Trace Metals in Man's Environment and their Determination by Atomic Absorption Spectroscopy

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SUMMARY

The role of trace metals in pollution and occupational diseases is discussed. The occurrence of these metals, their effects, and their detection by the method of atomic absorption spectroscopy are reviewed. A large number of references enables the reader to obtain a wide insight into the literature of this increasingly important field of medical science.

In Part I, three metals which are the chief sources of dangerous and worldwide environmental pollution are discussed with regard to their physiological significance and determination methods. The metals causing occupational diseases are discussed in Part II.

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The advent of techniques and instrumentation by means of which very small concentrations can be rapidly and accurately determined, has given great impetus to the knowledge regarding the physiological role played by undesirable elements in the human environment. These methods have also constituted a powerful tool in detecting and combating unhealthy environmental conditions. The technique largely responsible for the increase of knowledge in this particular field is atomic absorption spectroscopy. To a very large extent, it fulfils the requirements of low cost of equipment, sensitivity, accuracy, rapidity of analysis and ease of operation (routine work is often done by laboratory-trained personnel who are lacking in theoretical background). Furthermore, there is the possibility of partial or complete automation of procedures, so that large numbers of samples can be handled.

Atomic absorption spectroscopy is based on the phenomenon that atoms of an element can absorb radiation at specific frequencies (as well as emit it). If an atomic vapour with atoms of an element A present is illuminated with radiation from a source radiating the characteristic spectrum of A, then some of the radiation will be absorbed by the atoms of A in the atomic cloud. The degree of absorbance is proportional to the number of atoms of A in the atomic cloud.

To carry out an atomic absorption measurement, one therefore requires a source radiating the light of the analytical element, a means for producing an atomic cloud of the sample (which must be in solution), and a means for measuring the intensity of a particular wavelength band. The main components of the apparatus are therefore (*i*) the source (a hollow cathode lamp), (*ii*) the atomising unit, and (*iii*) the spectrometer and measuring unit.

The detailed principles of atomic absorption can be found in the books listed at the end of the bibliography.

In conventional atomic absorption, the samples and standards are atomised by aspirating them into a flame. In flameless atomic absorption, alternative means are used to obtain the atomic vapour. The sample is placed in a graphite tube, and atomisation is achieved by electrical resistance heating. With this modification, analytical sensitivities are, on the average, about a thousand times better than with the conventional method. Very small sample quantities are required for the flameless technique and samples need not necessarily be in solution. This technique does not replace the conventional flame method, but supplements it very well for special studies. The flame technique, being more rapid and generally more precise, will remain the method of choice for routine analyses especially where high precision and high speed analyses are required. Flame techniques are also more easily automated.

Because of the importance of the concepts of sensitivity and detection limits, these are defined as follows:

1. Sensitivity is the slope of the analytical curve, but the concentration giving an absorbance reading of 0,004 (absorption of 1%) is frequently given instead. To avoid confusion, the word sensitivity as a measure has been avoided in this article and the concentration/absorbance figures are given.

2. The limit of detection is defined in statistical terms and states the concentration which may be detected above the fluctuation of the background. The limit of detection is generally much lower than the concentration giving 0,004 absorbance because of the stability of the measurements, but for some analyses where the analytical conditions are noisy (e.g. tin), the limit of detection may be higher.

PART I.
METALS IN ENVIRONMENTAL POLLUTION

LEAD

Clinical Effects

The toxicity of lead compounds is due to a complex state of solubility in various structures of the organism, the cells and the organelles. This relationship affects the

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absorption, distribution and accumulation of lead in the body, as well as lead excretion from it.[1] Lead is a poison with markedly non-specific effects. The absorption of lead in the cells of many tissues has been reported. These include the red blood cells, bone, liver, brain, nerves, kidneys and blood vessels. Descriptions of the type of anaemia caused by lead poisoning are numerous.[2-4]

