

The biological behaviour and bioavailability of aluminium in man, with special reference to studies employing aluminium-26 as a tracer: review and study update†

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Until 1990 biokinetic studies of aluminium metabolism and biokinetics in man and other animals had been substantially inhibited by analytical and practical difficulties. Of these, the most important are the difficulties in differentiating between administered aluminium and endogenous aluminium—especially in body fluids and excreta and the problems associated with the contamination of samples with environmental aluminium. As a consequence of these it was not possible to detect small, residual body burdens of the metal following experimental administrations. Consequently, many believed aluminium to be quantitatively excreted within a short time of uptake in all, but renal-failure patients. Nevertheless, residual aluminium deposits in a number of different organs and tissues had been detected in normal subjects using a variety of techniques, including histochemical staining methods. In order to understand the origins and kinetics of such residual aluminium deposits new approaches were required. One approach taken was to employ the radioisotope ^{67}Ga as a surrogate, but this approach has been shown to be flawed—a consequence of the different biological behaviours of aluminium and gallium. A second arose from the availability, in about 1990, of both ^{26}Al —a rare and expensive isotope of aluminium—and accelerator mass spectrometry for the ultra-trace detection of this isotope. Using these techniques the basic features of aluminium biokinetics and bioavailability have been unravelled. It is now clear that some aluminium is retained in the body—most probably within the skeleton, and that some deposits in the brain. However, most aluminium that enters the blood is excreted in urine within a few days or weeks and the gastrointestinal tract provides an effective barrier to aluminium uptake. Aspects of the biokinetics and bioavailability of aluminium are described below.

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1 Introduction

The literature is replete with the description of a legion of studies that are concerned with some aspect or other of the behaviour of metal ions, such as aluminium ions, in the body. Most of these have concentrated on the measurement of chronically accumulated metal deposits, at the whole tissue and organ levels of complexity, and have commonly employed routine analytical techniques. A compilation of such data, for the tissue and organ contents/concentrations of 51 elements (including aluminium), is given in the ICRP publication “Reference Man”.¹ While these have helped us understand the distribution of metal ions—particularly those of the essential metals such as iron and calcium and chemically related metals—at the whole body level, they tell us little about the behaviour/kinetics of metals in intact biological systems, which may be affected by the body’s homeostatic mechanisms. In addition, many studies have been conducted looking at the behaviour of different metals within *in vitro* biochemical systems. These also may add little to our understanding of the behaviour of metals in the body.

In contrast, relatively few studies have been undertaken—using either qualitative morphological and histochemical techniques or quantitative techniques, such as electron probe microanalysis and quantitative autoradiography—which allow the description of the temporal distribution of metal ions *in vivo*. Similarly, relatively few pharmacokinetic studies have been undertaken that provide information on the whole-body retention and excretion patterns of different metals. It follows, that an in depth search of the literature will often fail to reveal basic information about the retention, distribution and redistribution patterns of metals in the body. Similarly, very

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little is known about the factors that determine the behaviour of different metals in the bodies and organs of children and adults, although the existence of a multiplicity of metabolic pathways for different essential metals is well documented, and these may be sufficiently non-specific to affect the biological disposition of a large number of "non-targeted" essential and non-essential, or even toxic, trace metals. This is a pity, as when the most effective treatment regimes are being devised, to counter the effects of either metal deficiency or to mitigate the consequences of metal overload and toxicity, it is the basic, simple and often absent, age-related, acute kinetic and distribution data for metals in the tissues of man that are often most required. Similarly, this data is required to devise appropriate biological assay techniques to determine metal body burden and to regulate metal intakes under situations of occupational exposure. Finally, such data can be used to construct mathematical models of metal deposition, redistribution and retention that are required for toxicology.

Until recently, many of the above remarks were as true for aluminium as for any other metal, in that data was only readily available concerning the relative concentrations of the metal in different organs and tissues, and little was understood concerning the biokinetics of aluminium retention and excretion. The review of Wilhelm *et al.*² summarises the state of knowledge concerning the toxicokinetics of aluminium at this time. In 1990 the following information was available:

(1) Aluminium intakes by adults within the western world normally ranged from about 2 to 20 mg day⁻¹, but that the use of pharmaceutical preparations containing aluminium could result in the daily ingestion of greater than 1g of the element.

(2) Both plasma aluminium levels and the quantity of aluminium excreted in the urine increased following the ingestion of most forms of aluminium—providing conclusive evidence that ingested aluminium is to some extent bioavailable.

(3) Aluminium welders showed elevated urinary excretion of aluminium indicating the transfer of inhaled metal to blood.

(4) Aluminium bioavailability (by ingestion), as determined either by balance studies, by aluminium excretion studies or by comparisons of integrated plasma aluminium concentrations lay within the uncertain range of 0.001 to 24%.

(5) Estimates of the level of protein binding of aluminium in plasma ranged from very little to 98%.

(6) Measured clearance half-times for plasma aluminium in man ranged from 14 h to 85 days and that the half-time measured was a function of the observation period.

(7) Most aluminium, perhaps even 100%, in blood is excreted *via* the kidneys in urine, but evidence also existed to suggest that an unknown fraction was excreted by the biliary route in faeces.

(8) When the body retained aluminium the sites of deposition were the liver, spleen, skeleton, kidneys and lungs.

It can be seen that for most biokinetic parameters the range of reported values was large, indicating substantial uncertainty. Indeed, at an earlier date, the International Commission on Radiological Protection,³ a body which reviews the metabolic data for all elements in man (in order to specify mathematical models which can be used for dose calculation purposes following intakes of radioactive isotopes) found few unambiguous data to support the formulation of a metabolic model for use in calculating radiation doses from internal deposits of ²⁶Al and ²⁸Al—aluminium's only significant, but rare, radioactive isotopes. The Commission proposed, as a working postulate, a model in which 30% of aluminium entering the body was deposited in the skeleton on bone surfaces and 70% became uniformly deposited throughout the remaining body tissues; systemic aluminium was assumed to be lost in urine, with a half time of 100 days. Clearly, this model, which was considered conservative, could be justified on the basis of the wide range of available data, but equally data is available to suggest a model

for which it is assumed that all aluminium intakes are rapidly excreted in urine.

More recently, however, the availability of ²⁶Al as a tracer, and of accelerator mass spectrometry (AMS) as an analytical tool has considerably facilitated our ability to conduct aluminium studies in man.⁴⁻⁶ As a consequence our knowledge of aluminium biokinetics has greatly increased during the last fifteen years.

2 Aluminium occurrence and chemistry

An outline knowledge of the occurrence and chemistry of aluminium is crucial to our understanding of its behaviour in biological systems—including the human body—and to understanding routes of exposure. While standard inorganic chemistry texts may be used to gain a general understanding of the chemical properties of this element, the reader is referred to texts such as those of Martin⁷ and Colombini⁸—in the book *Aluminium in Chemistry, Biology and Medicine*⁹—Elinder and Sjögren¹⁰ and Birchall and Chappell¹¹ for reviews of the aspects of aluminium chemistry relevant to its behaviour in biological systems.

Aluminium is the commonest metal in the earth's crust (8.1%) and is the third most common element. It was discovered in 1825 by the Danish chemist, Oersted. It is a soft, ductile, malleable, silvery metal. Its atomic number is 13 and it has one stable isotope, ²⁷Al, and one long-lived radioactive isotope, ²⁶Al. Aluminium belongs to Group IIIa of the Periodic Table, along with boron, indium, gallium and thallium. It is reactive and, therefore, never occurs naturally in its native form, but as insoluble compounds within minerals—including bauxite and clay minerals. In contrast, aluminium metal is effectively non-reactive—a consequence of the rapid formation (in the presence of oxygen) of a protective layer of oxide over any exposed surfaces. This protective layer, in addition to its light weight (density = 2.7 g cm⁻³) makes aluminium metal an ideal material for many applications in the construction industry, in the transport industry and in the packaging industry. Today, of all metals, that of steel only exceeds the global production of aluminium. In addition, aluminium compounds are widely utilised by industry.¹² They are used: in the paper industry; for water purification; in the dye industry; in solid-fuelled rocket motor fuels; in paints and pigments; in the textile industry; as a catalyst in oil refining; in the glass industry and as components of cosmetic and pharmaceutical preparations. Given its wide use, its putative potential for the contamination of workers and members of the public is high.

Aluminium is trivalent and forms trivalent ionic (Al³⁺) compounds, but has some covalent characteristics—a result of electron cloud polarisation resulting from aluminium's high ion charge: size ratio. In acidic solution aluminium dissolves to form Al³⁺ ions. Such ions are small, with an effective ionic radius, in six-fold co-ordination, of 54 pm (*cf.* Ga³⁺, 62 pm; Fe³⁺, 65 pm; Fe²⁺, 78; Ca²⁺, 100 pm). In other solutions a number of other complex ions may be formed including, Al(OH)²⁺, Al(OH)₂⁺ and Al(OH)₄⁻. These ions compete with the presence of Al³⁺ within aquatic systems and each predominates, in solution, within a characteristic range of pH (see Fig. 1). Consequently, the solution chemistry of aluminium is complicated. Aluminium is minimally soluble in water at about pH 6, when the Al(OH)₂⁺ ion dominates, but solubility increases at lower and higher pH values. At pH 7, and above, the most important ion is Al(OH)₄⁻, whereas, at low pH values Al³⁺ dominates. At near neutral pH aluminium is effectively insoluble and if acidic solutions containing Al³⁺ are neutralised then the vast majority of the aluminium hydrolyses to form an insoluble aluminium hydroxide (Al(OH)₃) precipitate. It is the formation of this, during water treatment to produce drinking

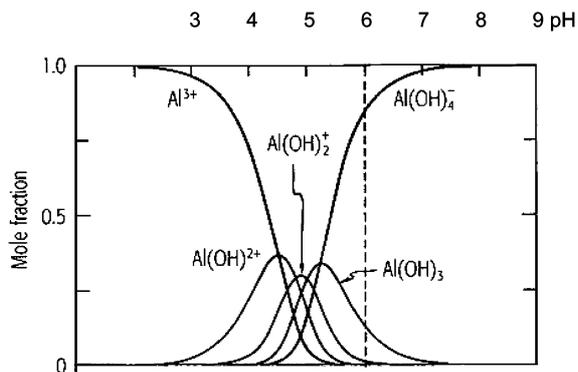


Fig. 1 Mole fraction of soluble aluminium ions as a function of solution pH in aqueous solution (modified from ref. 7).

water that results in the co-precipitation of ground water impurities, including bacterial spores, with the aluminium hydroxide.

Most natural waters contain very little dissolved aluminium (often $<10 \mu\text{g L}^{-1}$), reflecting the low solubility of minerals and the deposition of Al^{3+} in sediments as the hydroxide. Seawater contains only $1 \mu\text{g L}^{-1}$ of aluminium, and much of this is likely to be bound within the skeletons of diatoms (free-living single celled organisms). Where natural waters have either been acidified by acid rain or treated with aluminium sulfate to produce drinking water, the levels of the metal are higher. Concentrations in acidified lakes and rivers (up to $700 \mu\text{g L}^{-1}$) commonly exceed levels toxic to fish. This may cause a severe problem either in geographical areas where the soils are naturally acidic or where soils with a low buffering capacity have been acidified by anthropogenic acid rain—a consequence of sulfur dioxide and nitrogen oxides (NO_x) production during the burning of fossil fuels. In acidic well-water aluminium concentrations exceeding 1 mg L^{-1} occur. Aluminium concentrations in drinking water should not exceed $200 \mu\text{g L}^{-1}$ and within the UK average about $20 \mu\text{g L}^{-1}$.

Air concentrations of aluminium, in the form of suspended minerals, range from below 1 ng m^{-3} of air in rural environments to levels as high as 10 ng m^{-3} in urban, industrialised areas. The higher levels within urban areas result from the increased, dust creating, activities of man within towns and cities.

3 Use of aluminium-26 for biological studies

Prior to 1990, all studies of aluminium biokinetics and bio-availability in man and animals employed the stable isotope of aluminium, ^{27}Al . The use of the natural isotope imposed severe restrictions upon the studies that could be undertaken. This is for two main reasons. Firstly, since aluminium is present in the earth's crust at levels of about 10^{-1} g g^{-1} , but commonly only occurs in biological samples at levels ranging from 10^{-6} – 10^{-8} g g^{-1} the potential for sample contamination during the analysis of biological samples is high. It follows, that the study of the behaviour of aluminium in animals and man has been substantially inhibited by the difficulty of measuring small amounts of aluminium present within biological samples in an environment extensively contaminated with this element. This has made it impossible to rely upon the results generated by many studies—even though some centres seem better able to reproducibly measure aluminium than others.¹⁴ In this regard it is notable that successive studies of the levels of aluminium in tissues tend to report lower levels with time, these reflecting improvements in analytical techniques.^{15,16} Secondly, as aluminium is a normal component of the body the interpretation of biokinetic studies of either injected or ingested aluminium in man and animals is complicated by the investigator's inability to distinguish between aluminium from a test dose and that already present in the body. Most studies of stable

aluminium are thus of poor sensitivity. Yet biokinetic data on the retention and excretion of aluminium is vital in order to: interpret human bioassay data; to determine the bioavailability of ingested and inhaled aluminium compounds; to determine the effectiveness of chelation therapy to remove aluminium; to determine the long-term consequences of acute and chronic aluminium intake. It follows that tracer studies using either other aluminium isotopes or surrogate isotopes are required.

In all tracer studies the assumption is made that the body is unable to differentiate between the metal under investigation and the tracer employed and that as a consequence they behave identically in biological systems. For bioavailability studies it is further assumed that the fraction of the tracer absorbed will be the same as the fraction of the isotope under investigation. This is assumed to be true provided that the tracer and metal under investigation are similarly speciated.

To date, two main types of tracer have been used to study aluminium biokinetics. Firstly, some studies have employed gallium—commonly as the radioactive ^{67}Ga isotope—as a surrogate for aluminium. *In vitro*, the chemical properties of aluminium and gallium are very similar; it being very difficult to remove gallium from aluminium salts using chemical techniques. However, chemical reactions in the body, unlike in solutions, are very sensitive to ionic size^{17,18} and studies conducted using co-administered radionuclides, with the same speciation, at tracer levels have shown these elements behave quite differently in the body.¹⁹ It follows that gallium is normally unacceptable as a tracer for aluminium *in vivo* studies. In contrast it may be acceptable for *in vitro* studies.²⁰ The second approach employed the long-lived radioactive isotope of aluminium— ^{26}Al —in combination with the emerging accelerator mass spectrometry (AMS) technology. Unlike for gallium isotopes, there is no reason to doubt the suitability of ^{26}Al as a tracer.

Aluminium has only two isotopes with half-lives longer than a few minutes: ^{27}Al , which is stable, and ^{26}Al that is almost stable—it has the very long half life of 716 000 years. ^{26}Al decays either with the emission of a positron (β^+ -particle) (85%) or by electron-capture (15%). The decay is accompanied by the emission of two high-energy γ -photons at 1.82 (100%) and 1.1 (4%) MeV. In addition, each positron emitted is annihilated with the production of 511 keV X-rays. The radioisotope does occur naturally, but at such low levels that it can be reasonably regarded as wholly anthropogenic. It is produced in particle accelerators by bombarding a magnesium target with deuterons. Two reactions are employed: $^{26}\text{Mg}(\text{d}, 2\text{n})^{26}\text{Al}$ and $^{25}\text{Mg}(\text{d}, \text{n})^{26}\text{Al}$. Both of these reactions produce low yields of ^{26}Al in terms of radioactivity (a function of the long half-life and, therefore low specific activity of the isotope); consequently, magnesium targets have to be irradiated for long periods to produce the quantities of the radionuclide that are required for biological tracer studies—particularly those studies that rely on radiometric methods for ^{26}Al determination. It follows that ^{26}Al is expensive both to produce and to buy, and financial constraints may limit its experimental use.

Given that ^{26}Al is both radioactive and very long-lived it is possible to use both radiometric and mass spectrometry methods to determine its levels in biological samples. With respect to the former, it is possible to count either the 1.82 MeV γ -rays emitted when ^{26}Al decays to ^{26}Mg or the 511 keV X-ray quanta produced when the positron emitted by the decaying radionuclide nucleus is annihilated. The latter produces two quanta, at 180° to each other, which may be detected using coincidence-counting techniques. The use of ^{26}Al for biological experiments allows the use of a tracer that is normally absent from biological systems—this permits its unambiguous identification within studies. Also, when used in studies in combination with accelerator mass spectrometry it can be detected at such low levels within biological samples and over such a wide range of concentrations that it may be deployed both at relatively low radionuclide costs and without incurring

unacceptable radiation doses for human volunteer studies. This is important since ethical and regulatory requirements require the radiation doses received by volunteers be both as small as possible, consistent with the aims of the study, and below specified radiation dose limits.

Within the UK the limits that are applied by ethics committees considering human volunteer study applications and by the Department of Health Administration of Radioactive Substances Advisory Committee (ARSAC), which issues certificates approving studies, are those specified by the World Health Organisation.²¹ Under most circumstances the appropriate limit is the Category 1 dose limit corresponding to a 50-years committed radiation dose of 500 μSv . This value represents only about 1/2% of the corresponding background dose, but is still much higher than the doses received by subjects in volunteer studies using ^{26}Al . To date, the largest radiation dose received by any volunteer was that received by a subject (the author) following the intravenous administration of 510 Bq of ^{26}Al , namely 60 μSv .^{22,23,19} In some other studies the committed dose received by subjects has been as low as 0.2 μSv .²⁴ Such doses are trivial compared with regional variations in background radiation dose and are much lower than the doses normally received by the subjects in pharmacokinetic studies employing ^{14}C - and ^3H -labelled drugs (John Harrison, NRPB, Chilton, UK, personal communication). Consequently, ^{26}Al may be deployed for a wide a range of human studies.

4 Measurement of aluminium-26

For large samples and for ^{26}Al present within the body of human volunteers it is most convenient to count the 1.82 MeV γ -rays using conventional sodium iodide (NaI) detectors. The author and his colleagues using the Harwell Laboratory whole-body monitor have deployed this method. This facility comprises a heavily shielded (100 mm lead) room containing an array of six NaI(Tl) scintillation detectors, each 152 mm in diameter and 89 mm thick. In its standard configuration four of these detectors are above and two below a flat bed on which the subject lies. These are aligned along the mid-sagittal plane of the supine subject with a longitudinal separation of about 300 cm (Fig. 2).²⁵ This configuration invites calibration errors where the radioactive deposit is essentially confined to a specific



Fig. 2 Inside the Harwell Laboratory shielded, whole-body monitor showing the positions of the six NaI crystal γ -detectors that will be aligned above and below the supine subject.

anatomical region, as would be the case either following the inhalation of an insoluble radioactive aerosol or where radionuclide concentrates within a single organ *e.g.*, the liver or the kidneys. In the case of ^{26}Al this problem may be overcome in two ways. Firstly, whole body counts may be standardised to the known amount of administered radionuclide minus the amount excreted at different times after intake, until initial radionuclide redistribution is complete—a protracted procedure. Alternatively, the detector arrangement can be modified to simulate the response of a 54-crystal array that is insensitive to changes in the position of the radionuclide within the body.²⁴ To achieve this an initial γ -ray spectrum may be recorded in the standard arrangement, followed by eight additional spectra in each of which all detectors are displaced longitudinally and/or laterally, relative to their normal locations, by 150 mm in each direction. This system has the additional advantage that calibration may be achieved using a simple standardised point source in several locations in the mid-coronal plane of a water-filled elliptical cylinder.

