6. ANALYTICAL METHODS

The purpose of this chapter is to describe the analytical methods that are available for detecting, and/or measuring, and/or monitoring chloromethane, its metabolites, and other biomarkers of exposure and effect to chloromethane. The intent is not to provide an exhaustive list of analytical methods. Rather, the intention is to identify well established methods that are used as the standard methods of analysis. Many of the analytical methods used for environmental samples are the methods approved by federal agencies and organizations such as EPA and the National Institute for Occupational Safety and Health (NIOSH). Other methods presented in this chapter are those that are approved by groups such as the Association of Official Analytical Chemists (AOAC) and the American Public Health Association (APHA). Additionally, analytical methods are included that modify previously used methods to obtain lower detection limits, and/or to improve accuracy and precision.

6.1 BIOLOGICAL SAMPLES

Methods used to analyze biological samples for chloromethane are summarized in Table 6-1. S-methyl-cysteine may be a urinary metabolite of chloromethane in some humans (Nolan et al. 1985; van Doorn et al. 1980). S-methylcysteine can be analyzed by diluting urine with water and treating the resulting solution with a buffer and a phthaldialdehyde solution to derivatize the S-methylcysteine (DeKok and Antheunius 1981) Analysis is performed on a reversed-phase high-performance liquid chromatography (HPLC) column using methanol and sodium hydrogen phosphate buffer gradient elution with a fluorescence detector. The reported detection limit is 1 mg/L. S-methylcysteine, along with other methylthio- compounds, can also be analyzed as methanethiol following alkaline hydrolysis and acidification (van Doom et al. 1980).

Breast milk was analyzed for chloromethane by expressing a 60 mL sample into a wide-mouth bottle and then freezing until analysis (Pellizzari et al. 1982). Analysis was performed by warming the sample and then purging it with helium and directing the chloromethane and other volatilized compounds through a Tenax adsorbant. The analytes were thermally desorbed from the Tenax onto a gas chromatography (GC) column and analyzed by mass spectrometry (MS). No recovery or accuracy information was reported. A headspace analysis for chloromethane in blood has been described (Landry et al. 1983a) as has a method for chloromethane in exhaled air (Nolan et al. 1985). No limits of detection (LODs) or recovery information were available for these methods.

Table 6-1. Analytical Methods for Determining Chloromethane in Biological Samples

Sample matrix	Preparation method	Analytical method	Sample detection limit	Percent recovery	Reference
Urine (S-methyl-cysteine)	Dilution with water followed by derivatization with phthaldialdehyde.	HPLC/fluorescence	1 mg/L (ppm)	No data	DeKok and Antheunius 1981
Urine (S-methyl-cysteine and other methyl thio compounds)	Hydrolysis of 4 mL urine sample with 2 mL of 4 N sodium hydroxide at 100 °C followed by acidification to form methanethiol; introduction of aliquot of headspace into GC.	GC/FID	No data	No data	van Doorn et al. 1980
Exhaled air	Collection of breath into gas sampling bag followed by introduction of internal standard and introduction of aliquot into GC.	GC/ECD	No data	No data	Nolan et al. 1985
Rat blood (applied to human blood by Nolan et al. 1985)	Equilibration of sample at 37 °C for 3 minutes followed by injection of an aliquot of headspace into GC. (Nolan et al. 1985 added the step of heating to 100 °C to reduce rate of chloromethane loss.)	GC/ECD	No data	No data	Landry et al. 1983b
Breast milk	Warming of sample followed by purging with inert gas and adsorption of chloromethane onto Tenax; thermal desoprtion onto GC.	GC/MS	No data	No data	Pellizzari et al. 1982

ECD = electron capture detector; FID = flame ionization detector; GC = gas chromatography; HPLC = high-performance liquid chromatography; MS = mass spectrometry

