The Al-Zn of element toxicity: A summary of the toxicological information on 24 elements.

Introduction

This document is a summary of the available toxicological information on the elements included in the recent 2006 Food Standards Agency Total Diet Study (TDS). The information in this summary has been taken from toxicological reviews including Joint FAO/WHO Expert Committee on Food Additives and Contaminants (JECFA) monologues, Environmental Health Criteria, the Expert Group on Vitamins and Minerals Report, the WHO Drinking Water Guidelines, the US EPA Integrated Risk Information System, and ATSDR toxicological profiles. Literature searches using PubMed have also been carried out in an attempt to fill in any gaps in the data and to provide more recent references where necessary. The name of each element was used as a keyword when performing a search, including any synonyms or alternative spellings. Key studies identified by the reviews used in writing this summary are mentioned where possible. This summary has focussed where possible on the oral toxicity of each of the elements.

This summary includes any conclusions resulting from previous evaluations by COT, COM, COC, JECFA and any other relevant scientific advisory committees or regulatory bodies.

1. Aluminium (Al)

The key sources of the toxicological information in this summary were the evaluations by JECFA carried out in 2007, 1989 and 1977, the evaluation of aluminium intake by the European Food Safety Authority (EFSA, 2008), and Environmental Health Criteria 194 (WHO, 1997).

Chemistry and occurrence

Aluminium is too reactive to be found free in nature, where aluminium exists only in the oxidation state Al^{3+}. Aluminium can complex with a variety of ligands, and can form relatively strong bonds with water molecules and is often found as a hexavalent complex with water molecules. This complex behaves as a weak acid.

Being the third most abundant metal, aluminium is present in food and water as a natural contaminant. Aluminium in foods results from its natural occurrence, from the use of aluminium-containing food additives and from the presence of aluminium in food contact materials. In the UK, the Miscellaneous Food Additives Regulations 1995 permit the use of aluminium silicates in a limited number of foods at specified maximum levels, for example, up to 1% by weight in "sliced or grated hard, semi-hard, and processed cheese". Aluminium metal may also be used to *quantum satis* level (in line with good manufacturing practice) only for "external coating of sugar confectionery for the decoration of cakes and pastries" under The Colours in Food Regulations 1995. Foods that contain the highest concentration of aluminium include breads, cakes, pastries and biscuits (EFSA, 2008). Due to the design of the available human dietary studies and the analytical methods used, it is not possible to assess the contribution of aluminium from the various sources and thus only total aluminium exposure can be determined (EFSA, 2008).

The daily mean intake of aluminium from food in European adults ranges between 1.6 and 13 mg (EFSA, 2008). This is between 90 and 95% of total intake (WHO, 1997). Large individual variations in dietary exposure to aluminium can occur, with children representing the group with the highest potential exposure to aluminium per kg body weight (EFSA, 2008). Some over-the-counter medicinal products and consumer products such as anti-perspirants contain high levels of aluminium. Use of these products as well as occupational exposure to aluminium can result in far higher exposure to aluminium compared to that anticipated from the diet (EFSA, 2008).

Absorption and elimination

In healthy individuals aluminium is poorly absorbed from the digestive tract. This is probably due to the formation of insoluble aluminium phosphate that cannot be absorbed and is subsequently excreted in the faeces (Spencer & Lender, 1979). It is widely assumed that soluble aluminium compounds are more bioavailable than insoluble compounds (WHO, 2007). The net

absorption of food aluminium is approximately 1% (WHO, 1997, 2007). Urine is the major excretory route of absorbed aluminium.

Individuals with kidney damage or renal failure appear to have a reduced capacity for excreting aluminium, resulting in higher plasma levels and an increased risk of aluminium induced neurotoxicity (WHO, 1989).

Toxicity in animals

The toxicity of aluminium is influenced by the solubility. The acute toxicity of metallic aluminium and aluminium compounds is low, the reported oral LD_{50} values being in the range of several hundred to 1000 mg aluminium/kg body weight/day (WHO, 1997, 2007). Recent studies reviewed by JECFA indicated effects of aluminium compounds at doses lower than those reviewed previously by the Committee. Studies in rats, rabbits and monkeys have identified effects on enzyme activity, oxidative damage and calcium homeostasis in short-term studies with aluminium at oral doses in the region of 10-17 mg/kg body weight/day (WHO, 2007). Histopathological changes have been identified in the kidney and liver of rats administered aluminium sulphate by gavage, at a dose of 17 mg/kg body weight/day for 21 days; as well as in the kidney and brain of rats administered aluminium chloride in the drinking water at a dose of 30 mg/L for 6 months (WHO, 2007).

There is considerable evidence that aluminium is neurotoxic in experimental animals, although there is considerable variation among species (WHO, 1997, 2007). In susceptible species, toxicity following parenteral administration is characterised by progressive neurological impairment, resulting in death with status epilepticus (LD_{50} = 6 µg Al/g dry weight of brain in rabbits, the normal brain aluminium concentration in healthy rabbits is approximately 1.1 µg/dry weight). Morphologically, the progressive encephalopathy is associated with neurofibrillary pathology in large and medium size neurones predominantly in the spinal cord, brainstem and selected areas of the hippocampus. These tangles are morphologically and biochemically different from those that occur in Alzheimer's disease. Behavioural impairment has been observed in the absence of overt encephalopathy or neurohistopathology in experimental animals exposed to soluble aluminium salts (e.g. lactate, chloride) in the diet or drinking-water at doses of 200 mg aluminium/kg body weight/day or more (WHO, 2007). The neurotoxic potential of aluminium has received increasing attention. Studies in animals displaying effects have been conducted using parenteral administration and are thus of uncertain relevance for dietary exposure (WHO, 1997, 2007). The available data from studies using oral administration do not demonstrate definite neuropathological effects but indicate the potential for soluble aluminium compounds to cause neurobehavioural effects at doses in the region of 50-200 mg/kg body weight/day.

Osteomalacia, as it presents in man, is observed consistently in larger species (e.g. dogs and pigs) exposed to aluminium; a similar condition is

observed in rodents. These effects appear to occur in all species, including humans, at aluminium levels of 100 to 200 µg/g bone ash (WHO, 1997).

Aluminium can form complexes with DNA and cross-link chromosomal proteins and DNA, but it has not been shown to be mutagenic in bacteria or induce mutation or transformation in mammalian cells *in vitro.* However, chromosomal aberrations have been observed in bone marrow cells of exposed mice and rats (WHO, 1997).

Animal studies do not indicate that aluminium or aluminium compounds are carcinogenic via the oral route. Some studies have indicated that inhalation of aluminium-containing fibres and particles may induce carcinomas in the lung. However, in these cases it is likely that the toxicity reflects the physical properties of the particles/fibres (3.5 µm median diameter). Similarly, aluminium implanted subcutaneously has induced soft tissue carcinomas at the site of implantation, but in these cases also the effects are probably related to a chronic foreign body reaction rather than to the aluminium ion itself (WHO, 1997). No new studies of genotoxicity or carcinogenicity were identified in JECFA's most recent evaluation (WHO, 2007).

A complication of many animal studies is that the basal aluminium content of the diet is not taken into account before addition of the test material, and so total exposure is unknown (WHO, 2007).

Toxicity in humans

No acute pathogenic effects in the general population have been described after exposure to aluminium.

Long-term administration to humans of aluminium-containing antacids (resulting in aluminium doses much higher than those from food) such as aluminium hydroxide results in decreased plasma concentration of phosphorus because of decreased phosphorus absorption or increased deposition of phosphorus in bone as aluminium phosphate. Aluminium antacids may cause an inhibition of intestinal absorption of phosphorus and this may be followed by an increase in calcium loss (Spencer & Lender, 1979, WHO, 1989).

Exposure of patients with chronic renal failure to aluminium-containing dialysis fluids and pharmaceutical products, has been reported to cause encephalopathy, vitamin-D-resistant osteomalacia and microcytic anaemia.

It has been suggested that aluminium exposure is a risk factor for the development or acceleration of onset of Alzheimer's disease in humans. There are no reports of the toxicity of dietary aluminium to healthy individuals in the literature (Soni *et al.,* 2001). Soni *et al.* (2001) concluded that the present data do not support a causative role for dietary aluminium in Alzheimer's disease.

JECFA recently reviewed a number of epidemiological studies investigating the potential association of oral exposure to aluminium in water, food or antacids with Alzheimer's disease and cognitive impairment (WHO, 2007). Some studies of aluminium in drinking water suggested an association with Alzheimer's disease, but other studies did not confirm this association. None of the studies accounted for ingestion of aluminium in foods. There was minimal information on the association between intake of aluminium in food and neurological conditions. Epidemiological studies on the use of antacids did not capture dose information and did not demonstrate an association with neurological conditions. EFSA concurred, concluding that the implication of aluminium in the aetiology of Alzheimer's disease remains controversial but that based on the available scientific data, exposure to aluminium via food is not considered to constitute a risk for developing Alzheimer's disease (EFSA, 2008).

The International Agency for Research on Cancer (IARC) has not evaluated the carcinogenicity or genotoxicity of aluminium. However, EFSA concluded that aluminium is unlikely to be a human carcinogen at dietary relevant doses (EFSA, 2008).

