using a clean 14.6-cm (5.75-in.) disposable pasteur pipet for each sample into a smaller 7-mL clear vial containing 3 mL 0.2*N* H_2SO_4. Shake for 30 s by hand and let stand for approximately 5 min for phase separation. This wash step reduces the amount of PFBHA reagent and other interferants in the hexane extract. Do not skip this step. Draw off top hexane layer using another clean 14.6-cm (5.75-in.) disposable pasteur pipet for each sample and place in two 1.8-mL autosampler vials per sample. Store extra autosampler vials in a refrigerator at 4°C as a backup extract. Analyze extracts within 14 d of extraction.

e. Gas chromatography: Use the following operating conditions for the gas chromatograph: injector temperature 180°C; split valve open at 0.5 min; split flow at 50 mL/min; temperature program: 50°C for 1 min, rising at 4°C/min to 220°C and then at 20°C/min to 250°C; detector temperature: 300°C; carrier gas flow: 1.5 mL/min at 100°C; make-up gas flow: 27 mL/min.

At the beginning of each analysis, inject one hexane solvent blank to condition the GC and to verify that there are no interferences present. Inject 1 μL onto the splitless injector. See Figures 6252:1 and 6252:2 for examples of chromatograms obtained with the above GC conditions for both the analytical and confirmation columns. If dual-column analysis is unavailable, use the column specified in ¶ 3*j*3) above, but be aware of possible interferences. Table 6252:IV lists retention times for both columns.

Tabulate GC peak area response and concentration for each analyte and the internal standard. The formaldehyde peak will be much larger, for the same concentration, than the other analyte peaks. The formaldehyde peak may need to be attenuated on some instruments to avoid signal saturation.

For most asymmetrical carbonyl compounds derivatized with PFBHA, two isomers, referred to as (E) and (Z), are formed. Chromatographic resolution is usually obtained with the columns suggested for acetaldehyde, propanal, butanal, pentanal,

hexanal, heptanal, octanal, and crotonaldehyde. With dicarbonyl species such as glyoxal and methyl glyoxal, (E) and (Z) isomerism occurs during oxime formation, increasing the number of isomers. Use one of the following methods for both calibration and quantification of each method analyte.

1) Use the sum of the isomer peak areas for each constituent for both calibration and quantification.

2) Use peak area of each individual isomer to calculate independently a concentration for each isomer. Then calculate average amount for the two isomers to report one value for the analyte.

f. Calibration: Demonstrate acceptable initial calibration before analyzing any samples. Carry standards (¶ 4*l*) through entire derivatization and extraction procedure as above. This corrects for any recovery characteristics inherent in the method. Analyze the calibration standards under the same GC conditions as the samples.

Generate a calibration curve for each analyte by plotting the area ratios (A_a/A_{is}) against the concentration ratios (C_a/C_{is}) of the five calibration standards where:

A_a = peak area of analyte (or analyte isomer pair),
A_{is} = peak area of internal standard,
C_a = concentration of analyte, and
C_{is} = concentration of internal standard.

This curve must be forced through zero and can be defined as either first or second order. Forcing through zero allows for a better estimate of the background level of method analytes.

6. Quality Control

a. General considerations: PFBHA is a highly reactive *O*-substituted hydroxylamine. Like hydroxylamine, PFBHA reacts readily with a variety of carbonyl functional groups to produce corresponding oximes. The ease with which PFBHA reacts with carbonyl-containing compounds makes the potential contamination of samples a serious concern. Lower-molecular-weight aldehydes are commonly found in laboratory and outside air and can ultimately contaminate water samples, leading to incorrect calculation of aldehyde concentrations. As a further concern, PFBHA, especially in moist laboratory environments, can react to form oximes when directly exposed to aldehydes in air. For these reasons, exercise care to reduce the sources and exposure of samples, standard solutions, and PFBHA reagents to aldehyde contaminants. Consider storing PFBHA in a desiccator under an inert atmosphere, drying laboratory solvents with molecular sieves, using purified water, and making fresh derivatizing stocks on a regular basis. If, after analysis of appropriate sample blanks, contamination remains a problem, the source of the problem may be in the PFBHA reagent and solutions. Recrystallization of PFBHA may be necessary to remove oximes formed as a result of reagent contamination.