The lead atom has a tendency to react with various chemical groups, for example the SH groups. The inhibitory effect of lead on the sulph-hydryl-dependent enzymes is clearly shown in the disturbance caused in the biosynthesis of haem, an essential constituent of haemoglobin and of the cytochromes. According to Chisolm,[2] lead may be implicated in the formation of delta-aminolaevulinic acid (ALA) and in the conversion of coproporphyrinogen to protoporphyrin, but it undoubtedly interferes in the metabolism of ALA and in the final formation of haem from iron and protoporphyrin. Both of these steps are mediated by enzymes dependent on free sulph-hydryl groups for their activity. This aspect of lead toxicity is reviewed by Goldberg.[5]

The best-known chronic effect is chronic nephritis.[2] In Australia in 1929, Nye reported on a pattern of chronic nephritis and early death in Queensland. It was traced to children drinking rainwater collected by run-off from roofs painted with lead pigment.

Another known result of chronic over-exposure is peripheral nerve disease affecting primarily the motor nerves of the extremities. Fullerton of Middlesex Hospital, London, and other investigators have found that conduction of the nerve impulse may be impaired in the peripheral nerves of industrial workers after prolonged exposure to lead, without their showing symptoms of acute poisoning.[2]

Sources of Lead Pollution

Petrol: Danielson[1] summarised his comprehensive report 'Gasoline containing lead' for Swedish conditions as follows: 'Present trends indicate that the pollution of lead from passenger cars in Sweden will increase by 100 - 200% for the period ending 1977. The safety margin between the *present* lead absorption for a person living in the city and that absorption which gives rise to chronic damage in man is apparently very small, possibly completely non-existent'.

Danielson[1] also reported that from a series of measurements made around the earth, the average concentration of lead in the atmosphere of the big cities was a few $\mu g/m^3$ air, and perhaps as high as 10 $\mu g/m^3$. In Los Angeles, the average concentration was 29,3 $\mu g/m^3$ during the morning rush, 22,4 $\mu g/m^3$ during a workday and 22,2 $\mu g/m^3$ during the afternoon rush.[6]

He further reported that 80% of lead particles originating from petrol have a diameter such that they can remain suspended in the atmosphere for an average of 1 month. It is generally assumed that about 50% of airborne lead is absorbed on inhalation (in contrast to the 5 - 10% absorbed from lead-contaminated food and water[7]). It is interesting that Danielson[1] noted that in the USSR, the use of lead in petrol is forbidden in the major cities.

Food and beverages: Danielson[1] quoted a number of studies showing that increases in the lead content of animal and vegetable matter are due to lead originating from petrol combustion.

Food and beverages can become contaminated with lead from storage containers, especially earthenware improperly glazed with lead. The glaze on pottery is a thin surface film covering the ceramic material which may also contain significant amounts of lead. The glaze itself may cause poisoning, as was shown by Klein *et al.* who reported on two cases of childhood lead poisoning, one fatal, which were traced to an earthenware jug in which a continuously replenished supply of apple juice had been kept.

Lead-containing paints: The chipping paint in older buildings is the primary source of lead poisoning in children. Lead poisoning is not uncommon in young children and although there are restrictions on the use of lead in materials readily available to them, painted surfaces (particularly those in old houses) liable to be chewed, may contain lead-pigmented paints.

The determination of lead will be discussed in Part II of this review.

MERCURY

Clinical Effects

In 1950, a factory in Minamata, Japan, began discharging mercury-containing wastes into Minamata Bay and later into the Minamata River. From 1953 to 1960, 121 persons were reported poisoned after eating fish and shellfish from the contaminated areas. Among the 121, there were 19 congenital cases born of mothers who had eaten the same food; 46 deaths occurred.[8,9] A second outbreak occurred in 1964 - 1965 among fish-eaters living near Niigata, Japan. These poisonings totalled 47 cases with 6 deaths.[9] The active agent was reported to be methylmercury which had accumulated in fish and shellfish.

Ordonez *et al.*[10] in 1966 reported unusual illnesses involving the central nervous system. Originally thought to be encephalitis, the disease was shown to be mercury poisoning, as autopsy samples were found to contain high concentrations of this metal. The deceased had ingested wheat seed (treated with a pesticide) which contained 17 ppm of mercury.