Previous evaluations

Having previously set an ADI 'not specified' for aluminium, in 1989 JECFA set a Provisional Tolerable Weekly Intake (PTWI) for aluminium and its salts of 0-7 mg/kg body weight expressed as aluminium (WHO, 1989). More recently, the PTWI was revised because aluminium compounds were considered to have the potential to affect the reproductive system and developing nervous system at doses lower than those used in establishing the previous PTWI. The Committee based its evaluation on evidence from several dietary studies with LOELs in the region of 50-75 mg/kg body weight/day for different species (mice, rats, dogs). An uncertainty factor of 300 was applied to the lower end of this range to allow for inter- and intra-species variation (100) and deficiencies in the database (3), resulting in a PTWI of 1 mg/kg body weight, which applies to all aluminium compounds, including additives (WHO, 2007). The Committee noted that the PTWI is likely to be exceeded by some population groups.

EFSA also based its evaluation on the combined evidence from several studies showing adverse effects on testes, embryos and the developing and mature nervous system following dietary administration. EFSA derived the same TWI of 1 mg/kg body weight and concluded that this TWI is likely to be exceeded in a significant part of the European population (EFSA, 2008).

The (P)TWI of 1mg/kg bw is based on the most recent information and therefore is considered to be the most relevant.

Previous COT evaluatons

In 1985 the COT considered aluminium in food. The estimated intake from food by adults was approximately 0.39 mg/kg body weight/week and the estimated intakes from food by infants (3-6 kg) were 0.32 to 0.62 mg/kg body weight/week (consuming soya-based food) and 0.035 to 0.06 mg/kg body weight/week (consuming cows milk-based food). The COT commented that the levels of aluminium in food were not a cause for concern. The Committee also stated that 'the relationship between aluminium and Alzheimer's disease remains unclear and is the subject of continuing research. There is no evidence that aluminium in food affects the occurrence of Alzheimer's disease.'

In 2003, the COT evaluated aluminium in food when the results of the 2000 TDS were considered (COT, 2003). The estimated population dietary exposure to aluminium from the 2000 TDS was 4.7 mg/day, approximately 78 µg/kg body weight/day for a 60 kg adult. The Committee noted that the estimated exposures were below the then current JECFA PTWI of 7mg/kg bw and concluded that current dietary exposures to aluminium were unlikely to be of any toxicological concern for consumers.

References

COT (2003). Statement on twelve metals and other elements in the 2000 Total Diet Study. COT Statement 2003/07. Annual Report 2003 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Foods, Consumer Products and the Environment.

EFSA (2008). Safety of aluminium from dietary intake. Scientific Opinion of the Panel on Food Additives, Flavourings, Processing Aids and Food Contact Materials (AFC). *The EFSA Journal*, **754**, 1-34.

Katz, A.C. (1981). A 6-month subchronic dietary toxicity study with Levair (sodium aluminium phosphate, acidic) in beagle dogs. Unpublished report by Stauffer Chemical Co., Farmington, Connecticut. Submitted to WHO by US FDA, 1982.

Soni, M.G., White, S.M., Flamm, W.G. and Burdock, G.A. (2001). Safety evaluation of dietary aluminium. Regulatory Toxicological Pharmacology 2001 **33(1)**: 66-79

Spencer, H. & Lender, M. (1979). Adverse effects of aluminium- containing antacids on mineral metabolism. *Gastroenterology,* **76**: 603-606.

WHO (1977). Summary of toxicological data of certain food additives. WHO Food Additives Series 12.

WHO (1989). Evaluation of certain food additives and contaminants. Thirty-third Report of the Joint FAO/WHO Expert Committee on Food Additives. *WHO Technical Report Series* No. **776**. World Health Organization, Geneva.

WHO (1997). Environmental Health Criteria 194: Aluminium. World Health Organisation, Geneva.

WHO (2007). Safety evaluation of certain food additives and contaminants. WHO Food Additives Series 58. Prepared by the Expert Committee on Food Additives (JECFA). World Health Organization, Geneva.

2. Antimony (Sb)

The key source of information was the WHO drinking water guideline document (WHO, 2003). A review of antimony produced by Elinder and Friberg (1979) was also used.

Chemistry and occurrence

Antimony (Sb) is in Group V, sharing some chemical properties with lead, arsenic, and bismuth. The most stable valency states of antimony are Sb^{3+} (trivalent) and Sb^{5+}(pentavalent). Numerous inorganic and organic compounds of antimony are known. Most of the common antimony compounds are slightly/readily soluble in water. The compounds are used in flame proofing, paints, ceramics, glass, pottery, rubber technology and dyestuffs. Subcutaneous injections of pentavalent antimony (sodium stibogluconate and meglumine antimoniate) are also used to treat leishmaniasis in both animals and humans.

There is no information on the forms of antimony that would be found in food.

Absorption and elimination

Antimony, even in soluble forms, is not readily absorbed from the gastrointestinal tract, irrespective of its valency state. Absorption rates between 5% and 20% have been observed in animals. However, estimation of actual absorption rate is complicated by simultaneous gastrointestinal excretion (Elinder and Friberg, 1979). Examination of four persons after involuntary acute intoxication with antimony potassium tartrate revealed a human absorption rate of 5% (WHO, 2003).

In experimental animals, after absorption, antimony is bound to the red blood cells and then transported mainly to the spleen, liver and bone, and to some extent into skin and hair. It is unknown to what extent inorganic and organic Sb^{5+} may be reduced to Sb^{3+} *in vivo* (WHO, 2003).

Sb^{3+} in the form of $Sb(OH)_3$ can react strongly with cell membranes, especially erythrocyte membranes, due to its lack of electrical charge. This seems to be the reason for its longer elimination half time (94h) compared to Sb^{5+} (24h) (WHO, 2003).

Toxicity in animals

In a lifetime study, rats given 5 mg/L antimony potassium tartrate in drinking water from the time of weaning showed significantly shortened survival compared with controls (Schroeder *et al.,* 1970).

In a limited lifetime study in which rats received antimony in drinking-water at a single dose level of 0.43 mg/kg body weight/day, effects observed were decreased longevity and altered blood levels of glucose and cholesterol. No effects were observed on the incidence of benign or malignant tumours (Schroeder *et al.* 1970).

In a study by Poon *et al.* (1998), rats given antimony in drinking water showed no overt signs of toxicity in any of the treatment groups apart from a marked but reversible loss of body weight gain in the males of the highest treatment group (60 mg/kg body weight/day). However Poon *et al*. (1998) identified a NOAEL of 0.06 mg/kg body weight/day based on subtle histopathological changes in the thyroid glands (increased epithelial height, decreased follicular size) of male rats.

Lynch *et al*. (1999) questioned the authors evaluation of the otherwise "generally well designed study" pointing to the reversible/adaptive nature of the "critical" thyroidal and the other biochemical and histological observations in this study, the absence of any quantitative dose/response-relationship although a more than 1,000-fold dose range was applied, and the high physiological variability and/or treatment-related occurrence of the observed "critical" effects. Instead of 0.06 mg antimony/kg body weight/day, Lynch et al. (1999) proposed a subchronic NOAEL of 6.0 mg antimony/kg body weight/day. This NOAEL was based on the decreased body weight gain and reduced food and water intake observed at the LOAEL of 60 mg antimony/kg body weight/day.

In *in vivo* carcinogenicity studies, both antimony trioxide and antimony trisulphide produced a significant increase in the incidence of lung tumours (scirrhous and squamous cell carcinomas and bronchioloalveolar tumours) in females following inhalation exposure. No tumours were seen in male rats. In contrast, studies in animals exposed to antimony via the oral route showed no evidence of carcinogenicity (WHO, 2003).

There are conflicting data regarding the mutagenicity and clastogenicity of antimony compounds *in vivo* and *in vitro*. *In vivo* studies of the clastogenicity of antimony trioxide have shown that it is not genotoxic, despite a positive result from one study, although the purity of the compound used has been questioned. Antimony potassium tartrate and bilharcid (piperazine antimony tartrate), two important antischistosomal drugs, were reported to be genotoxic after acute and subacute application to rats, however the application of these drugs was via the intraperitoneal route (WHO, 2003). There is no information on whether these are mutagenic or clastogenic via the oral route.

Toxicity in humans

Much of the information in humans is related to occupational exposure and therefore either inhalation or dermal exposure. However acute antimony

poisoning via the oral route has been reported to result in vomiting, nausea, abdominal cramps and diarrhoea (Elinder and Friberg, 1979).

There have been suggestions that antimony leaching from cot mattresses could be linked to Sudden Infant Death Syndrome (SIDS). However, in 1998, the Expert Group on Cot Death concluded that there is no evidence to suggest that antimony compounds used as fire retardants in polyvinyl chloride and other cot mattress materials are a cause of SIDS (DH, 1998).

Repeated oral exposure to therapeutic doses of Sb^{3+} was associated with optic nerve destruction, uveitides and retinal bleeding. Specific symptoms of intoxication are generally accompanied by headache, coughing, anorexia, troubled sleep and vertigo (Stemmer, 1976). Antimony-containing compounds may also produce alterations in cardiac function and autopsy studies have shown that cardiac toxicity was the cause of death in patients treated with antimonial drugs (Klaassen *et al.,* 1995)

There is no information on the oral carcinogenicity of antimony and compounds in humans. However based on inhalation data in rats and *in vitro* data, the IARC concluded that antimony trioxide is possibly carcinogenic to humans (Group 2B), and antimony trisulfide is not classifiable as to its carcinogenicity to humans (Group 3) (IARC 1989).

Previous evaluations

JECFA has not evaluated antimony.

In 1991 the US Environmental Protection Agency derived an oral reference dose for antimony of 0.4 µg/kg body weight/day (US EPA, 1991). This was based on the rat study of Schroeder *et al.* (1970), in which rats were administered antimony potassium tartrate in drinking water (5 mg/L). The concentration of 5 mg/L was expressed as an exposure of 0.35 mg/kg body weight/day and represented a LOAEL due to effects on longevity, blood glucose and cholesterol. An uncertainty factor of 1000 (100 for inter- and intra-species variation and 10 for the use of a LOAEL instead of a NOAEL) was applied to the LOAEL of 0.35 mg/kg body weight/day, giving a reference dose of 0.4 µg/kg body weight/day.