The effects of chromatographic and analytical conditions on E/Z ratios of the oximes have not been fully explored. The possibility of changing E/Z ratios under differing analytical conditions, such as injection temperature, requires that analytical conditions be carefully controlled. E/Z ratios may change as a function of time; therefore analyze samples as soon as possible after preparation and within groups. Use the sum of the isomer peak areas for each constituent for both calibration and quantification. With dicarbonyl species such as glyoxal, E/Z isomerism occurs from oxime formation with both carbonyl groups, increasing the number of possible isomers. Formation of the mono-derivatives from these di-carbonyl species may pose a problem if analytical conditions do not favor the complete derivatization of both carbonyl groups. Mono-derivatives have been shown to have similar retention and mass spectral characteristics as single carbonyl-containing oxime derivatives, potentially leading to incorrect identification and underestimation of the amounts of di-carbonyl species present in water samples. This method has been validated for the recovery of oxime derivatives of aldehydes from organic-free water; the recovery of the surrogate standard from this matrix appears to reflect method performance. Consequently, compare the *RRF* of surrogate standard extracted from aqueous samples to the value obtained when building the calibration curve. If these values are outside the range for accepted mean recovery values of 30% (see Section 6251B.7*c*), authentic oxime standards may have to be used to validate the method for the new matrix. In this case, if pure standards are unavailable, recognize that analyses of aldehydes are semi-quantitative; report as such.

b. Monitoring for interferences:

1) Solvent blank—Analyze each reagent bottle of hexane containing internal standard before it is used. If there are any spurious peaks in the chromatogram, solvent purity has been compromised. Remake the working solvent.

2) Method blank—See Section 6251B.7*b*.

3) Travel or shipping blanks—Prepare blanks for each sampling location in the laboratory by filling 40-mL vials, as described above, with organic-free water and containing the same reagents present (if any) in the sample vials. Ship to the sampling site and back to the laboratory with the sample bottles. Do not open these bottles in the field.

c. Internal standard assessment: Injections of the hexane extracts are acceptable if the area counts of the internal standard peak do not vary more than ±20% from the mean of all the samples analyzed with the same batch of PFBHA. Reanalyze samples that do not meet this precision. If, after reinjection, criteria are still not met, the sample holding time has not been exceeded, and the same working solvent used for constructing the calibration curve is still available, the second vial may be analyzed.

d. Verification of calibration standard materials: Analyze a laboratory-fortified blank from standard materials from a source other than those used to prepare the initial calibration curve. Calculate the concentration of this quality control sample (QCS) from the calibration curve. The calculated concentration of the QCS must be within 60 to 140% of its true value. This step validates the calibration standard materials before sample analysis.

e. Ongoing calibration: Time, temperature, pH, and PFBHA concentration affect the rate, efficiency, and reproducibility of the derivatization reaction. Calibration frequency depends on the laboratory's ability to control these parameters so that continuing calibration check standard criteria can be met. After a successful initial calibration, maintain an ongoing calibration by either of the following options:

- Verify initial calibration daily using a minimum of two calibration standards. Prepare a minimum of one low-level calibration standard (suggested concentration 2 to 5 μg/L) and one mid-level calibration standard (suggested concentration 10 to 30 μg/L) with each batch of samples. Using these two standards, verify calibration before analyzing the sample extracts from the

batch. In addition, reanalyze one of these two standard extracts after every tenth sample extract, and after the last sample in an analysis batch to ensure instrument stability throughout the analysis batch. Recovery must be within 70 to 130% of the true value for the mid-level standard, and within 50 to 150% of the true value for the low-level standard.

• Calibrate method daily with all five calibration standards. Some laboratories may find it more productive to prepare and analyze a calibration curve with each batch of samples.

f. Surrogate standard recovery: Add the surrogate (2,3,5,6-tetrafluorobenzaldehyde) directly to the 20-mL aqueous sample portions before reagent addition to monitor constituent recovery from the sample matrix. If the surrogate area is low or absent, there is likely to be a problem with derivatization or extraction that needs to be resolved before quantification can be undertaken (see Section 6251B.7*e*). A sample extract is acceptable if the area counts of the surrogate peak (or the *RRF* values compared to an acceptable internal standard area) do not vary more than 30% from other samples analyzed with the same batch of PFBHA.

g. Sample quantification: See Section 6251B.7*f*.

h. Matrix additions: See Section 6251B.7*g*.

i. Replicate analysis: See Section 6251B.7*h*.

7. References

1. Sclimenti, M.J., S.W. Krasner, W.H. Glaze & H.S. Weinberg. 1990. Ozone disinfection by-products: Optimization of the PFBHA derivatization method for the analysis of aldehydes. *In* Advances in Water Analysis and Treatment, Proc. 18th Annu. AWWA Water Quality Technology Conf., Nov. 11–15, 1990, San Diego, Calif., p. 477. American Water Works Assoc., Denver, Colo.
2. Forsberg, K. 1989. Chemical Protective Clothing Performance Index Book. John Wiley & Sons, New York, N.Y.
3. American Society for Testing and Materials. 1989. Annual Book of ASTM Standards: Water and Environmental Technology. Vol. 11.01. American Soc. Testing & Materials, Philadelphia, Pa.
4. Cancilla, D.A., C.C. Chou, R. Barthel & S.S. Que Hee. 1992. Characterization of the *O*-(2,3,4,5,6-pentafluorobenzyl)-hydroxylamine hydrochloride (PFBOA) derivatives of some aliphatic mono- and dialdehydes and quantitative water analysis of these aldehydes. *J. Assoc. Offic. Anal. Chem. Int.* 75:842.