When the whole-body monitor is used a γ -ray spectrum is collected that comprises three components: a counter background response, derived mainly from natural sources of ambient γ -radiation; a response associated with the body's normal radioactivity, principally due to ^{40}K , which emits 1.46 MeV photons; a response derived from the ^{26}Al tracer (principally photons at 0.511 and 1.82 MeV). It follows that to derive the content of ^{26}Al present on each occasion it is necessary to deconvolute the spectrum. To achieve this, reference spectra are provided for the three constituents and the proportions of each are calculated such that, when combined, they provide the best least-squares fit to the subject's spectrum. The reference spectrum for counter background can be derived by recording the response of the counter in the presence of an inactive, water-filled phantom simulating the human form and providing comparable scattering and absorption of ambient radiation. For the response to normal body radioactivity a spectrum may be collected from the subject prior to exposure to ^{26}Al tracer and that for the tracer may be collected using standardised point sources of ^{26}Al tracer.

It can be seen from Table 1 below that the detection limit for ^{26}Al in the human body using a whole-body monitor is about 5–10 Bq, depending upon its distribution within tissues and organs. In contrast, much lower detection limits, down to about 0.1 Bq per sample, may be achieved by counting the ^{26}Al , 511 keV annihilation quanta under favourable counting geometry and using co-incidence counting techniques. This technique may be used to determine the ^{26}Al -content of standardised small samples counted using reproducible geometry. Such samples are commonly processed *e.g.*, ashed urine and faeces samples, or are unprocessed liquids *e.g.*, blood. The counting system deployed at Harwell comprised two NaI(Tl) crystal scintillation detectors operating in coincidence. Each crystal was 152 mm in diameter and 89 mm in thickness. They were located with their faces horizontally parallel at a separation of 16 mm. For counting, samples were loaded into Petri dishes and these were then accurately and reproducibly positioned between the crystal detectors. Standardisation was achieved using a Petri dish, reference source consisting

Table 1 The detection limits achievable using different methods to determine ^{26}Al

Method	Detected entity	Detection limit/Bq
Whole-body monitor	1.82 MeV ^{26}Al γ -emission	5–10
Coincidence counting	511 keV β^+ annihilation quanta	0.1
Accelerator mass spectrometry	Accelerated Al ions	10^{-8} (10^{-17} g)

of a known activity of ^{26}Al suspended in gelatine. The response to coincident 511 keV annihilation quanta achieved was $0.11 \text{ counts s}^{-1} \text{ Bq}^{-1}$ above a background of $0.012 \text{ counts s}^{-1}$. This low background is achieved because only simultaneous events in both detectors are counted, but most background events will occur in one detector only. In contrast, when the ^{26}Al positron is annihilated it is unlikely that only one of the two 511 keV quanta released will be detected.

Despite the superior detection limits achieved using coincidence counting, experience has shown that ^{26}Al levels in excreta and blood fall so rapidly following intake that they rapidly become uncountable. To determine the levels of ^{26}Al in such samples accelerator mass spectrometry (AMS) is deployed. Under favourable conditions this method has been used to detect as little as 10^{-18} g of the nuclide in geological samples, but for biological samples the detection limit is higher. The application of AMS to the measurement of ^{26}Al in biological samples has been described by Elmore,²⁶ Barker *et al.*,²⁷ Day *et al.*,²⁸ and by Flack and Elmore.²⁹ The method was developed from earlier applications of AMS for the measurement of the isotope in mineralogical specimens—where its presence is used to determine the time that surface rocks have been exposed to cosmic radiation. Unlike radiometric methods that detect the radiations produced when radionuclides decay, AMS—like other mass spectrometric methods—relies upon the unambiguous detection of ions of a specified mass-to-charge (m/z) ratio. AMS is employed to measure the abundance of ions of different atomic mass in samples that are commonly no larger than 1 mg in weight. In the case of aluminium, AMS is used to measure the relative frequency of ^{26}Al and ^{27}Al in samples. Where the amount of ^{27}Al in the sample is known then knowledge of this ratio allows the calculation of the mass of ^{26}Al present. It follows that in order to calculate the amount of ^{26}Al in a sample, a known quantity of ^{27}Al is added and the ratio measured. For the purposes of most studies the amount of stable spike added to each sample is 1 mg. This amount is much larger than the amount of endogenous aluminium present in the sample, such that the latter can be ignored for the purposes of ^{26}Al determination. This is true for the vast majority of biological samples, which normally contain very low levels of aluminium. Where samples are suspected to contain larger amounts of endogenous aluminium, these levels must be determined separately using atomic absorption spectrometry (most commonly, graphite-furnace atomic absorption spectrometry, GF-AAS) prior to AMS.

Compared to other mass spectrometry methods AMS has the advantage that it eliminates confounding molecular species with the same mass as the isotope ions being detected. For example, inductively coupled plasma mass spectrometry (ICP-MS) results are confounded by the difficulty in separating ^{26}Al ions from those of ^{26}Mg and those produced from molecular species of the same mass, such as $^{13}\text{C}_2$, $^{25}\text{Mg}^1\text{H}$ and $^{12}\text{C}_2^1\text{H}_2$. These problems are particularly important when measuring low masses of tracer aluminium and effectively preclude the use of ICP-MS (and other MS methods, such as thermal ionisation MS) for ultra-trace determinations of ^{26}Al . During AMS, magnesium interference is eliminated in the ion source of the accelerator. A variety of negative metal and metal oxide ions are produced within the ion source by sputtering with caesium ions. In the case of aluminium the Al^- ion is selected for acceleration and measurement since the corresponding Mg^- is unstable and unavailable for acceleration. In addition, potentially confounding molecular species are disaggregated and broken down, by a process known as Coulomb explosion, in the region of the high voltage terminal of the accelerator. This occurs when the accelerated ion beam intercepts and traverses a thin carbon foil. At the approximate detection limit of the technique, and assuming that 1 mg of ^{27}Al tracer has been added to the sample the measured ratio of $^{26}\text{Al}:$ ^{27}Al will be about 1×10^{-14} .



Fig. 3 The accelerator building at the Department of Nuclear Physics building at the Australian National University, Canberra, showing the tower containing the 14UD tandem accelerator.

AMS for ^{26}Al , in biological specimens, is currently undertaken at a small number of global locations. Of these, the laboratories with the most experience are those at the Australian National University, Canberra, Australia (Fig. 3) and at Purdue University, Lafayette, USA. At both laboratories the accelerator employed is a tandem Van de Graff accelerator. The layout of the Australian accelerator is shown in Fig. 4. For ^{26}Al determinations by AMS, a specimen comprising equal volumes of aluminium oxide and powdered silver is sputtered with caesium ions within the ion source. A

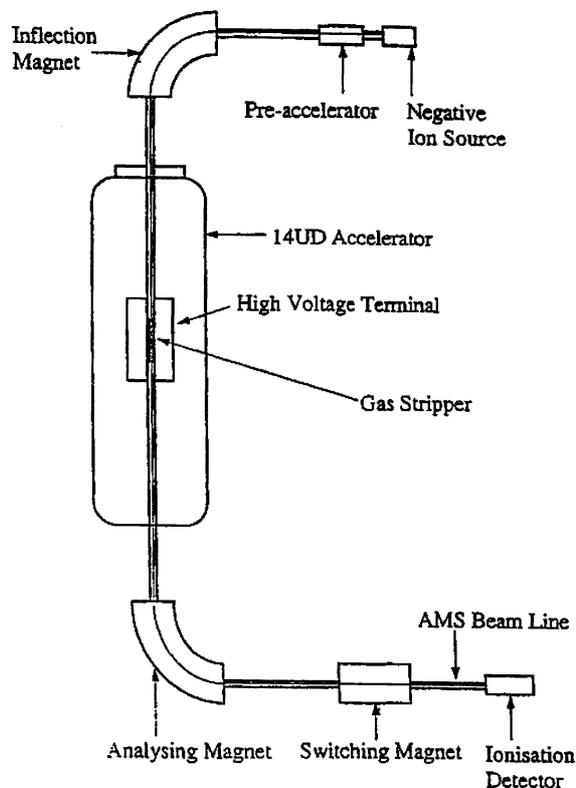


Fig. 4 Diagram showing the layout of the 14UD accelerator used for AMS at the Department of Nuclear Physics, the Australian National University, Canberra. The pre-accelerator accelerates Al^- to 100 kV and the high voltage terminal operates at about 11 MV. $^{26}\text{Al}^{7+}$ ions are detected in the ionisation detector. $^{27}\text{Al}^{7+}$ ions are detected in a Faraday cup (not shown) that intercepts the beam before the ionisation detector. Note: for ^{26}Al determinations a carbon foil, rather than gas stripper, is employed and a velocity filter is incorporated into the AMS beam line.

beam of negative ions, including Al^- ions, at 5 kV is produced and this is accelerated to about 100 kV before entering the first inflection magnet. Within this magnet ions with a mass of either 26 or 27 are selected for passage by varying the current supplied to the magnet, which changes the strength of the magnetic field applied within the range 2.9–3.0 kG. Ions of non-selected masses fail to negotiate the magnet's 90° bend and are removed from the beam. The selected Al^- ions (and other ions with the same mass-to-charge ratio) are then accelerated towards the positive high voltage terminal (~ 11 MV) where they interact with a thin carbon foil ($3 \mu\text{g cm}^{-2}$). At this point any molecular species with the same m/z ratio as the selected ion are eliminated and the selected aluminium ions are stripped of some electrons producing positive ions. A spectrum of ions is produced, e.g., Al^{6+} , Al^{7+} and Al^{8+} . These are then repelled by the positive charge at the high voltage terminal and are further accelerated towards a second 90° analysing magnet—hence tandem acceleration. This magnet operates at a fixed field strength of ~ 7.7 kG and selectively allows the passage of Al^{7+} ions. These ions are then deflected, by a switching magnet (~ 9.9 kG), through a Wein filter, which reduces background by allowing only the passage of ions with a given velocity, along the AMS beam line to a detector where, in the case of ^{26}Al , they are counted. For ^{27}Al the beam current is measured using a Faraday cup placed in the beam line immediately before the ion detector. The ratio ^{27}Al to ^{26}Al is calculated. Typical count times are a few minutes for each isotope, although, much longer count times are possible. The precision of the ^{26}Al mass determinations made using this method is *circa* 2%.

The availability of ^{26}Al as a tracer and of AMS for its detection has significantly enhanced the ability of workers to undertake biokinetic and bioavailability studies of aluminium. It follows, that since 1990, our knowledge of the behaviour of aluminium in biological systems has rapidly increased.

In all cases where AMS is employed to measure low-levels of ^{26}Al in biological samples, chemical processes must be employed to isolate and concentrate the aluminium (both ^{27}Al tracer and ^{26}Al) present in the samples. Such techniques have been described, in detail, by Day and his co-workers²⁷ and others have been described, in outline, by Flack and Elmore.²⁹ In general, the selectivity of the AMS process is such that the presence of small amounts of other metals in the aluminium oxide used to prepare the ion sources usually causes few problems, however, the presence of relatively massive amounts of some elements (such as boron- $^{10}\text{B}^{16}\text{O}$) can cause problems. It follows that the best AMS results are obtained when aluminium has been selectively removed from the tissues to be analysed using either the selective precipitation of aluminium salts from solution or their concentration using cation/anion ion exchange resins. Typical protocols are outlined in Fig. 5, which has been developed from the publication of Flack and Elmore.²⁹

5 Aluminium deposition in body tissues and organs

It is normal for aluminium to enter the body either from ingested material in the intestinal tract or following the dissolution of an inhaled deposit containing the metal.³⁰ The relative importance of these two routes of intake is variable. For example, in aluminium workers it is thought that inhalation will be most important (e.g., ref. 31), but members of the general population—except under exceptional circumstances—derive most of their aluminium body burdens from ingested materials.^{32–33} Whichever, the aluminium will enter the blood stream in a soluble form. It will then become distributed around the body and may be either deposited in organs and tissues or excreted. The extent to which metals deposit in different tissues depends upon their affinity for the metal. Similarly, its speciation within blood and other tissue

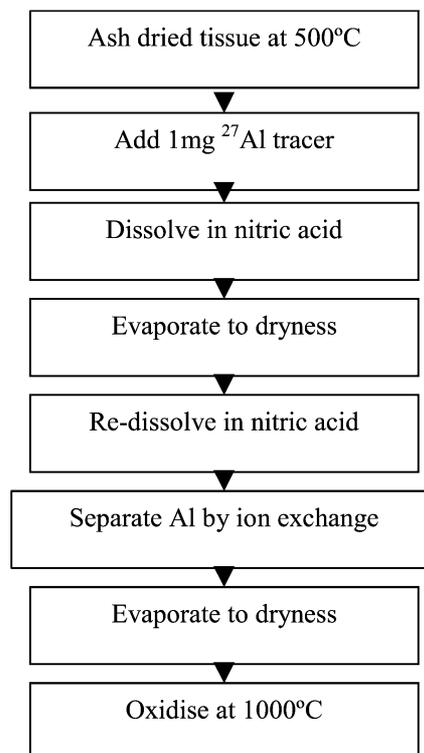


Fig. 5 Typical procedures employed in the chemical preparation of biological samples for AMS (adapted from ref. 29).

fluids will depend upon its chemistry and affinity for tissue components.

5.1 Aluminium in blood

Most of the metals in groups I and II of the periodic table—including the monovalent ions of sodium and potassium and the divalent ions of magnesium and calcium—are present in blood as ionic species, although, complexes with low molecular weight organic molecules, such as citrate and bicarbonate, are formed.¹⁸ The chemistry of aluminium and of many other polyvalent metals precludes such a speciation, since at physiological pH their ions normally undergo hydrolysis and form polymeric species, e.g., $\text{Al}(\text{OH})_3$. However, such hydrolytic reactions are modified by the presence, in blood, of complexing anions and/or other binding species and in the case of aluminium these normally completely inhibit hydrolysis. Consequently, in blood and tissue fluids aluminium ions are found complexed with organic molecules. As for many other metals¹⁸ a common site of complexation is the iron transport protein transferrin. Chromatographic separation studies,^{2,34} utilising endogenous ^{27}Al and equilibrium speciation models suggest that about 80–94% of aluminium will be associated with this protein.^{6,35–36}

A study conducted by Day and Priest,^{27–28,37} using blood from a subject that had been injected with ^{26}Al citrate, showed 99% of the blood-aluminium to be present in plasma at 1 h post-injection. Of this, 95% was protein-associated, with 80% bound to transferrin (molecular mass 72 kDa), 10% to albumin and 5% to a low molecular weight protein fraction. At the later time of 880 days post-injection the fraction of aluminium present in the plasma fraction had fallen to 86%, with the remainder associated with erythrocytes (red blood cells). This fraction is close to the 89% reported by Bowen,³⁸ who measured stable ^{27}Al , by neutron activation analysis.

It is widely accepted that aluminium binds to the iron binding sites on the transferrin molecule³⁹ and, like iron, aluminium requires the presence of bicarbonate to bind. In doing so it does not replace iron, since the strength of the aluminium bond is

known to be much weaker than that of iron. Nevertheless, according to Birchall and Chappel¹¹ it is sufficiently strong to inhibit the binding of aluminium to other ligands present in plasma, including OH⁻, phosphate and silicic acid. According to these authors the binding affinity of iron to transferrin is about 10² times greater than that of aluminium to this molecule. Martin⁷ suggests that the difference is hugely greater (10⁹-association constant for iron 10²² M⁻¹ and for aluminium 10¹⁴ M⁻¹) and that aluminium binds to the molecule only because plasma transferrin is about 50 μM deficient in iron, *i.e.*, unsaturated. Van de Vyver³⁹ assumes that 40 μM L⁻¹ is available for binding to aluminium and showed that blood transferrin is able to bind about 1 mg Al³⁺ L⁻¹-this mass is much larger than any conceivable blood aluminium loading. Martin⁷ also suggests that, based on consideration of stability constants, serum albumins are unlikely to be a significant binding site for plasma aluminium and that this element will bind more strongly to both transferrin and citrate. Consequently, it is predicted that while transferrin will be the predominant high molecular binding site for aluminium in blood, citrate will be the predominate small molecule Al³⁺ binder.⁷ However, it should be recognised that Martin's calculations take no account of the kinetics of aluminium binding to different binding sites. In practice kinetic considerations may be as important as thermodynamic considerations in determining speciation. Calculations by Harris (cited by Yokel and McNamara⁶) suggest that the percentage of aluminium species associated with transferrin, citrate, free hydroxide and phosphate in the blood are 91%, 7-8%, <1% and <1%, respectively. Citrate occurs at a level of 0.1 mM in blood plasma and normally occurs as the Ca²⁺ complex. Aluminium easily replaces calcium (and magnesium) in such complexes. Unlike protein-bound aluminium, that bound to citrate (and other small molecular weight species) is filterable within the kidneys.

A consequence of the presence of competitive binding sites is that aluminium will equilibrate between high and low molecular weight fractions in blood and in adjacent tissue fluids. The latter is important since experiments with ²⁶Al and ⁶⁷Ga suggested the rapid loss of about half of blood aluminium, but not gallium, to extravascular tissue fluids.¹⁹ Subsequently, since 45-75% of injected ²⁶Al is excreted in the first 24 h post-injection,⁴⁰ much of that lost must have migrated back into the blood prior to removal by the kidneys. This is possible since there is a net loss of blood fluids to tissue fluids in the arterial regions of capillaries where the hydrostatic blood pressure exceeds the osmotic pressure produced by proteins (particularly albumin) dissolved in the blood. Aluminium attached to low molecular weight species will then leave the capillaries with the serum. In the venous regions of capillaries the osmotic pressure of the "concentrated blood" exceeds the reduced hydrostatic pressure resulting in a net flow of tissue fluids back into capillaries. In these regions aluminium complexed to citrate may be expected to return to blood. As a consequence of these transfers aluminium will rapidly equilibrate between blood proteins and low molecular weight species in blood and tissue fluids. Given the low concentration of dissolved proteins in tissue fluids most aluminium will be complexed to citrate. For example, the percentage of aluminium associated with citrate extracellular fluids in the brain is 90%.⁶ The loss of aluminium, but not gallium, from the blood was interpreted by Priest and his co-workers to result from the stronger binding of gallium to blood proteins. Such a suggestion is also consistent with the much longer retention half time of ⁶⁷Ga in the blood of the subject. Finally, it is possible that, since the collection of blood isolates it from its environment, including surrounding tissue fluids, speciation studies on isolated blood samples may provide an unreliable indicator of normal *in vivo* aluminium speciation. Nevertheless, it is clear that compared with many other polyvalent cations (including actinides, lanthanides and

gallium⁴¹⁻⁴²), aluminium binds rather weakly to blood components maximising its potential both for transfer to binding sites within tissues and to urine, *via* the kidneys. For example, whereas 16% of plutonium, which binds strongly to plasma proteins, remains in human blood at 24 h post-injection (and even after 15 days about 1% remains in blood)⁴³ and 10% of injected ⁶⁷Ga remains in blood at 24 h, only ~0.5% of ²⁶Al remains at this time.^{19,22}

The studies of Fifield,⁴⁴ Priest *et al.*⁴⁵ and Bowen³⁸ suggested that about 10% of blood aluminium is associated with erythrocytes. The association of aluminium with these cells has been studied in more detail by Day and his co-workers⁴⁴ using a volunteer fed 1 μg of ²⁶Al in a citrate-containing drink. Erythrocyte and plasma ²⁶Al levels were then measured for times up to 3 months post-administration (Fig. 6). Blood plasma levels peaked at about 1 h after administration and then fell rapidly, initially with a retention half time of a few hours and subsequently with a progressively slower half time. In contrast, the peak erythrocyte aluminium level was noted 1.1 days after the corresponding plasma peak. The retention half-time of ²⁶Al in the erythrocyte fraction was 1-2 days for the first few days and then 20-40 days thereafter. It follows that the retention time of aluminium in the red blood cell fraction is much longer than in the plasma, but shorter than the life expectancy of individual red blood cells (120 days). Day⁴⁴ speculates that the red blood cells are likely to have incorporated aluminium by slow, direct exchange processes in blood, rather than during erythrocyte formation in the bone marrow.