In 1993 the WHO proposed a TDI for antimony in order to determine a provisional guideline for an acceptable level of antimony in water (WHO 1993). The basis for the TDI was the rat study of Schroeder *et al.* (1970) in which rats were administered 0.43 mg antimony/kg body weight/day in drinking water. An uncertainty factor of 500 (100 for inter- and intra-species variation and 5 for the use of a LOAEL instead of a NOAEL) was applied to the LOAEL of 0.43 mg/kg body weight/day, giving a TDI of 0.86 µg/kg body weight/day.

The Committee on Mutagenicity (COM) considered mutagenicity data (*in vivo* and *in vitro*) on antimony trioxide in 1997. They concluded that antimony had been adequately investigated for its mutagenic potential *in vitro*. Negative results were obtained on Salmonella assays for gene mutation and in a mouse lymphoma assay. There was limited evidence of questionable significance for genotoxicity in DNA repair assays and some evidence for a clastogenic effect was documented in a metaphase analysis using human peripheral lymphocytes. There was no evidence for clastogenicity in a bone marrow micronucleus test using single and repeated oral doses of antimony trioxide, and negative results were also obtained from a rat liver UDS assay. The COM considered that these data suggested an absence of mutagenic activity following oral administration but Members noted that no indication had been given as to whether material had reached the bone marrow. Information on this was needed before definite conclusions could be drawn regarding other routes of exposure.

In 2003 the WHO produced a revised drinking water guideline (0.02 mg/L) in which a TDI of 6 μg/kg body weight/day was proposed (WHO, 2003). This guideline was maintained in the latest edition of the Guidelines for Drinking Water Quality (WHO, 2006) and was based on the NOAEL of 6 mg/kg body weight/day from a sub-chronic drinking water study (Poon *et al.,* 1998; Lynch *et al.,* 1999) and an uncertainty factor of 1000 (100 for intra- and inter-species variation and 10 for the use of a subchronic study). The WHO agreed with Lynch *et al*. (1999) regarding their evaluation of the sub-chronic drinking water study, choosing to use the NOAEL of 6 mg/kg body weight/day and not the NOAEL (0.06 mg/kg body weight/day) proposed by Poon *et al.* (1998). No reason was given for this decision.

This TDI is based on the most recent information and therefore considered to be the most relevant.

Previous COT evaluations

In 1998, the COT evaluated antimony in food when the results of the 1994 TDS were considered (COT, 1998). The estimated population dietary exposure to antimony from the 1994 TDS was 0.003 mg/day, approximately 0.05 μg/kg body weight/day for a 60 kg adult. The Committee concluded that there was no evidence to suggest that the estimated intake of antimony should be a cause for concern.

References

COT (1998). Statement on the results of multielement surveys in various items of the diet. COT Statement 1998. Annual Report 1998 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Food, Consumer Products and the Environment.

DH (1998). Expert group on cot death Final report. Department of Health. Available at: http://www.doh.gov.uk/limer.htm.

Elinder, C-G. and Friberg, L.G. (1979). Antimony. *In: Handbook on the Toxicology of Metals,* Editors; Friberg, L.G., Nordberg, F. and Vouk V.B., Elsevierorth-Holland Biomedical Press, New York, pages 283-292.

IARC (1989). Antimony trioxide and antimony trisulfide. IARC Monographs on the Evaluation of the Carcinogenic Risks to Humans. International Agency for Research on Cancer. World Health Organization.

Klaassen, C.D., Amdur, M.O. and Doull J. (eds.) (1995). Casarett and Doull's Toxicology. The Basic Science of Poisons. 5th edition. New York, NY: McGraw-Hill, 726

Lynch, B.S., Capen, C.C., Nestmann, E.R., Veenstra, G. and Deyo, A. (1999). Review of subchronic/Chronic Toxicity of Antimony Potassium Tartrate. Reg. Toxicol. Pharmacol. **30**: 9 – 17.

Poon, R., Chu, I., Lecavalier, P., Valli, V.E., Foster, W., Gupta, S. and Thomas, B. (1998). Effects of antimony on rats following 90-day exposure via drinking water. Food and Chemical Toxicology **36 (1)** :21-35.

Schroeder, H.A., Mitchener, M. and Nason, A.P. (1970). Zirconium, niobium, antimony, vanadium and lead in rats: life term studies. *J. Nutr.* **100:** 59-68.

Stemmer, K.L. (1976). Pharmacology and toxicology of heavy metals: antimony. Pharmac. Ther. A. **1**: 157 – 160.

US EPA (1991). Integrated Risk Information System (IRIS): Antimony. US Environmental Protection Agency National Center for Environmental Assessment.

WHO (1993). Guidelines for Drinking-Water Quality, 2nd ed., volume I: Recommendations. World Health Organization, Geneva.

WHO (2003). Guidelines for Drinking-Water Quality. Volume 1 Recommendations. Third edition. World Health Organization, Geneva.

WHO (2006). Guidelines for Drinking-Water Quality. Volume 1 Recommendations. First addendum to the third edition. World Health Organization, Geneva.

3. Arsenic (As)

The key sources of information for this summary were Environmental Health Criteria 224 (WHO, 2001), and the JECFA monographs on arsenic (WHO, 1983 and 1989).

Chemistry and occurrence

Arsenic has complex chemistry and forms a number of inorganic and organic compounds. It can exist in many oxidation states, the most common being the tri- and pentavalent forms. A variety of arsenates (AsO_4^{3-}, pentavalent arsenic) and arsenites (AsO_3^{3-}, trivalent arsenic) are found in water and at low levels in food.

Organic arsenic compounds such as monomethylarsenic (MMA) and dimethylarsenic (DMA) occur naturally in the environment, and are subject to bacterial demethylation to inorganic arsenic. Most arsenic in fish (>90%) is in the form of arsenobetaine which is also the main form found in crustaceans and bi-valve molluscs (Kohlmeyer *et al.*, 2002); the remainder is arsenocholine and a small amount of inorganic arsenic (usually ≤ 1%). Fish is the main source of arsenic in the diet; arsenobetaine is therefore the main form of arsenic present in food.

Absorption and elimination

Inorganic arsenic compounds (arsenite and arsenate) are well absorbed by the oral route in humans with absorption values reported to be from 50% to >95% (DEFRA/EA 2002). They are metabolised by methylation and then excreted in the urine with a half-life of 3 to 5 days (DEFRA/EA, 2002).

The fate of organic arsenic in man has not been clearly defined. In general. organoarsenicals are less extensively metabolised than inorganic arsenic and more rapidly excreted (WHO, 2001). Yamauchi *et al.* (1990) calculated biological half-lives after administration of organoarsenicals to hamsters, reporting a 6.1 hour half-life for arsenobetaine. In humans, exposure to arsenobetaine through consumption of plaice resulted in 69 to 85% of the arsenobetaine being excreted in the urine within 5 days (Luten *et al.* 1982). There are no data on tissue distribution of arsenic in humans following ingestion of arsenic present in fish and seafood. Animal studies show that following an intravenous dose of arsenobetaine the highest tissue concentrations were found in kidney, liver and pancreas, respectively, in both mice and rabbits (Vahter *et al*., 1983), and liver, kidney, spleen, muscle, skin and brain, in hamsters (Yamauchi *et al.,* 1986). Limited data indicate that organic arsenic compounds such as arsenobetaine and arsenocholine are not converted to inorganic arsenic (WHO, 2001).

Toxicity in animals

Since inorganic arsenic is considered to be less toxic in animals than humans and due to the lack of an appropriate animal model of toxicity, specifically the carcinogenicity, in man (DEFRA/EA, 2002), only the toxic effects in humans will be discussed.

There have been very few studies of the toxicity of organic arsenic in animals. In one study on organic (fish) arsenic, weanling rats were fed diets containing fish that provided a dose of approximately 3 mg/kg body weight/day organic arsenic for 42 days. No treatment-related toxic effects were reported in the limited range of endpoints studied (Siewicki, 1981).

Toxicity in humans

The organic forms of arsenic are generally considered to be less toxic than the inorganic compounds although few data are available (DEFRA/EA, 2002; WHO, 1989). There are no reports of toxicity in man from the consumption of organoarsenicals in seafood. The only information on toxicity in man comes from reports of populations who consume large quantities of fish resulting in intakes of organic arsenic of about 0.05 mg/kg body weight/day, with no subsequent reports of ill health effects (WHO, 1989).

Inorganic arsenic is clastogenic in *in* vitro and *in vivo* assays (some evidence suggests clastogenicity in humans) (IARC, 1980, 1987, 2002).

Arsenic in drinking-water (primarily inorganic, as arsenate and to a lesser extent arsenite) was evaluated as 'carcinogenic to humans' (Group 1) on the basis of 'sufficient evidence' for an increased risk for cancer of the urinary bladder (IARC, 2004), lung and skin (IARC, 1980, 1987). Increased risks of lung and bladder cancer and of arsenic-associated skin lesions and other skin changes (such as hyperkeratosis and pigmentation changes) have been reported to be associated with ingestion of drinking water at concentrations ≤ 50 µg arsenic/litre (WHO, 2001).

Chronic exposure to arsenic in drinking water has also been associated with cardiovascular diseases and peripheral vascular diseases such as blackfoot disease (WHO, 2001).