6410 EXTRACTABLE BASE/NEUTRALS AND ACIDS*

6410 A. Introduction

1. Sources and Significance

The semivolatile compounds covered by this section include many classes of compounds, each characterized by different sources. The compounds include polynuclear aromatic hydrocarbons, often as by-products of petroleum processing or combustion; phthalates, used as plasticizers; phenolics, found most often in wood preservatives; organochlorine pesticides, found most often in agricultural runoff or in wastewaters draining such areas; and PCBs (also see Section 6431A). Many of the listed compounds are toxic or carcinogenic. However, they generally are relatively insoluble in water so they do not occur frequently in potable waters or most wastewaters.

* Approved by Standard Methods Committee, 2000.

2. Selection of Method

Method 6410B is a broad-spectrum gas chromatographic/mass spectrometric (GC/MS) packed- or capillary-column method for detection of these compounds following liquid-liquid extraction. Although this method can be used to determine all the listed compounds, it is not the most sensitive method for individual classes of compounds, which are detected at lower concentrations by GC methods such as those listed in 6420C (phenols), 6440B and C (polynuclear aromatic hydrocarbons), and 6630C and D (organochlorine pesticides and PCBs). In some cases, notably the pesticides, the GC method is substantially more sensitive than the GC/MS method. In other cases, such as the phenols, there is less difference between the methods.

6410 B. Liquid-Liquid Extraction Gas Chromatographic/Mass Spectrometric Method

This method[1] is applicable to the determination of organic compounds that are partitioned into an organic solvent and are amenable to gas chromatography,* in municipal and industrial discharges.

1. General Discussion

a. Principle: A measured volume of sample is extracted serially with methylene chloride at a pH above 11 and again at a pH below 2. The extract is dried, concentrated, and analyzed by GC/MS.[2,3] Qualitative compound identification is based on retention time and relative abundance of three characteristic masses (*m/z*). Quantitative analysis uses internal-standard techniques with a single characteristic *m/z*.

b. Interferences:

1) General precautions—See Section 6010C. Method interferences may be caused by contaminants in solvents, reagents, glassware, and other sample-processing hardware that lead to discrete artifacts and/or elevated base lines in detector output. Routinely demonstrate that all materials are free from interferences under the conditions of the analysis by running laboratory reagent blanks as described in Section 6200A.5*c*3).

Clean all glassware thoroughly[4] as soon as possible after use by rinsing with the last solvent used in it, followed by detergent washing with hot water and rinsing with tap water and distilled water. Drain glassware dry and heat in a muffle furnace at 400°C for 15 to 30 min. Some thermally stable materials, such as PCBs, may not be eliminated by this treatment. Solvent rinses with acetone and pesticide-quality hexane may be substituted for the baking. Thorough rinsing with such solvents usually eliminates PCB interference. Do not heat volumetric ware in a muffle furnace. After drying and cooling, seal and store glassware in a clean environment to prevent accumulation of dust or other contaminants. Store inverted or capped with aluminum foil.

Use high-purity reagents and solvents to minimize interference. Purification of solvents by distillation in all-glass systems may be required.

Matrix interferences may be caused by coextracted contaminants. The extent of matrix interferences will vary considerably depending on the sample.

2) Special precautions—Benzidine can be lost by oxidation during solvent concentration. Under the alkaline conditions of the extraction step, α-BHC, γ-BHC, endosulfan I and II, and endrin are subject to decomposition. Hexachlorocyclopentadiene is subject to thermal decomposition in the inlet of the gas chromatograph, chemical reaction in acetone solution, and photochemical decomposition. *N*-nitrosodimethylamine is difficult to separate from the solvent under the chromatographic conditions described. *N*-nitrosodiphenylamine decomposes in the gas chromatographic inlet and cannot be separated from diphenylamine. Other methods may be preferred for these compounds.[1]

The base-neutral extraction may cause significantly reduced recovery of phenol, 2-methylphenol, and 2,4-dimethylphenol. Results obtained under these conditions are minimum concentrations.

The packed gas chromatographic columns recommended for the basic fraction may not be able to resolve certain isomeric pairs including the following: anthracene and phenanthrene; chrysene and benzo(a)anthracene; and benzo(b)fluoranthene and benzo(k)fluoranthene because retention time and mass spectra for these pairs are not sufficiently different to make unambiguous identification possible. Use alternative techniques, such as the method for polynuclear aromatic hydrocarbons (Section 6440B), to identify and quantify these compounds.