Curley *et al.*[11] reported a case of indirect mercury poisoning in humans in the USA, caused by the ingestion of contaminated meat from hogs which had consumed mercury in waste seed grain.

Methylmercury poisoning has been shown to cause atrophy of the cerebellar granule cells and preferential injury to the calcerine and other cortical regions.[12,13] Clinical symptoms start with numbness of the extremities, lips and tongue, followed by dysarthria, ataxia of the gait, dysphagia, deafness and blurring of vision associated with constriction of the visual field.[12,14,15] These symptoms develop 1 - 2 months after ingestion of or exposure to methylmercury.[9] Alkylmercury compounds have the ability

to cross the placental barrier as well as the brain-blood barrier. Congenital neurological injuries have been well documented and the reports were reviewed by Löfroth.[9] Spyker *et al.*[16] argued against using only obvious neurological dysfunction when setting standards for the allowable daily intake, as was done in the USA. According to these authors, consideration should also be given to possible behavioural, biochemical, carcinogenic or other subtle effects, such as lowered intellectual capacity and premature senility, as well as to effects detrimental to the fetus.

Löfroth[9] stated that nothing is known about the methylmercury concentration which can cause irreversible damage in single brain cells. He pointed out that even a low frequency of brain cell damage over a long period of time has an effect, since the number of available cells for each brain function is limited and such damage may have serious effects in later life.

Sources of Mercury Pollution

Agriculture: Mercury compounds have been widely used as fungicides and seed preservatives but were taken off the market in this country in 1974 as a result of a promulgation by INDAC (Interdepartmental Advisory Committee for the safeguarding of the public against poisonous substances; Head Office: Department of Agricultural Technical Services).

Industry: Mercury compounds find industrial application in pulp and paper mills and mercury metal is used extensively in electrolytic processes in the manufacture of chlorine and caustic soda. According to a survey made in Canada in 1969,[17] some 100 000 kg of mercury were required annually as make-up in Canadian chlorine-alkali plants. Most of the lost mercury probably found its way into rivers and lakes. The Minamata poisonings were caused by a vinylchloride and acetaldehyde plant which discharged large quantities of mercury through waste water.

Oceans, lakes and rivers: Although, where pollution occurs, these waters have in the first instance been contaminated by human activities, and although the coastal and especially the inland waters of this country have not yet been seriously polluted by mercury, the Republic would do well to learn from experiences in other parts of the world, where these waters themselves have become the sources of pollution in the aquatic food chain.

Mercury is strongly held by the bottom sediments of natural water courses and several binding mechanisms have been suggested.[18] As has been shown by Jensen and Jernelöv,[19] mercury, discharged into the waters in any form (e.g. inorganic mercury), can be methylated by micro-organisms. Methylmercury then accumulates in fish and other biota. In 1970 the Canadian government announced a ban on the sale and export of fish taken from the waters of Lake St Clair because these fish were mercury-contaminated.

Pike (a freshwater fish) have been used as an indicator of mercury contamination in the environment.[17] Swordfish and tuna were confiscated on a large scale in the USA as a consequence of the establishment of a maximum permissible level of 0,5 ppm for mercury in fish. Ganther *et al.*[20] described the surprising finding that tuna in the diet decreased the toxicity produced with high concentrations of mercury. They presented evidence that the selenium present in tuna may be responsible for this.

Mercury Determination

Mercury has an appreciable vapour pressure, so that the conventional flame is not needed. Instead, use is made of an absorption cell placed in the path of the hollow cathode beam. The sample is chemically treated to release its mercury which is then swept through the absorption cell by aeration. The 2 537 Å line is commonly used. The bandpass should be fairly narrow (± 2 Å) for maximum sensitivity, so as to minimise the effect of the nearby 2 535 Å line. The bandpass is the product of the dispersion and the slit width, e.g. a monochromator having a dispersion of 32 A/mm operated with a 100-μm slit width, has a bandpass of 3,2 Å.