It is not known whether ²⁶Al is incorporated within the cells or is merely bound to transferrin receptors on the cell membrane. Whichever, aluminium concentrations in erythrocytes are likely to be much more stable than the plasma concentrations, which are subject to huge daily variations resulting from both variations in dietary intake and from its rapid elimination from plasma by the kidneys. Consequently, it is possible that aluminium levels in blood erythrocytes, unlike levels in plasma and urine, could prove to be a useful indicator of the average size of aluminium intakes over periods of weeks and months. For this reason, the possibility of using red blood cell aluminium for human occupational bioassay is currently being investigated. Similarly, such measurements could be used as an indicator of aluminium exposure and intake for epidemiological studies of possible aluminium toxicity.

All studies conducted, to date, have shown that the rate of aluminium clearance from blood decreases with increasing time after intake. Consequently, retention times for this element in blood cannot be adequately expressed by a single exponential function of time. Priest *et al.*¹⁹ have shown that, following the

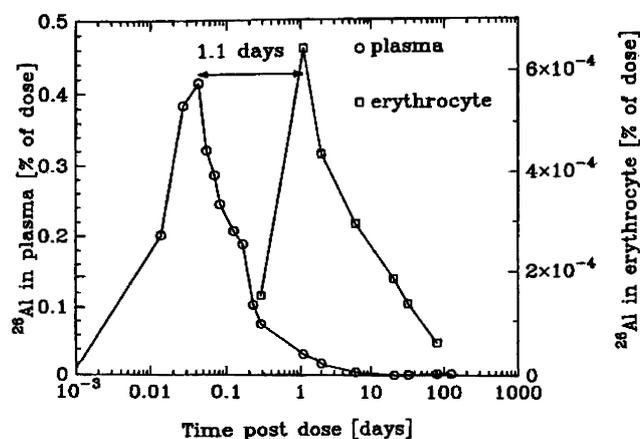
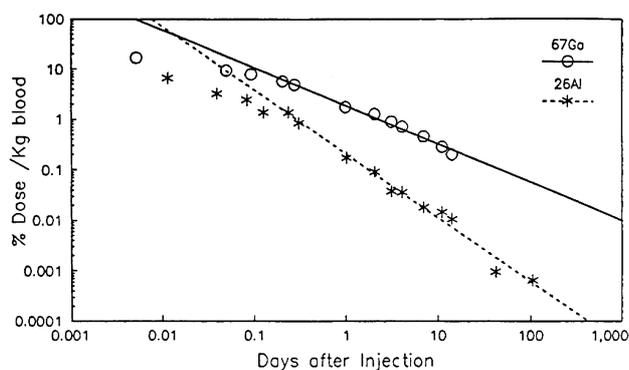


Fig. 6 ²⁶Al measured in plasma and red blood cells following the ingestion of the isotope as aluminium citrate. The uptake by red blood cells is delayed by about 1 day (from ref. 44).



67Ga results by gamma spec.
26Al results by Acc. Mass spec.

Fig. 7 Power function curves fitted for the clearance of ^{26}Al and ^{67}Ga from blood by linear regression. The much faster elimination of aluminium is evident. (From ref. 22).

intravenous injection into a male subject, for both ^{26}Al and ^{67}Ga the clearance pattern of each radionuclide from blood, for times after 0.2 days post-injection and until about 1200 days could be adequately expressed by single power functions of time (Fig. 7). For ^{67}Ga the concentration (A_{Ga}) of radionuclide in the blood stream of the subject (expressed as % injected activity kg^{-1} blood) at any time, t days, after injection could be described by the following formula:

$$A_{\text{Ga}} = 1.84 t^{-0.751}$$

For ^{26}Al (A_{Al}) the corresponding equation was:

$$A_{\text{Al}} = 0.206 t^{-1.250} \text{ (for times to 1200 post-injection)}$$

or

$$A_{\text{Al}} = 0.181 t^{-0.993} \text{ (for times to 5 days post-injection)}$$

In a second experiment,^{40,46} inter-subject variations in early aluminium excretion were measured in a further six male volunteers injected with a smaller mass of ^{26}Al citrate. Measurements of blood aluminium, made at times up to 5 days post-injection showed that the early loss of ^{26}Al from the blood of all subjects was qualitatively similar, but an approximate order of magnitude difference was found between absolute levels. For example, at 5 days post-injection only $0.02\% \text{ L}^{-1}$ of the injected ^{26}Al remained in the blood of one subject while $0.16\% \text{ L}^{-1}$ remained in the blood of another. At 1 day the fraction retained varied from 0.1 to $0.6\% \text{ L}^{-1}$. The fitted function for the mean retention of ^{26}Al for times t days (between 0.4 and 5 days) was:

$$A_{\text{Al(early)}} = 0.37 t^{-0.90}$$

The corresponding power functions showed less variation and ranged within the band $t^{-0.9}$ to $t^{-1.1}$ for all but one anomalous subject (for which the indicated rate of loss of ^{26}Al from the blood was much slower $t^{-0.6}$). In comparison, the rate of loss of ^{26}Al from blood during the first few days after the ingestion of ^{26}Al citrate was slower, $t^{-0.82}$ (calculated from the data of Day⁴⁴). This is consistent with a tail of protracted intake of aluminium from the gut, as indicated by the results of other bioavailability experiments.⁴⁷⁻⁴⁸

5.2. Tissue distribution of aluminium

Whereas, some metal ions quickly become widely and rather uniformly distributed throughout a wide range of tissues (*e.g.*, sodium and potassium) most metal ions are deposited to a much greater extent than the average in a few tissues and

organs. In this context the most important tissues are the skeleton, liver and kidneys, although, the fraction of the total body burden deposited in any of these is very variable and depends upon many factors, including the element concerned and the age, sex and species of animal. Nevertheless, the major deposition site for many divalent and other polyvalent metals is the skeleton, and for this reason these metals are often referred to as bone-seekers.

Two main types of bone-seeker can be identified—these are reviewed elsewhere.¹⁸ Firstly, the divalent alkaline earth elements, including strontium, barium and radium, which all deposit in the skeleton by substituting for calcium on bone surfaces and within the bone mineral crystal. This group includes the divalent uranyl ion UO_2^{2+} that in its hexavalent state fails to behave like other polyvalent actinides. All the metals in this category fail to be deposited and retained within other tissues to any significant extent—a feature of their failure to ride “piggy-back” on the metabolic pathways of iron and, more generally, to bind to proteins. Consequently, alkaline earth elements and their analogues not only deposit almost exclusively in the skeleton, but they are also rapidly cleared from the blood.

The second main group of metals comprises most transition metals, the lanthanides and the actinides (except uranium). These metals are polyvalent and bind strongly to proteins including those involved in the metabolism of iron. Consequently, they may deposit at sites of iron utilisation, deposition and storage such as the liver, spleen and bone marrow. These metals are also deposited in the skeleton, where they are thought to become attached to the organic components (particularly phosphoproteins) of the extracellular bone matrix rather than to the calcium hydroxyapatite bone mineral. It follows that the skeleton and soft tissues, notably the liver, provide competitive binding sites for polyvalent metals. The extent, to which any metal is deposited in the skeleton or soft tissues, at least in part, seems to be a function of the size of the metal ion.

Research conducted at the Lawrence Berkeley Laboratory, by Durbin and her associates soon after the last world war, has shown that, in rats, there is a progressive shift in the initial distribution pattern of these elements, which appears to be related to ion size.^{17,49} The actinide and lanthanide elements show a contraction in ionic size with increasing atomic number. This decrease—known as actinide and lanthanide contraction, respectively—accompanies the filling of the four electron shells of the atom and is associated with increased hydrolysis of the aqueous ions, with a decrease in the solubility of compounds and with a greater stability of complex ions.¹⁷ The relationship found is shown in Fig. 8. It can be seen that liver deposition is high and skeletal deposition is lower for the lighter, trivalent lanthanides of larger ionic size, ranging from cerium ($r = 103 \text{ pm}$) to gadolinium ($r = 94 \text{ pm}$); whereas for the heavier smaller lanthanide ions, spanning terbium ($r = 92 \text{ pm}$) to lutetium ($r = 85 \text{ pm}$) skeletal deposition is high and liver deposition is lower. Durbin showed that qualitatively all the trivalent metals tested behaved similarly in rats. Why ion size is so important is unclear. However, as binding to plasma proteins has been shown to be dependent on ionic size^{17,50-52} and as the uptake of non-colloidal cations by the liver involves the active transport of metals across cell membranes, which is also likely to be an ion size-dependent process, Durbin speculated that these represent the distribution determining processes. In contrast, the uptake of metals by bone surfaces has been described as a less specific, passive process, independent of ion size.⁵³ Consequently, it may be much less important in determining the final deposition of a contaminating metal. As yet data for the stable, group IIIa, trivalent metals aluminium, gallium and indium have not been collected, but it would be interesting to speculate that a similar relationship exists for these. If so, on the basis of their ion sizes, aluminium with an

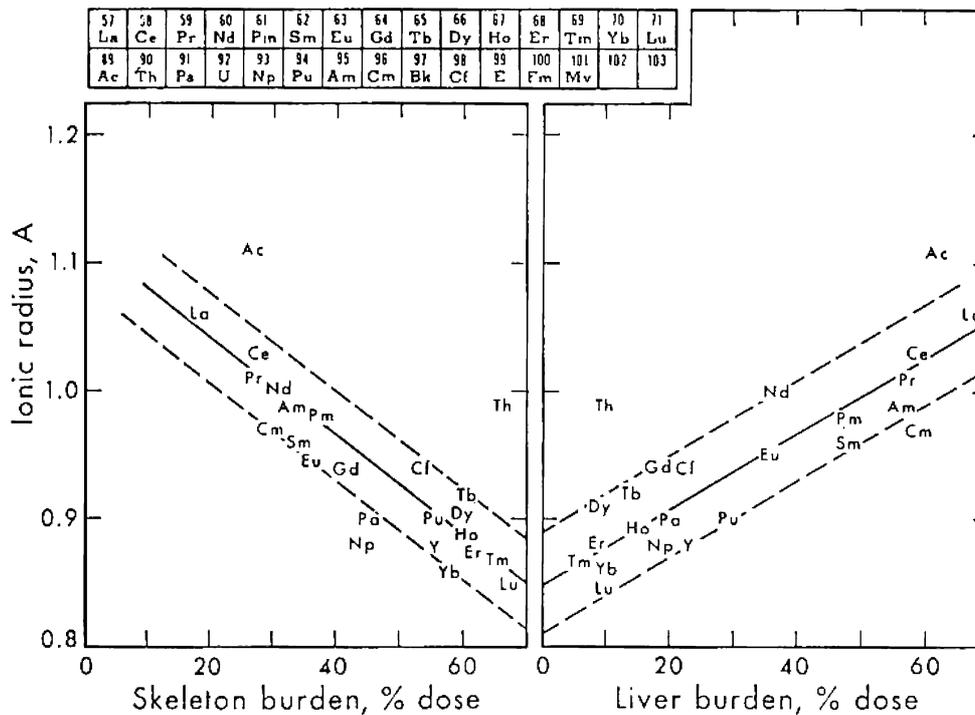


Fig. 8 Graphs showing the relationship between ion size and metal deposition in the skeleton and liver of rats. It can be seen that large ions preferentially deposit in the liver (from ref. 17).

ion size of 54 pm, and gallium with an ion size of 62 pm, should not deposit to any significant extent in the rat liver.

The situation in man is not clear since observations suggest that for the actinides and lanthanides examined more metal deposits in the liver than would be predicted based on the rat data. Nevertheless, the predictions would be consistent with the general trend of observations—namely that gallium deposits in the liver and skeleton,¹⁹ but that aluminium deposits in the liver are much less important.

Published data concerning the aluminium content of organs and tissues were collated by the ICRP for Reference Man—a summary of these is shown in Table 2. This Table excludes data for the lung and lymph nodes, which contain un-dissolved particulate-aluminium deposits that should not be regarded as part of the systemic body burden of the metal. Of the remaining tissues listed by the ICRP only the liver, connective tissues, skin and skeleton seem to concentrate more aluminium than the average for the body as a whole. Furthermore, it should be noted that the data reported for skin is of questionable veracity given the likelihood (rather than possibility) of skin contamination by unabsorbed surface deposits of contaminating environmental aluminium. In other tissues such as the muscle and central nervous system the reported tissue concentrations of aluminium are much lower than those in the skeleton and liver, at about one third of the average concentration for the body as a whole. Overall, the data presented in this table

suggests that only the skeleton concentrates significant quantities of aluminium, with this organ containing about 54% (or even more if the skin content is disregarded) of the total body content.

Subsequently, further data on aluminium levels in tissues have been collated by the International Programme on Chemical Safety.⁵⁴ For its publication tissue content data were reviewed for both normal members of the public and for those occupationally exposed to aluminium. A total of 13 publications were listed that had been published after the publication of Reference Man in 1975. Of these, about 6 provide information for a range of tissues. The findings of these studies are listed in Table 3. It can be seen that for normal subjects the levels reported are broadly consistent with those collated by the ICRP. However, for some workers the levels are at variance with those expected. For example, Teraoka⁵⁵ presented data for a stonemason that had died of silicosis. High concentrations of aluminium were found in the liver, but none in the skeleton. For this subject the concentrations of aluminium in the lung were 2000 mg kg⁻¹ and it is likely that the high liver content resulted from the migration of macrophages (scavenger cells) containing aluminium, as particles, from the lung to the liver, *via* regional lymph nodes. Such migration has been demonstrated in miners exposed to coal dust and has been suggested to occur in radiation workers that have inhaled insoluble, radioactive particles.¹⁸ In the same way the high liver

Table 2 Aluminium content of different organs and tissues (adapted from ICRP, 1975)—all tissues containing >1% of total deposit

Tissue	Mass/g	Al content/g	Content/g	Concentration factor	%
Adipose	1.25E+04	1.90E-03	1.52E-07	0.25	5
Blood and vessels	4.42E+03	1.44E-03	3.26E-07	0.54	4
Connective tissues	1.60E+03	1.10E-03	6.88E-07	1.14	3
Central nervous system	1.43E+03	3.40E-04	2.38E-07	0.39	1
GI tract	1.20E+03	6.30E-04	5.25E-07	0.87	2
Liver	1.80E+03	1.20E-03	6.67E-07	1.11	3
Muscle	2.80E+04	5.60E-03	2.00E-07	0.33	14
Skeleton	1.00E+04	2.10E-02	2.10E-06	3.48	54
Skin	2.60E+03	5.20E-03	2.00E-06	3.32	13

Table 3 Recently reported tissue concentrations of aluminium/mg kg^{-1a}

Subject	Bone	Muscle	Kidney	Liver	Brain
Normal males ⁵⁵	<15d	nd	11d	19d	nd
Stonemason ⁵⁵	nd	nd	16d	130d	nm
Ball-mill aluminium worker ⁵⁶	30	nd	nd	90	5
Normal controls ⁶⁰	10.6	23.6	17.5	15.8	11.9
Dialysis encephalopathy ⁶⁰	272.7	13.8	156.5	610.2	66.1
Healthy controls ⁶¹	3.3d	1.2d	nm	4.0d	2.2d
Dialysis encephalopathy ⁶¹	281d	15d	nm	301d	24.5d
Normal controls ⁶²	2.4d	nd	nd	nd	nd
Al welders ⁵⁷	18–29d	nd	nd	nd	nd

^ad = mg kg⁻¹ dry weight; nm = not measured; nd = none detected.

content in the aluminium ball-mill worker⁵⁶ is consistent with a correspondingly high lung burden (340–430 mg kg⁻¹ wet weight). It follows that the data generated for aluminium workers may be difficult to interpret where the workers have been exposed to insoluble aluminium-containing aerosols. In contrast, the situation may be simpler where worker exposures are to inhaled aluminium fume, which because of the small particle size of condensation fumes is likely to be soluble. Elinder *et al.*⁵⁷ report the aluminium distribution in aluminium welders. In this study measurable aluminium deposits were only found in the skeleton. Despite this, the measurements of Triger and Singh⁵⁸ suggest that the normal level of aluminium in the liver, in the UK, is $5.05 \pm 1.24 \mu\text{g g}^{-1}$. Interestingly, levels in the livers of patients with cirrhosis were lower ($1.73 \pm 0.87 \mu\text{g g}^{-1}$). Similarly, the elevated levels of aluminium excreted, *via* the biliary route, in the faeces of subjects given the chelating agent desferrioximine (DFO), are consistent with significant levels of aluminium in this organ.⁵⁹

The data generated for normal subjects also shows considerable variability between studies and is, therefore, difficult to interpret. To a large extent, this reflects the previously discussed difficulties in measuring very small amounts of aluminium in samples that may easily become contaminated with environmental aluminium. That this is likely can be deduced from the mismatch that exists between body burdens extrapolated from such tissue concentration data and body burdens calculated using biokinetic data.¹⁹ It follows that the best indication of the relative affinity of different organs and tissues for aluminium may be better deduced from the distribution data available for dialysis encephalopathy patients where tissue contents are high and the likely effects of sample contamination small. The data produced for such patients by Flendrig *et al.*⁶⁰ suggests a body burden of about 2.7 g aluminium in the skeleton, about 1 g in the liver, about 400 mg in muscle and about 100 mg in the brain. These relative concentrations are not inconsistent with those measured in rabbits following the intra-muscular injection of ²⁶Al-labelled adjuvants.⁶³ In these animals ²⁶Al concentrations were five to ten times lower in the liver and spleen than in the kidney. In turn these concentrations were about ten times higher than those measured in the heart, lymph nodes and brain. The data of Alfrey,⁶¹ after corrections for dry weight, suggests a different distribution with a smaller fraction of body aluminium in the liver (skeleton ~1.4 g; liver ~54 mg, muscle ~42 mg; brain 3 mg). However, the uncertainty associated with either extrapolation is large.

In addition to the above, it is possible that underlying disease may have affected the measured distributions. For example, it is possible that liver deposits of aluminium are accumulated as a result of the breakdown of aluminium-contaminated red blood cells and the levels of red blood cell aluminium in renal dialysis patients will be abnormally high due to failed filtration within the kidneys. If so, then it is possible that some of the aluminium in the liver of normal subjects represents secondary

deposits accumulated as a result of blood breakdown, rather than to the accumulation of metal from plasma.

The data collected by Flendrig *et al.*⁶⁰ and by Alfrey⁶¹ also seem to show relatively high levels of aluminium in the diseased kidneys of the dialysed renal failure patients, in the kidneys of normal controls and lower, but still elevated levels in the brains of both groups. With respect to the latter, these results show that some aluminium is able to penetrate the blood brain barrier.⁶⁴ Moreover, the ICRP data and that of Alfrey⁶¹ are both consistent with concentrations of aluminium in the brain that are about ten times lower than those in the skeleton. Animal studies with ²⁶Al suggest a higher factor of about 100.⁶ In contrast, the kidney results tell us little, since in the normal control tissues all aluminium concentrations were similar—consistent with each becoming contaminated with a similar level of environmental aluminium—and in the dialysis patients' aluminium retention may be associated with the inability of the kidneys to excrete this metal.

5.3 Skeletal aluminium deposits

Skeletal deposits of aluminium are important for three reasons. Firstly, such deposits are a reservoir for the long-term retention of aluminium within the body. Secondly, knowledge of the pattern of deposition and retention of aluminium in the skeleton is important for the development of mathematical models of aluminium biokinetics. Finally, if sufficient aluminium accumulates then the bone formation process is disrupted and a fracturing osteomalacia, AIBD (Aluminium-Induced Bone Disease) results.