Previous evaluations

In 1983 the JECFA estimated a Provisional Maximum Tolerable Daily Intake (PMTDI) for inorganic arsenic of 2 µg/kg body weight/day (WHO, 1983). JECFA noted there was epidemiological evidence of an association between the overexposure of humans to inorganic arsenic from drinking-water and increased skin cancer risk: 0.2 mg/L was associated with a 5% increase in the

lifetime risk of skin cancer. However they also noted that skin cancer does not occur in the absence of other toxic effects due to arsenic (WHO, 1983).

JECFA considered that the available epidemiological evidence allowed the tentative conclusion that arsenicism could be associated with water supplies containing an upper arsenic concentration of 1 mg/L or greater, and that concentrations of 0.1 mg/L may give rise to presumptive signs of toxicity. The chemical species of arsenic present in the drinking-water were not clearly determined but it would be reasonable to consider them to be inorganic arsenic. Assuming a daily water consumption of 1.5 litres, intakes of 1.5 mg/day of inorganic arsenic were considered likely to result in chronic arsenic toxicity and daily intakes of 0.15 mg or 2 μg/kg body weight (for a 70 kg individual) may also be toxic in the long term to some individuals.

This evaluation was reviewed in 1989, and a Provisional Tolerable Weekly Intake (PTWI) for inorganic arsenic of 15 μg/kg body weight was assigned (WHO 1989). However JECFA acknowledged that there was a narrow margin between the PTWI and intakes reported to have toxic effects in epidemiological studies. In 1989 JECFA also considered organic arsenic present in seafood (WHO 1989). It commented that further investigations of the type and levels of organic arsenic compounds naturally occurring in marine products and further animal studies on these specific compounds would be highly desirable. The available data were not sufficient for JECFA to set a PTWI for organic arsenic in food. However it was noted that organic arsenic intakes of about 50 μg/kg body weight/day produced no reports of ill effects, and that organoarsenicals found in fish, although almost completely absorbed, are excreted rapidly by humans.

In 1993 the US Environmental Protection Agency derived an oral reference dose for inorganic arsenic of 0.3 μg/kg body weight/day (US EPA, 1993). This was based on a human epidemiological study in which a chronic oral NOAEL of 0.8 μg/kg body weight/day was identified based on effects of hyperpigmentation, keratosis and possible vascular complications. An uncertainty factor of 3 (to account for both the lack of data to preclude reproductive toxicity and to account for some uncertainty in whether the NOAEL accounts for all sensitive individuals) was applied to the NOAEL of 0.8 μg/kg body weight/day, giving a reference dose of 0.3 μg/kg body weight/day.

Previous COT evaluations

In 2003 the COT considered a survey of arsenic (total and inorganic) in the diet (COT 2003a). They noted that the JECFA PTWI was established in 1989 using an approach which would not now be considered appropriate in view od the genotoxicity and carcinogenicity data and concluded that there are no relevant tolerable intakes or reference doses by which to assess the safety of either inorganic or organic arsenic in the diet. Inorganic arsenic is genotoxic and a known human carcinogen, therefore exposure to inorganic arsenic from all sources should be ALARP.

The COT noted that fish is the major contributor to dietary exposure to arsenic, and the predominant form of arsenic in fish is organic. There is a general assumption that organic arsenic is less toxic than inorganic arsenic but this is based on an extremely limited database. However there is no evidence that exposure to organic arsenic through high levels of fish consumption has resulted in harmful effects, which indicates that the dietary exposure to organic arsenic identified in this survey is unlikely to constitute a hazard to health. They also noted that the average population dietary exposure to total arsenic was lower than that estimated for previous years providing reassurance that exposure to total arsenic through food is not increasing.

Also in 2003, the COT evaluated arsenic in food when the results of the 2000 TDS were considered (COT, 2003b). The estimated population dietary exposure to total arsenic from the 2000 TDS was 0.055 mg/day, approximately 0.9 µg/kg body weight/day for a 60 kg adult. The Committee recommended that future surveys should measure both total and inorganic arsenic and include consideration of other sources of exposure such as water. This is because the degree of toxicity of arsenic is dependent on the form (e.g. inorganic vs. organic). The Committee noted that although the survey measured total arsenic only, the data appeared consistent with survey data reviewed previously. The Committee concluded that dietary exposure to arsenic at that time was unlikely to constitute a hazard to health, and exposure to inorganic arsenic should be ALARP.

Arsenic has been included in the rolling revision of the WHO Guidelines for Drinking Water Quality and an updated factsheet is due to be published in the second addendum to the third edition (WHO, 2008). The evidence from epidemiological studies suggests that consumption of elevated levels of arsenic through drinking water is causally related to the development of cancer at several sites, particularly skin, bladder and lung. In the third edition of the Guidelines for Drinking Water Quality, a provisional guideline value for arsenic was set at the practical quantification limit of 10 µg/L (WHO, 2006). However, some recent publications indicate that there is not any quantifiable bladder cancer risk from exposure to low levels of arsenic in water (<50 µg/L) and that studies from Taiwan and China may have been misinterpreted, leading to overestimates of the projections to low-dose exposures (WHO, 2008).

The European Commission has requested that the EFSA evaluate the risks to human health related to the presence of arsenic in foodstuffs (including drinking water). In addition the Commission also asked EFSA to assess:

- the typical ratios between inorganic and organic arsenic forms in different groups of foodstuffs;
- the contribution of different foodstuffs to human exposure for total arsenic and inorganic arsenic, including the contribution from drinking water; and

- the exposure of specific population groups (e.g. high consumers, infants and children, people following specific diets, etc.) and to provide an indication of the age group in which children would be most exposed to the toxic effects of arsenic.

There is currently an open call for data with the objective to collect all available data analysed during the time period from January 2003 to November 2008 (EFSA, 2008). These data will then be used to produce the EFSA opinion on arsenic in food.

References

COT (1998). Statement on the results of multielement surveys in various items of the diet. COT Statement 1998. Annual Report 1998 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Food, Consumer Products and the Environment.

COT (2003a). Statement on arsenic in food; Results of the 1999 Total Diet Study on Arsenic. (2003-01). Committee on Toxicity of Chemicals in Foods, Consumer Products and the Environment. Available at: http://www.food.gov.uk/multimedia/pdfs/ArsenicStatement.PDF.

COT (2003b). Statement on twelve metals and other elements in the 2000 Total Diet Study. COT Statement 2003/07. Annual Report 2003 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Foods, Consumer Products and the Environment.

DEFRA/EA (2002). Contaminants in soil: Collation of Toxicological Data and Intake Values for Humans. Arsenic. Environment Agency Publications. Bristol.

EFSA (2008). Open Calls for data: Request for data on arsenic levels in food and water. The European Food Safety Authority. Available at: http://www.efsa.europa.eu/EFSA/efsa_locale-1178620753812_1211902010663.htm

IARC (1980). International Agency for Cancer Research. Arsenic and Arsenic Compounds. In *Evaluation of carcinogenic risk of chemicals to humans*, vol. 2, IARC Monograph, IARC, Lyon, pp. 48-73.

IARC (1987). International Agency for Cancer Research. Overall evaluations of carcinogenicity: an updating of IARC Monographs 1 to 42. In *Evaluation of carcinogenic risk of chemicals to humans*, supplement 7, IARC Monograph, IARC, Lyon.

IARC (2004). International Agency for Cancer Research. Some Drinking water disinfectants and contaminants, including Arsenic. Vol 84, October 2002.

Kohlmeyer U, Kuballa J. and Jantzen E. (2002). Simultaneous Separation of 17 inorganic and organic arsenic compounds in marine biota by means of high performance liquid chromatography/inductively coupled plasma mass spectrometry. *Rapid Communications in Mass Spectrometry.* **16**: 965-974.

Luten, J.B., Riekwel-Booy, G. & Rauchbaar, A. (1982). Occurrence of arsenic in plaice *(Pleuronectes platessa),* nature of organo- arsenic compound present and its excretion by man. *Environ. Health Perspect.,* 45, 165-170.

MAFF (1998). Lead, Arsenic and other Metals in Food. *Food Surveillance Paper No.* **52**. Ministry of Agriculture, Fisheries and Food.The Stationery Office, London.

Siewicki, T.C. (1981). Tissue retention of arsenic in rats fed witch flounder or cacodylic acid. *J. Nutr.,* **111**: 602-609.

US EPA (1993). Integrated Risk Information System (IRIS): Arsenic, inorganic. US Environmental Protection Agency National Center for Environmental Assessment.

Vahter, M., Marafante, E. and Dencker, L. (1983). Metabolism of arsenobetaine in mice, rats and rabbits. *Sci Total Environ,* **30:** 197-211.

WHO (1983). Toxicological Evaluations of Certain Food Additives and Contaminants, 27th Report of the JECFA, WHO Food Additives Series No 18.

WHO (1989). Toxicological Evaluations of Certain Food Additives and Contaminants, 33rd Report of the JECFA, WHO Food Additives Series No 24.

WHO (2001). Environmental Health Criteria 224. Arsenic and Arsenic Compounds (Second Edition).

WHO (2006). Guidelines for Drinking-Water Quality. Volume 1 Recommendations. First addendum to the third edition. World Health Organization, Geneva.

WHO (2008). Plan of work for the rolling revision of the WHO Guidelines for Drinking Water Quality. World Health Organization website: http://www.who.int/water_sanitation_health/gdwqrevision/en/index.html. Accessed July 2008.

Yamauchi, H. *et al.* (1986). Metabolism and excretion of orally administered arsenobetaine in the hamster. *Bull. Environ. Contam. Toxicol.* **36:** 350 – 355.