Water samples: By means of the absorption cell method, mercury can be accurately determined down to 1 ppb in solution according to the method originally described by Hatch and Ott.[21] The mercury in seawater can be concentrated easily to a level well above 1 ppb by means of extraction and by using a chelating agent. Bailey and Lo[22] described the handling of large numbers of samples by means of an automated version of the method of Hatch and Ott. Chau and Saitoh[17] used a combination of dithizone extraction and the aeration-atomic absorption technique to determine mercury in lake waters. The concentration range of their samples was 0,048 - 0,478 ppb and they attained a limit of detection of 0,008 ppb.

Biological samples: Jeffus *et al.*[23] determined mercury in hog tankage, wheat, treated wheat (5,64 ppm), shellfish and fish. In the analysis of biological samples, the digestion prior to determination is the critical step. Such samples cannot be dry-ashed as the mercury will be lost at the temperatures required for ashing. Even with low temperature ashing, using radio frequency power, Pillay *et al.*[24] recorded mercury losses of 81 - 98%. An excellent discussion of the release of mercury by volatilisation under wet oxidation conditions is to be found in a publication by Gorsuch.[25]

Lindstedt[26] determined mercury in urine by means of the technique under discussion and used overnight digestion of 1-ml urine samples at room temperature with sulphuric acid and potassium permanganate.

In their report on mercury determination in water, fish, urine and organomercury compounds, Hwang *et al.*[27] used accepted methods of digestion and investigated the other analytical variables. Magos[28] described the selective determination of inorganic mercury and methylmercury in undigested biological samples.

CADMIUM

Clinical Effects

In a review of the literature on the toxicity of cadmium, Nilsson[29] listed the following effects in man: amino-

is 2 $\mu g/m^3$, yet even the lowest value is well within the capabilities of the abovementioned instrumentation.

IRON

Workers in the iron and steel industries exposed to iron dust may develop conjunctivitis, choroiditis, retinitis and siderosis of tissues. Iron oxide fumes are generated in welding operations and continued exposure to concentrations above 30 mg/m^3 air can cause chronic bronchitis and fresh iron oxide fumes can cause metal fume fever (Sax,[4] p. 905). Sullivan[89] listed pneumoconiosis as one of the effects on humans.

Determination of Iron

Because of other non-absorbing lines very close to the most sensitive 2 483 Å absorbing line, a narrow bandpass, not greater than 2 Å, should be used. The signal-to-noise ratio with this line is not as good as with other lines and more precise measurements can be made with the 3 720 Å line. An oxidising air/acetylene flame should be used. Organic solvents have been used to increase sensitivity.[70] For the determination of iron in serum, Zettner *et al.*[71] recommended extracting the iron-bathophenanthroline complex into MIBK which concentrated the iron and eliminated interferences. Other reports are those of Rodgerson and Helfer[72,73] and Zaino.[74] Welz and Wiedeking[75] determined iron in 2-μl samples of serum using the flameless technique. Zettner and Mensch[76] proposed the use of atomic absorption for the measurement of the iron content of haemoglobin.

Urinary iron may be determined directly, which is particularly useful in the treatment of patients with haemochromatosis whose urine may contain the iron-chelating compound desferrioxamine. Zettner and Mansback[77] recommended that for very accurate work, a urine pool be used to prepare the iron-calibrating solutions.

CHROMIUM

Chromium metal is essentially non-toxic, but workers in the chromium-plating, chromium-tanning pigments and chromium-alloy industries may be subjected to the effects of chromic acid and its salts. These have a corrosive action on the skin and mucous membranes, lesions being confined to the exposed parts. The characteristic lesion is a deep, penetrating ulcer which usually does not tend to suppurate, but is slow in healing. On the mucous membrane of the nasal septum, the ulcers are usually accompanied by purulent discharge and crusting. Continued exposure may result in perforation of the nasal septum, but produces no deformity of the nose. Chromate salts have been associated with chronic catarrh, emphysema, and cancer of the lungs.[78]

Apart from the toxicity aspect, physiological interest in chromium has recently been stimulated by the identification by Schwarz and Mertz[79] of chromium (III) as the active ingredient of the glucose tolerance factor in the rat. Work along these lines was also done by Glinsman *et al.*[80] who studied the chromium concentration in plasma in conjunction with a glucose tolerance test.