6.2 ENVIRONMENTAL SAMPLES

Methods for the determination of chloromethane in environmental samples are presented in Table 6-2. In air, chloromethane can be analyzed by NIOSH Method 1001 (NIOSH 1994). This method involves drawing a 0.4-3 L sample through a coconut charcoal tube followed by methylene chloride desorption and analysis by GC with flame ionization detection (FID). The method has a working range of 66-670 mg/m^3 for a 1.5 L sample and an LOD of 0.01 mg/tube. The method of Oliver et al. (1996) also uses a preconcentration approach, but analyte recovery is accomplished via thermal desorption. The large sample concentration factor combined with the sensitivity of the ion trap detector (ITD) provides for an LOD of less than 1 ppb. Chloromethane can also be trapped cryogenically from an aliquot of air collected into an evacuated canister followed by determination using GC with either electron capture or mass spectrometric detection (EPA 19888). LODs were reported to be in the low ppb range. Loss of chloromethane from air samples stored in canisters can impact the accuracy of the determination. Kelly and Holdren (1995) reported a 17% loss for chloromethane at 2.1 ppb stored for 33 days. On the other hand, Brymer et al. (1996) showed a loss of approximately 5% over a 30-day period for chloromethane in a canister at 2.3 ppb (v/v). They also reported a method detection limits of 0.82 ppbv and a recovery of 124%. Potential changes in analyte concentration as function of time after sample collection indicates that field control samples should be used. Field controls are always appropriate regardless of the collection approach used. Fukui and Doskey (1996) reported using a canister-based approach to collect chloromethane and other volatile compounds emitted from grasslands. Extreme care must be taken, especially at very low air concentrations, to ensure that no contamination is introduced into the sampling and analysis method; method blanks must always be used to verify the cleanliness of the sample collection and analysis system.

Chloromethane can be analyzed in municipal and industrial waste water by EPA Test Method 601-Purgeable Halocarbons or EPA Test Method 624Purgeables (EPA 1982a). Both methods are adequate for measuring chloromethane in waste waters. However, care must be exercised during sample collection because chloromethane is volatile and some of the chemical might be lost during the sampling process. Method 601 involves purging the sample with an inert gas and passing the gas through a trap containing 2,6-diphenylene oxide polymer (Tenax GC), silica gel, and coconut charcoal to adsorb the purged chloromethane and other halocarbons (called the “purge and trap” method). After the purging is complete, the trap is heated to desorb the chloromethane. The desorbed chloromethane is analyzed by GC using an electrolytic conductivity (EC) or microcoulometric detector. Method 624 is similar to Method 601, but the trap material is made of 3% methyl silicone (OV-1) on packing material, 2,6-diphenylene oxide polymer

Table 6-2. Analytical Methods for Determining Chloromethane in Environmental Samples

Sample matrix	Preparation method	Analytical method	Sample detection limit	Percent recovery	Reference
Air	Pumping of air through solid sorbent tubes (coconut shell charcoal) followed by elution using dichloromethane.	GC/FID (Method 1001)	0.003 mg/m^3 (1.5 ppb)	104	NIOSH 1994
Air	Collection of air into evacuated canister; introduction of aliquot of air and internal standard onto cryotrap; thermal desorption.	GC/FID/ECD, GC/SIM (Method TO-14)	low ppb	No data	EPA 1988g
Air	Pumping of air through multisorbent trap followed by a dry helium purge; thermal desorption onto a cryotrap followed by thermal desorption onto GC column.	GC/ITD	0.53 ppb (0.0011 mg/m^3)	approx. 95% (4.9% RSD at 5 ppb)	Oliver et al. 1996
Water	Addition of internal standards and purging of water sample with inert gas; adsorption of chloromethane onto adsorbent trap followed by thermal desorption.	GC/MS (Method 6210D)	No data	101% (4.7% RSD) at 0.5 µg/L (ppb) using narrow-bore GC column	Greenberg et al. 1992a
Waste water (municipal and industrial discharges)	Addition of internal standards and purging of water sample with inert gas; adsorption of chloromethane onto adsorbent trap followed by thermal desorption.	GC/EC (Method 6230B)	0.08 µg/L (0.08 ppb)	95% at 1 µg/L	Greenberg et al. 1992b
Water	Purging of sample with inert gas and trapping vapors onto adsorbent with subsequent thermal desorption of trap.	GC/EC (Method 502.1)	0.01 µg/L (0.01 ppb)	93% (8.5% RSD) at 0.40 µg/L	EPA 1989a
Water	Addition of surrogate standards followed by purging of sample with inert gas and trapping vapors onto adsorbent with subsequent thermal desorption of trap.	GC/MS (Method 524.2)	0.13 µg/L (0.13 ppb) using wide bore column	93–110% (9% RSD) at 0.1 to 10 µg/L	EPA 1989b