Yamauchi, H. *et al.* (1990). Toxicity and metabolism of trimethylarsine in mice and hamster. *Fundam Appl. Toxicol.* **14:** 399 – 407.

4. Barium (Ba)

The key sources of information for this summary were the recent WHO Concise International Chemical Assessment Document on Barium (WHO, 2001), Environmental Health Criteria document 107 (WHO, 1990) and the WHO Guidelines for Drinking Water Quality (WHO, 1996).

Chemistry and occurrence

There is no information on the uptake of barium from food sources, nor of the chemistry and physical form of barium in food.

Apart from medical uses, intake from food is the major source of exposure to barium - a study in the UK by Schroeder *et al.* in 1972 estimated a total daily intake of 1.3 mg; with 1.2 mg arising from food (93%), 0.086 mg from water and 0.001 mg from ambient air. Natural barium levels in public drinking water supplies are negligible. Generally, levels in groundwater in the UK are between 20 and 100 µg/L (BGS, 1989). Barium is not considered to be an essential element for human nutrition (Schroeder *et al.,* 1972).

Absorption and elimination

The oral toxicity of barium is influenced by the solubility of the ingested compound and hence the bioavailability of the Ba^{2+} ion. Nitrate, chloride and sulphide salts are readily soluble and are absorbed (e.g. 3% -11% for $BaCl_2$ in drinking water). Other salts such as the sulphate and carbonate are relatively insoluble but even these less soluble forms can be absorbed to some extent (EPA, 1985).

Absorbed barium is cleared rapidly from plasma and soft tissue but accumulates in the bone (preferentially the active areas of bone growth), pigmented parts of the eye and in cardiac muscle. Metabolism closely parallels that of calcium. Adults contain approximately 22 mg of barium, 90% of which is located in the bone (EPA, 1985). The principal route of elimination following oral administration is in the faeces (WHO, 2001).

Toxicity in animals

The oral LD_{50} of barium chloride in rats is reported to be 118 mg/kg body weight. In repeat dose studies, the kidney is the most sensitive target organ (WHO, 2001).

Weanling female Long-Evans rats were exposed to barium at 1, 10 or 100 ppm in their drinking water for up to 16 months (equivalent doses were calculated to be 0.051, 0.51 and 5.1 mg barium/kg body weight/day. Mean systolic pressure remained unchanged in animals exposed to the lowest dose for 16 months, at the intermediate dose there were mean increases in blood pressure of 4-7 mm Hg at 8 months which persisted thereafter. At the highest dose, significant and persistent rises in blood pressure were recorded after 1 month, increasing to 16 mm Hg by the end of the study. The WHO evaluation (1996) considered the increase in systolic blood pressure of 4-7 mm Hg to be

"small enough not to constitute an adverse effect" and subsequently identified a NOAEL of 0.51 mg/kg body weight/day. In concluding this figure was a NOAEL, the WHO took into consideration calculations made for the USA population that a small insignificant increase in blood pressure of 5 mm Hg at age 35 could become a difference of 10 mm Hg at age 65, and as such could increase the risk of heart attacks by 14% (Wilkins and Calabrese, 1985).

McCauley *et al.* (1985) studied the histological and cardiovascular effects of barium in male Sprague-Dawley rats via the drinking water with concentrations up to 250 mg barium/L (35 mg/kg body weight/day) for 36 and 68 weeks, and at concentrations up to 1000 mg/L (140 mg/kg body weight/day) for 16 weeks. Females were exposed to 250 mg/L for 46 weeks. Electrocardiogram studies (involving challenges with arrhythmagenic doses of l-norepinephrine) and blood pressure measurements studies were conducted. With the exception of ultrastructural changes in kidney glomeruli and the presence of myelin figures at 1000 mg/L, no other adverse effects were noted. No barium related changes were observed in normal and hypertensive strains of rats exposed for 16 weeks to 1000 ppm barium (140 mg/kg body weight/day).

Schroeder and Mitchener (1975 a,b) exposed rats and mice to 5 mg/L barium in their drinking water for a lifetime (dose equivalents of 0.25 mg/kg body weight/day in rats and 0.825 mg/kg body weight/day in mice). No adverse effects were noted but blood pressure was not measured.

As part of the NTP carcinogenicity evaluation, preliminary 15-day and 13-week toxicity studies of barium chloride were conducted in F344/N rats and $B6C3F_1$ mice. In 15-day studies barium chloride dihydrate produced minimal and biologically insignificant effects in rats and mice even at the highest concentrations of 2000 ppm and 692 ppm, respectively (NTP, 1994). Consequently, higher doses were used in the 13-week studies. Barium chloride was administered to groups of 20 male and 20 female mice ($B6C3F_1$) and 20 male and 20 female rats (Fischer 344/N) in drinking water at doses ranging from 125 ppm to 4000 ppm over 92 days. The highest dose corresponded to 436 – 562 mg barium/kg body weight/day in mice and 120 – 136 mg barium/kg body weight/day in rats (males and females, respectively). Serum electrolyte determinations, histopathology, and behavioural tests were conducted. Cardiovascular studies were conducted only during the 13-week study in rats on days 0, 45 and 91, comprising heart rate and systolic arterial pressure measurements and analysis of ECG recordings. The no effect level from these observations was considered by the authors to be 2000 pm (i.e. 165-166 mg barium/kg body weight/day in mice; 61 – 81 mg barium/kg body weight/day in rats). The main pathological finding at the highest dose was renal toxicity possibly from the deposition of insoluble barium crystals in renal tubules, although a physiological mechanism involving hypokalaemia was not excluded by the authors. Additionally the known cardiovascular effects of barium could also have nephrotoxic consequences. Effects noted in previous animal toxicity studies (hypokalaemia, cardiovascular effects) were not seen in these studies, although doses were higher than those used in other studies (NTP, 1994; Dietz *et al.,* 1992).

Exposure levels of 0, 500, 1250 or 2500 ppm barium chloride dihydrate were selected for 2 year NTP carcinogenicity bioassays in F344/N rats and B6C3F_1 mice (NTP 1994). 60 males and 60 females were allocated to each dose group. No cardiovascular studies were conducted during the course of these investigations. In rats, no increased neoplasm incidences could be attributed to barium dihydrate administration but in male rats there was a dose related decrease in mononuclear cell leukaemia (0 ppm: 35/50; 500 ppm: 25/50; 1250 ppm: 26/50; 2500 ppm: 6/50) and in benign and malignant pheochromocytomas of the adrenal medulla (0 ppm: 13(2 malignant)/49; 500 ppm: 11(2)/50; 1250 ppm: 12(2)/49; 2500 ppm: 6(0)/50). It is postulated that effects could be due to a change in hormonal profile of the rats arising from the calcium like effects of barium upon the pituitary and adrenals, although no measurements of hormone levels were taken. In mice no carcinogenic effects attributable to treatment were recorded. However, the incidence of hepatocellular adenoma was significantly decreased in 2500 ppm males. Survival rates of 2500 ppm female mice were significantly reduced and attributable to barium related nephropathy, similar to that seen in the 13-week studies. The difference in response between rats and mice may be due to the 2 to 4 - fold higher intake of barium by mice as compared to rats. (i.e. 2500 ppm barium chloride in drinking water over two years is calculated to be approximately 250 mg barium/kg body wieight/day in mice and 82.5 mg barium /kg body weight/day in rats).The conclusion of the NTP studies was as follows:

> "Under the conditions of these 2-year drinking water studies, there was *no evidence of carcinogenic activity* of barium chloride dihydrate in male or female F344/N rats that received 500, 120 or 2500 ppm. There was *no evidence of carcinogenic activity* in male or female B6C3F, mice that received 500, 1250 or 2500 ppm. There were chemical-related increased incidences of nephropathy in male and female mice".

No mutagenic effects in *Bacillus subtilis* or on viral DNA transcription have been reported (WHO, 1996).

In the Ames test, barium chloride dihydrate at 100 to 10,000 µg/plate did not induce mutations in *S typhimurium* strains TA100, TA1535, TA1537, TA97 and TA98 with and without Arochlor 1254 induced rat or hamster S9 mix (NTP, 1994).

In the mouse lymphoma assay barium chloride dihydrate at 250 µg/mL and above, induced gene mutations in the $TK^{+/-}$ locus of L5178Y mouse lymphoma cells but only in the presence of Arochlor 1254 induced rat liver S9 mix (NTP, 1994).

Barium chloride dihydrate did not induce sister chromatid exchanges or chromosomal abberations in cultured Chinese Hamster Ovary cells with or without Arochlor 1254 induced rat liver S9 (NTP, 1994).

No *in vivo* mutagenic studies have been reported in the published literature.

Toxicity in humans

The critical effects in humans resulting from exposure to barium are hypertension and renal function (WHO, 2001). At high concentrations barium causes vasoconstriction (by direct stimulation of arterial muscle), peristalsis (direct action on smooth muscle) and convulsions and paralysis (stimulation of the central nervous system). Symptoms of barium poisoning in man are excessive salivation, vomiting, colic, diarrhoea, convulsive tremors, slow hard pulse and elevated blood pressure (Beliles, 1994).

The mode of toxic action upon muscle results from hypokalaemia following a barium-stimulated uptake of potassium by cells. The mechanism is thought to involve the transfer of potassium from extracellular to intracellular compartments rather than to actual urinary or gastrointestinal losses of potassium. Consequently in the heart, extra-nodal cells begin to depolarise spontaneously and premature ventricular contractions and abnormalities in the ECG become evident. Patients receiving potassium-depleting diuretics are at higher risk of hypokalaemia from barium poisoning.