Determination of Chromium

A good analytical sensitivity can be obtained for chromium, the hollow cathode lamps being very stable so that scale expansion can be used. The line at 3 579 Å is the most sensitive, and with aqueous solutions, a concentration of 0,05 ppm gives an absorbance reading of 0,004.

Chromium is relatively interference-free. Williams *et al.*[81] reported that calcium, silicon and phosphorus depressed the chromium absorption. The addition of calcium eliminated phosphate interference, and calcium and silicate were added to the standard solutions to compensate for their effects. Silicate should, however, not be a consideration of importance in biological analyses. Iron markedly suppresses chromium absorption,[82,83] but given the right flame conditions, ammonium chloride can effectively reduce this suppression. The chromium 3 579 Å line lies at the edge of the MgOH absorption bands, as noted by Koirtyohann and Pickett,[56] but this is not likely to cause serious interference and may only become significant when scale expansion is used. Feldman and Purdy[84] undertook an investigation regarding optimal conditions for chromium determination.

The atomic absorption limit of detection of 0,05 ppm for chromium in aqueous solution is inadequate for the levels present in biological samples and preconcentration steps must be taken to determine the natural levels of chromium in samples such as plasma and tissues. Analyses of biological material have been reported.[60,65,81]

No preconcentration is required when the flameless technique is used. A technique for the determination of chromium in blood plasma, urine and other biological materials was developed by Davidson and Secrest.[85] They obtained a detection limit of about 2 picograms. For routine analysis of plasma, the method is effective with either pre-ashed samples or the direct analysis of samples of the order of 20 - 200 μl in volume.

MANGANESE

The main sources of manganese pollution are mining, the iron and steel industry, fuel oil, incineration, coal and dry cell batteries. Continued exposure may lead to chronic manganese poisoning and manganese pneumonia.[86] Chronic manganese poisoning is clearly characterised as resulting from the inhalation of fumes or dusts of manganese. The central nervous system is the chief site of damage. When well established, the disease results in permanent disability (Sax,[4] p. 956).

Manganese Determination

The atomic absorption method is particularly well suited for the determination of manganese. A high degree

of sensitivity is obtainable and there are very few interferences. A concentration of 0,01 ppm gives 0,004 absorbance, and the detection limit is of the order of 0,001 ppm. A convenient working range is 2 - 20 ppm. With the most sensitive 2 795 Å line, a narrow bandpass of 2 Å or less should be used for maximum sensitivity so as to exclude the 2 801 Å line. The latter may also be used for analysis with about half the sensitivity. Scale expansion is easily possible because of the stability of the manganese lamp. The air/acetylene flame gives the highest sensitivity. Organic solvents (e.g. acetone)[87] enhance absorption, but when these are used, the burner height is more critical. When using an air/acetylene flame, the only serious interference comes from silicon which depresses absorption. Allan[88] tested for possible interference effects from 3 000 ppm potassium or calcium, 1 000 ppm sodium or magnesium or 500 ppm phosphorus, all on 10 ppm manganese, and found none.

Mahoney *et al.*[89] used flame atomic absorption techniques to analyse serum and reported the normal level for fasting adults as 0,024 ± 0,007 μg/ml. Joyner and Finley[90] extracted and concentrated manganese using sodium diethyldithiocarbamate chelate and extracting into MIBK. Because of its sensitivity, the flameless technique has the advantage of not requiring extraction. This technique is especially suited to serum analysis, as has been described by Bek *et al.*[91]

VANADIUM

Sources of vanadium pollution and/or poisoning are mines, coal, fuel oil and vanadium concentration industries.