Table 6-2. Analytical Methods for Determining Chloromethane in Environmental Samples (continued)

Sample matrix	Preparation method	Analytical method	Sample detection limit	Percent recovery	Reference
Water (standards)	Addition of standard compounds and internal standard to saturated salt water; immersion of SPME fiber in water for 5 minutes with stirring; removal of fiber from water and insertion of fiber into injection port for thermal desorption.	GC/MS	<25 ppb	No data	Shirey 1995
Aqueous culture medium (phytoplankton culture)	Purging of chloromethane with helium from 10 mL with adsorption onto Poropak-Q followed by thermal desorption onto GC.	GC/ECD	7 pM (0.35 ppt)	No data	Tait and Moore 1995
Water, soil, solid waste	Purging of sample with inert gas with adsorption of chloromethane onto adsorbent with subsequent thermal desportion onto GC column.	GC/EC (EPA Method 8010B)	12.5 µg/kg[a] (ppb) for high concentration soil, sludge	95% at 1 µg/L[a]	EPA 1986b
Water, soil, solid waste	Purging of sample with inert gas with adsorption of chloromethane onto adsorbent with subsequent thermal desorption onto GC column.	GC/FID/EC (EPA Method 8021A)	0.03 µg/L[a] (water)	96% (9.3% RSD)[a]	EPA 1986c
Water/waste water	Purging of sample with inert gas with adsorption of chloromethane onto adsorbent with subsequent thermal desorption onto GC column.	GC/EC (EPA Method 601) GC/MS (EPA Method 624)	0.08 µg/L (0.08 ppb) No data	91.4% 99±24%	EPA 1982a
Water (river, sea)	Injection of 1 mL of sample into flow injection analysis system.	MIMS/ITD	10 ppb	No data	Bauer and Solyom 1994

[a] Detection limits and recoveries will vary depending on the particular matrix.

EC = electrolytic conductivity; ECD = electron capture detector; FID = flame ionization detector; GC = gas chromatography; ITD = ion trap detector (mass spectrometer); MIMS = membrane introduction mass spectrometry; MS = mass spectrometry; RSD = relative standard deviation; SIM = selected ion monitoring; SPME = solid-phase microextraction

(Tenax GC), and silica gel; analysis is made by GC/MS. Overpurging the sample may result in loss of some chloromethane. The average recovery from reagent water and effluents was 91.4±13.4% for Method 601 and 99±24% from waste water for Method 624. The Contract Laboratory Program analytical method involves screening the sample for component concentrations by rapidly transferring the room temperature sample to a volumetric flask; adding hexadecane; extracting the volatiles, including chloromethane, for 1 minute; and then qualitatively analyzing the sample by GC/FID (EPA 1988a). The quantitative analysis method for the sample is by GC/MS and is essentially identical to EPA Method 624 (EPA 1982a).

Three additional purge-and-trap approaches with LODs as low as 0.01 μg/L (0.01 ppb) have also been described for drinking water: Standard Method 6210D (Greenberg et al. 1992a), Method 502.1 (EPA 1989a), and Method 524.2 (EPA 1989b). A purge-and-trap approach to the determination of chloromethane in an aqueous culture medium provided an LOD of 0.35 ppt (Tait and Moore 1995). A technique known as solid-phase microextraction (SPME) has been demonstrated to be applicable to low ppb chloromethane concentrations in a water matrix (Shirey 1995). In this method, a polymer-coated fiber is equilibrated in a water sample until the chloromethane partitions into the polymer coating. The fiber is withdrawn and inserted into the hot injection port of a GC, where the analyte is thermally desorbed onto the GC column.