AIBD is frequent only in renal patients, most of who were dialysed with fluids containing aluminium, but seems to occur at lower body burdens than encephalopathy. That this is so is indicated by the absence of dementia, but presence of bone disease in patients without kidney disease receiving long-term treatment with parenteral nutrition fluids that were heavily contaminated with aluminium.⁶⁵ Similarly, the results of a further two studies⁶⁶ showed that 25–30% of dialysed uremic patients having no parenteral exposure to aluminium and no dementia, but routinely receiving oral phosphate binders containing aluminium, had aluminium associated bone disease.

Within the skeleton aluminium is deposited on bone surfaces.^{39,67} Such deposits have been visualised using a variety of different histochemical staining methods: aluminon method; Morin method; naphthochrome green B method; solochrome blue-black R method; solochrome azurine method; acid solochrome cyanine method (these have been reviewed by Verbueken *et al.* and by Van de Vyver,^{68–69}) and subsequently confirmed by microanalytical techniques such as LAMMA^{68,70} and EPXMA.⁷¹ Aluminium deposits at the mineralised bone front on both growing and resting bone surfaces—often in association with other metals such as iron.^{69,72} In this way the pattern of aluminium deposition is essentially the same as that found for a wide range of other polyvalent metals.¹⁸

Three main mechanisms of aluminium deposition are possible. Firstly, the metal may become attached to bone surfaces by heterionic exchange with calcium that is present either in the bone mineral crystal or, more likely, in the hydration shell that surrounds each of these. This shell contains many entrapped ions, including those of calcium, phosphate, and even organic complexes such as citrate, and may also bind other ions such as those of radium and uranium.^{73–74} Ionic exchange within the hydration shell is likely to be a necessary precursor to exchange within the crystal matrix itself and is also likely to be easily reversed. Aluminium trapped in this way is, therefore, likely to be mobile and subject to back exchange—firstly with complexes in the tissue fluids close to bone surfaces and then with transferrin in plasma. Compared with exchange with calcium and phosphate in pre-existing mineral, it is likely that exchange within the hydrated layer surrounding each bone

mineral crystal will be much less specific allowing the entry of ions that on the basis of valency or size would normally be excluded from the hydroxyapatite crystal lattice.

In addition to the above, it is likely that aluminium will be co-precipitated with calcium at sites of bone mineral formation and incorporated into the structure of the developing hydroxyapatite crystal.⁷⁵ Aluminium trapped in the mineral lattice at the time of its formation at sites of bone growth and accretion may be expected to resist back exchange with tissue fluids and be relatively fixed.

Finally, aluminium may become attached to bone surfaces by complexation with components of the organic, rather than inorganic, bone matrix. It is to these components of bone that actinide metals, including trivalent americium, have been reported to bind.⁷⁶ Possible organic receptor molecules include the glycosaminoglycans, acid glycoproteins and acidic matrix proteins.^{77–78} Which of these are sites of aluminium complexation, if any, is unclear although studies have demonstrated the metal binding potential of phosphate groups, such as those present in nucleic acids and phosphoproteins.⁷⁹ Also, phosphoproteins have been shown, *in vitro*, to strongly bind lanthanum—a trivalent element with many characteristics in common with aluminium.⁸⁰ These molecules are concentrated at the bone osteoid interface, but are largely absent from un-mineralised osteoid. This may be important since it has been shown that most metals, *e.g.*, iron, aluminium, plutonium and uranium, do not deposit in bone osteoid, but rapidly migrate through it to deposit at the mineralising matrix front (commonly referred to as the *lamina limitans*).

Which of the above is most important for aluminium is unclear, but the presence of this metal at the *lamina limitans* is associated with inhibited mineralisation within the osteoid seam,⁸¹ leading to the vitamin D-resistant osteomalacia characteristic of AIBD (Fig. 9). *In vitro* tests have shown that aluminium ions have a concentration-dependent suppressive effect upon hydroxyapatite formation.⁷⁵ Also, the presence of aluminium citrate inhibits the growth of calcium phosphate crystals in saturated aqueous solutions (Blumenthal and Posner, 1984; Meyer and Thomas, 1982).^{82–83} These effects may be due to an inhibition of mineralisation produced by the precipitation of aluminium hydroxide.⁸⁴ Alternatively, effects of aluminium on the function of the bone matrix producing cells, the osteoblasts, may have produced the osteomalacia. High levels of aluminium have been shown to inhibit the replication of osteoblasts, damage collagen formation^{85–86} and inhibit the removal of pyrophosphate.⁶⁹

Despite the above, at lower, non-pathogenic, levels of aluminium contamination bone mineralisation proceeds normally—even on contaminated bone surfaces—and tetracycline studies consistently show the presence of stainable aluminium in areas

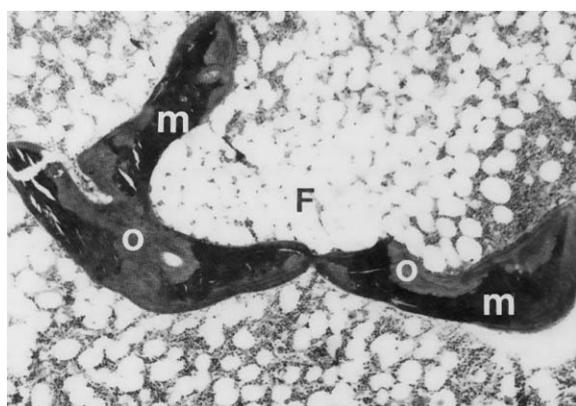


Fig. 9 The characteristic osteomalacia of aluminium-induced bone disease (AIBD). o = un-mineralised osteoid, m = mineralised bone, F = fatty bone marrow. (Photomicrograph by J. Humphreys, AEA Technology plc, Harwell).

of growing bone labelled with this fluorescent antibiotic.⁸⁷ It follows, that the majority of aluminium retained on growing bone surfaces can expect to become buried by the apposition of new bone onto the contaminated osteoid seam.

Eastwood *et al.*⁸⁸ showed buried lines of aluminium, stained using the solochrome azurine method, at depths of between 15 and 40 μm below the bone surface at 7–8 months after exposure to aluminium in drinking water at Camelford, UK. These buried lines were smooth, superimposed upon the normal lamellar structure of the bone and suggested no effect of the aluminium on bone metabolism. This is to be expected since the bulk level of aluminium in the bone samples assayed was within the normal range of 1.5–13.3 $\mu\text{g g}^{-1}$.⁸⁹ A logical consequence of the above is that all bone matrices may be expected to be contaminated with aluminium at some level—since aluminium is an ubiquitous component of tissue fluids and blood, but that due to the pattern of normal bone remodelling concentrations of this metal on bone surfaces may be higher. Overall, there is little reason to doubt that aluminium cycles within the skeleton in the same way as trivalent actinide and lanthanide elements. Moreover, the observation that aluminium deposits, which can be stained with solochrome azurine,⁹⁰ are present in marrow macrophages suggests that aluminium, like actinides, is retained in these cells as a result of bone resorption.

The cycle of heavy metals in the skeleton, as developed for plutonium, is shown in Fig. 10. This was developed, from autoradiographic evidence, to describe the behaviour of plutonium in the skeleton,¹⁸ but all the available evidence suggests that this model is appropriate for aluminium. The cycle comprises pathways that result from both bone growth and bone resorption and these have been used to model plutonium (and aluminium) retention in the skeleton (see section 7). If aluminium is substituted for plutonium then the first pathway shown in the figure represents the transfer of aluminium, bound to transferrin (or citrate), from the bloodstream, through tissue fluids, to bone surfaces where it becomes attached. In this way aluminium, like the trivalent actinides deposits on all bone surfaces, including resting and remodelling internal (endosteal) and external (periosteal) surfaces, trabecular bone surfaces and the surfaces of the vascular canals that permeate compact bone. Subsequently, the aluminium will either become buried to form a volume deposit within the bone matrix or will be transferred to osteoclasts (bone resorbing cells) during bone resorption. It is also likely that some aluminium will be released from bone surfaces, either by heterionic exchange or by bone resorption, and this will become immediately available for either local re-deposition or for excretion. However, these pathways have not been demonstrated experimentally for plutonium and are not shown in the figure. The next pathway shown in the figure represents the transfer of metal to macrophages within the bone marrow. Finally that lost from the macrophages, almost

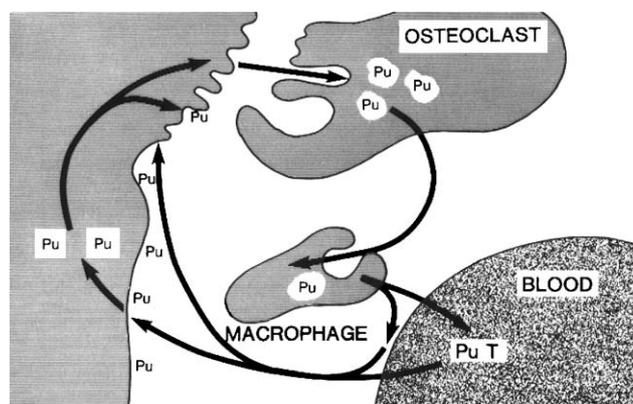


Fig. 10 The cycle of metals in bone as developed for plutonium. Burial and resorption pathways are shown. The macrophage acts as a temporary store of metal within the bone marrow. (From ref. 18).

certainly in association with transferrin, is shown as either being re-deposited locally onto bone surfaces or as re-entering the bloodstream. The metal re-entering the bloodstream will either be redistributed around the body and deposited in tissues, including the skeleton, or (more likely in the case of aluminium) excreted *via* the kidneys.

As a result of the cycling described above aluminium will become distributed throughout the skeleton, but surface concentrations will remain prominent, as volume-distributed aluminium that is removed by bone resorption will be deposited on bone surfaces. Fig. 11 shows a stained section of iliac bone removed from a renal failure patient that had accumulated aluminium. Aluminium was stained using the solochrome azurine method and both surface and volume deposits are seen.

Given that the skeleton is the major site of aluminium deposition in the body, and that bone turnover rates are low in adults (about 3% per year in compact bone and 20% per year in trabecular bone) it may be expected that the residence time of some aluminium in the body will be long—typically bone seeking elements are retained in the skeleton with half-times of several tens of years.¹⁸ Those elements that are strongly bound to proteins in the blood and tend to be re-deposited rather than excreted (*e.g.*, plutonium, thorium, americium and lanthanum) have long effective retention half times in the skeleton—typically 50 years. In contrast, those elements (*e.g.*, barium, radium, strontium, uranium and aluminium) that are poorly bound to blood proteins and are effectively filtered by the kidneys are retained for shorter periods—typical half times being 10–20 years. When modelling the retention of aluminium in the body (see section 7) a best fit to overall retention was obtained when long-term skeletal retention half-times ranging from 1.4 to 29 years were assumed.

5.4 Aluminium in the brain

Unlike for the skeleton, comparatively little is known about the deposition of polyvalent metals in the brain. Consequently, we can learn little about the likely behaviour of aluminium by reference to trivalent actinide and lanthanide analogues—although our knowledge of the influx and efflux of other metals such as iron and manganese may be of more help. In part, this is because the blood–brain barrier acts as an effective barrier to the passage of these metals into the brain,^{6,91–92} such that the small deposits accumulated have attracted little attention. Indeed, if it were not for the link between aluminium and dialysis dementia^{93–94} and the suggested,⁹⁵ but unlikely,^{96–97}

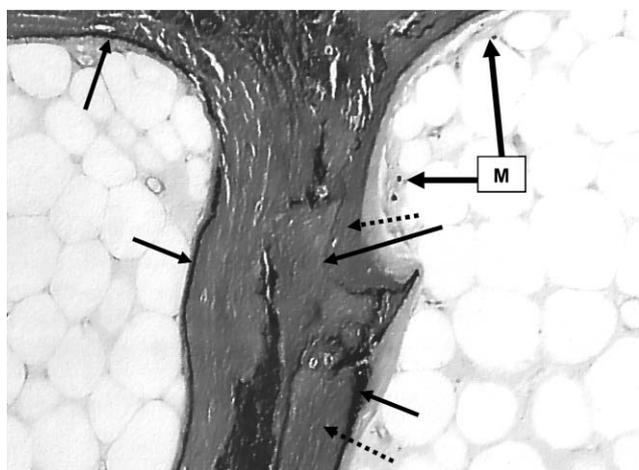


Fig. 11 Iliac bone removed from a renal failure patient with a high body burden of aluminium. Stained with solochrome azurine, it shows surface and buried aluminium line deposits (arrows), volume deposits (dotted arrows) and aluminium deposits in macrophages (M) within the bone marrow. (Photograph supplied by J. Humphreys, AEA Technology plc, Harwell.)

link with Alzheimer's disease (AD)^{98–99} aluminium deposits in the brain would also be of little interest, since their small size means that they little impact upon the retention and distribution of the metal in man. However, interest in these encephalopathies has resulted in extensive research on the interaction between aluminium, the central nervous system in general and the brain in particular. These studies have tended to be either histological studies using advanced imaging techniques such as secondary ion mass spectrometry (SIMS), studies of bulk tissue concentrations or studies with the tracer ⁶⁷Ga—although studies¹⁰⁰ have demonstrated the transfer of ²⁶Al from the gut to brain in rats. Most of these studies have employed either diseased or normal brain tissues removed at autopsy. More recently, research interest has been stimulated again by the indications that aluminium body-burdens may be associated with impaired cognition/neurobehavioural deficits.^{101–103} Kilburn¹⁰⁴ discusses these in relation to occupational exposures to aluminium dust. The most important effects, should they be confirmed, seem to relate to cognitive function and memory.

The bulk tissue studies have suggested that normal human brain aluminium levels fall within the range of <1 to ~5 mg kg⁻¹ with most reported results being towards the lower end of this range, *e.g.*, 0.5 mg kg⁻¹,^{105a} 1.4 mg kg⁻¹, dry weight,^{105b} 2.2 mg kg⁻¹, dry weight,⁶¹ 0.467 mg kg⁻¹.¹⁰⁶ Higher estimates such as those of Flendrig *et al.*⁶⁰, 11.9 mg kg⁻¹, are difficult to reconcile with other data, although individual variation may be high. For example, Crapper *et al.*¹⁰⁷ found a mean level of aluminium in the cortical and sub-cortical regions of the brain of 1.9 ± 0.7 µg g⁻¹, dry weight, but the range was large (0.4–14.0 µg g⁻¹). The levels of aluminium in the brains of patients with renal failure are much higher, but considerable controversy exists concerning the presence or absence of elevated levels of aluminium in AD patients. This controversy has been detailed by Bjertness *et al.*,⁹⁶ who undertook a further study designed to avoid many of the methodological deficiencies in previous studies. These authors reported aluminium levels in the brains (plus liver and femoral heads) of 92 confirmed AD patients and controls. Using well-controlled GF-AAS analyses they reported the following aluminium levels in normal and AD patients: brain frontal cortex (1.8 ± 0.7 vs. 1.7 ± 0.7 µg g⁻¹ dry weight); brain temporal cortex (1.4 ± 0.3 vs. 1.5 ± 0.5 µg g⁻¹ dry weight); liver (2.0 ± 1.3 vs. 2.0 ± 1.2 µg g⁻¹ dry weight); head of femur (2.4 ± 1.6 vs. 2.2 ± 1.0 µg g⁻¹ ash weight). The authors conclude that there is no elevation of bulk aluminium in either the brain or the body of AD patients. Neither was a correlation found between bulk aluminium concentrations and the number of the senile plaques and neurofibrillary tangles that are characteristic of the disease. Yokel *et al.*⁶⁴ suggest that brain aluminium is inconsistently raised during Alzheimer's disease. As a result of these studies and others it is now generally accepted that there is no causal link between aluminium and AD.^{108–109} Nevertheless, under certain conditions when aluminium enters the brain, cognition can be affected and this metal can cause dementia—it follows that aluminium is neurotoxic at high concentrations. It remains an issue of contention as to whether, in the absence of overt disease, aluminium intakes resulting from environmental and workplace exposures can be large enough to cause neurological effects,^{56,104} although some calculations suggest that toxic levels of aluminium in the brain would only be reached under such circumstances after 100–150 years—too long a period to be important.⁹¹

The distribution of aluminium in the brain has been studied in renal failure patients with elevated aluminium body burdens by staff at the MRC Neurochemical Pathology Unit using imaging SIMS at the Harwell Laboratory.¹¹⁰ Using SIMS it is possible to determine the distribution of aluminium deposits, in specimens prepared from frozen bone slices, within the brain at levels down to its distribution within individual cells—using this technique, and fluorescence microscopy, aluminium deposits

have been demonstrated in the Golgi apparatus of cultured cells,¹¹¹ Candy *et al.*¹¹⁰ found aluminium to be present within the brains of former chronic renal failure patients as numerous focal deposits within cortical pyramidal neurones. It is speculated¹¹² that this uptake pattern corresponds to the distribution of transferrin receptor sites within the brain cortex. In support of this hypothesis the authors demonstrated that areas with high densities of transferrin receptors, as determined by autoradiography with ¹²⁵I-Fe-transferrin, such as the *insula* cortex and *amygdala* show much higher densities of aluminium-containing cells than those regions that are low in transferrin receptors, including the *putamen* and *globus pallidus*. Similar aluminium deposits were not demonstrated in the brains recovered from normal control patients. The cells rich in transferrin receptors are metabolically active and have a high requirement for iron during the synthesis of respiratory chain enzymes.

It would seem from the above, therefore, that the entry of aluminium into the brain *via* the blood-brain barrier is, at least in part, a function of the ability of aluminium to ride "piggyback" on a metabolic pathway that has evolved to ensure an adequate supply of iron to the brain. Studies using aluminium have been undertaken to confirm this hypothesis using ⁶⁷Ga as a surrogate. These¹¹² have demonstrated a unidirectional entry of gallium into the rat brain with a permeability of $2.48 \times 10^{-6} \text{ mL min}^{-1} \text{ g}^{-1}$. Assuming a similar blood-brain barrier permeability for aluminium, the authors estimate that the human brain will accumulate approximately 8 µg per year, an amount that they considered consistent with the observed content of aluminium after lifetime exposure.

Two mechanisms of aluminium uptake by the brain have been suggested.⁶ First, Roskams and Connor¹¹³ demonstrated the uptake of aluminium, as for iron and manganese, across the blood-brain barrier by transferrin-receptor mediated endocytosis (TfR-ME)—as implied by Edwardson *et al.*¹¹² This process, which involves the binding of aluminium-transferrin to cell membranes and the transfer of aluminium to cells (neuroblastoma cells and oligodendroglia) has been demonstrated *in vitro*¹¹⁴ and in rats using ²⁶Al.⁶⁴ In the latter study 0.001% of the injected dose per g of brain was transferred by 4 h post-injection. Second, Allen *et al.*¹¹⁵ demonstrated a more rapid uptake of aluminium by the brain from plasma in the presence of excess citrate. Their results suggest the uptake of aluminium, bound to low molecular weight species, directly across the blood-brain barrier. The results produced by Yokel *et al.*⁹¹ suggest the operation of a non-diffusive mechanism of aluminium uptake. A candidate mechanism for aluminium influx into the brain is the monocarboxylate transporter (MCT). The MCT is located at the blood-brain barrier surface and is believed to facilitate the active transport of mono carboxylates across the barrier such as lactate and pyruvate. It is suggested that the free carboxylate of the aluminium citrate complex provides a binding site for the MCT. The report of Allen *et al.*¹¹⁵ also suggests that citrate may also facilitate the loss of aluminium from the brain. It is possible that such aluminium is present in extracellular tissue fluids, but not in cells. In contrast transferrin-receptor mediated endocytosis provides a mechanism for aluminium uptake by brain cells.

