6. ANALYTICAL METHODS

The purpose of this chapter is to describe the analytical methods that are available for detecting and/or measuring and monitoring methyl mercaptan in'environmental media and in biological samples. The intent is not to provide an exhaustive list of analytical methods that could be used to detect and quantify methyl mercaptan. Rather, the intention is to identify well established methods that are used as the standard methods of analysis. Many of the analytical methods used to detect methyl mercaptan in environmental samples are the methods approved by federal agencies 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 refine previously used methods to obtain lower detection limits, and/or to improve accuracy and precision.

As a volatile material boiling at only 5.95°C (Windholz 1983), methyl mercaptan, CH_3SH, also called methanethiol, is readily determined by gas chromatographic analysis. The sensitivity and selectivity of detection are increased by the use of sulfur-selective detectors; in one study, substitution of a sulfur-selective flame photometric detector for a flame ionization detector reduced the detection limit from 20 to 2 μg/unit sample (Knarr andRappaport 1980). Hydrogen sulfide (H_2S), dimethyl sulfide ($[CH_3]_2S$), and dimethyl disulfide ($[CH_3]_2S_2$) are other reduced sulfur compounds that may occur along with methyl mercaptan and are commonly determined along with it. Normally, methyl mercaptan is collected from the gas phase or from vapor evolved from the sample matrix on a column of solid sorbent, such as Tenax@. Collection on molecular sieve is also possible (Kangas and Ryosa 1988), although problems are encountered from incomplete desorption of methyl mercaptan from molecular sieve. Cryogenic (low temperature) collection may also be possible and is less likely to lead to alterations of the analyte in the collection apparatus (Brettell and Grob 1985). The presence of water can result in reduced sorption capacity for methyl mercaptan as well as decomposition during thermal desorption. Sorption efficiency is improved markedly by removal of water from the air stream with calcium chloride (Tangerman 1986). Purge-and-trap techniques are used to collect methyl mercaptan from water (Badings et al. 1985). Headspace vapor in equilibrium with the sample in a closed container may also be subjected to gas chromatography.

6.1 BIOLOGICAL MATERIALS

Methods for detection of methyl mercaptan in biological materials are summarized in Table 6-l.

Methyl mercaptan has been determined in a variety of biological materials as shown in Table 6-l. Normally,'for determination in biological samples, methyl mercaptan is released from the sample matrix and collected on a column of solid sorbent, cryogenically or as headspace gas. As a result of

TABLE 6-1. Analytical Methods for Determining Methyl Mercaptan in Biological Materials

Sample matrix	Preparation method	Analytical method	Sample detection limit	Accuracy	Reference
Bacterial cultures	Mix with potassium carbonate, collection of headspace vapor at 60°C	GC/MS	No data[a]	No data[a]	Hayward et al. 1977
Clostridium bacterial cultures	Collection of headspace vapor under vacuum	GC/MS	1-10 ppb[b]	No data	Rimbault et al. 1986
Blood, brain tissue	No data	GC/SSD	No data	No data	Al Mardini et al. 1984
Collagen	Collection of headspace vapor	GC	No data	No data	Johnson and Tonzetich 1985
Blood and serum	Release from metabolites, collection on Tenax®	GC/SSD	<0.3 µmol/L	92±7%-99±4%	Tangerman et al. 1985
Breath	Trapping from breath onto Tenax®, desorption	GC/SSD	0.2 ng/L (0.1 ppb)	97±5%	Tangerman et al. 1983

[a]Analytical results were reported as "peak areas", not concentrations.
[b]Level in headspace gas.

GC = gas chromatography; MS = mass spectrometry; SSD = sulfur-specific detector

6. ANALYTICAL METHODS

Phase II metabolic reactions in biological systems (Manahan 1989), methyl mercaptan may be bound as conjugates from which it must be released prior to analysis. Two such bound fractions of methyl mercaptan have been identified in human serum (Tangerman et al. 1985). In one fraction, from which the methyl mercaptan is released by acid, the methyl mercaptan is thought to be bound as methyl-beta-D-thioglucuronide. In another fraction methyl mercaptan is covalently bound to proteins in a disulfide linkage and is released by reaction with dithiothreitol.

