6. ANALYTICAL METHODS

6.1 BIOLOGICAL MATERIALS

No study was located regarding the analysis of isophorone in human biological materials, but animal studies (see Section 2.6) suggest that methods are available. In general, isophorone was extracted from the urine using ether (continuous extraction for 48 hours) followed by evaporation of the ether (Dutertre-Catella et al. 1978; Truhaut et al. 1970). The resulting residue was subjected to gas chromatography with flame ionization detector using the retention time as the indicator for the presence of isophorone or metabolites. In distribution studies, isophorone was extracted from minced tissues with dichloromethane, and the extract was analyzed by gas chromatography using the flame ionization detector (Dutertre-Catella 1976). In addition to these methods for analyzing isophorone in mammalian urine and tissues, Ozretich and Schroeder (1986) described a method for analyzing isophorone in fish tissue (Table 6-l).

6.2 ENVIRONMENTAL SAMPLES

Isophorone can be analyzed in municipal and industrial wastewater by EPA Test Method 609 - Nitroaromatics and Isophorone, or by EPA Test Method 625 Base/Neutrals and Acids (EPA 1982; Shafer 1982). These methods are adequate for measuring isophorone in most wastewaters, although interfering compounds may be present in some wastewaters. Method 609 involves the extraction of isophorone with methylene chloride followed by solvent exchange to hexane and analysis by gas chromatography (GC) using a flame ionization detector (FID). Method 625 is similar to Method 609, but the extraction is performed at pH 11 and is followed by concentration (without solvent exchange) and GC/MS analysis. The Contract Laboratory Procedure (EPA 1987a) is essentially identical (Table 6-l). The average recovery from reagent water and effluents was 49-67% for Method 609 and 75 $\pm$ 33% from reagent water for method 625, Method 609 shows a pronounced negative bias (the concentration detected by the method is lower than the true concentration present) (Kinzer et al. 1984). Table 6-l presents accuracy and detection limit data for the methods. In air, isophorone can be analyzed by NIOSH Method 2508 (NIOSH 1984). The method involves drawing a 2 to 25 liter air sample through a petroleum based charcoal tube followed by carbon disulfide desorption and analysis by GC-FID. The method has a range of 0.2-10 mg per tube and a detection limit of 0.02 mg per tube. Table 6-l presents accuracy information for this method.

The method for analyzing soil in the EPA Contract Laboratory Program involves the extraction of isophorone using methylene chloride followed by analysis by GC/MS. The usual detection limit is 330 ppb, although the exact detection limit is matrix dependent.

TABLE 6-1. Analytical Methods for Isophorone

Sample Matrix	Sample Preparation	Analytical Method	Sample Detection Limit	Accuracy[a]	Reference
Air	Charcoal tube collection and CS_2 desorption	GC-FID[b]	2 mg/m^3	104.9	NIOSH 1984
Water	Methylene chloride extraction, hexane solvent exchange, concentration	GC-FID	5.7 µg/L	49-67[c]	EPA 1982 Kinzer et al. 1984
	Methylene chloride extraction and concentration	GC/MS[d]	2.2 µg/L	75 ± 33[e]	EPA 1982 EPA 1987a (CLP)
Soil	Methylene chloride extraction and concentration	GC/MS	330 µg/kg	NS	EPA 1987a (CLP)
Fish Tissue	Macerate tissue mixed with anhydrous Na_2SO_4, extract with acetonitrile by sonocation. Concentrate extract, clean-up by column chromatography	GC/MS	NS	61	Ozretich and Schroeder 1986

[a] Average percent recovery
[b] Gas chromatography flame ionization detector
[c] Laboratory water and effluents
[d] Gas chromatography mass spectrometry
[e] Laboratory water
NS, not specified

6. ANALYTICAL METHODS

6.3 ADEQUACY OF THE DATABASE

Section 104 (i) (5) of CERCLA, 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 isophorone is available. Where adequate information is not available, ATSDR, in cooperation with the National Toxicology Program (NTP), is required to assure the initiation of a program of research designed to determine these health effects (and techniques for developing methods to determine such health effects). The following discussion highlights the availability, or absence, of exposure and toxicity information applicable to human health assessment. A statement of the relevance of identified data needs is also included. In a separate effort, ATSDR, in collaboration with NTP and EPA, will prioritize data needs across chemicals that have been profiled.

6.3.1 Data Needs

Methods for Parent Compound and Metabolites in Biological Materials. No information is available concerning the analysis of isophorone in biological materials. If information were available, it would allow both investigators and reviewers to assess the accuracy and uncertainty of the methods used. Furthermore, the ready availability of tested analytical methods would permit a standardized approach to the analysis of biological materials and allow a comparison of the levels of exposure with the possible health effects in humans.
Methods for Biomarkers of Exposure. No methods are available for the analysis of isophorone biomarkers of exposure in biological materials. If a method for the determination of the level of a specific biomarker were available in a biological medium, it could be used to indicate the level of exposure and the possible resultant health effect.

Methods for Parent Compound and Degradation Products in Environmental Media. Adequate methods appear to be available for the analysis of isophorone in groundwater, surface water, soil, and workplace air. No methods were found for the analysis of isophorone in ambient air, where concentrations are expected to be much lower than in workplace air. If the parent compound is stable as in the present case, it is essential that its concentrations in different environmental media be known so that the level of its exposure can be estimated.

No adequate methods appear to be available for the analysis of isophorone degradation products in environmental media. In cases where a degradation product of a chemical is toxic, it is important that its concentration in the environment be known. In certain instances, monitoring the level of a degradation product may be used as an indirect measurement of the parent compound in the environment.

6.3.2 On-going Studies

No studies were located regarding on-going analytical methods development for isophorone.