Barium induced hypertension appears to result from a direct action of barium on the heart or the resistance vessels, and Ba^{2+} is assumed to act as a Ca^{2+} agonist in these tissues. The barium ion also increases the secretion of calcitonin and adrenal catecholamines, seemingly by entry into secretory cells where it mimics the stimulatory effects of the calcium ion.

All signs of acute barium toxicity in man except hypertension are alleviated by intravenous potassium administration. The cardiotoxic properties of barium chloride are blocked by the calcium antagonist, verapamil (Matilla *et al.,* 1986).

Evidence of adverse effects of barium upon health are available from epidemiological investigations of an unspecified number of Illinois communities with high concentrations of barium in drinking water (2 - 10 mg/L) against those with low barium concentrations (<0.2 mg/L) which reported significantly higher sex- and age- adjusted death rates for "all cardiovascular disease" and "heart disease" at higher barium concentrations (Brenniman *et al.,* 1979). However, as water higher in barium tends to be higher in both calcium and magnesium (i.e. "harder"), these findings may be confounded by this and other factors (e.g. population mobility, use of water softeners) which were not controlled for in these studies.

A follow up cross sectional study (Brenniman and Levy, 1985) compared the prevalence of cardiovascular disease in 1175 adult residents of West Dundee, Illinois (mean barium concentration in drinking water 7.3 mg/L: range 2 – 10 mg/L) with 1203 residents of McHenry, Illinois (mean barium concentration in drinking water 0.1 mg/L). Many of the possible confounding factors not considered in earlier studies were accounted for. There were no significant differences between the two populations in the prevalence of hypertension, stroke and heart and kidney disease. The authors concluded that blood pressure in adults did not appear to be adversely affected even following

prolonged ingestion of drinking water containing 7 mg barium per litre (equivalent to 14 mg/day).

In a further study by Wones *et al.* (1990), 11 healthy male volunteers were exposed to barium chloride in drinking water (0 mg/L for 2 weeks, 5 mg/L for the next 4 weeks and 10 mg/L for the last 4 weeks). The barium content of the diet was not determined but the authors considered it likely to be in the region of 0.011mg Ba/kg bw/day. Volunteers consumed 1.5 litres of water per day, equating to a high intake of 15 mg/day. No consistent indication of any adverse effects was found upon cardiovascular risk factors and the authors conclude that the effects of high concentrations of barium upon hypokalaemia and hypertension were not seen at the lower concentrations investigated. There was a trend however towards an increase in serum calcium between 0 and 5 mg/L which persisted at 10 mg/L. As half the total calcium in blood is protein bound (principally to albumin) a recalculated value adjusted for plasma albumin revealed a statistically significant increase in total calcium. The magnitude of this finding was small (about a 2% rise) and the authors concluded that it was not clinically important but suggested that studies are required to confirm this effect. Blood pressures were not significantly affected; an NOAEL of 0.21 mg barium/kg body weight/day was reported. The lack of adverse effects observed in this work may be attributable to the small number of subjects included or the short period of exposure.

Previous evaluations

In 1993 the WHO recommended a drinking water guideline value for barium of 0.7 mg/L (WHO, 1993). This value was confirmed in the most recent edition of the Guidelines (WHO, 2006). The Guideline value was set based on concern regarding the potential of barium to cause hypertension. A NOAEL of 7.3 mg/L was identified in humans (from a study by Brenniman & Levy, 1985) and an uncertainty factor of 10 was applied to account for intraspecies variation to arrive at the guideline value. This value is equivalent to an intake of 1.4 mg/day or 0.02mg/kg bw/day in a 60kg adult, for exposure from drinking water, assuming consumption of 2 litres per day.

Subsequently, the WHO used the NOAEL of 0.21 mg/kg body weight/day from the study of Wones *et al.* (1990) to propose a tolerable intake for barium (WHO, 2001). Applying an uncertainty factor of 10 to allow for database deficiencies and differences between humans, resulted in a tolerable intake of 0.02 mg/kg body weight/day.

The USA Environmental Protection Agency's evaluation of the oral toxicity of barium derived an oral reference dose of 0.2 mg/kg body weight/day (US EPA, 2005). This reference dose was based on a $BMDL_{05}$ of 63 mg/kg body weight/day for nephropathy from the 2-year NTP drinking water study. An uncertainty factor of 300 was applied: 100 for inter- and intra-species variation and 3 to allow for uncertainty in the deficiencies of the database.

An IPCS Environmental Health Criteria Document for Barium published in 1990 concluded that for the general population, barium, at the usual concentrations found in food, air and water (especially water), does not

constitute any significant health risk (WHO, 1990). The evaluation warned however that for specific sub-populations (elderly or potassium deficient individuals), and under specific circumstances (high water concentrations, occupational exposures), a potential for adverse health effects may exist.

Previous COT evaluations

The 1994 Total Diet Study (MAFF, 1998) provided the following results: average dietary intake of 0.58 mg/day with an upper range of exposure (97.5%ile) of 1.33 mg/day. Highest levels of barium were reported in nuts (56 mg/kg). Bread contained 1 mg/kg and other foodstuffs contained levels lower than this. The results also indicated that marine fish and shellfish are unlikely to contribute more than 0.027 mg/day. The results of the 1994 TDS were considered by the COT in 1998 which concluded that there was no evidence to suggest that current dietary intakes of barium in the UK may be harmful to health (COT, 1998). Barium was not analysed in the 2000 TDS.

References

Beliles RP (1994). The Metals - Barium in Patty's Industrial Hygiene and Toxicology Ed Clayton GD and Clayton FE Vol 2C p 1925-1930, John Wiley & Sons Inc.

BGS (1989). Trace element occurrence in British groundwaters.

British Geological Survey Resaerch Report SD/89/3, 1989.

Brenniman GR, Nametkata T, Kojola WH, Carnow BW & Levy PS (1979). Cardiovascular disease death rates in communities with elevated levels of barium in drinking water. Environ Res 20 p318-324.

Brenniman GR & Levy PS (1985). Epidemiological study of barium in Illinois drinking water supplies. In Calabrese EJ, Tuthil RW Condie L eds Inorganics in water and cardiovascular disease. Princeton Sci Pub p231-240.

COT (1998). Statement on the results of multielement surveys in various items of the diet. COT Statement 1998. Annual Report 1998 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Food, Consumer Products and the Environment.

Dietz DD, Elwell MR Davis Jr, WE & Meirhenry EF (1992). Subchronic toxicity of barium chloride dihydrate administered to rats and mice in the drinking water. Fund Appl Tox 19 p527-537.

EPA (1985). Environmental Protection Agency USA Office of Drinking Water. Drinking Water Criteria Document on Barium. Washington DC.

Matilla MJ Anyos K & Puisto E-L (1986). Cardiotoxic actions of doxepin and barium chloride in conscious rabbits. Arch Tox (Suppl) 9 205-208.

MAFF (1997). 1997 MAFF Multielement study of dietary supplements.

McCauley PT, Douglas BH, Laune RD and Bull RJ (1985). Investigations into the effect of drinking water barium in rats in Advance in modern environmental toxicology, Princeton Publishing Vol IX p197-210.

NTP (1994). Toxicology and carcinogenesis studies of barium chloride dihydrate in F344/N and B6C3F_1 mice . US Department of Health and Human Services. National Toxicology Report Series No 432 , January 1994.

Schroeder HA, Tipton IH & Nason AP (1972). Trace metals in man: strontium and barium. J Chron Dis 25 491-517.

Schroeder HA & Mitchener M (1975a). Life time studies in rats : Effects of aluminium, barium, beryllium and tungsten. J Nutr 105 p 421-427.

Schroeder HA & Mitchener M (1975b). Life time effects of mercury, methyl mercury and nine other trace metals on mice. J Nutr 105 p452-458.

US EPA (2005). Integrated Risk Information System (IRIS): Barium and compounds. US Environmental Protection Agency National Center for Environmental Assessment.

WHO (1990). IPCS Environmental Health Criteria 107. Barium. World Health Organisation, Geneva.

WHO (1993). World Health Organisation Guidelines for Drinking Water Quality, Volume 1 p42-43, Geneva

WHO (1996). World Health Organisation Guidelines for Drinking Water Quality, Volume 2 Barium p173- 183, Geneva.

WHO (2001). Concise International Chemical Assessment Document 33: Barium and barium compounds. World Health Organization, Geneva.

WHO (2006). Guidelines for Drinking-Water Quality. Volume 1 Recommendations. First addendum to the third edition. World Health Organization, Geneva.

Wilkins JR and Calabrese (1985). Health Implications of a 5 mm Hg increase in blood pressure. In: Inorganic in Drinking water and Cardiovascular Disease; Advances in Modern Envrionmental Toxicology Volume IX, Ed Calabrese EJ, Tuthill RW, and Condie L Princeton Sci Pub 1985 and also published in Toxicology and Industrial Health p13-28.

Wones RG, Stadler BL and Frohman LA (1990). Lack of effect of drinking water barium on cardiovascular risk factors. Env Health Perspec 85 p355-359.

5. Bismuth (Bi)

Bismuth has not been the subject of any international review by WHO or other relevant bodies (e.g. EFSA, EPA etc).

Chemistry and occurrence

The chemical element bismuth is a soft, brittle, highly lustrous metal belonging to the same group in the periodic table as arsenic, ie Group VA. Its atomic number is 83. The average abundance of bismuth in the earth's crust is about 0.00002 per cent. Bismuth forms compounds in the 3+ and 5+ oxidation states; the 3+ state is the more stable of the two.