An alternative method of entry of aluminium into the brain has been suggested by Perl and Good¹¹⁶ and by Zatta *et al.*¹¹⁷ These authors suggest the transfer of aluminium into the central nervous system along nasal-olfactory pathways from the nose; the aluminium crosses the nasal epithelium and reaches the brain by axonal transport. This implicates inhaled aluminium as a source of brain aluminium, but the importance of this route in man has not been established.

The rate of aluminium loss from the brain is slow—and possibly consistent with the slow rate of cell turnover in the brain. In rats a retention time in excess of 100 days was determined using ²⁶Al.⁶⁴ It is likely in man that aluminium is

retained in the brain for times that are either similar to or exceed those in the skeleton. It follows that aluminium will continue to accumulate in the brain throughout life.¹¹⁸

5.5 Aluminium and iron homeostasis

As previously described the linkage between aluminium and the metabolic pathways that exist for iron is well established—such that aluminium will follow many of the metabolic pathways that exist for this element. Similar linkages exist for other elements, *e.g.*, plutonium, that are able to bind to iron transport and iron receptor molecules. A consequence of this is that the biological behaviour of these metal ions may be influenced by the iron status of the individual. In this way it was shown that the rate of plutonium loss from macrophages in the rat liver is increased in iron-deficient rats and decreased in iron-overloaded rats.¹¹⁹ It was concluded that plutonium excretion levels would be higher and retained body burdens lower in women that are commonly iron-deficient. Recent evidence suggests that aluminium distribution and retention by rats can be influenced in the same way. Nolte *et al.*¹²⁰ have shown that 24 h following the *per oral* administration of ²⁶AlCl₃ to normal rats, iron deficient rats and iron overload rats both the tissue levels of the tracer and the amount excreted were iron status dependent. In the iron deficient animals the concentrations of ²⁶Al in plasma, liver and spleen, bones and urine were all enhanced compared with the control animals, whereas the opposite trend was seen for the iron-loaded animals. These results may be explained, at least in part, if aluminium is unable to bind to iron-saturated sites on transferrin and becomes attached to alternative binding sites in tissues.

The above study does not, however, provide evidence to suggest that iron metabolism is influenced by aluminium intake status. Other studies do suggest this. For example, a study of aluminium dust-exposed workers¹²¹ shows that while there were no differences in manganese, copper, zinc and lead levels in the blood of control workers compared with aluminium workers the levels of iron in blood were elevated by a similar extent (approximately doubled) to those of aluminium in serum. Since, the worker and control populations were well matched this suggests that aluminium exposure increases the uptake of iron by workers. De Kom *et al.*¹²² also saw a similar, but much less convincing, increase in iron-blood levels of aluminium workers. Ward *et al.*¹²³ have provided a possible explanation for these observations; these authors administered aluminium gluconate to rats to produce an aluminium "overload" model. The increased concentrations of aluminium in tissues were paralleled by increases in their iron content. Increased levels of these elements were detected in a wide variety of tissues: liver, kidney, heart, spleen, and different regions of the brain. Increases in iron uptake were attributed to aluminium-induced changes in the regulatory control of iron metabolism. It is thought that the iron regulatory proteins (IRP-1 and IRP-2) control iron uptake and homeostasis. Under low iron conditions these bind to iron regulatory elements present on transferrin receptor mRNA stimulating the production of transferrin receptors that, in turn, result in increased uptake of iron by cells. Under conditions of high iron status IRP-1 is deactivated, by binding to iron, while IRP-2 is degraded. The loss of active IRP results in decreased transferrin receptor production and decreased iron uptake by cells. Ward *et al.*¹²³ speculate that aluminium binds strongly to IRP-2 inhibiting its degradation and thus stimulating the continued uptake of iron (and aluminium) by cells. Also, under high iron conditions the production of the iron storage protein, ferritin (which also has an iron regulatory element to which IRP can bind), is stimulated such that more iron is stored reducing the toxicity of the excess intracellular iron content. The authors showed, using ²⁶Al, that ferritin is unable to bind significant aluminium—providing a possible mechanism for the

accumulation of toxic levels of active aluminium within neurones. This is not unexpected since the binding of iron to ferritin involves a reduction stage, from Fe^{3+} to Fe^{2+} , which cannot be mimicked by the aluminium ion.

6 Aluminium biokinetics

6.1 Aluminium retention by man

As stated above, the demonstrated deposition of aluminium in the skeleton will result in a long-term component to its retention within the human body—this being the result of the stability of metal deposits in the skeleton and of the low rate of their release due to bone turnover in the adult skeleton. It follows, that previously published assertions that aluminium is not accumulated within the body, but is quantitatively excreted within a short period of its intake are not tenable.

To date, only one study has been undertaken to determine the long-term biokinetics of aluminium in the human body. This study was a single volunteer study using ^{26}Al and was initiated at the Harwell Laboratory in 1991.^{19,22} In addition, a multi-volunteer study was initiated using a further six adult male subjects to examine inter-subject variability in the short-term retention of aluminium.^{40,46} For both studies ^{26}Al was intravenously injected as ultra-filtered, citrate solutions, pH 6.5. In the case of the multi-volunteer study approximately 60 Bq of ^{26}Al was injected into each subject, but for the single male adult volunteer study 510 Bq of ^{26}Al was employed. In addition, for this study the volunteer was also injected with 222 kBq of ^{67}Ga to assess the usefulness of the isotope as an aluminium surrogate in biokinetic studies. The studies showed that during the first five days after injection the average amount of aluminium retained by all seven volunteers was 25.7 ± 2.8 (SEM)%. Overall the mean percentage amount retained (R_{te}) for times $t \geq 1$ to 5 days could be expressed ($r^2 = 0.99$) by the power function:

$$R_{te} = 38.2t^{-0.25}$$

However, intersubject variability was significant with a range of residual body burdens at 5 days post-injection ranging from about 16 to 37% of the injected activity. Most of this variability resulted from differences in the levels of aluminium excreted during the first day of the experiments (Fig. 12). This suggests early inter-subject differences in excretory clearance rate, such as those that might arise from variability in the kinetics of aluminium speciation within the blood following injection.

Long-term retention of aluminium has only been studied in one volunteer, but observations of this subject have been

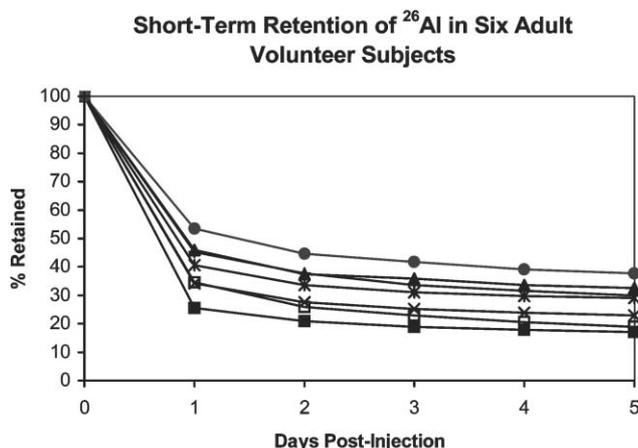


Fig. 12 Graph showing the percentage retention of ^{26}Al as a function of time post-injection. About 70% of aluminium is excreted in the first five days post-administration. (Based on ref. 46)

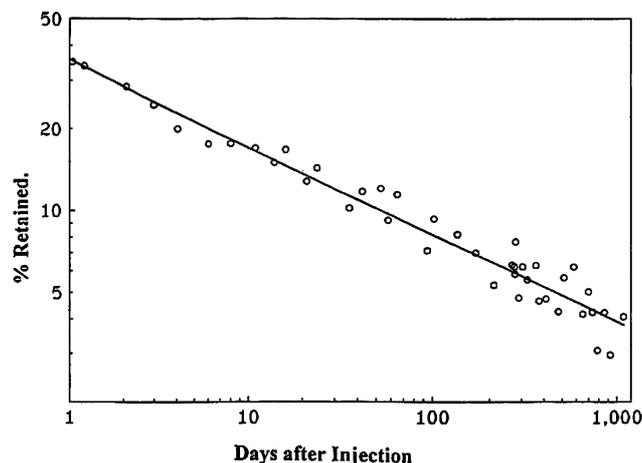


Fig. 13 Retention of ^{26}Al in the volunteer injected with 510 Bq of the isotope, as determined by whole-body, γ -spectrometry. The curve shown is a regression power function plot. Later measurements show the retention of about 2% of injected aluminium at ~ 3000 days. (From ref. 22)

maintained for about twelve years post-injection. In the most recent publication¹⁹ ^{26}Al retention (R_t) to time (t) = 1178 days post-injection (Fig. 13) was described as conforming to the power function:

$$R_t = 35.4t^{-0.32}$$

More recently the retention pattern has been re-analysed (D. Newton, AEA Technology, personal communication) using whole-body monitoring data collected for times up to 3000 days post-injection. It was found that these could also be represented adequately by a three-component exponential function of time:

$$R_t = 29.3e^{-0.595t} + 11.4e^{-0.172t} + 6.5e^{-0.000401t}$$

where the rate constants in the exponents correspond to clearance half-lives of 1.4, 40 and 1727 days. The future trend in the retention is unpredictable, but a reduction in the fractional loss rate is expected, below that implied by the 1727-days half-life of the final term of exponential equation. This is because losses, which are related to bone resorption processes exhibit progressively longer half-lives. Indeed, unpublished excretion data suggest that the current retention half time for this volunteer is in the region of 50 years (see excretion data below). If so, this suggests that aluminium is being retained in the body with a pattern similar to that seen for the heavier alkaline earth elements, which are retained almost entirely within the skeleton. Indeed, the overall pattern of ^{26}Al retention is not dissimilar to the retention of ^{133}Ba in the same volunteer (Fig. 14). Nevertheless, differences are evident with higher aluminium than barium retention within the first year post-injection, but lower retention at the latest times (Fig. 15). Overall, it would seem that the pattern of aluminium retention in the body is more complicated (perhaps because of the retention of some aluminium in other tissues) than that of barium.

In Priest *et al.*¹⁹ the authors used an extrapolation of an integrated form of the power equation to give an estimate of the accumulated body-burden of systemic aluminium from long-term steady exposure. After 55 years, this corresponded to 440 times the daily systemic intake and model predictions made using data from this subject suggest a factor of 417 (see Section 7). Integration of the multi-exponential function suggests, with some reservations, a smaller deposit of about 170 times the daily intake. However, the latest excretion data suggest that this lower factor is too conservative.

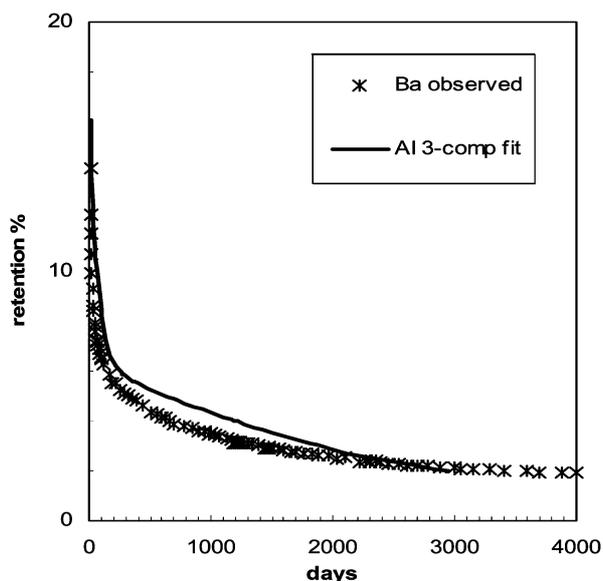


Fig. 14 A comparison of the long-term clearance patterns of aluminium-26 and barium-133—a bone-seeking element—from a volunteer injected with both isotopes, but at different times.

Owing to the relatively short radioactive half-life of ^{67}Ga (3.26 days) it was only possible to make accurate measurements of gallium body content for 21 days after its injection. Nevertheless, at the end of this period the equivalent of about 50% of injected stable isotope remained in the subject²² demonstrating clear differences in the kinetics of aluminium and gallium—with gallium being cleared much more slowly. It follows that ^{67}Ga is not an appropriate surrogate for aluminium in biokinetic studies.

6.2 Aluminium excretion

6.2.1 Urine. Aluminium has been variously described as being excreted both as wholly in urine⁶⁹ and as mostly, *via* the biliary route, in faeces.¹²⁴ Nevertheless, consensus existed that most aluminium was excreted in urine with an unknown fraction in faeces.² Reported values for the concentration of aluminium in urine range from 0.68 to 8 $\mu\text{g L}^{-1}$ ^{1125–126}—with recent estimates tending to be lower. However, given the variations in the volume of urine excreted by different individuals then the mass of aluminium excreted per day in urine is a more informative statistic. The values quoted in the

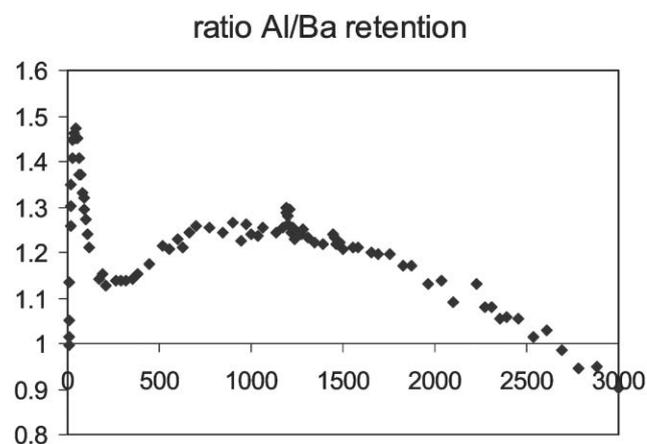


Fig. 15 Graph showing the ratio of ^{26}Al to ^{133}Ba retention for different times after their injection into the same volunteer. Aluminium is initially retained more than barium, but after 2500 days, post-injection it is lost more rapidly.

above publications are 1.8–9.3 μg per day and 4–12 μg per day, respectively. Confirmation of the importance of the urinary route and relative unimportance of the biliary route to faeces has been obtained using injected ^{26}Al citrate.^{6–19}

The selection of citrate for such injection studies is critical and needs to be discussed in the context of urinary excretion. In order to undertake a valid biokinetic study using an injected radionuclide it is necessary to inject it in a bioactive form and in such a way that its behaviour may be reasonably expected to simulate that of the element following entry into the bloodstream by natural routes. One common method is to inject soluble salts such as either chloride or nitrate in acid solution.¹²⁷ However, as previously stated, in the case of aluminium, this would result in the formation of insoluble colloids that are rapidly removed from the bloodstream by cells comprising the reticulo-endothelial system. Indeed, in rats ^{28}Al (the short-lived aluminium isotope) intravenously injected as the chloride, was rapidly taken up by the liver—almost certainly as a consequence of the formation, within the blood, of insoluble colloids. Other studies have employed metal ions bound by a strong chelator such as EDTA, DTPA or desferrioximine (DFO); in these cases the ion is maintained in solution, but it is bound so strongly that almost complete excretion occurs.¹²⁸ It follows, that the injection solution of choice should be capable of stabilizing the aluminium in solution, but should not bind it so strongly that it substantially affects its biokinetic properties.

Of the possibilities available, citrate solutions were used for the present studies. Citrate has the advantages that it is a normal major component of blood and a demonstrated binding site for aluminium within blood. In addition, it is possible that aluminium entering from the gut enters the bloodstream as a citrate complex,^{129–130} although, this is contested by Taylor *et al.*¹³¹ who point out the different kinetics of aluminium and citrate uptake into and loss from blood. It has the disadvantage that it is a carrier for metals excreted in urine. The possibility exists, therefore, that the levels of citrate injected intravenously, 35 mg, in the ^{26}Al injection studies,²² may have stimulated excretion and resulted in excess aluminium loss. Nevertheless, many studies using a variety of metal ions have demonstrated the similar behaviour of metals after injection as citrate, after administration as a complex with blood proteins, after entry into blood from a deposit in muscle/subcutaneous tissue and after natural entry. Moreover, citrate is rapidly excreted, at a rate of about 45 mg h^{-1} in urine, its average entry rate into blood from tissue fluids and diet must be equal to this and its rate of entry when citrate is ingested must be larger. Consequently, the injection of 35 mg of citrate is unlikely to have produced either a significant or persistent perturbation in citrate excretion. Indeed, Priest *et al.*²² report that their results showed no evidence of any major (>factor of 2) or sustained (more than 1 h) elevation in excretory clearance from blood.

The studies undertaken^{19,40} have shown that the majority of injected aluminium is excreted within the first few days post-injection in urine (Fig. 16). Subsequently, the amount lost per day falls with the ^{26}Al body burden. At 3350 days post-injection the daily loss of ^{26}Al by the single volunteer that had been injected with 510 Bq of this isotope had fallen by about 6 orders of magnitude to $3.9 \times 10^{-5}\%$ per day (Priest, unpublished data)—this demonstrating the power of AMS as an analytical tool. A power function for daily aluminium loss in urine (U_t) at t days, based on excretion data to 14 days plus the data at the later date has been calculated, $r^2 = 0.98$:

$$U_t = 24.28t^{-1.6951}$$

Given that the estimated retention of aluminium in the subject at this time was about 2% of the injected activity, the measured excretion rate of $3.9 \times 10^{-5}\%$ per day suggests that the contemporary body-burden is being lost with a very long half-time.

Cumulative ^{26}Al Excretion in Urine

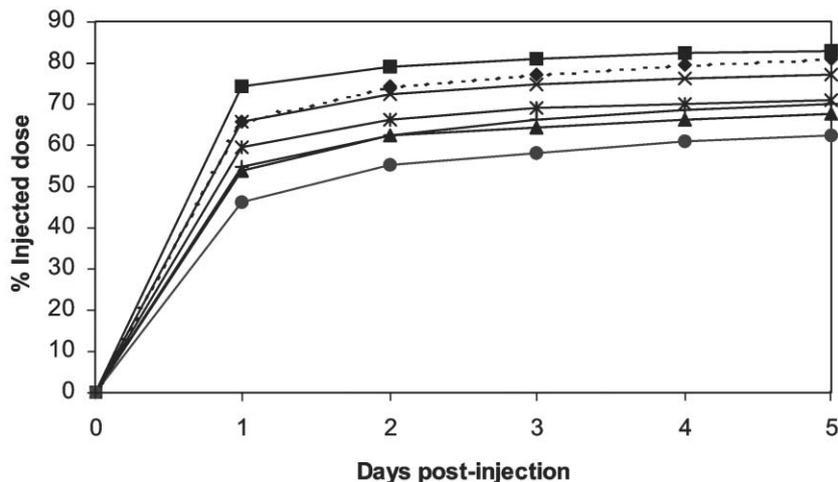


Fig. 16 Multi-volunteer study.⁴⁶ Cumulative urinary excretion of ^{26}Al to five days post-injection for six adult male subjects plus subject from single volunteer experiment (dotted line).²²

Estimates of renal clearance rate (litres of whole blood per day) were derived from the contemporary blood and urine ^{26}Al levels for all seven subjects.⁴⁰ A six-fold variation in renal clearance was found in the multi-volunteer group, encompassing the value of 32 L per day reported for the single volunteer. The calculated mean value (16 ± 10 L per day) was roughly an order of magnitude lower than would be expected from typical glomerular filtration rates, suggesting either that excretion of the tracer was, at this stage, inhibited through binding to blood components, such as transferrin, or that aluminium undergoes selective tubular reabsorption. The former is considered more likely and is consistent with the known binding of aluminium to transferrin. By contrast, the rate reported was an order of magnitude greater than the renal clearance rates suggested by data¹³² for normal aluminium excretion. Talbot *et al.*⁴⁰ also report that, over the first five days post-injection, urinary excretion of ^{26}Al declined much more rapidly than the levels of this tracer in blood. The authors conclude that during this period it is likely that the speciation of aluminium in the blood continues to adjust.

