6. ANALYTICAL METHODS

The purpose of this chapter is to describe the analytical methods that are available for detecting and/or measuring and monitoring vanadium 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 vanadium. 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 vanadium in environmental samples are the methods approved by federal agencies such as EPA and 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.

6.1 BIOLOGICAL MATERIALS

A number of analytical techniques have been used to determine ppm to ppt levels of vanadium in biological materials. These include neutron activation analysis (NAA), graphite furnace atomic absorption spectrometry (GFAAS), spectrophotometry, isotope dilution thermal ionization-mass spectrometry (IDMS), and inductively coupled plasma atomic emission spectrometry (ICP-AES). Table 6-l summarizes the analytical methods for determining vanadium in biological materials.

In general, biological and environmental samples may be prepared prior to quantification of vanadium by acid digestion with nitric acid. Sample dilution with nitric acid or other agents to solubilize vanadium from the sample matrix can also be employed. If the concentration of vanadium in the dissolved sample is very low, preconcentration techniques such as chelation or extraction may be used. Chelation and extraction efficiency will vary with the technique used.

Owing to its high sensitivity, the NAA technique has been widely used to measure trace elements (including vanadium) in biological samples (Allen and Steinnes 1978; Lavi and Alfassi 1988; Mortin and Chasteen 1988; Mousty et al. 1984). The NAA technique is based on the interaction of the nuclei of vanadium atoms in the sample with thermal neutrons, resulting in the emission of photons (gamma rays). The resultant gamma ray is detected with a highresolution lithium-drifter germanium detector. The concentration of vanadium is determined through its short-lived (half-life = 3.75 minutes) radionuclide 52V. Detection limits of low- to sub-ppb (μg/L) levels of vanadium in blood and urine samples were obtained (Allen and Steinnes 1978; Lavi and Alfassi 1988; Mousty et al. 1984). The advantages of the NAA technique are its sensitivity and multi-elemental capability. The disadvantages of this technique include its high cost and the limited availability of nuclear facilities for NAA analysis.

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

Sample matrix	Preparation method	Analytical method	Sample detection limit	Percent recovery	Reference
Blood and urine	Digest sample and evaporate; redissolve in acid; extract with MIBK; evaporate; redissolve in acid	NAA	~1 µg/L (blood); 2-4 µg/L (urine)	No data	Allen and Steinnes 1978
	Digest sample and evaporate; extract and concentrate with BTA, evaporate in benzene; dissolve in acid	GFAAS	0.08 µg/L (serum); 0.06 µg/L (urine)	90.3% (serum); 90.8% (urine)	Ishida et al. 1989
	Digest sample, evaporate; redissolve in water; extract with CPCHA in chloroform	Spectrophotometry	0.8 mg/L	No data	Paul and Gupta 1982
	Digest sample	ICP-AES	µg/L levels	No data	Kawai et al. 1989
Serum	Coprecipitate sample with lead nitrate or bismuth nitrate; dry and irradiate	NAA	0.7 µg/L	No data	Lavi and Alfassi 1988
Serum and liver	Digest sample and evaporate; redissolve in acid, concentrate on cation exchange column evaporate; redissolve in ammonium acetate	IDMS	2.6 µg/L (serum); 0.0987 µg/g (liver)	No data	Fassett and Kingston 1985

BTA = benzoyl-N-(o-tolyl)hydroxyl amine; CPCHA = chlorophenylcinnamohydroxamic acid; GFAAS = graphite furnace atomic absorption spectrometry; ICP-AES = inductively coupled plasma atomic emission spectrometry; IDMS = isotope mass spectrometry; MIBK = methyl isobutyl ketone; NAA = neutron activation analysis

GFAAS has also been used for measuring trace levels of vanadium in the serum and urine of humans and animals (Ishida et al. 1989; Mousty et al. 1984). Detection limits of 0.08 μg/L in serum and 0.06 μg/L in urine were achieved (Ishida et al. 1989). The GFAAS technique is as sensitive as NAA, and is also rapid, simple, relatively free from interference, and relatively inexpensive (Ishida et al. 1989; Krishnan et al. 1976).

Spectrophotometry has been extensively used to measure vanadium in environmental and biological samples (Abbasi 1981 and 1987; Agrawal 1975; Gupta and Tandon 1973; Jha 1979; Paul and Gupta 1982). The method typically involves dry or wet ashing of the sample, followed by preconcentration via an extraction procedure using chloroform and a chelating agent specific for vanadium. The spectrophotometric technique is convenient, rapid, selective, and sensitive for measuring ppm (mg/L) levels of vanadium in blood and urine samples (Paul and Gupta 1982).