In samples containing many interferences, use chemical ionization (CI) mass spectrometry to make identification easier. Tables 6410:I and II give characteristic CI ions for most compounds covered by this method. Use of CI mass spectrometry to support electron ionization (EI) mass spectrometry is encouraged but not required.

c. Detection levels: The method detection level (MDL) is the minimum concentration of a substance that can be measured and reported with 99% confidence that the value is above zero.[5] The MDL concentrations listed in Tables 6410:I and II were obtained with reagent water.[6] The MDL actually obtained in a given analysis will vary, depending on instrument sensitivity and matrix effects.

d. Safety: The toxicity or carcinogenicity of each reagent has not been defined precisely. Benzo(a)anthracene, benzidine, 3,3′-dichlorobenzidine, benzo(a)pyrene, α-BHC, β-BHC, δ-BHC, γ-BHC, dibenzo(a,h)anthracene, *n*-nitrosodimethylamine, 4,4′-DDT, and polychlorinated biphenyls (PCBs) have been tentatively classified as known or suspected, human or mammalian carcinogens. Prepare primary standards of these compounds in a hood and wear a NIOSH/MESA-approved toxic gas respirator when handling high concentrations.

2. Sampling and Storage

Collect grab samples in 1-L amber glass bottles fitted with a screw cap lined with TFE. Foil may be substituted for TFE if the sample is not corrosive. If amber bottles are not available, protect samples from light. Wash and rinse bottle and cap liner

---

* *Base/neutral extractables*: acenaphthene, acenaphthylene, anthracene, aldrin, benzo(a)anthracene, benzo(b)fluoranthene, benzo(k)fluoranthene, benzo(a)pyrene, benzo(ghi)perylene, benzyl butyl phthalate, β-BHC, δ-BHC, bis(2-chloroethyl) ether, bis(2-chloroethoxy) methane, bis(2-ethylhexyl) phthalate, bis(2-chloroisopropyl) ether more correctly known as 2,2-oxybis (1-chloropropane), 4-bromophenyl phenyl ether, chlordane, 2-chloronaphthalene, 4-chlorophenyl phenyl ether, chrysene, 4,4′-DDD, 4,4′-DDE, 4,4′-DDT, dibenzo(a,h)anthracene, di-*n*-butylphthalate, 1,3-dichlorobenzene, 1,2-dichlorobenzene, 1,4-dichlorobenzene, 3,3′-dichlorobenzidine, dieldrin, diethyl phthalate, dimethyl phthalate, 2,4-dinitrotoluene, 2,6-dinitrotoluene, di-*n*-octylphthalate, endosulfan sulfate, endrin aldehyde, fluoranthene, fluorene, heptachlor, heptachlor epoxide, hexachlorobenzene, hexachlorobutadiene, hexachloroethane, indeno(1,2,3-cd)pyrene, isophorone, naphthalene, nitrobenzene, *N*-nitrosodi-*n*-propylamine, PCB-1016, PCB-1221, PCB-1232, PCB-1242, PCB-1248, PCB-1254, PCB-1260, phenanthrene, pyrene, toxaphene, 1,2,4-trichlorobenzene.

Acid extractables: 4-chloro-3-methylphenol, 2-chlorophenol, 2,4-dichlorophenol, 2,4-dimethylphenol, 2,4-dinitrophenol, 2-methyl-4,6-dinitrophenol, 2-nitrophenol, 4-nitrophenol, pentachlorophenol, phenol, 2,4,6-trichlorophenol.

The method may be extended to include the following compounds: benzidine, α-BHC, γ-BHC, endosulfan I, endosulfan II, endrin, hexachlorocyclopentadiene, *N*-nitrosodimethylamine, *N*-nitrosodiphenylamine.

with acetone or methylene chloride, and dry before use. Follow conventional sampling practices[7] but do not rinse bottle with sample. Collect composite samples in refrigerated glass containers. Optionally, use automatic sampling equipment as free as possible of plastic tubing and other potential sources of contamination; incorporate glass sample containers for collecting a minimum of 250 mL. Refrigerate sample containers at 4°C and protect from light during compositing. If the sampler includes a peristaltic pump, use a minimum length of compressible silicone rubber tubing, but before use, thoroughly rinse it with methanol and rinse repeatedly with distilled water to minimize contamination. Use an integrating flow meter to collect flow-proportional composites.

Fill sample bottles and, if residual chlorine is present, add 80 mg sodium thiosulfate per liter of sample and mix well. Ice all samples or refrigerate at 4°C from time of collection until extraction.

Extract samples within 7 d of collection and analyze completely within 40 d of extraction.