Vanadium compounds act chiefly as irritants to the conjunctivae and respiratory tract. Prolonged exposure may lead to pulmonary involvement.[4] Symptoms and signs of poisoning are pallor, greenish-black discoloration of the tongue, paroxysmal cough, conjunctivitis, dyspnoea, pain in the chest, bronchitis, rales and rhonchi and radiographic reticulation.[4] Cardiovascular disease and carcinogenesis have also been listed under the effects of vanadium on human health.[92]

Vanadium Determination

All the reports mention the use of highly reducing (i.e. fuel-rich) acetylene/nitrous oxide flames. These reducing conditions are necessary to aid in the dissociation or to prevent the formation of the very stable vanadium oxides in the flame. It is not possible to determine vanadium in an air/acetylene flame. Using the acetylene/nitrous oxide flame and the most sensitive line at 3 184 Å, Amos and Willis[93] obtained an absorbance reading of 0,004 for 1,5 ppm in aqueous solution. Sachdev *et al.*[94] investigated the determination of vanadium. They tested a large number of ions (in fivefold excess) and some acids (diluted 5 : 100) for interference. Some of these substances enhanced vanadium absorption. Those with an enhancing effect which are present in biological materials were Na^+, K^+ and Fe^{3+}. Other substances with the same effect were acetic acid and phosphoric acid. These workers recommended the addition of 300 ppm aluminium to standards and samples for a 10 - 100 ppm vanadium concentration range to suppress interferences. For a further increase in sensitivity, the addition of 6% diethylene glycol diethyl ether was recommended. The ratio and pressure of the flame gases and the flame position are critical.

Although no work has been reported on the determination of vanadium by means of the flameless technique, the indications are that this method should be suitable. Because of the presence of carbon, the atmosphere is a reducing one and this, together with the use of an argon-filled gas chamber, should make the determination of very small quantities of vanadium entirely feasible.

NICKEL

Workers in steel and nickel-alloy and in nickel-plating industries may be subjected to nickel poisoning, the symptoms of which are dermatitis and respiratory disorders.[95] Nickel carbonyl is sometimes found as one of the constituents in town gas. Although the metal itself may generally be described as relatively non-toxic, inhaled nickel carbonyl is deposited in decomposed form in extremely fine particles in the lung and is very toxic.[60] Extended exposure is capable of producing lung carcinoma.[60,95] Sunderman[96] studied the carcinogenesis of nickel carbonyl and its effects on RNA. D'Alonzo and Pell[97] found serum nickel levels raised to 0,5 μg/ml in patients with acute myocardial infarction and Schroeder[98] reported that urine of human hypertensives contains increased amounts of nickel. Sunderman[99] reported a mean serum nickel concentration of 0,022 μg/ml and a range of 0,001 - 0,077 μg/ml.

Nickel Determination

The most sensitive line is the one at 2 320 Å (0,13 ppm required for 0,004 absorbance), but a nearby non-absorbing line at 2 319,8 Å cannot be resolved, resulting in a marked curvature of the calibration graph at higher concentrations. A bandpass of 1 Å or less should be used for maximum sensitivity, but the 2 320 Å line is very weak, so that a narrow slit results in a low signal-to-noise ratio.

Willis[54] preferred to use the 3 415 Å line for determination of nickel in urine. Even though this line is fifteen times less sensitive, scale expansion can be used more conveniently at this wavelength. He found that 0,5 - 10 ppm nickel could be determined by directly aspirating the urine and using fourfold scale expansion. For more dilute concentrations, an extraction technique was applied. Standard additions were used for calibration by adding nickel to a separate portion of the urine sample, 0,05 - 0,06 ppm nickel in urine being recovered satisfactorily. This is just above the expected maximum normal concentration in urine and 2 - 3 times the average, so that this method can be used only for determination of elevated levels of nickel.