6.2 ENVIRONMENTAL SAMPLES

For the determination of methyl mercaptan in air, the analyte is usually trapped and concentrated from a large volume of air on a solid sorbent such as Tenax®, activated carbon, or molecular sieve from which it is released thermally for subsequent measurement. It is advisable to dry the air sample with calcium chloride prior to collection of methyl mercaptan to preventanalyte decomposition on the collection medium (Kangas and Ryosa 1988; Tangerman 1986). For aqueous and solid waste samples, methyl mercaptan is purged with an inert gas and collected on a solid such as Tenax®, or cryogenically, followed by thermal desorption and measurement. Gas chromatography using sensitive and highly specific mass spectrometry (MS) or highly sensitive flame photometric detection (FPD) for detection is the analytical method of choice for the determination of methyl mercaptan in environmental samples.

Methods for the determination of methyl mercaptan in environmental samples are summarized in Table 6-2.

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 methyl mercaptan is available. Where adequate informationis not available, ATSDR, in conjunction with the NTP, is required to assurethe initiation of a program of research designed to determine the health effects (and techniques for developing methods to determine such health effects) of methyl mercaptan.

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 or eliminate the uncertainties of human health assessment. In the future, the identified data needs will be evaluated and prioritized, and a substance-specific - research agenda will be proposed.

TABLE 6-2. Analytical Methods for Determining Methyl Mercaptan in Environmental Samples

Sample matrix	Preparation method	Analytical method	Sample detection limit	Accuracy	Reference
Ambient air	Removal of water by calcium chloride, adsorption on molecular sieve, thermal desorption	GC/FPD	<0.01 cm^3/m^3	No data	Kangas and Ryosa 1988
Air	Removal of water by calcium chloride, trap from air onto Tenax® at -196°C, desorption at 200°C	GC/FPD	<12 ppt	93±7%[a]	Tangerman 1986
Air[b]	Retention by activated carbon	GC	No data	No data	ASTM 1987
Air	Collection on a glass fiber filter impregnated with mercuric acetate	GC/FPD	17 $\mu g/m^3$	98.4±0.22%[c]	Knarr and Rappaport 1980
Flue gases	Collection on sample loop, direct injection	GC/FPD	<1 ppm	No data	De Souza 1987
Water	Purge, cryogenic trap	HRGC	No data	No data	Badings et al. 1985
Water[d]	Direct injection	GC	1 mg/L	No data	ASTM 1988
Waste water	Purge from water by helium, collect on Tenax®, thermal desorption	GC/MS	No data	No data	Van Langenhove et al. 1985
Soil[e]	Purge by helium; collection on solid, thermal desorption	GC/MS	5 µg/kg	No data	EPA 1986

[a]As recovery from Tenax®
[b]Absorption characteristics for sampling atmospheric vapor with activated carbon for subsequent analysis by GC.
[c]At 1 ppm
[d]General method for the determination of volatile organic matter in water using flame ionization detection. Selectivity and sensitivity for methyl mercaptan can be greatly enhanced with sulfur-selective detection.
[e]Also applicable to sediment and solid waste.

FPD = flame photometric detector; GC = gas chromatography; HRGC = high resolution gas chromatography; MS = mass spectrometry; ppt = parts per trillion

6.3.1 Data Needs

Methods for Determining Biomarkers of Exposure and Effect. Sensitive and selective methods are available for the qualitative and quantitative measurement of methyl mercaptan in biological materials, after it is separated from its sample matrix (Al Mardini et al. 1984; Hayward et al. 1977; Johnsonand Tonzetich 1985; Rimbault et al. 1986; Tangerman et al. 1983, 1985). However, there are currently no methods available to qualitatively or quantitatively correlate exposure to methyl mercaptan with biomarkers in tissue or fluid. As discussed previously (Section 2.5), tissue and fluid levels of methyl mercaptan can be independent of exogenous exposure to that compound.