3. Apparatus

a. Separatory funnel, 2-L, with TFE stopcock.

b. Drying column, chromatographic, 400 mm long × 19 mm ID, with coarse frit filter disk.

c. Concentrator tube, Kuderna-Danish, 10-mL, graduated.† Check calibration at volumes used. Use ground-glass stopper to prevent evaporation.

d. Evaporative flask, Kuderna-Danish, 500-mL.‡ Attach to concentrator tube with springs.

e. Snyder column, Kuderna-Danish, three-ball macro.§

f. Snyder column, Kuderna-Danish, two-ball micro.‖

g. Vials, 10- to 15-mL, amber glass, with TFE-lined screw cap.

† Kontes K-570050-1025 or equivalent.
‡ Kontes K-570001-0500 or equivalent.
§ Kontes K-503000-0121 or equivalent.
‖ Kontes K-569001-0219 or equivalent.

TABLE 6410:I. CHROMATOGRAPHIC CONDITIONS, METHOD DETECTION LEVELS, AND CHARACTERISTIC MASSES FOR BASE/NEUTRAL EXTRACTABLES

Compound	Retention Time *min*	Method Detection Level *µg/L*	Characteristic Masses					
			Electron Impact			Chemical Ionization		
			Primary	Secondary	Secondary	Methane	Methane	Methane
1,3-Dichlorobenzene	7.4	1.9	146	148	113	146	148	150
1,4-Dichlorobenzene	7.8	4.4	146	148	113	146	148	150
Hexachloroethane	8.4	1.6	117	201	199	199	201	203
bis(2-Chloroethyl) ether	8.4	5.7	93	63	95	63	107	109
1,2-Dichlorobenzene	8.4	1.9	146	148	113	146	148	150
bis(2-Chloroisopropyl) ether*	9.3	5.7	45	77	79	77	135	137
N-Nitrosodi-*n*-propylamine			130	42	101			
Nitrobenzene	11.1	1.9	77	123	65	124	152	164
Hexachlorobutadiene	11.4	0.9	225	223	227	223	225	227
1,2,4-Trichlorobenzene	11.6	1.9	180	182	145	181	183	209
Isophorone	11.9	2.2	82	95	138	139	167	178
Naphthalene	12.1	1.6	128	129	127	129	157	169
bis(2-Chloroethoxy) methane	12.2	5.3	93	95	123	65	107	137
Hexachlorocyclopentadiene†	13.9		237	235	272	235	237	239
2-Chloronaphthalene	15.9	1.9	162	164	127	163	191	203
Acenaphthylene	17.4	3.5	152	151	153	152	153	181
Acenaphthene	17.8	1.9	154	153	152	154	155	183
Dimethyl phthalate	18.3	1.6	163	194	164	151	163	164
2,6-Dinitrotoluene	18.7	1.9	165	89	121	183	211	223
Fluorene	19.5	1.9	166	165	167	166	167	195
4-Chlorophenyl phenyl ether	19.5	4.2	204	206	141			
2,4-Dinitrotoluene	19.8	5.7	165	63	182	183	211	223
Diethyl phthalate	20.1	1.9	149	177	150	177	223	251
N-Nitrosodiphenylamine†	20.5	1.9	169	168	167	169	170	198
Hexachlorobenzene	21.0	1.9	284	142	249	284	286	288
α-BHC†	21.1		183	181	109			
4-Bromophenyl phenyl ether	21.2	1.9	248	250	141	249	251	277
γ-BHC†	22.4		183	181	109			
Phenanthrene	22.8	5.4	178	179	176	178	179	207
Anthracene	22.8	1.9	178	179	176	178	179	207
β-BHC	23.4	4.2	181	183	109			
Heptachlor	23.4	1.9	100	272	274			
δ-BHC	23.7	3.1	183	109	181			
Aldrin	24.0	1.9	66	263	220			
Dibutyl phthalate	24.7	2.5	149	150	104	149	205	279

TABLE 6410:I. CONT.