A procedure has been developed for the determination of vanadium in biological materials using IDMS. In this procedure, the vanadium isotope 5% in the sample was increased by adding a ^{50}V-enriched spike solution, and ion counting detection was employed (Fassett and Kingston 1985). Isobaric interferences caused by chromium and titanium in the matrix require efficient dissolution and clean-up procedures prior to mass spectrometric measurements. These include wet digestion and dry-ashing of the sample matrix, followed by separation of vanadium from the matrix by chelation on an ion-exchange chromatography column. Detection limits of 2.6 μg of vanadium/L of serum and 0.0987 μg of vanadium/g of liver were obtained (Fassett and Kingston 1985). Although the sensitivity for measuring vanadium by the IDMS technique is in the ppb range in biological and environmental samples, it is a time-consuming and expensive technique.

6.2 ENVIRONMENTAL SAMPLES

Many of the analytical methods for detecting vanadium in biological samples have also been used to measure vanadium in environmental samples. They are detailed in Table 6-2. These include GFAAS, spectrophotometry, IDMS, and ICP-AES. Other techniques employed for measuring vanadium in environmental samples are flame atomic absorption spectrometry (FAAS) and direct current plasma-atomic emission spectrometry (DCP-AES). The most widely used methods utilize some modification of atomic adsorption spectrometry (AAS). In general, similar methods are employed for preparation and clean up of environmental and biological samples prior to quantification of vanadium (see Section 6.1).

Both AAS and AES methods are commonly used to detect vanadium in air. Trace levels of vanadium (as vanadium pentoxide) have been detected in air samples by GFAAS (Quickert et al. 1974). A detection limit of 0.25 ng of vanadium/m^3 of sample for an air sample of 2,000 m^3 was achieved. A method

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

Sample matrix	Preparation method	Analytical method	Sample detection limit	Percent recovery	Reference
Air	Collect air sample on acetyl-cellulose filter; digest and evaporate; redissolve in acid	ICP-AES	Working range of 5.0-2,000 $\mu g/m^3$ for a 500-L air sample	No data	NIOSH 1984b (Method 7300)
	Collect sample on filter; extract with trichloro-ethylene; digest; evaporate; redissolve in acid	DCP-AES	4 $\mu g/m^3$ for a 25-L sample	100%	Pyy et al. 1983
	Collect sample on glass fibre filter; digest; evaporate; redissolve in acid	GFAAS	0.25 ng/m^3 for an air sample of 2,000 m^3	No data	Quickert et al. 1974
Water	Acidify sample; extract and concentrate with HC PMC PTH in chloroform	Spectrophotometry	0.07 µg/sample	No data	Jha 1979
Water and waste water	Digest sample; evaporate; redissolve in acid	GFAAS and FAAS	4 µg/L (GFAAS); 200 µg/L (FAAS)	No data	EPA 1986a (Methods 7911 and 7910)
Water, plants, soils, and rocks	Acidify sample; oxidize with potassium permanganate; extract with DAMNHA in chloroform	Spectrophotometry	0.05 µg/sample	No data	Abbasi 1981
Citrus leaves and oyster tissue	Digest sample; evaporate; redissolve in acid; concentrate cation on exchange column; elute with HCl/H_2O_2; evaporate; redissolve in ammonium acetate	IDTI-MS	2.316 µg/g (oyster tissue); 0.245 µg/g (citrus leaves)	No data	Fassett and Kingston 1985
Soil and cabbage leaves	Digest sample; evaporate; redissolve in water; extract with N-o-CPCHA in chloroform	Spectrophotometry	0.8 µg/g	No data	Paul and Gupta 1982

DAMNHA = N-(p-N, N-dimethylanilino-3-methoxy-2-napho)hydroxamic acid; DCP-AES = direct current plasma atomic emission spectrometry; FAAS = flame atomic absorption spectrometry; GFAAS = graphite furnace atomic absorption spectrometry; HCl = hydrochloric acid; H_2O_2 = hydrogen peroxide; HC PMC PTH = N-hydroxy-N-p-chlorophenyl-N-(2-methyl-S-chloro)phenyl-p-toluamide hydrochloride; ICP-AES = inductively coupled plasma atomic emission spectrometry; IDTI-MS = isotope dilution thermal ionization-mass spectrometry; N-o-CPCHA = N-o-chlorophenylcinnamohydroxamic acid

for the determination of vanadium in workplace air using DCP-AES was reported by Pyy et al. (1983). A detection limit of 4 μg of vanadium/m^3 of sample and a practical working range of 0.01-100 μg of vanadium/m^3 of sample were obtained. DCP-AES was shown to have a sensitivity and precision equal to or greater than commonly used AAS techniques. No limitations of this method were noted by the authors. NIOSH has recommended,ICP-AES (Method 7300) for detecting vanadium and other elements in air. A working range of 5-2,000 μg of vanadium/m^3 of sample in a 500-L air sample was obtained (NIOSH 1984b).