In the analysis of methyl mercaptan in biological materials, capillary gas chromatography, also commonly known as high-resolution gas chromatography (HRGC), has greatly facilitated the analysis of compounds such as methyl mercaptan that can be measured by gas chromatography and has resulted in vast improvements in resolution and sensitivity. The instrumental capability to separate volatile analytes by HRGC is, for the most part, no longer the limiting factor in their analysis.

It would be useful to have the means to transfer analytes that have been isolated from a biological matrix, quantitatively and in a narrow band, to the HRGC, and to identify and accurately measure the quantity of compounds in the HRGC peaks. Mass spectrometric detection and Fourier transform infrared spectroscopy (FTIR) may prove to be the most useful methods for these functions.

There is a lack of standard methods for the measurement of metabolites of methyl mercaptan in biological materials and development of these methods would facilitate their determination in routine practice.

Specific methods for biomarkers that correlate levels of methyl mercaptan or its metabolites with toxic effects in exposed populations are not available. These methods would be helpful in defining the potential health risks of certain tissue or fluid levels of these compounds, independent of their source.

Methods for Determining Parent Compounds and Degradation Products in Environmental Media. Methods for determining the parent compound, methyl mercaptan, in water, air and waste samples with excellent selectivity and sensitivity are well developed (ASTM 1987, 1988; Badings et al. 1985; De Souza 1987; EPA 1986; Kang as and Ryosa 1988; Knarr and Rappaport 1980; Tangerman 1986; Van Langenhove et al. 1985), so the database in this area is good andundergoing constant improvement. For example, research is on-going to develop a "Master Analytical Scheme" for organic compounds in water (Michael et al. 1988), which includes methyl mercaptan as an analyte. The overall goal is to detect and quantitatively measure organic compounds at 0.1 μg/L in drinking

water, 1 ug/L in surface waters, and 10 μg/L in effluent waters. Analytes are to include numerous nonvolatile compounds and some compounds that are only "semi-soluble" in water, as well as volatile compounds (bp<l50"C).

Improved methods are needed for the determination of methyl mercaptan in solid environmental samples, including soil and sediments. The standard EPA Method 8240 for gas chromatography/mass spectrometry of volatile organics in wastes, soils, and sediments (EPA 1986) could be tested thoroughly for methyl mercaptan analysis and optimized for this application.

Sampling methodologies for compounds such as methyl mercaptan continue to pose problems such as nonrepresentative samples, insufficient sample volumes, contamination, and labor-intensive, tedious extraction and purification procedures (Green and Le Pape 1987). It is desirable to have means to measure organic compounds such as methyl mercaptan in situ in water and other environmental media without the need for sampling and extraction procedures to isolate the analyte prior to analysis.

Degradation products of methyl mercaptan in environmental media are difficult to determine because these products may come from a number of sources other than methyl mercaptan.

6.3.2 On-going Studies

The Environmental Health Laboratory Sciences Division of the Center for Environmental Health and Injury Control, Centers for Disease Control, is developing methods for the analysis of methyl mercaptan and other volatile organic compounds in blood. These methods use high resolution gas chromatography and magnetic sector mass spectrometry which gives detection limits in the low parts per trillion range.

Examination of the literature suggests that other studies are underway to improve means for determining methyl mercaptan and other reduced sulfur compounds in biological samples and environmental media. Improvements continue to be made in chromatographic separation and detection, including supercritical fluid extraction and supercritical fluid chromatography (Smith 1988). Fourier transform infrared flow cell detectors are sensitive and selective for the detection of compounds such as methyl mercaptan that have been separated by fluid chromatography (Wieboldt et al. 1988). Immunoassay methods of analysis are very promising for the determination of various organic pollutants and toxicants, and it is reasonable to assume that methyl mercaptan, and particularly its metabolites such as methyl-beta-Dthioglucuronide are candidates for this type of analysis.