Sunderman[99] concentrated the nickel in biological samples and found a mean nickel concentration in 24-h collections of urine for 17 normal subjects of 0,018 μg/ml, with a range of 0,004 - 0,031 μg/ml. The mean urinary excretion was 19,8 μg/day. Koirtyohann and Feldman[100] determined nickel in tissue ash and prepared standards in a solution of a synthetic soft tissue ash to avoid errors arising from matrix effects. Although not yet reported, very small samples could probably be analysed using some kind of pre-oxidation step with the flameless technique. The sensitivity should thus be adequate for most biological samples.

ZINC

Workers in zinc smelting and refining plants and in zinc galvanising industries may be subjected to zinc oxide fumes. These fumes, when freshly inhaled, can cause a disease known as 'brass founders' ague' or 'brass chills'. Fatalities have also resulted from lung damage caused by the inhalation of high concentrations of zinc chloride fumes.[4] Exposure to these fumes can cause damage to the mucous membrane of the nasopharynx and respiratory tract and give rise to a pale grey cyanosis. It is stated in Sax's book[4] that zinc oxide which is not freshly formed is virtually innocuous and that as far as can be determined, the continued administration of zinc salts in small doses has no effect on man except that of constipation and disordered digestion. However, in his comprehensive study of zinc pollution, Athanassiadis[101] listed hypertension and arteriosclerotic and heart disease as long-term effects.

Zinc Determination

With the exception of silicon, zinc is free from significant interference of other elements.[102] The 2 139 Å line is the most sensitive. With conventional instruments, the concentration required for 0,004 absorbance is of the order of 0,025 ppm in aqueous solutions. Detection limits have been reported as 0,002 - 0,015 ppm.[103] Allan[104] increased the sensitivity about five times by complexing and extraction.

The air/acetylene flame absorbs about 25% of the radiation at the 2 139 Å line and it is thus essential that the flame should not vary.

Sulphuric[105] and perchloric[104] acids depress zinc absorption and should be present in the standards in the same concentrations as in the digested samples.

Methods for measurement of plasma zinc have been described using both protein precipitation[106] and direct determination on diluted serum.[107] A detailed study on serum zinc by Reinhold *et al.*[108] illustrated the importance of the internal diameter of capillary tubing used for aspirating the sample into the burner. This finding is of significance for all trace metals in serum. Dawson and Walker[109] described the determination of zinc in whole blood, plasma and urine. The first two types of sample were considerably diluted. Willis[110] determined urinary zinc. Harrison *et al.*[111] discussed the measurement of zinc in human hair as a possible aid to toxicological and pathological studies.

ARSENIC

In this country, the agricultural uses of arsenic are limited to the use of arsenic compounds in cattle dips and in sprays used in the citrus industry. Other sources of arsenic pollution are smelters processing arsenical ores and in some instances the sulphuric acid manufacturing industry. Iron pyrite from which sulphuric acid is manufactured is sometimes associated with arsenopyrite. Arsenopyrite is also found in the ores associated with gold mining.

The fairly common acute allergic reactions to arsenic compounds used in medical therapy are well-known to the medical profession, as are the symptoms of acute poisoning. Chronic arsenic poisoning, whether through ingestion or inhalation, may cause disturbances of the digestive system, the blood, kidneys and nervous system.[4] Chronic poisoning can also cause bronchitis[112] as well as a variety of skin abnormalities including itching, pigmentation and cancerous changes (Sax,[4] p. 464).

Determination of Arsenic

The destruction of organic matter prior to arsenic determination is usually accomplished by wet ashing, although Evans and Bandemer[113] dry-ashed biological material at 600° with magnesium nitrate as ashing aid. The latter is important since Gorsuch[35] found that after dry ashing with no ashing aid, the arsenic recovery was only 88%; with the ashing aid it was 99%.

The arsenic lines most suitable for absorption are located below 2 000 Å (1 890, 1 937 and 1 972 Å), a region in which air and most conventional flames absorb radiation strongly. For example, as much as 60 - 75% of the source radiation at 1 937 Å may be absorbed by the various molecular species in the air/acetylene flame. Kirkbright and Ranson[114] overcame this difficulty by using a slightly fuel-rich nitrous oxide/acetylene flame and by separating the flame by nitrogen shielding, i.e. separating the primary from the secondary combustion region. In this way they obtained a much higher degree of transparency of the flame at the desired wavelength. A detection limit of 1 ppm was obtained.