GFAAS and FAAS are the techniques (Methods 7911 and 7910) recommended by EPA's Office of Solid Waste and Emergency Response for measuring low levels of vanadium in water and waste water (EPA 1986a). Detection limits of 4 μg of vanadium/L of sample and 200 μg of vanadium/L of sample were achieved using GFAAS and FAAS techniques, respectively. Spectrophotometry has also been employed to measure ppm levels of vanadium in aqueous media (Abbasi 1981; Jha 1979).

Spectrophotometry is the method commonly employed to analyze for the presence of vanadium in soil. Detection of low ppm concentrations in the soil have been reported (Abassi 1981; Paul and Gupta 1982). IDMS and spectrophotometry have been used for measuring low ppm (μg/g) levels of vanadium in plant and marine animal tissues (Abbasi 1981; Fassett and Kingston 1985; Paul and Gupta 1982).

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 vanadium 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 vanadium.

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.

6.3.1 Data Needs

Methods for Determining Biomarkers of Exposure and Effect. AAS methods are the most widely used to measure low- to sub-ppb (μg/L) levels of vanadium in blood, urine, tissues, and other biological media (Ishida et al. 1989; Krishnan et al. 1976; Mousty et al. 1984). Other methods include NAA (Allen and Steinnes 1978; Lavi and Alfassi 1988; Morten and Chasteen 1988; Mousty et al. 1984), AES (Kawai et al. 1989), and ITDI-MS (Fassett and Kingston 1985). These methods have all been demonstrated to be sensitive, precise, and reliable methods to measure metals as long as sample preparation procedures are rigidly followed and cleanliness and purity of all vessels and reagents are maintained. Background levels of vanadium have been reported with these techniques, but discrepancies and problems have occurred because of the extremely low levels of vanadium found in many samples. Contamination from reagents and containers used in analyses can approach background levels and interfere with results. All of these techniques provide sensitive and reliable measures of exposure with the capability of correlating environmental levels with tissue and urinary levels of vanadium. Improvements in sample preparation techniques that simplify sample preparation while maintaining or increasing reliability of analyses would be advantageous. Since there are no well-documented biomarkers of effect specific for vanadium, it is difficult to assess the sensitivity and reliability of these methods for measuring levels associated with health effects.

A decreased level of cystine (or cysteine) in hair and fingernails has been historically linked to vanadium exposure and suggested as a monitor of exposure (Mountain 1955). Accurate and precise methods exist for measuring these amino acids, but their depletion in hair and fingernails is not specific for vanadium exposure. More recent research did not indicate a correlation between cystine (or cysteine) depletion and vanadium exposure. There does not appear to be a need for additional research on this topic.

Methods for Determining Parent Compounds and Degradation Products in Environmental Media. AAS methods are the most widely used to measure levels of vanadium in environmental media (EPA 1986a; Quickert et al. 1974). In addition, AES techniques (NIOSH 1984b; Pyy et al. 198j), IDTI-MS (Fassett and Kingston 1985), and spectrophotometry (Abbasi 1981; Jha 1979) are available to measure vanadium in air, water, wastewater, soil, plants, and marine tissues. As with biological material, these methods are sensitive and reliable as long as sample preparation and cleanup are carefully done.

Since the most significant exposure to vanadium occurs in workers occupationally exposed to airborne vanadium, and significant absorption occurs by the inhalation route, air is the media of concern for potential human exposure. The GFAAS technique has sufficient accuracy and precision to measure background levels of vanadium in air as well as levels at which health effects might occur (Quickert et al. 1974). At hazardous waste sites, exposure is mostly likely to occur by ingestion of contaminated water, soil,

or plants, although absorption via this route is small. The techniques recommended by EPA, GFAAS and FAAS (Methods 7910 and 7911), provide sensitive and reliable measures of ppb levels of vanadium in water and waste water (EPA 1986a). Spectrophotometry is commonly used to detect vanadium in soil accurately (Abbasi 1981; Paul and Gupta 1982). IDTI-MS and spectrophotometry are used to measure low-ppm levels of vanadium in plants as well as marine tissues (Abbasi 1981; Fassett and Kingston 1985; Paul and Gupta 1982). No additional methods for detecting vanadium in environmental media appear to be necessary at this time; however, improvements in sample preparation could increase sensitivity and reliability.

6.3.2 On-going Studies

No on-going studies regarding methods for measuring vanadium in biological and environmental samples were located.