EPA Method 5030 for analysis of chloromethane in soil and solid waste (EPA 1986b) involves the direct purge-and-trap method for low-level samples or the methanolic extraction for high-level samples, based on a hexadecane extraction as described above. For low-level samples, the soil and solid waste are placed in a purge impinger, mixed with water, purged with an inert gas, and trapped on a Tenax GC and silica gel (EPA 1988a) or on a OV-1, Tenax GC, and silica gel column (EPA 1986b). The trap column is heated and purged to desorb the chloromethane and other volatiles onto the GC column. For medium-level samples, the soil and solid waste are mixed with methanol and shaken. An aliquot of the methanol is removed, diluted with water, and purged as described above for water samples. Overpurging the sample may result in loss of some chloromethane. Analysis is performed by EPA Method 8000 (Gas Chromatography/Mass Spectrometry for Volatile Organics) and Method 8010B (Halogenated Volatile Organics) or by Method 8240 (GC/MS for Volatile Organics) (EPA 1986b). Method 8010 uses GC with an electrolytic conductivity detector. EPA Method 8021A uses analysis by GC with photoionization detection and electron capture detection in series (EPA 1986c). LODs range from 0.03 μg/L with chloromethane in water (Method 8021A) (EPA 1986c) to 12.5 μg/kg for high-concentration soils and sludges (Method 8010B) (EPA 1986b). Other method characteristics are shown in Table 6-2.

No methods for chloromethane in foods were found. However, a purge-and-trap method applicable to the determination of trihalomethanes in liquid and viscous foods has been published by researchers at the U.S. Food and Drug Administration (FDA) (McNeal et al. 1995). This method is a modification of EPA Method 524.2 (EPA 1989b) and should be applicable to the determination of chloromethane in foods. However, this method has not been validated for chloromethane.

6.3 ADEQUACY OF THE DATABASE

Section 104(i)(5) of CERCLA, as amended, directs the Administrator of ATSDR (in consultation with the Administrator of EPA and agencies and programs of the Public Health Service) to assess whether adequate information on the health effects of chloromethane is available. Where adequate information is not available, ATSDR, in conjunction with the NTP, is required to assure the initiation of a program of research designed to determine the health effects (and techniques for developing methods to determine such health effects) of chloromethane.

The following categories of possible data needs have been identified by a joint team of scientists from ATSDR, NTP, and EPA. They are defined as substance-specific informational needs that if met would reduce the uncertainties of human health assessment. This definition should not be interpreted to mean that all data needs discussed in this section must be filled. In the future, the identified data needs will be evaluated and prioritized, and a substance-specific research agenda will be proposed.

6.3.1 Identification of Data Needs

Methods for Determining Biomarkers of Exposure and Effect. No biomarker that can be associated quantitatively with exposure to chloromethane has been identified (see Section 2.6). Methods are available for the analysis of chloromethane in blood, expired air, and breast milk. In addition, a method exists for the analysis of the metabolite S-methylcysteine in urine. Quantitative relationships have not been established between exposure and measurement of chloromethane or S-methylcysteine in these biological media. The observed variability of metabolism (see the discussion of the metabolism of chloromethane in Section 2.3.3) suggests that a correlation of chloromethane levels in tissues with levels of chloromethane exposure is not likely to be found. It may be possible to use levels of yet unidentified metabolites in blood or urine as biomarkers of exposure. If reliable biomarkers of exposure were available, it would allow both investigators and reviewers to assess the accuracy and uncertainty of the methods used in toxicological

studies. Furthermore, the ready availability of tested analytical methods for the biomarkers, including sample preservation, would permit a standardized approach to the analysis of biological materials to assist in measuring human exposure and monitoring effects in humans. Thus, methods for biomarkers of exposure and effect are needed.

Methods for Determining Parent Compounds and Degradation Products in Environmental Media. Methods appear to be available for the analysis of chloromethane in all environmental media. Methods for drinking water, groundwater, surface water, and waste water (Bauer and Solyom 1994; EPA 1982, 1989a, 1989b; Greenberg et al. 1992a, 1992b; Shirey 1995) have LODs as low as 0.01 ppb; methods for soil and solid waste (EPA 1989b, 1989c), and for workplace and ambient air (EPA 19888; NIOSH 1994; Oliver et al. 1996) have LODs in the 0.5 to 1.5 ppb range. The MRL for chronic inhalation exposure to chloromethane is 0.05 ppm and all of the methods reported for air are adequate. No MRLs have been established for ingestion exposures. No methods were identified for chloromethane in foods; the need for analytical methods would be driven by oral MRLs. Chloromethane degrades to a number of products in the environment, including methanol and formaldehyde, both of which are natural products. While analytical methods exist for these compounds, they cannot be used as indicators of chloromethane degradation since methanol and formaldehyde have large natural sources.

6.3.2 Ongoing Studies

No ongoing studies were located in which new methods for chloromethane might be developed.