Compound	Retention Time *min*	Method Detection Level *µg/L*	Characteristic Masses					
			Electron Impact			Chemical Ionization		
			Primary	Secondary	Secondary	Methane	Methane	Methane
Heptachlor epoxide	25.6	2.2	353	355	351			
Endosulfan I†	26.4		237	338	341			
Fluoranthene	26.5	2.2	202	101	100	203	231	243
Dieldrin	27.2	2.5	79	263	279			
4,4′-DDE	27.2	5.6	246	248	176			
Pyrene	27.3	1.9	202	101	100	203	231	243
Endrin†	27.9		81	263	82			
Endosulfan II†	28.6		237	339	341			
4,4′-DDD	28.6	2.8	235	237	165			
Benzidine†	28.8	44	184	92	185	185	213	225
4,4′-DDT	29.3	4.7	235	237	165			
Endosulfan sulfate	29.8	5.6	272	387	422			
Endrin aldehyde			67	345	250			
Butyl benzyl phthalate	29.9	2.5	149	91	206	149	299	327
bis(2-Ethylhexyl) phthalate	30.6	2.5	149	167	279	149		
Chrysene	31.5	2.5	228	226	229	228	229	257
Benzo(a)anthracene	31.5	7.8	228	229	226	228	229	257
3,3′-Dichlorobenzidine	32.2	16.5	252	254	126			
Di-*n*-octyl phthalate	32.5	2.5	149					
Benzo(b)fluoranthene	34.9	4.8	252	253	125	252	253	281
Benzo(k)fluoranthene	34.9	2.5	252	253	125	252	253	281
Benzo(a)pyrene	36.4	2.5	252	253	125	252	253	281
Indeno(1,2,3-cd)pyrene	42.7	3.7	276	138	277	276	277	305
Dibenzo(a,h)anthracene	43.2	2.5	278	139	279	278	279	307
Benzo(ghi)perylene	45.1	4.1	276	138	277	276	277	305
N-Nitrosodimethylamine†			42	74	44			
Chlordane‡	19–30		373	375	377			
Toxaphene‡	25–34		159	231	233			
PCB 1016‡	18–30		224	260	294			
PCB 1221‡	15–30	30	190	224	260			
PCB 1232‡	15–32		190	224	260			
PCB 1242‡	15–32		224	260	294			
PCB 1248‡	12–34		294	330	262			
PCB 1254‡	22–34	36	294	330	362			
PCB 1260‡	23–32		330	362	394			

*The proper chemical name is 2,2′-oxybis(1-chloropropane).
†See introductory section of text.
‡These compounds are mixtures of various isomers. (See Figures 6410:2 through 12.)
Column conditions: Supelcoport (100/120 mesh) coated with 3% SP-2250 packed in a 1.8 m long × 2 mm ID glass column with helium carrier gas at 30 mL/min flow rate. Column temperature held isothermal at 50°C for 4 min, then programmed at 8°C/min to 270°C and held for 30 min.

h. Continuous liquid-liquid extractor, equipped with TFE or glass connecting joints and stopcocks requiring no lubrication.#

i. Boiling chips, approximately 10/40 mesh. Heat to 400°C for 30 min or extract in a Soxhlet extractor with methylene chloride.

j. Water bath, heated, with concentric ring cover and temperature control to ±2°C. Use bath in a hood.

k. Balance, analytical, capable of accurately weighing 0.0001 g.

*l. Gas chromatograph:*** An analytical system complete with a temperature-programmable gas chromatograph and all required accessories including syringes, analytical columns, and gases. Use chromatograph with the injection port designed for on-column injection when packed columns are used and for splitless injection when capillary columns are used.

1) *Column for base/neutrals*, 1.8 m long × 2-mm ID glass, packed with 3% SP-2250 on Supelcoport (100/200 mesh) or equivalent. This column was used to develop the detection level and precision and bias data presented herein. Guidelines for the use of alternate columns (e.g., DB-5 fused silica capillary) are provided in ¶ 5*b*.

2) *Column for acids*, 1.8 m long × 2-mm ID glass, packed with 1% SP-1240DA on Supelcoport (100/120 mesh) or equivalent. The detection level and precision and bias data presented herein were developed with this column. For guidelines for the

Hershberg-Wolf Extractor, Ace Glass Co., Vineland, NJ, P/N 6841-10, or equivalent.
** Gas chromatographic methods are extremely sensitive to the materials used. Mention of trade names by *Standard Methods* does not preclude the use of other existing or as-yet-undeveloped products that give *demonstrably* equivalent results.

TABLE 6410:II. CHROMATOGRAPHIC CONDITIONS, METHOD DETECTION LEVELS, AND CHARACTERISTIC MASSES FOR ACID EXTRACTABLES

Compound	Retention Time *min*	Method Detection Level *µg/L*	Characteristic Masses					
			Electron Impact			Chemical Ionization		
			Primary	Secondary	Secondary	Methane	Methane	Methane
2-Chlorophenol	5.9	3.3	128	64	130	129	131	157
2-Nitrophenol	6.5	3.6	139	65	109	140	168	122
Phenol	8.0	1.5	94	65	66	95	123	135
2,4-Dimethylphenol	9.4	2.7	122	107	121	123	151	163
2,4-Dichlorophenol	9.8	2.7	162	164	98	163	165	167
2,4,6-Trichlorophenol	11.8	2.7	196	198	200	197	199	201
4-Chloro-3-methylphenol	13.2	3.0	142	107	144	143	171	183
2,4-Dinitrophenol	15.9	42	184	63	154	185	213	225
2-Methyl-4,6-dinitrophenol	16.2	24	198	182	77	199	227	239
Pentachlorophenol	17.5	3.6	266	264	268	267	265	269
4-Nitrophenol	20.3	2.4	65	139	109	140	168	122