Following other studies, utilising stable aluminium, it has been suggested that glomerular filtration rate increases as a function of plasma-aluminium-loading.¹³³ This may indicate that at high levels of aluminium in plasma a higher than average fraction of the metal is complexed to low molecular weight plasma components. Lastly, some studies have shown that the levels of aluminium excreted in the urine of men is greater than that excreted by women.¹²⁶ Stauber *et al.*¹³⁴ suggest that this is a function of the higher dietary intake of aluminium in men.

6.2.2. Faeces. Fig. 17 shows the pattern of faecal excretion of aluminium tracer during the first days following injection as described by Talbot *et al.*⁴⁶ It can be seen that very little ^{26}Al excretion occurs in the first day following injection and that the mean level excreted increases rapidly at the beginning of each day, then a plateau is reached until the beginning of the next day. The delay in excretion results from the time taken for aluminium released into the upper regions of the small intestine to traverse the length of the remaining small and large intestines prior to voiding. The stepwise pattern results from the tendency of the volunteers to void faeces at the beginning of each day.

Overall, 1.2 ± 0.3 (SD)% (range 0.9–1.8%) of the injected ^{26}Al was excreted by this route in five days. This confirms the

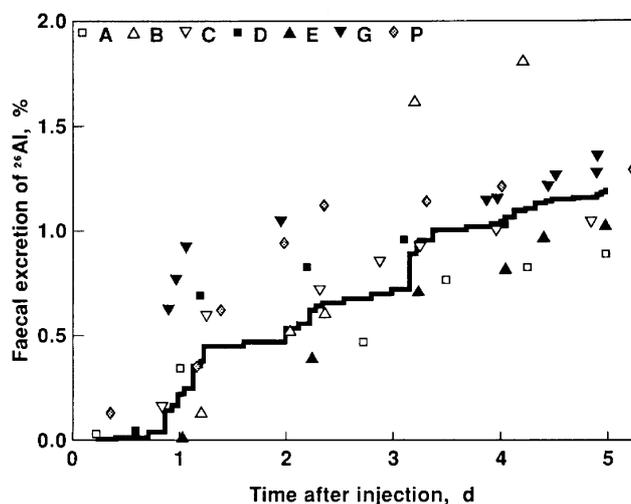


Fig. 17 Cumulative faecal excretion of ^{26}Al by seven volunteers (designated A–E, G and P) injected with ^{26}Al -citrate. The injections were administered each morning. Most voiding also occurred in the mornings resulting in a step-like pattern of aluminium excretion. (Reproduced from ref. 46.)

relative unimportance of faecal, relative to urinary, excretion at early times after injection. However, the results of the single volunteer study²² showed that, whereas the levels of ^{26}Al excretion in urine decreased rapidly during the period 4–14 days post-injection, faecal excretion during this period remained relatively constant at about 0.06% per day. These studies report no measurements of ^{26}Al excretion in faeces after 14 days post-injection. Therefore, it was thought possible that at longer times after intake, when most of the initial body intake has been excreted in urine, faecal excretion of aluminium may become progressively more important and even exceed the levels of ^{26}Al excretion in urine. If so, it would be likely that such an increase would be related to the increase seen in the levels of aluminium in red blood cells during the first days post-intake⁴⁴ and, perhaps, to a progressive transfer of aluminium to the liver. However, recent unpublished data by Priest and his co-workers have shown that faecal excretion levels fall in line with decreases in the level of urinary excretion. At 3200 to 3250 days post-injection of ^{26}Al the amount of the injected dose excreted in the faeces was $3.5 \times 10^{-6} \pm 1.8 \times 10^{-6}$ % per day, *cf.* $3.8 \times 10^{-4} \pm 1.3 \times 10^{-4}$ % per day in urine.

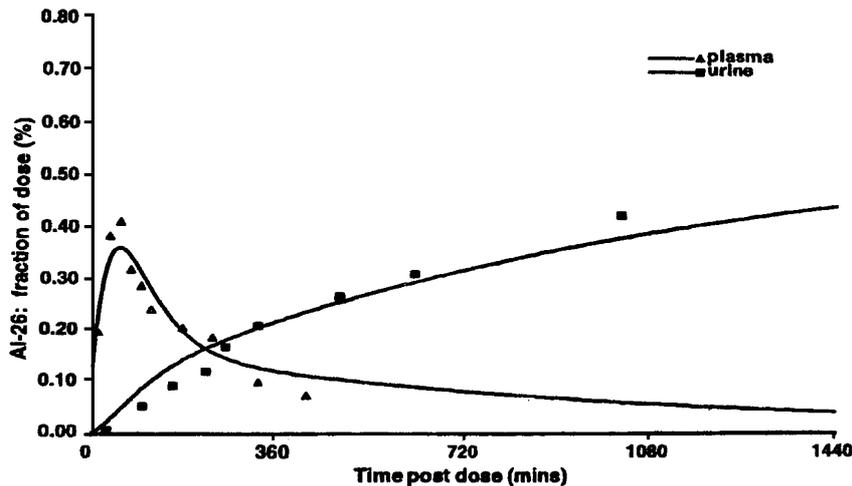


Fig. 18 The results of Day's three-compartment box model fit to ^{26}Al data for ingested aluminium citrate. (Modified from^{ref. 44}).

7 Modelling aluminium biokinetics in man

Until recently, the only comprehensive physiological-data-based pharmacokinetic model of the behaviour of aluminium in man that had been published is that published by the ICRP in its Publication 30/Part 3.¹³⁵ For this model, it was assumed that 30% of aluminium leaving the transfer compartment (blood) is translocated to bone mineral and 70% is uniformly distributed throughout all other organs and tissues of the body. Aluminium deposited in any organ or tissue was taken to be retained with a biological half-life of 100 days. Clearly this model is at variance with the data generated by studies using ^{26}Al . This is not surprising given the paucity of data available to the ICRP at the time of model formulation.

Using ^{26}Al data Day and his co-workers have attempted limited modelling to describe the early pharmacokinetics (plasma and urine) of ingested aluminium citrate.⁴⁴ A three-compartment box model was employed to model the kinetics of aluminium uptake and excretion for times up to about a month

post-intake. The model consisted of a compartment for plasma and two others for tissues. Unlike in the ICRP model the tissue compartments employed by Day were not representative of specific tissues or organs, but were defined in terms of aluminium kinetics with retention half times of 10.5 and 105 h, respectively. The model parameters employed provided a reasonably good fit to the data (Fig. 18).

Recently, a model based on the results obtained for subject P, who was injected with $> 500 \text{ Bq } ^{26}\text{Al}$ -citrate and followed for 10+ years, has been developed at Middlesex University. This model is much more complex than that of Day and employs 8, tissue-associated compartments (Fig. 19). The model produces a good fit to the total body retention of aluminium by the subject to 1000 days post-intake, as determined by whole-body gamma-spectrometry (Fig. 20), and to the levels of urinary and faecal excretion to > 3000 days, as determined by radiochemical analyses of samples. Given that no data exists for the distribution of aluminium within the body the box descriptors employed are considered indicative—although they are consistent

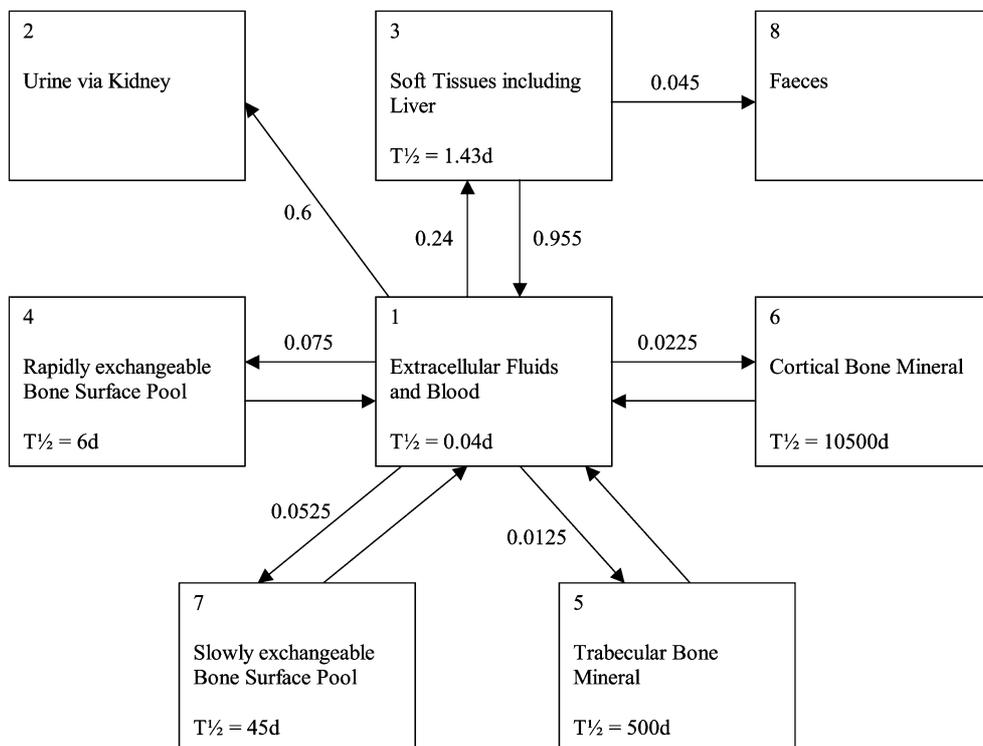


Fig. 19 The Middlesex University biokinetic model of aluminium retention—as developed to fit data collected for a subject injected with about 500Bq of ^{26}Al .

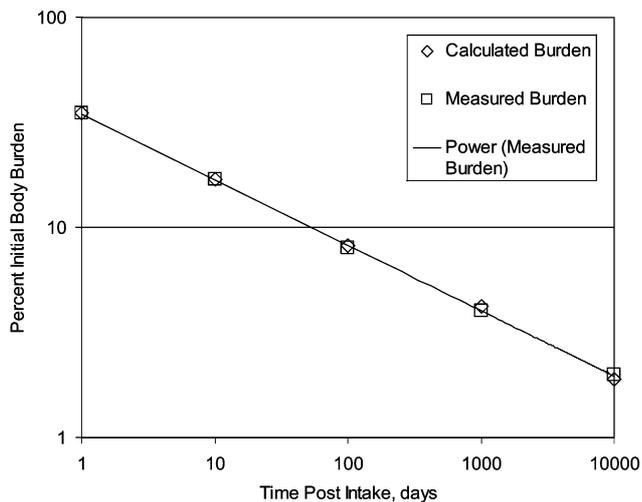


Fig. 20 Middlesex University model prediction of the retained body-burden of aluminium at different times after an acute single intake of aluminium compared with data collected by whole-body monitoring for ^{26}Al retention to 1000 days post-intake and by extrapolation of the determined power function of retention to 10000 days.

with a bone-seeking nuclide that is lost from the skeleton by ion exchange and bone resorption processes. In most of such models the central compartment represents the blood pool of nuclide. However, reproducing the levels of aluminium in the blood of subject P is not possible using this model. This is because of uncertainties in the kinetics of aluminium transfer from blood to tissue fluids and because the speciation of aluminium in blood seems to change—as indicated by changes in the excretory clearance rate of aluminium.¹⁹ Moreover, neither the size of the blood pool nor of the tissue fluid pool have been determined such that would allow the amount of ^{26}Al in blood samples and model predictions to be precisely compared. When further data becomes available it is planned to add additional model compartments to represent the amount of aluminium in red blood cells and the brain.

The biokinetic data for subject P were also used to develop an open compartment model for describing aluminium biokinetics in man and rats.¹²⁰ This has a central compartment comprising four pools of aluminium (plasma transferrin, plasma citrate, interstitial fluid transferrin, and interstitial fluid citrate) and three peripheral compartments for organs, muscle and bone and a GI tract compartment. Unlike the Middlesex University model, the model of Nolte and co-workers is able to predict, with reasonable accuracy, the levels of ^{26}Al in the blood plasma of subject P to 400+ days after injection and in faeces and urine to 5 days post-injection. The model was also able to simulate changes seen in rats following iron deficiency and iron overload. In iron deficiency aluminium ingestion resulted in increased levels of ^{26}Al in plasma, liver and spleen, bone and in increased levels of aluminium excretion relative to control and iron overload animals. The changes in tissue levels and aluminium excretion could be predicted using the model.

When deployed in chronic accumulation mode the Middlesex University model may be used to predict terminal body burdens of aluminium following any pattern of aluminium intake during the integration period. For continuous, level intake over 50 years the model predicts a retained body burden equal to 417 times the daily aluminium intake into the blood. During this period only the amount of aluminium in the cortical bone compartment is predicted to increase continuously. For other compartments their aluminium content is predicted to rise to an equilibrium position - after a period that depends upon the relevant chosen half-time of retention, e.g., after about 3 years for aluminium in trabecular bone mineral.

Use of the model in this mode shows that despite a large increase in body burden over a 50-year period the amount of aluminium excreted in the urine only increases from an amount equivalent to 64% of the daily intake at day 1 to an amount equivalent to 96% of the daily intake at day 18263. Also, that despite the larger body burden at 50 years if aluminium intake ceases then the amount of aluminium excreted rapidly falls so that after a further 100 days the amount excreted has fallen to 7% of the previous daily intake. These calculations show the insensitivity of aluminium excretion levels, either at the time of intake or post-intake, to the size of the extant body burden and the probable inability of urine analyses to detect significant body burdens of this metal. Clearly, other methods have to be deployed to determine the size of likely body burdens. Such methods might include a chelation challenge if the fraction of the body burden that could be removed by a chelating agent, such as DFO, could be established. If such a method were established then it would be equivalent to the EDTA method used to estimate lead body burdens.¹³⁶

8 Aluminium bioavailability following ingestion

Given the ubiquitous nature of aluminium in the environment, it is surprising the human body contains, at most, only a few tens of milligrams of this element and that it has no known essential function. This low level results both from the insolubility, at neutral pH, of most natural aluminium compounds and from the protective barrier that the body's gut wall presents to the uptake from food of potentially toxic metal ions. Nevertheless, some aluminium overcomes these barriers and enters the body. While these levels are likely to be small for most environmental aluminium compounds, such as aluminosilicate—the essential component of rock and soil minerals—they could be considerably greater for some manufactured compounds, which may be of high solubility—leading to increased uptake.

Metal ions enter the body by one of three main routes: *via* the gut wall, by inhalation and through wounds (Fig. 21). All of these may be important for aluminium. Probably the most important route is *via* the gut wall, even so, by far the greatest fraction of ingested aluminium passes through the intestinal tract without being absorbed. By comparison with the bioavailability of other trivalent metal ions, including the lanthanide and actinide elements, which have been much studied

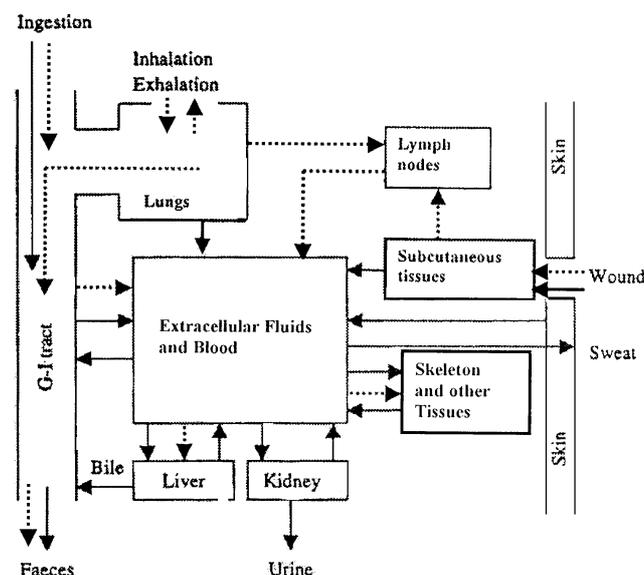


Fig. 21 Diagram showing intake and re-distribution possibilities for soluble and particulate aluminium within the body. Transfers involving particles are shown as dotted arrows.

because of their importance in the nuclear industry,¹³⁷ it may be predicted that only about 1.0×10^{-4} (or less) of the insoluble species (such as aluminium oxides and aluminosilicates), depending upon their physicochemical properties, will be absorbed. For the more soluble species, studies on other polyvalent metals would suggest that, fractional intakes will be higher.¹³⁸

Studies of aluminium absorption by man consistently show enhanced aluminium uptake when the metal is present in the gut in association with citrate—although the relationship may be complex.¹³⁹ In early studies, reviewed by Molitoris *et al.*¹³⁰ aluminium uptake from aluminium hydroxide, by uremic patients not receiving dialysis, was shown to be enhanced by up to about an order of magnitude by the co-ingestion of Shoal's solution (a citric acid–sodium citrate buffer). Similar effects have been noticed for lactate¹⁴¹ and may be related to the ability of many carboxylic acid derivatives to bind aluminium, as soluble complexes, within the gut, *e.g.* ascorbate¹⁴² and gluconate, malate, oxalate and tartarate.³² Using a perfused rat intestine model¹⁴⁰ the fractional absorption of aluminium was found to depend upon the nature of the ligands associated with the Al^{3+} ions in the gastrointestinal fluid and uptake decreased in the order citrate > tartarate, gluconate, lactate > glutamate, chloride, sulfate and nitrate. How aluminium absorption is enhanced by citrate is unknown. One possibility is that aluminium binds to citrate in the gut, then crosses the intestinal barrier bound within this complex and enters the bloodstream where it dissociates. However, others^{7,129,131} have suggested that it is far more likely that enhanced aluminium absorption, in the presence of citrate, takes place in the upper GI tract *via* paracellular pathways that are opened by citrate-induced loosening of the tight junctions between gastrointestinal mucosal cells—this process seems to be a passive diffusion process that requires no energy. In support of this suggestion Taylor *et al.*¹³¹ demonstrated that the uptake kinetics of aluminium and citrate from the diet are quite different: serum aluminium peaked at 90 min post-administration and citrate at 32 min post-administration. These authors conclude, however, that the data that they collected did not exclude the possibility that citrate facilitates the absorption of aluminium by maintaining it, within the gut, in a form available to other mechanisms of absorption. In addition, there is some disputed evidence to suggest that some aluminium may be absorbed across the gut wall by an energy-dependent, active transport process through cell walls.¹⁴⁰

In contrast to citrate, the co-ingestion of high concentrations of phosphate, fluoride and silicic acid will reduce the bio-availability of ingested aluminium by the formation of insoluble precipitates within the gut.^{143–144} Factors affecting the gastrointestinal absorption of aluminium are reviewed by Powell and Thompson¹⁴⁵ and by Yokel and McNamara.⁶

8.1 Food and beverages as sources of aluminium

The potential for aluminium uptake from food is limited as most foods contain little aluminium.^{146–147} With respect to plants, reports describe diverse levels in different foods and reported values vary for similar foods. It is likely that much of this variation results either from the inadequate removal and/or contamination of foods with soil, prior to analysis, or the use of poor analytical techniques. Most un-cooked plant foods contain $<5 \mu\text{g g}^{-1}$. Reported values for some typical food plants are as follows. Un-cooked plant foods: barley, $5\text{--}7 \mu\text{g g}^{-1}$; wheat, $4\text{--}16 \mu\text{g g}^{-1}$; green beans, $8 \mu\text{g g}^{-1}$; carrots, $3.8 \mu\text{g g}^{-1}$; onions, $5\text{--}10 \mu\text{g g}^{-1}$; tomatoes, $0.2\text{--}1.1 \mu\text{g g}^{-1}$; apples, $0.2\text{--}0.9 \mu\text{g g}^{-1}$; oranges, $<0.4 \mu\text{g g}^{-1}$; rhubarb, $0.8\text{--}4.8 \mu\text{g g}^{-1}$; peanuts; $<2 \mu\text{g g}^{-1}$. Processed plant foods: white sugar, $<2 \mu\text{g g}^{-1}$; whole wheat bread, $5.4 \mu\text{g g}^{-1}$; corn flakes, $<2 \mu\text{g g}^{-1}$; spaghetti, $<2 \mu\text{g g}^{-1}$; molasses, $110 \mu\text{g g}^{-1}$; olive oil, $0.1\text{--}0.4 \mu\text{g g}^{-1}$. However, reported concentrations in herbs

and spices are sometimes much higher, ranging from low concentrations of $5\text{--}10 \mu\text{g g}^{-1}$ for mustard, nutmeg and sesame seed, to high values of $500\text{--}1000 \mu\text{g g}^{-1}$ for marjoram and thyme. Where high values are found it is suggested that these often result either from the dehydrated state of the food or from their content of aluminium-containing grinding materials.