Bismuth occurs naturally as the free metal or in combined forms in such ores as bismuthinite, Bi_2Si_3, which commonly contains copper or iron, and as the oxide bisnite. The principal deposits are in Bolivia and Peru (Beliles, 1994).

There is no published information on levels of bismuth in water or air in the UK, but in the USA, drinking water contains on average 0.01 mg/L and ambient air levels range from < 0.002 to 0.03 µg/m^3. Based on these limited data, a daily intake of 20 µg has been estimated. The body burden of bismuth in soft tissues of a 70 kg reference man is < 230 µg. Levels reported in various food include: dairy produce (1 µg/kg), fish (0.9 µg/kg) and green vegetables (0.7 µg/kg) (Snyder *et al.,* 1975).

Bismuth has a long history of pharmaceutical use in Europe and North America. Both inorganic and organic salts have been used both internally (for diarrhoea and other gastrointestinal complaints) and externally (astringent and antibacterial properties). Insoluble bismuth salts have also been used in cosmetics. Medicinal use of bismuth salts declined following poisoning episodes in France and Australia in the 1970s characterised by a unique encephalopathy.

There is no information on the forms of bismuth primarily found in foods.

Absorption and elimination

Most bismuth salts are insoluble and poorly absorbed when given orally or applied to the skin. However, soluble bismuth medicinal preparations given orally are absorbed promptly, giving urinary excretion curves closely resembling those of intramuscular injections of aqueous and oil solutions. However even with these soluble bismuth preparations, urinary excretion amounts to about 0.5 per cent of the administered dose. Excretion of insoluble bismuth salts is almost wholly faecal (Beliles, 1994).

Despite the large variation in bismuth compounds used in human and animal studies, the data on distribution of the metal in tissues are in remarkably close agreement. The highest concentration is always found in the kidneys, where retention is also longer than other tissues - in the rat, 144 hours after injection of bismuth citrate (^{206}Bi), 12% of injected dose remained in the kidney, 0.9% in the bone, but little or no levels were detected in other organs (Slikkerveer

and de Wolff, 1989). Because newborn animals from bismuth-treated mothers show the same concentration as adults, it is inferred that bismuth passes into the placental circulation. Bismuth is also excreted into the milk, but in lower concentrations than into the urine (Beliles, 1994).

In man, the normal concentration of bismuth in the blood is between 1 and 15 µg/L, but absorption from oral medicinal preparations produces a significant rise. Distribution of bismuth in the organs is largely independent of the compound or the route of administration and the concentration in the kidney is always the highest. It is bound to a bismuth-metal binding protein in the kidney, the synthesis of which can be induced by the metal itself. Elimination takes place by urinary or faecal routes but the exact proportion contributed by each route is still unknown. Elimination from the blood displays multicompartment pharmacokinetics - the shortest half-life in humans being 3.5 minutes and the longest 17-22 years. Elevated levels of bismuth in blood have been detected following poisoning episodes, and a safety level of 50 µg/L and an alarm level of 100 µg/L have been proposed (Slikkerveer and de Wolff, 1989), although the supporting scientific basis for these values is weak.

Toxicity in animals

The toxicity of bismuth from bismuth preparations is dependent upon many factors including the solubility of the compound (most inorganic compounds are poorly soluble), the physical form of bismuth (differences have been observed in uptake from different particle sizes of bismuth subnitrate), the nature of the associated anionic component dosage (there is lower percentage bioavailability at higher doses in animal studies), the extent of protein binding, etc.

The acute toxicity of soluble bismuth pharmaceuticals results in LD_{50} values ranging from 13 - 82 mg/kg body weight. This contrasts with the low oral toxicity of bismuth oxychloride (an insoluble inorganic pigment) which has a quoted LD_{50} value of 21.5 g/kg body weight - equivalent to 17.2 g Bi/kg body weight (quoted by Preussmann and Ivankovic, 1975).

In studies reported very briefly in the published literature, bismuth oxychloride (BiOCl) was administered to groups of 40 male or 40 female BD rats at doses of 1, 2 or 5% in the diet over 2 years; doses corresponding to 350, 700 and 1750 mg BiOCl/kg body weight/day in male rats (equivalent to 280, 560 and 1400 mg Bi/kg body weight/day) and 280, 560 and 1400 mg BiOCl/kg body weight/day in female rats (equivalent to 224, 448 and 1120 mg Bi/kg body weight/day). 60 animals of each sex served as controls. Mean body weights did not differ from controls, but mean survival times for treated animals were generally 10% lower than controls with larger deviations around the means value. No macroscopic or histological findings could be attributed to BiOCl. The incidence of tumours was closely comparable in test and control groups. The authors concluded that, as the fibroadenomas and hypophyseal adenomas are spontaneous tumours characteristic of this strain, "BiOCl seems to be non-carcinogenic in rats". A chronic NOAEL in the region of 1400 - 1750 mg/kg body weight/day for BiOCl (equivalent to 1400 -1120 mg Bi/kg

body weight/day) was inferred from this work (Preussmann and Ivankovic, 1975).

Bismuth citrate given to pregnant rats and rabbits at maternal doses of 200 and 1200 mg/kg body weight/day (equivalent to 105 and 613 mg Bi/kg body weight/day) did not impair foetal viability in either species, and there were no abnormalities in rat postnatal development (Secker, 1993).

Although the absorption of bismuth would not comprise more than a small fraction of that ingested, absorbed bismuth has been shown to be concentrated in the placenta and readily transferred to the foetus (Thompson *et al* 1941).

Toxicity in humans

A number of toxic effects in humans have been attributed to bismuth compounds: nephropathy, encephalopathy, osteoarthropathy, gingivitis, colitis, stomatitis and hepatitis. However these adverse effects on different organ systems are associated with different bismuth compounds e.g. neurotoxic effects are attributable to so-called insoluble bismuth compounds, whereas bismuth compounds used for the treatment of syphilis caused kidney and bone disease, but not neurotoxicity.

An epidemic of bismuth encephalopathy occurred in France and was associated with the intake of inorganic salts including bismuth subnitrate, subcarbonate and subgallate. In the prodromal phase patients developed problems in walking, standing or writing, deterioration of memory, changes in behaviour, insomnia and muscle cramps, together with several psychiatric symptoms. The manifest phase started abruptly and was characterised by changes in awareness, myoclonia, memory disturbances, writing disturbances and dysarthia. Patients recovered spontaneously after discontinuation of bismuth. A small outbreak of poisoning was also seen in Australian patients who had undergone a colostomy or an ileostomy and taken oral bismuth subgallate. A so far unidentified additional factor besides bismuth was held responsible for these intoxications. Despite many theories on enhanced intestinal absorption, the exact aetiology of bismuth encephalopathy remains a mystery (Slikkerveer and de Wolff, 1989).

In 41 patients on bismuth therapy no symptoms were reported at dosages of 10 - 20 g/day, which gave rise to median blood levels of 13 µg/L (range 5 - 100 µg/L). A group of 63 other patients with bismuth encephalopathy had median concentrations of 680 - 700 µg/L (range 100 - 2850 µg/L). On the basis of this study a blood level of > 100 µg/L was considered to be an arbitrary `alarm' level and 50 µg/L was considered as a safety level. Blood bismuth concentrations in persons not receiving bismuth therapy are reported to be 1 - 15 µg/L (Slikkerveer and de Wolff, 1989). Behrendt *et al.* (1991) reported levels of 3 µg/L for non-exposed individuals. Gavey *et al.* (1989) found median steady-state plasma concentrations of 17 µg/L (i.e. just slightly outside the values of reported endogenous levels) in a group of patients receiving De-Nol tablets (2 tablets twice daily, containing 108 mg bismuth/tablet as colloidal bismuth citrate, equivalent to a total bismuth intake

of 7.2mg/kg bw/day in an average 60 kg adult). Gavey *et al.* (1989) estimated that only 0.2% of the ingested bismuth was absorbed thus indicating absorption of 0.8 mg bismuth/day (i.e. 4 x 0.2 mg per day). This may be taken as an indication of a daily oral uptake of bismuth from a bioavailable form (ie 800 µg bismuth/day) that is without adverse effect.

Previous COT evaluations

In the 1994 total diet study (MAFF, 1997a), average and mean intakes of bismuth from the diet were both estimated at 0.4 µg/day - with an upper (97.5%-ile) intake of 0.7 µg/day. Highest levels were reported for dairy produce (1 µg/kg), fish (0.9 µg/kg) and green vegetables (0.7 µg/kg). High-level consumers of fish and shellfish could ingest 0.4 µg/day from this source alone. However in the 1994 multielement study of dietary supplements (MAFF, 1997b) an intake of 6 µg/day was calculated. This is an order of magnitude above levels of bismuth that might be ingested from the diet.

The results of the 1994 TDS were considered by the COT in 1998 who concluded that *despite a lack of information on the effects of low doses of bismuth upon man, and no detail of the likely bioavailability of bismuth from food sources, the level of bismuth in the diet and in dietary supplements does not indicate a cause for concern* (COT, 1998). Bismuth was not analysed in the 2000 TDS.

References

Behrendt WA Groger C, Kuhn D Schulz H-U and Topfmeier P (1991). A study relating to the bioavailability and renal elimination of bismuth after oral administration of basis bismuth nitrate. Int J. Clin Pharm Ther. Tox 29 p357-360.

Beliles RP (1994). Bismuth In: Patty's Industrial Hygiene and Toxicology, Clayton GD and Clayton FE Volume IIC p1948-1954

COT (1998). Statement on the results of multielement surveys in various items of the diet. COT Statement 1998. Annual Report 1998 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Food, Consumer Products and the Environment.