Column conditions: Supelcoport (100/120 mesh) coated with 1% SP-1240DA packed in a 1.8-m-long × 2-mm-ID glass column with helium carrier gas at 30 mL/min flow rate. Column temperature held isothermal at 70°C for 2 min then programmed at 8°C/min to 200°C.

use of alternate columns (e.g., DB-5 fused silica capillary) see ¶ 5*b*.

m. Mass spectrometer, capable of scanning from 35 to 450 amu every 7 s or less, utilizing 70-V (nominal) electron energy in the electron impact ionization mode, and producing a mass spectrum that meets all the criteria in Table 6410:III when 50 ng of decafluorotriphenyl phosphine [DFTPP; bis(perfluorophenyl) phenyl phosphine] is injected through the GC inlet.

n. GC/MS interface: Any GC to MS interface that gives acceptable calibration points at 50 ng or less per injection for each of the compounds of interest and achieves all acceptable performance criteria may be used. GC to MS interfaces constructed of all glass or glass-lined materials are recommended. Glass can be deactivated by silanizing with dichlorodimethylsilane.

o. Data system: See Section 6200B.2*f*.

4. Reagents

a. Reagent water: See Section 6200B.3*a*.

b. Sodium hydroxide solution, NaOH, 10*N:* Dissolve 40 g NaOH in reagent water and dilute to 100 mL.

c. Sodium sulfate, Na_2SO_4, granular, anhydrous. Purify by heating at 400°C for 4 h in a shallow tray.

TABLE 6410:III. DFTPP KEY MASSES AND ABUNDANCE CRITERIA

Mass	*m/z* Abundance Criteria
51	30–60% of mass 198
68	Less than 2% of mass 69
70	Less than 2% of mass 69
127	40–60% of mass 198
197	Less than 1% of mass 198
198	Base peak, 100% relative abundance
199	5–9% of mass 198
275	10–30% of mass 198
365	Greater than 1% of mass 198
441	Present but less than mass 443
442	Greater than 40% of mass 198
443	17–23% of mass 442

d. Sodium thiosulfate, $Na_2S_2O_3 \cdot 5H_2O$, granular.

e. Sulfuric acid, H_2SO_4, 1 + 1: Slowly add 50 mL conc H_2SO_4 to 50 mL reagent water.

f. Acetone, methanol, methylene chloride, pesticide quality or equivalent.

g. Stock standard solutions: Prepare from pure standard materials or purchase as certified solutions. Prepare by accurately weighing about 0.0100 g of pure material, dissolve in pesticide-quality acetone or other suitable solvent, and dilute to volume in a 10-mL volumetric flask; 1 µL = 1.00 µg compound. When compound purity is assayed to be 96% or greater, use the weight without correction to calculate concentration of the stock standard. Use commercially prepared stock standards at any concentration if certified by the manufacturer or by an independent source.

Transfer stock standard solutions into TFE-sealed screw-cap bottles. Store at 4°C and protect from light. Check stock standard solutions frequently for signs of degradation or evaporation, especially just before preparing calibration standards. Replace stock standard solutions after 6 months, or sooner if comparison with check standards indicates a problem.

h. Surrogate standard known-addition solution: Select a minimum of three surrogate compounds from Table 6410:IV. Prepare a surrogate standard solution containing each selected surrogate compound at a concentration of 100 µg/mL in acetone. Adding 1.00 mL to 1000 mL sample is equivalent to a concentration of 100 µg/L of each surrogate standard. Store at 4°C in TFE-sealed glass container. Check solution frequently for stability. Replace solution after 6 months, or sooner if comparison with quality-control check standards indicates a problem.

i. DFTPP standard: Prepare a 25-µg/mL solution of DFTPP in acetone.

j. Calibration standards: Prepare calibration standards at a minimum of three concentration levels for each compound by adding appropriate volumes of one or more stock standards to a volumetric flask. To each calibration standard or standard mixture, add a known constant amount of one or more internal standards (such as those listed in Table 6410:IV), and dilute to volume with acetone. Prepare one calibration standard at a con-

TABLE 6410:IV. SUGGESTED INTERNAL AND SURROGATE STANDARDS

Base/Neutral Fraction	Acid Fraction
Aniline-d_5	2-Fluorophenol
Anthracene-d_{10}	Pentafluorophenol
Benzo(a)anthracene-d_{12}	Phenol-d_5
4,4′-Dibromobiphenyl	2-Perfluoromethyl phenol
4,4′-Dibromooctafluorobiphenyl	
Decafluorobiphenyl	
2,2′-Difluorobiphenyl	
4-Fluoroaniline	
1-Fluoronaphthylene	
2-Fluoronaphthylene	
Naphthalene-d_8	
Nitrobenzene-d_5	
2,3,4,5,6-Pentafluorobiphenyl	
Phenanthrene-d_{10}	
Pyridine-d_5	

centration near, but above, the MDL and others corresponding to the expected range of sample concentrations or defining the working range of the GC/MS system.

k. Quality control (QC) check sample concentrate: Obtain a check sample concentrate containing each compound at a concentration of 100 µg/mL in acetone. Multiple solutions may be required. PCBs and multicomponent pesticides may be omitted. If such a sample is not available from an external source, prepare using stock standards prepared independently from those used for calibration.