Similarly, the concentration of natural aluminium in animal-derived foods is low and is influenced by the low concentration of aluminium in animal feeds, by the poor biological uptake of “food-aluminium” by food-animals and by the limited ability of body-aluminium to transfer to products such as eggs and milk. Consequently, most animal-based foods contain $<1 \mu\text{g g}^{-1}$ of aluminium. Typical values reported in the literature are: freshwater and marine fish, $<1 \mu\text{g g}^{-1}$; beef, $<1\text{--}8.4 \mu\text{g g}^{-1}$; lamb and pork, $<1 \mu\text{g g}^{-1}$; eggs, $0.2\text{--}1.4 \mu\text{g g}^{-1}$; chicken, $<1 \mu\text{g g}^{-1}$; milk, $0.1\text{--}0.7 \mu\text{g g}^{-1}$; Swiss cheese, $19 \mu\text{g g}^{-1}$.

Of the remaining miscellaneous foods in common usage few contain significant aluminium. Beer stored in aluminium cans, for up to one year only contains $<29 \mu\text{g mL}^{-1}$ and the maximum level recorded in beverages within aluminium cans is $1.5 \text{ mg per } 375 \text{ mL can}$. Tea, which when dry, contains large quantities of aluminium (1.28 mg g^{-1}), contains relatively little when steeped ($2.8 \mu\text{g mL}^{-1}$). For comparison, brewed coffee contains less than $0.4 \mu\text{g mL}^{-1}$. The aluminium content of drinking water and other beverages, and their contribution to aluminium intake is described by López *et al.*¹⁴⁸

It follows that natural diets are likely to contain little aluminium. However, a diet containing either cooked or processed foods may contain more. For example, cooking in aluminium vessels may increase the aluminium concentration of acidic foods. In contrast, the cooking of non-acidic food products in aluminium utensils has little effect upon their aluminium content. Similarly, the use of aluminium foil for wrapping foods adds little or nothing to their metal content. Also, many aluminium compounds are permitted food additives.¹⁴⁹ While most are present in foods as trace components, others may be present in significant quantities. For example, aluminium-based baking powders, employing acidic sodium aluminium phosphate (SALP), may contain more than 10 mg g^{-1} of aluminium, and bread or cake made with these may contain $5\text{--}15 \text{ mg}$ of the element per slice. American processed cheese may contain as much as 50 mg of aluminium per slice, resulting from the addition of basic sodium aluminium phosphate (Kasel)—an emulsifying agent. Pickled cucumbers, in the US, have been reported to contain 10 mg of aluminium per fruit, when alum has been employed as a firming agent. Aluminium anti-caking agents may also be present in significant quantities in common table salt.

Estimates of aluminium in the total diet (excluding aluminium from drinking water) have been made for the Food and Drugs Administration in the United States, by Pennington,^{146,150} and for the Ministry of Agriculture Fisheries and Food in the UK, by Sherlock.¹⁴⁷ These suggest daily aluminium intakes of between 0.7 and 14 mg —depending upon age and sex. In Finnish, Japanese, Swiss and UK studies,⁵⁴ daily intake of aluminium from food was calculated to be 6.7 , 4.5 , 4.4 and 3.9 mg , respectively. In general, it is likely that daily intakes of aluminium in North America are higher than in Europe, due to a higher utilisation of SALP and Kasel in the preparation of processed foods. As for measurements of the aluminium content of individual foods, the measurement of total daily diets is complicated by the problems of analysis and of sample contamination. However, the WHO data are generally regarded as reliable. Aluminium concentrations are also higher in manufactured infant milk formulas than in human breast milk—although concentrations are very variable and product specific—suggesting a possible cause for concern.¹⁵¹

To aluminium intakes from food, should be added a component to allow for the daily consumption of drinking

water. At aluminium levels of $20 \mu\text{g L}^{-1}$ and with an average daily consumption of 1.5 L of drinking water per day, intakes from this source would average about $30 \mu\text{g}$ —i.e., about 0.3% of the average US intake from food. At aluminium levels in drinking water of $200 \mu\text{g L}^{-1}$, corresponding to the maximum level recommended by the World Health Organisation,¹³ aluminium from this source would account for about 3% of average total dietary intake in the USA and perhaps a little more in Europe. In a Spanish study of the aluminium content of beverages¹⁴⁸ the average dietary uptake of aluminium from beverages was calculated to be $156 \mu\text{g}$ per day—somewhat lower than from intakes of water at the WHO limit. This result was calculated using the mean daily consumption of different beverages, containing different levels of aluminium, in Spain: water (4.2 – $165.3 \mu\text{g L}^{-1}$); fruit juices (49.3 – $1144.6 \mu\text{g L}^{-1}$); soft drinks (44.6 – $1053.3 \mu\text{g L}^{-1}$).

Given the above data it is reasonable to assume daily aluminium intakes in the diet of about 10 mg per day. Using this figure, an estimated retention of absorbed aluminium of 2% (derived from ref. 152) and estimated terminal aluminium body-burdens, ranging from about 5 mg^{19} to 60 mg^1 —these include the central estimates of 35 – 40 mg^{132} —it is possible to estimate the likely range of values of the fractional absorption of dietary aluminium. Ignoring contributions from sources other than dietary aluminium, for a terminal body burden of 5 mg, accumulated over a period of 50 years, the daily contribution to body burden, from the diet must be in the region of $2.74 \times 10^{-4} \text{ mg}$. If over this period an average of 98% of the aluminium taken up by blood is excreted (see above) then this corresponds to a daily uptake of $1.4 \times 10^{-2} \text{ mg}$, which, in turn, corresponds to an absorbed fraction of 1.4×10^{-3} (0.14%). The corresponding calculation for an assumed terminal body burden of 60 mg is 1.6×10^{-2} (1.6%) and for central estimates would be about 0.5–0.8%. Given that sources other than diet (e.g., vaccinations, pharmaceuticals, inhaled dusts, toiletries) will contribute to aluminium body burden⁶ it would be reasonable to assume that these estimates are upper limits.

An alternative approach to the determination of fractional intakes is to use the biokinetic data generated by the ²⁶Al studies in combination with the known levels of aluminium excretion. If the ingested amount is 10 mg and the average daily excretion of aluminium in urine is $7.5 \mu\text{g}$ (unpublished data, AEA Technology plc), the average intake (allowing for retention and faecal excretion) is $7.8 \mu\text{g}$ corresponding to an absorbed fraction of 7.8×10^{-4} (0.08%). Using a higher daily excretion level of $15 \mu\text{g}^{153}$ the absorbed fraction would be higher at 1.5×10^{-3} (0.15%). Similar calculations have been made by Stauber *et al.*,¹²⁶ who undertook a major bioavailability study using 21 male and 8 female volunteers ranging in age from 36 to 76 years. For this study the volunteers were fed controlled diets containing a known mass of aluminium, plus either alum-treated drinking water or a low-aluminium control water. Strict quality control of analyses was maintained. The absorbed fractions calculated, using the ²⁶Al excretion data of Priest and co-workers, were 0.28–0.64% for aluminium in food and 0.37% for aluminium in alum-treated water.

The above fractions accord well with expectations based on the results of other human bioavailability studies employing stable aluminium preparations (see reviews: US DHSS,¹⁵⁴ WHO,⁵⁴ and Gardner and Gunn¹⁵⁵). For example, the Toxicological Profile for Aluminium—update¹⁵⁴ reports that most estimates of average gastrointestinal absorption of aluminium under normal dietary conditions lie in the range of 0.1–0.3%. This publication cites a representative study by Greger and Baier¹⁴¹ who measured aluminium absorption in eight subjects fed either a control diet (5 mg Al per day) or a diet supplemented with aluminium lactate (120 mg Al per day) for 20-day periods. The absorbed percentage fractions measured

were 0.09 and 0.78%, respectively. This study demonstrated the effect of lactate on enhancing the uptake of the metal.

Despite the above, studies employing ²⁷Al cannot in general be used to measure the bioavailability of specific ingested aluminium species following single low-level exposures and some have failed to identify increments in aluminium in urine and/or blood following intakes, (e.g., Turnquest and Hallenbeck,¹⁵⁷ and Drewitt *et al.*,¹⁵⁶). This is because of the difficulties in measuring small amounts of aluminium in biological samples and the impossibility of distinguishing excreted endogenous aluminium from aluminium administered in the test food/fluid. They have the advantage, however, that the speciation of the aluminium administered is appropriate—this would be difficult to establish for ²⁶Al-labelled foods.

8.2 Medical exposures to ingested aluminium

While, the ingestion of food and water only results in small aluminium uptakes, the use of aluminium-containing pharmaceuticals could result in relatively larger uptakes. Consequently, they provide the most likely source of excess aluminium body-burden in members of the general population. The major cosmetic/pharmaceutical uses of aluminium compounds are: aluminium hydroxide as an antacid; aluminium hydroxide as an effective, non-absorbed phosphate-binder for patients with long-standing kidney failure; as a component of buffered aspirin; aluminium hydroxide and monostearates as components of some vaccines/injection solutions (e.g., the combined MMR (measles, mumps, rubella) vaccine); aluminium chloride, aluminium zirconium glycine complex and aluminium chlorohydrate as the active ingredients of antiperspirants. In addition, aluminium is present as a contaminant in total parenteral nutrition solutions. Many of these applications are under review and their use is discouraged where alternatives of equal efficacy are available and where the potential for high aluminium uptakes exist. For example, calcium carbonate is a suitable alternative to the long-term use of aluminium hydroxide for the treatment of chronic acid indigestion in some patients and lanthanum compounds have been used as phosphate binders. The use of carboxylic acids in combination with aluminium compounds is also discouraged.

Studies of the bioavailability of ingested pharmaceutical and medicinal preparations, using stable aluminium preparations (particularly aluminium hydroxide) have been described by a number of authors. Most of these^{153,158–159} indicate low levels of absorption despite ingested masses (typically multi-gram) that may be several orders of magnitude greater than those employed for food studies. However, the co-administration of citrate and other chelating molecules (e.g., lactate and ascorbate) increased absorption. For example, following the ingestion of approximately 1 g of aluminium hydroxide suspended either in water, in orange juice or in citric acid, the measured absorptions were 0.004, 0.03 and 0.2%, respectively.¹⁶⁰ Given that 1 g of aluminium was ingested these results suggest uptakes of 0.04 mg, 0.3 mg and 2 mg, and long-term daily aluminium retentions of 0.8 μg , 6 μg and 40 μg per day, respectively. It is clear that chronic ingestion of multi-gram amounts of aluminium hydroxide-containing OTC drugs, with or without citrate, has the potential to build aluminium body-burdens at a rate inconceivable either by the ingestion of food and drink or following most occupational exposures to aluminium (Table 4).

8.3 Bioavailability studies using ²⁶Al

The first reported bioavailability study using ²⁶Al was reported by Day *et al.*,³⁷ in a landmark study that demonstrated the veracity of the use of this isotope, in combination with AMS, for human studies of aluminium biokinetics. For this study Day drank a solution containing ²⁶Al, in the presence of excess

Table 4 Predicted terminal body burdens of aluminium after 30 years continuous exposure

Intake Source	Assumed daily uptake/ μg	Body burden at 50 years ^a	
		Low estimate	High estimate
Low environmental	1	85 μg	880 μg
Environmental	6	510 μg	5.3 mg
Occupational	40	3.4 mg	35 mg
High occupational	220	19 mg	200 mg
High pharmaceutical	2000	170 mg	1.76 g

^a Upper and Lower Estimates based on Results of ²⁶Al Retention Studies^{40,45}

citrate. Aluminium uptake was estimated from the concentration of the radioisotope in blood—a method that had been successfully deployed to measure the bioavailability of a number of other metals, including lead.¹⁶¹ The estimated absorbed fraction was 0.01 (1%). This method, which has also been employed by Edwardson and his colleagues in Newcastle, is problematic since the levels of ²⁶Al in blood reach no convenient plateau for intake estimation^{19,44,47} and aluminium absorption from the gastro-intestinal tract may be protracted over many hours or days.⁴⁵

Other studies of ²⁶Al absorption have been conducted in rats, where the absorbed fraction is easier to measure. Schönholzer *et al.*¹⁶² measured absorbed fractions of 0.1, 0.7, 5.1 and 0.1% in rats gavaged with ²⁶Al-labelled aluminium hydroxide, aluminium citrate, aluminium citrate (plus additional sodium citrate) and aluminium moltolate, respectively. Other published studies include those of Zafar *et al.*,¹⁶³ which found a fractional absorption of 8×10^{-3} for ²⁶Al ingested by rats in saline and that of Jouhannau *et al.*¹⁶⁴ that measured absorbed fractions of 4×10^{-4} for ²⁶Al ingested in both water and citrate solution. These studies produce good data for rats and, while some studies show reasonable accord between the levels of GI absorption in rats and man (*e.g.*, ref. 33) their significance for man is uncertain given possible interspecies differences in bioavailability.¹³⁸

Ideally, studies to measure the bioavailability of aluminium intakes by man should be conducted in man and employ dual tracer methodologies. Such studies normally require the simultaneous administration of two different tracer isotopes—one administered by injection and the other by the test route, *i.e.*, ingestion. The bioavailability is then calculated from the ratio of the integrated tracer levels in either blood or urine over the period of absorption. However, dual isotope studies are not possible for aluminium given the availability of only one unique isotope—²⁶Al. Nevertheless, an approximation to this type of study can be made if the results of ingestion studies are compared, for the purposes of uptake measurement, with the results of separate injection studies. In this way the integrated blood/urine levels can be compared without the use of a second isotope.

Using these methods the Harwell group have undertaken three aluminium bioavailability studies—two following ingested aluminium and one following inhaled aluminium. All of these studies were conducted to GLP (good laboratory practice) standards and were subject to quality control. They also measured (using gamma-spectrometry) retained ²⁶Al in administration tubes/vessels *etc.*, to confirm the administered dose. The first of the ingestion studies was undertaken to: determine the bioavailability of ingested aluminium, as aluminium citrate and as the hydroxide; measure the effects of the simultaneous ingestion of citrate on aluminium absorption; determine the kinetics of aluminium clearance from the digestive tract.^{47,165} Aluminium citrate and aluminium hydroxide were chosen for the experiments since experience with a wide range of other metals and the available literature for aluminium, suggested that the soluble, aluminium citrate complex would be amongst

the most bioavailable of ingested aluminium compounds, while the hydroxide, as a relatively insoluble suspension, would be amongst the least bioavailable. Two male volunteers were used for this study. They were fed, through a paediatric feeding tube (inserted into the stomach *via* the nose) to ensure the precise delivery of ~ 115 Bq of ²⁶Al in the appropriate chemical form to the stomach. Urine, faecal and blood samples were collected for 5 days after administration of the labelled aluminium preparations.

The results showed differences between the kinetics of aluminium uptake in the two volunteers as determined by blood and excretion data. Also, unabsorbed ²⁶Al was retained for about 1 day longer within the gastro-intestinal tract, prior to excretion in faeces, in one volunteer (Fig. 22). It follows that the higher aluminium uptake that was reported for this volunteer was probably due to the longer period available for uptake—a suggestion supported by his more protracted period of aluminium excretion and higher blood concentrations at 24 h post-intake. This is possible since the most likely site for aluminium retention in the gut is within the small intestine associated with the walls of the villi—a site where many metals may be temporarily retained. Inter-subject differences in the kinetics of ²⁶Al transfer to blood were reported and the impossibility of obtaining meaningful gut transfer factors from the ²⁶Al in blood data was highlighted.

The co-administration of citrate with the aluminium hydroxide suspension was found to enhance the uptake of the radiotracer in both volunteers. Using a urinary excretion factor based on the results of the earlier published injection studies, the absorbed fraction for each of the doses were estimated: aluminium as aluminium citrate, 5.2×10^{-3} (0.5%); aluminium as aluminium hydroxide, 1.0×10^{-4} (0.01%); aluminium hydroxide with citrate, 1.4×10^{-3} (0.1%). It can be seen that the range of absorbed fractions observed, from approximately 1% to 0.01%, closely matches expectations based on the results of either studies using stable aluminium or calculations based on biokinetic measurements. It is also consistent with the study of Hohl *et al.*¹⁶⁶ that administered 100 ng of ²⁶Al in the presence of 100 μg of ²⁷Al carrier as an aluminium chloride solution. These authors measured uptakes of 0.1 and 0.24% in two subjects, respectively.

The second ingestion study was designed to measure aluminium uptake from drinking water.^{48,167} For this study young, adult, male volunteers received a single oral dose of drinking water, supplied by the Sydney (Australia) Water

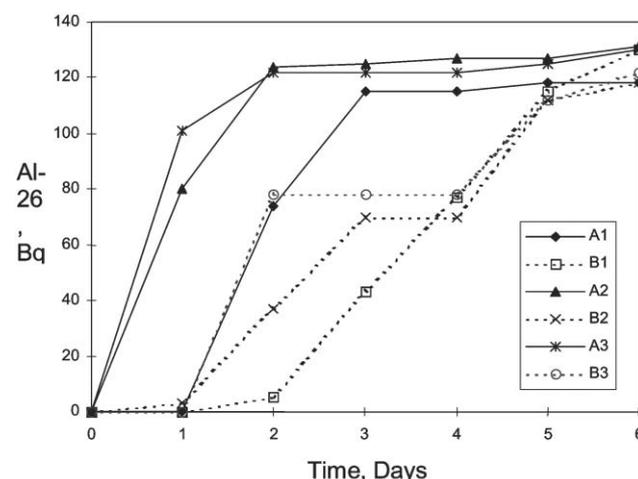


Fig. 22 Ingestion study. Cumulative faecal excretion of ²⁶Al for volunteers (A and B) following the administration of labelled aluminium hydroxide (1), labelled hydroxide in the presence of citrate (2) and labelled citrate (3). It can be seen that, compared with A, volunteer B consistently retained aluminium within his gut for 1 to 2 days longer—allowing time for extra uptake. (From ref. 47)

Board, and containing 100 Bq of ^{26}Al . The doses were given in the morning after overnight fasting. Blood, urine and faeces were collected and analysed for ^{26}Al using either γ -spectrometry or AMS. The levels in faeces were used to confirm the administered dose of ^{26}Al . Based on the urinary output of ^{26}Al previously given to other subjects by intravenous injection, a fractional absorption of 2.2×10^{-3} (0.22%) for aluminium in drinking water was calculated. As in the previous study, one of the volunteers retained aluminium in the gut for about 24 h longer than the other volunteer and exhibited a slightly higher level of aluminium uptake (0.26 cf. 0.18%). The results of this study, particularly since it was undertaken under fasted conditions (when absorption may be higher) show that drinking water is unlikely to be a major source of aluminium in the body. A similar conclusion was reached following another study using stable aluminium¹²⁶ where the absorbed fraction of aluminium in drinking water was 0.36%. Consequently, it is difficult to reconcile the above with suggestions that exposure to aluminium in drinking water may result in neurotoxicity.⁹⁸⁻⁹⁹ A recent publication¹⁰¹ purports to describe cognitive changes in people exposed to drinking water contaminated with aluminium sulfate during the Camelford incident, but this paper has been widely criticised. Finally, a rat study using ^{26}Al assessed the bioavailability of aluminium in drinking water to be $\sim 0.3\%$ —consistent with human data.