Chu *et al.*[115] described a flameless atomic absorption method which involves the chemical conversion of arsenic to arsine. The arsine evolved was swept into an absorption tube by means of an argon carrier gas. The relative standard deviation for 5 replicates of 0,4 μg arsenic standards was 0,36%.

Massmann[116] described the determination of arsenic by means of the furnace technique, with a limit of detection of 1 ng, or in terms of concentration, 0,1 μg/ml.

BERYLLIUM

Beryl ($3BeO \cdot Al_2O_3 \cdot 6SiO_2$), although mined in this country and in South West Africa, is not yet processed here.

Some beryllium compounds, however, find a number of applications in industry. The mineral, beryl, probably has little toxicity in itself. It has not been known to cause pneumoconiosis (Sax,[4] p. 501). However, metallic beryllium is toxic in very small amounts.[117] The industrial extraction of beryllium from its ore causes man to be exposed to acid salts of the metal, particularly to the fluoride, the ammonium fluoride and the sulphate, and also to beryllium oxide and hydroxide. Exposure to the oxide also occurs when beryllium alloys are cast and in operations with beryllia ceramics. In the manufacture of fluorescent powders, for fluorescent lamps and sign tubes, there may be exposure to beryllium carbonate and to more complex salts.

Exposure to certain beryllium compounds in finely divided form may result in dermatitis of an oedematous and papulovesicular type, chronic skin ulcers, rhinitis, nasopharyngitis, epistaxis, bronchitis and, in severe cases, in an acute pneumonitis with severe cough, low-grade fever, rales, dyspnoea and substernal pain. Radiographs show diffuse haziness throughout both lungs, followed by the appearance of soft, ill-defined opacities. The condition usually occurs while the worker is exposed, sometimes within a month of starting work, and as a rule recovery occurs within two months, though radiographic changes sometimes persist for longer periods, with an occasional failure of complete resolution, followed by fibrosis (Sax,[4] p. 502). A delayed form of lung disease, for which the prognosis is poor, has also been described.[4]

Under the effects of beryllium on human health, Durocher[118] listed chemical ulcer and carcinogenesis. Beryllium in the diet causes severe rickets[119,120] and since this type of rickets is not cured by vitamin D, it has been suggested that this metal may have a local toxic action on the bone calcification mechanisms, especially on alkaline phosphatase.[121] Beryllium inhibits serum alkaline phosphatase, the action of which is prevented *in vitro* and *in vivo* by chelating agents.[122]

Beryllium Determination

A method for preparing lung tissue for analysis has been described by Hayes *et al.*[123] and the preparation of urine samples by Smith *et al.*[124] Because of the formation of refractory beryllium oxides, hotter flames than the air/acetylene flame are required to atomise beryllium. Willis[125] obtained an absorbance reading of 0,004 for 0,03 ppm in aqueous solutions using a nitrous oxide/acetylene flame. Ramakrishna *et al.*[126] did investigations with both the nitrous oxide/acetylene and air/acetylene flames and found determinations to be free from interferences to a large extent. Bokowski[127] could detect 2 ppb of beryllium in urine. Here again, with some form of pre-ashing this determination could be done more rapidly, and probably with greater sensitivity, by means of the flameless technique.[94]

CONCLUSION

The progress in trace and macro metal analyses by the use of atomic absorption spectroscopy has aided greatly in defining the precise functions of metals in metabolism, in diagnosing pathological conditions and in defining maximum allowable concentrations with regard to some aspects of pollution.

Many excellent atomic absorption instruments are commercially available at relatively modest cost — especially when compared with other types of analytical apparatus. These instruments are usually accompanied by books of standard methods and analyses can accordingly be carried out by most technicians. However, it is recommended that some knowledge of the principles concerned is necessary to obtain the maximum benefit from these instruments.

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