5. Procedure

a. Extraction: Extraction by means of a separatory funnel, ¶ 1), is most common, but if emulsions will prevent acceptable solvent recovery, use continuous extraction, ¶ 2).

1) Separatory funnel extraction—Normally use a sample volume of 1 L. For sample volumes of 2 L, use 250-, 100-, and 100-mL volumes of methylene chloride for the serial extraction of the base/neutrals and 200-, 100-, and 100-mL volumes of methylene chloride for the acids.

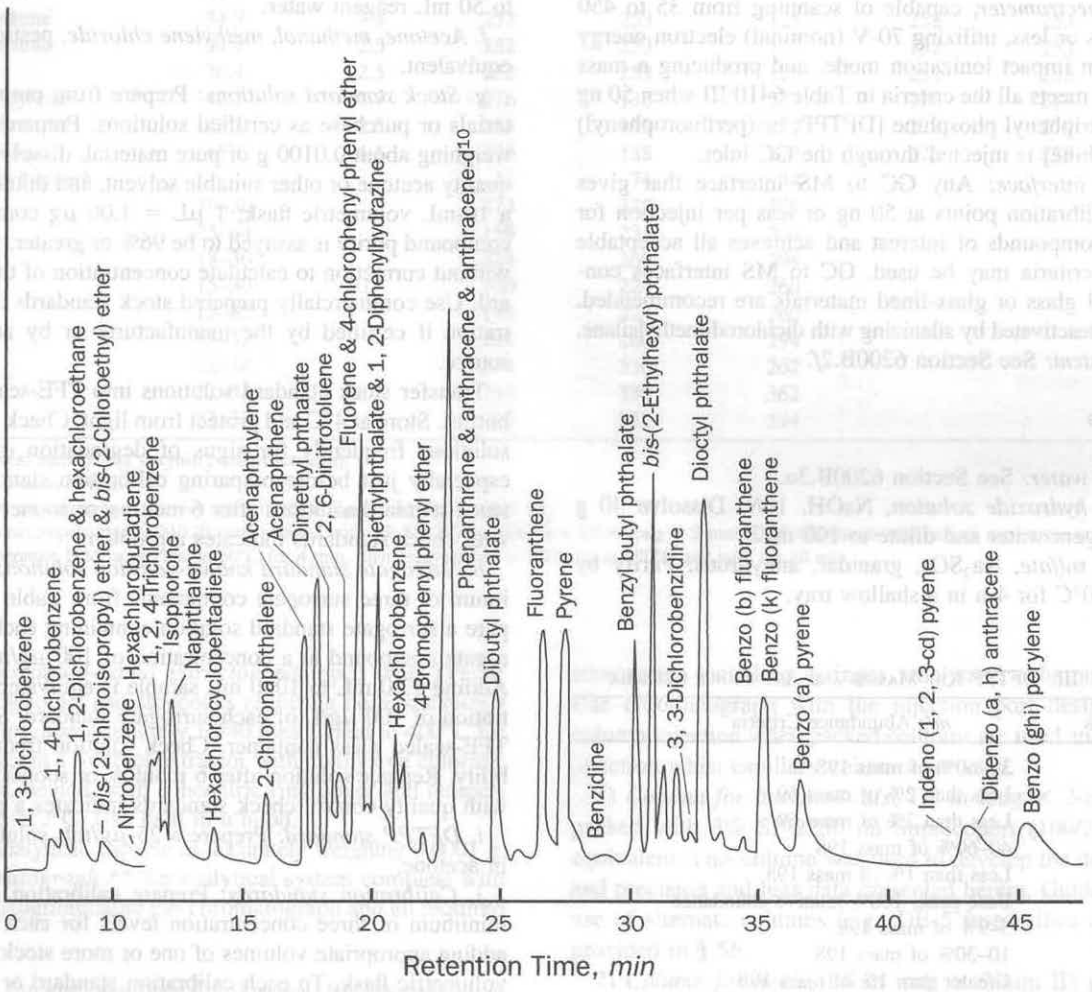

Figure 6410:1. Gas chromatogram of base/neutral fraction. Column: 3% SP-2250 on Supelcoport; program: 50°C for 4 min, 8°C/min to 270°C; detector: mass spectrometer.