Other studies have been undertaken to determine the effect of the co-ingestion of silicon on aluminium absorption¹⁶⁸ and the relative levels of aluminium uptake in normal subjects and Down's syndrome patients (who develop AD in adult life).^{28,169} The results produced for silicon are presented in Fig. 23. This indicates a marked effect—an approximately 5-fold reduction in uptake, of $100 \mu\text{M L}^{-1}$, of dissolved silicon in $75 \text{ ng } ^{26}\text{Al}$ in orange juice for all subjects tested. In addition, it is suggested that silicon intakes, as orthosilicic acid, may facilitate the renal excretion of aluminium.¹⁷⁰ Consequently, the co-ingestion of silicon with aluminium may both reduce aluminium bioavailability by the formation of insoluble aluminosilicate complexes within the gut contents and increase the rate of excretion of that aluminium which crosses the gut barrier. Using the same methods an excess ^{26}Al uptake was seen in Down's syndrome patients, compared with controls and a 1.6 times excess ^{26}Al uptake has been described in Alzheimer's patients compared with age-matched controls.¹⁷¹ However, all of these studies employed the levels of ^{26}Al in blood at either 1, 4 or 6 h post-administration to assess uptake. As described previously, this

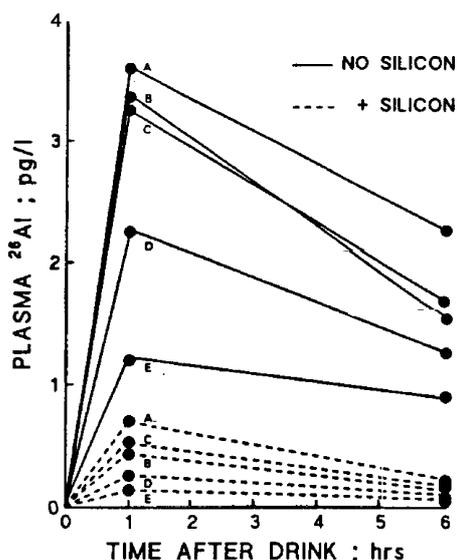


Fig. 23 A comparison of the plasma concentration of ^{26}Al in volunteers at either 1 or 6 h following the ingestion of this isotope in orange juice. (From ref. 168).

method is unlikely to yield an accurate result—they do, however, produce valuable relative data. Nevertheless, T. I. Lidsky (personal communication) has suggested that the result seen for AD patients, together with a disrupted blood–brain barrier in such patients¹⁷² could explain the excess aluminium that has been observed in the brains of these patients. The results of studies using ^{26}Al in man are shown in Table 5. The presence of calcium and magnesium carbonates in hard water does not appear to modify the oral bioavailability of aluminium in drinking water.⁶

To date, no studies have been conducted to investigate the oral bioavailability of aluminium in pharmaceutical preparations—other than those concerned with aluminium hydroxide. It follows that the bioavailability of aluminium in the antacid sucralate and in dental products—as aluminium lactate—has yet to be established, although it is likely that any such investigations would show uptakes within the range previously measured for other substances.

9 Measurement of bioavailability following inhalation

9.1 Exposures to inhaled aluminium

While the gut wall presents a substantial barrier to the uptake of aluminium, the lung-blood barrier presents little, if any, obstacle to the uptake of aluminium in solution. Inhaled aerosols are either exhaled, deposited in the nose, trachea and bronchioles, or are deposited within the alveolar tissues of the lung in fractions varying according to their size.³ Particles deposited in the airways are mostly rapidly cleared and swallowed, presenting little chance for their dissolution and absorption, but material deposited in the alveolar region is not cleared mechanically. Instead it is engulfed by macrophages, which are the lung's scavenger cells and is mostly retained until it is dissolved, whereupon it leaves the cells and enters the blood stream; a few macrophages, with their particles, may migrate, via the lymphatic system, to the blood stream—these will mostly deposit in the regional lymph nodes and liver (see Fig. 21). In general, the mass of aluminium-containing particles in the ambient atmosphere is small (estimated daily intake by inhalation is $4.4 \mu\text{g}$) and many are likely to be either too large or too small for efficient deposition in the deep lung. Furthermore, most particles will be essentially insoluble and, therefore, add little to the body's aluminium burden. Consequently, under normal circumstances the uptake of aluminium by the inhalation route can be ignored as a significant source of body contamination. In contrast evidence exists that some workers may have accumulated substantial amounts of

Table 5 Absorption of ingested ^{26}Al compounds by man (data obtained by extrapolation from blood samples are shown in italics)

Subject	Administered form	Absorbed fraction	Reference
Normal	Aluminium citrate	5.2×10^{-3}	Ref. 47
Normal	Aluminium hydroxide	1.0×10^{-4}	Ref. 47
Normal	Aluminium hydroxide plus citrate	1.4×10^{-3}	Ref. 47
Normal	Drinking water	2.2×10^{-3}	Ref. 48
Normal	Drinking water	1.7×10^{-3}	Ref. 166
Normal	Aluminium citrate	1.0×10^{-2}	Ref. 37
Normal	Orange juice	1.5×10^{-4}	Ref. 168
Normal	Orange juice plus silicon	2.0×10^{-5}	Ref. 168
Normal	Orange juice	9.0×10^{-5}	Ref. 28
Down's syndrome	Orange juice	4.7×10^{-4}	Ref. 28
Normal	Orange juice	6×10^{-4}	Ref. 171
Alzheimer's disease	Orange juice	1×10^{-3}	Ref. 171

aluminium in their lungs and bodies¹⁷³ as a consequence of their employment

9.2 Occupational exposure to aluminium by inhalation

It is likely that a number of occupational groups are currently exposed to aluminium and/or aluminium compounds as a consequence of their employment. These groups include workers on rocket launch pads—exposed to the alumina powders exhausted by solid-fuel rocket motors, aluminium arc sprayers, and welders—exposed to aluminium vapour condensation fumes,^{57,174} and workers within the aluminium production industry—exposed to a range of aluminium species, ranging from bauxite ore to aluminium fume.^{31,122,175–177} All of these may excrete much higher than normal levels of aluminium. In addition, during the early part of the last century, hard-rock miners in Canada and elsewhere were deliberately exposed to a black powder comprising ultra-fine aluminium powder and powdered aluminium minerals.¹⁷⁸ The inhalation of this mixture, known as McIntyre powder, by workers at the end of each work shift was believed to prevent lung silicosis—a crippling lung disease that was common in hard-rock miners. It can be concluded that, while aluminium uptakes by the general public result mostly from the ingestion of the metal, many uptakes of aluminium by workers result from inhalation.

Of the above groups, workers within the modern aluminium industry have been most extensively studied, with respect to their exposure to aluminium and aluminium intakes. Within the aluminium industry exposures to aluminium have been monitored, by measuring the levels of this metal that are either excreted each day or are present in plasma, as a function of occupation type. The results obtained in one major study of the French aluminium industry¹⁷⁶ are presented in Fig. 24. This shows that compared with the general population, which excretes $<10 \mu\text{g g}^{-1}$ creatinine (equivalent to about $6 \mu\text{g day}^{-1}$) of aluminium, a few aluminium workers may excrete as much as $100+ \mu\text{g g}^{-1}$ creatinine. In general, the workers with the highest aluminium excretion rates (and plasma aluminium levels) were employed either within the potrooms, where aluminium is smelted, or on aluminium powder production facilities. It is speculated that high levels of aluminium excretion result either from the inhalation of ultra-fine condensation aerosols of aluminium or from the inhalation of soluble compounds of aluminium. The inhalation of high masses of aluminium welding fume—an ultra-fine condensation aerosol—sometimes results in even greater levels of aluminium

excretion.^{57,174,177,179} In one study¹⁸⁰ the median urinary aluminium concentration, in a group of welders, was $40.5 \mu\text{g L}^{-1}$ (range $19\text{--}130 \mu\text{g L}^{-1}$). Over a working lifetime intakes of this size could result in persistent aluminium body burdens within the range $100\text{--}800 \text{ mg}$. At such intake levels/body burdens neuropsychological effects are expected¹⁸¹—although other studies have failed to identify a problem.¹⁸²

9.3 Bioavailability of inhaled ²⁶Al oxide

Given the importance of inhalation, within the context of the aluminium industry, as a source of aluminium intake a study was undertaken to determine the bioavailability of an inhaled “industrial-type” aerosol. The objectives of the study²⁴ were: to determine the retention and absorption characteristics of inhaled ²⁶Al-labelled transitional aluminium oxide deposited in the respiratory tract of male human volunteers; to determine the fraction of the inhaled aluminium that is transferred from lungs to blood and then excreted in urine; to estimate, by the assay of faeces, the fraction of the inhaled aluminium that is mechanically removed from the lung *via* the upper respiratory tract, swallowed and excreted.

A solution of aluminium nitrate, containing ²⁶Al, was dispersed to produce an aerosol, which was dried in a drying tube and calcined in a furnace. The oxide particles produced had a mean aerodynamic diameter of $1.2 \mu\text{m}$ and comprised transitional oxides as determined by X-ray diffraction. Particles were re-suspended and inhaled by two subjects. After inhalation the estimated intakes of ²⁶Al were $16 \pm 5 \text{ Bq}$ for one subject and $6 \pm 5 \text{ Bq}$ for the other. After short-term mechanical clearance, $9 \pm 5 \text{ Bq}$ (13 ng) and $4 \pm 5 \text{ Bq}$ remained in the subjects, respectively. ²⁶Al levels in urine were determined. Measurements made at different times post-inhalation showed that by 900+ days all measurable ²⁶Al excretion had ceased (Fig. 25). The masses of ²⁶Al excreted by both volunteers were little changed during the first months of measurement—with about 0.02% of the ILD cleared each day. The corresponding retention half time, for loss by dissolution, was about 3000 days (8.25 years). In contrast, the amount of aluminium tracer excreted each day decreased with a half time of 87 days in subject A and 89 days in subject B. This, in the absence of significant dissolution, was taken to indicate that most of the oxide particles deposited in the lung were mechanically cleared from the body—before they had time to dissolve. The calculated value for the fraction of the initial respiratory deposit of aluminium that is transferred to blood

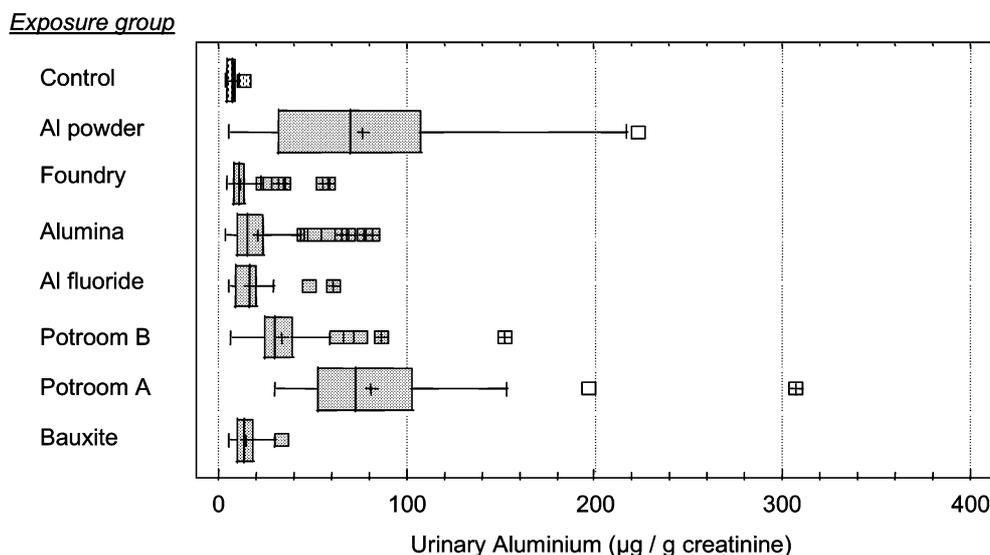


Fig. 24 Urinary aluminium concentrations for 41 volunteers working in French aluminium plants, by occupation/exposure type. It can be seen that aluminium powder and flake workers excrete most aluminium. (From ref. 176). NB. Adult males excrete about 1.7 g of creatinine per day.

Combined Data - Urinary Excretion

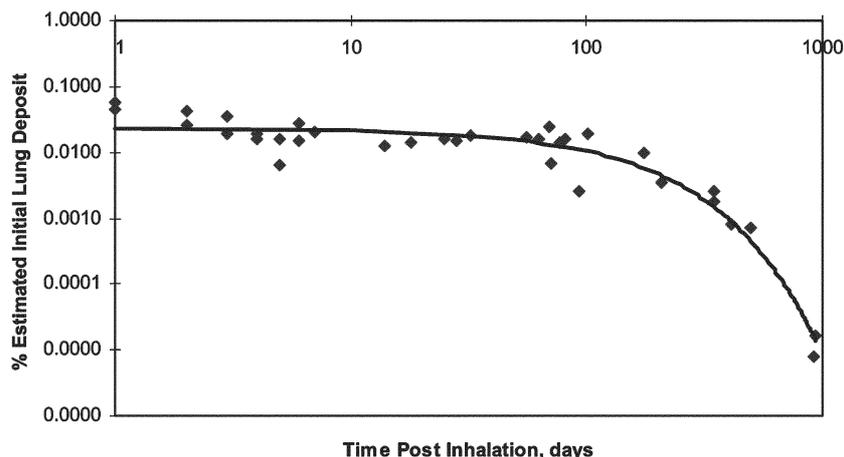


Fig. 25 Graph showing the combined urinary excretion data for subjects A and B plotted as a function of time post-inhalation of ^{26}Al -labelled transitional aluminium oxide. The low excretion levels indicate that most aluminium oxide was lost from the lungs by mechanical clearance.⁴⁵

was 1.9%. This value is completely consistent with the fractional aluminium uptake by workers of about 1.5 to 2.0% estimated from aluminium in air and aluminium in urine data.^{103,175,183} Given that approximately 98% of the aluminium that enters the blood is excreted in urine the amount retained in the body was much lower. It was calculated that for every mg of aluminium, inhaled as 1 μm MMAD transitional alumina particles, only about 500 ng of aluminium would be retained by the body.

This study demonstrated that inhaled aluminium oxide was too insoluble to effectively contribute to the aluminium body-burden of workers and it was concluded that where high levels of aluminium excretion were seen in workers these must have resulted from the inhalation of other, more soluble forms of aluminium. Similarly, it is likely that the inhalation of insoluble, environmental aluminium-containing aerosols, such as mineral dusts, will result in little aluminium transfer to blood. On the other hand, the inhalation of more soluble aluminium compounds, e.g., aluminium chlorohydrate antiperspirant aerosols, may result in larger fractional transfers, but to date, no studies have examined this possibility.

10 Bioavailability following intakes by other routes

Aluminium salts present in aerosol antiperspirant and "roll on" gel antiperspirant preparations—typically aluminium chlorohydrate—may enter the body either by trans-dermal absorption or from skin wounds caused by the removal of pubic and axillary hair. A study using ^{26}Al -labelled aluminium chlorohydrate undertaken using two volunteers at the Perdue University¹⁸⁴ showed the uptake of 0.012% of the tracer applied. It is also known that the spraying of under-arm antiperspirants onto abraded skin produced during the process of razing axillary hair results in the intake of some aluminium: Freemont and his colleagues have described granulomas as resulting from this practice.¹⁸⁵ Similarly, granulomas have been described following the injection of the triple vaccine.¹⁸⁶

The uptake of aluminium through wounds is generally considered to be of little consequence except in the situation where aluminium is injected into the body as a vaccine adjuvant. Such adjuvants are added to vaccines to potentiate the immune response of the antigen. One of two adjuvants is commonly employed, either aluminium hydroxyphosphate or aluminium oxyhydroxide—both are insoluble and adsorb charged antigens at physiological pH 7.4. As these are injected directly into the body then they circumvent the body's protective barriers and when they dissolve they can directly

enhance the body burden of aluminium. For example, twenty injections, each containing 0.5 mg of aluminium, could release 10 mg of aluminium into the body (it might take 4000 days to reach the same level of intake from normal dietary intakes) of which 0.2 mg would be retained as a persistent body burden—making vaccines an important source of body aluminium; this is particularly true for children. For example, body burden calculations were made assuming hepatitis B vaccinations (each containing 0.25 mg Al) at birth, 2 months, and 6 months and diphtheria plus tetanus toxoids (each containing about 0.5 mg Al) at 2 months, 4 months, 6 months and 12 months by Keith *et al.*¹⁸⁷ These suggest that at age one year the body burden of aluminium (~ 0.2 mg) will exceed that resulting from the ingestion of breast milk (~ 0.1 mg), but is less than that calculated assuming aluminium intakes at the ATSDR minimal risk level for oral intake (2 mg Al per day) ~ 2 mg. Studies using ^{26}Al in rabbits⁶³ indicate that aluminium hydroxyphosphate dissolves approximately three times faster than aluminium oxyhydroxide. However, the dissolution rate of both were slow with average rates of $8.1 \times 10^{-4} \text{ mg h}^{-1}\text{g}^{-1}$ and $2.7 \times 10^{-4} \text{ mg h}^{-1}\text{g}^{-1}$, respectively, during the first 28 days after intra-muscular injection. Undissolved particles will be phagocytosed by macrophages.

In addition, a new type of histopathological lesion—macrophagic myofasciitis—has been described following MMR and other injections.¹⁸⁸ This lesion, which is described in detail by Gherardi *et al.*,¹⁸⁹ is associated with macrophages loaded with insoluble aluminium salt deposits. The lesion is not, in itself, serious and is probably not caused by bioavailable aluminium, but it may, however, be linked with diffuse arthromyalgias and fatigue. This condition has been described in France following the administration of hepatitis A, hepatitis B and tetanus toxoid vaccinations. Other sources of wound uptake, such as the entry of aluminium-containing clays into cuts and grazes, are unlikely to add substantially to the body burden of this metal, even in the most adventurous of little boys.

The situation regarding the release of aluminium from some bone-joint prostheses has not been investigated—although, these do wear, releasing black wear debris into surrounding tissues, some of which will dissolve and add to the aluminium body burden.

11 Conclusion

The literature demonstrates that the biokinetics and bioavailability of aluminium and its compounds are typical of those for

other trivalent metals, including the lanthanides and some actinides. Such metals are of low solubility, form insoluble hydroxides at neutral pH, have a low bioavailability and are retained by the skeleton. In the case of aluminium, studies with ^{26}Al have shown that about 2% of aluminium entering the blood is retained within the body for years, but that the remainder is excreted—the vast majority in urine. Within blood, most aluminium is bound to the iron transport protein transferrin—but the strength of binding is low and the metal is readily removed from blood in the kidneys. As a consequence of the retention of some aluminium it is predicted that aluminium body-burdens will increase as a function of time. Incremental increases in body aluminium will be largest for aluminium workers—inhalng soluble aluminium fumes—and in patients given large oral doses of aluminium-containing antacids and phosphate binders.

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