Gavey CJ, Szeto M-L, Nwoloko CU, Sercombe J and Pounder RE (1989). Bismuth accumulates in the body during treatment with potassium dicitrato bismutate. Alim Pharmancol & Therap 3 p21-28.

MAFF (1997a). 1994 Total Diet Study: Metals and Other Elements. Food Surveillance Information Sheet No 131.

MAFF (1997b). 1997 MAFF Multi-element Study of Dietary Supplements.

Preussmann R and Ivankovic S (1975). Absence of carcinogenic activity in BD rats after oral administration of high doses of bismuth oxychloride. Fd Cosmet Toxicol 13 P543/544.

Secker RC (1993). Effect of bismuth citrate on pregnant rats and rabbits. *Teratol* 48 p33A

Slikkerveer A and de Wolff FA (1989). Pharmacokinetics and Toxicity of Bismuth Compounds. Med Toxicol Adv. Drug Exp. 4 p303-323.

Snyder WS, Cook MJ, Nasset ES, Karhausen LR, Howells GP and Tipton IH, (1975). International Commission on Radiological Protection (IRCP). Report of the Task Group on Reference Man, New York. ICRP Publ. 23.

Thompson et al (1941). The transfer of bismuth into the fetal circulation after maternal administration of solbisminol. Am J Syph 25 p725 – 730.

6. Cadmium (Cd)

The key sources of information for this summary were the 2003 JECFA review of cadmium (WHO, 2003) and Environmental Health Criteria 134 (WHO, 1992).

Chemistry and occurrence

Cadmium is most stable as Cd^{2+}, and reacts with all halides, oxygen, and sulphur. It is used in electroplating, the production of alloys, batteries, and atomic reactors. Its compounds are used as pigments and as phosphors in colour television tubes. The main routes of exposure are inhalation (occupational exposure) and oral.

Edible free-living food organisms such as shellfish, crustaceans, and fungi are natural accumulators of cadmium. Staple foods such as cereals and potatoes can also accumulate cadmium and these foods contribute significantly to the dietary intake or cadmium.

Cadmium found in food is likely to have originated from mining or other industrial activities. No information is available on the forms of cadmium found in foods.

Absorption and elimination

Many factors influence the absorption of ingested cadmium; animal species, cadmium compound, dose, frequency, age, stage of development, pregnancy and lactation, nutritional status and interactions with various nutrients. For example, cadmium absorption is enhanced when the iron status of the body is sub optimal. It has been reported that on average 5% of ingested cadmium is absorbed in humans (can be up to 15%) (Flanagan *et al*., 1978).

Absorbed cadmium is transported in blood mainly in erythrocytes and is bound to proteins of low and high relative molecular mass. The low molecular mass protein is similar to metallothionein (Miles *et al.,* 2000). There is little transfer of cadmium across the placental barrier once fully formed (Ahokas and Dilts, 1979).

Long term exposure to cadmium leads to accumulation in the liver and renal cortex (75% of total body burden found in these organs) (Friberg *et al.,* 1985).

Cadmium has an extremely long half-life in animals (200 days to 22 years) due to its very slow excretion in the urine (Friberg *et al.,* 1985). In man the half-life is estimated to be 17 years or longer (WHO, 2003).

In man urinary excretion of cadmium is related to body burden, recent exposure, and renal damage. Urinary excretion of cadmium is increased in

individuals with renal damage and those exposed to excessive levels of cadmium. Gastrointestinal excretion is approximately equal to urinary excretion but cannot be easily measured. Other excretory routes such as lactation, sweating or placental transfer are insignificant (WHO, 2003).

Toxicity in animals

Oral LD_{50} values for animals range from 225 to 890 mg/kg body weight for elemental cadmium, 63 to 88 mg/kg body weight for cadmium chloride, 72 mg/kg body weight for cadmium oxide, and 590 to 1125 mg/kg body weight for cadmium stearate (USAF, 1990).

Exposure of rabbits to 0.013 mg/kg body weight/day cadmium chloride in drinking water produced histological alterations in the liver but no clinical signs of toxicity (Stowe et al., 1972). In a study by Kotsonis and Klaassen (1978), rats exhibited proteinurea after receiving 3.1 and 8.0 mg/kg body weight/day as cadmium chloride in the drinking water for six weeks. Rats given 1.2 mg/kg body weight/day as cadmium chloride in the drinking water exhibited no renal effects even after 24 weeks, although higher exposure levels induced proteinurea after six weeks exposure.

There is evidence for cadmium-induced immunotoxicity in animals. Koller et al. (1975) noted a decrease in the number of spleen plaque-forming cells in mice receiving cadmium at 0.6 mg/kg body weight/day for 10 weeks. Blakley (1985) reported a dose-dependent suppression of the humoral immune system in mice receiving cadmium in drinking water at concentrations of 5 to 50 mg/L for three weeks. These immune system effects occurred at kidney tissue concentrations of 0.3 to 6.0 µg/g - lower than those associated with renal toxicity.

Toxicity in humans

Long-term exposure to cadmium affects the kidneys, bones, and calcium metabolism; it can also result in carcinogenicity, neurodevelopmental and neurobehavioral effects.

The kidney is the critical organ in humans exposed to cadmium for long periods (WHO 1989, 1992). In 2003 JECFA reviewed some new epidemiological data evaluating the relationship between exposure to cadmium and renal dysfunction. The Committee concluded that the new data were consistent with the hypothesis that low-level environmental exposure to cadmium is associated with an increased prevalence of renal proximal tubular dysfunction, as assessed by biomarkers (WHO, 2003). Cadmium causes tubular dysfunction possibly due to the formation of a cadmium-metallothionine complex, although unbound cadmium may also play a part in the toxicity (Liu *et al.,* 1999). This dysfunction first manifests as low molecular weight proteinuria but as exposure increases so does the damage to the kidney. It is suggested that a renal concentration of between 100 and 200

μg/g is likely to be the critical concentration of cadmium in the kidney for about 50% of the population (corresponding to a dietary intake of 175 μg Cd/day for 50 years) (WHO, 1989); above this concentration nephropathy may occur.

One of the most well known effects of long-term excessive intake of cadmium is a severe bone disease known as Itai-itai. This manifests as osteoporosis and osteomalacia and is associated with severe cadmium nephropathy. The disease is probably due to a combination of the toxic effects of cadmium, renal disease and nutritional deficiencies (Nogawa and Nishijo, 1999; Aoshima and Kasuya, 1991). Cadmium is also a potential neurotoxin in animals and humans, but some level of protection is provided by metallothionein in the brain (Choudhuri *et al*., 1996).

The IARC (1993) has classed cadmium and its compounds as 'carcinogenic to humans' (Group 1). The human evidence leading to this classification consisted of occupational inhalation exposure to cadmium and its compounds as dust, which is mainly associated with lung cancer although increases in prostate cancer have also been reported. Cadmium is genotoxic, damaging the DNA of human cells *in vitro* (DNA strand breaks, mutations, chromosomal damage and cell transformation). Cadmium compounds also inhibit the repair of DNA damage by other agents, thereby enhancing their genotoxicity.

In 2003 JECFA concluded that there was no evidence that cadmium is carcinogenic to humans exposed by the oral route (WHO, 2003).

Previous evaluations

In 1989 JECFA concluded that because of the risk of kidney damage, levels of cadmium should not exceed 50 μg/g in the renal cortex. Assuming an absorption rate of 5% and a daily excretion of 0.005% of body burden, total intake should therefore not exceed about 1 μg/kg body weight/day continuously for 50 years. The provisional tolerable weekly intake for cadmium was therefore set at 7 μg/kg body weight (WHO, 1989). JECFA confirmed this PTWI of 7 μg/kg body weight/week in 2001 even though there was some evidence that a proportion of the general population may be at greater risk of tubular dysfunction when exposed to the current PTWI. In 2003, the PTWI was maintained and JECFA concluded that no excess prevalence of renal tubular dysfunction would be predicted to occur at this PTWI (WHO, 2003). In 2006 JECFA evaluated the impact of different maximum levels on the overall intake of cadmium and concluded that the effect would be very small (WHO, 2006a). Based on the JECFA PTWI of 7 μg/kg body weight, WHO derived a drinking water guideline value of 3 μg/L (assuming a 60 kg adult drinking 2 litres of water per day and allocating 10% of the PTWI to drinking water; WHO, 2006b).

In 1994 the US Environmental Protection Agency derived an oral reference dose for cadmium in food of 1 μg/kg body weight/day (US EPA, 1994). This was based on a NOAEL of 0.01 mg/kg body weight/day for proteinuria and an

uncertainty factor of 10 (to allow for intraspecies differences). This NOAEL was predicted by applying a toxicokinetic model to data from human chronic studies.

Previous COT evaluations

In 1995 the COT considered the levels of cadmium, mercury and other metals in food. They commented that *'the intake of cadmium is below levels known to cause renal toxicity, although we note that the margin between average cadmium intakes and levels associated with toxicity is smaller than for most other metals. We are reassured that the average dietary intake of cadmium did not increase between 1982 and 1991, as we would view any increase with concern. We note that cadmium is recognised as a human carcinogen via inhalation of cadmium dusts and fumes in the workplace. There is inadequate evidence to assess the carcinogenicity of dietary cadmium'*.

In 2003, the COT evaluated cadmium in food when the results of the 2000 TDS were considered (COT, 2003). The estimated population dietary exposure to cadmium from the 2000 TDS was 0.009 mg/day, approximately 0.15 µg/kg body weight/day for a 60 kg adult. The Committee concluded that current dietary exposures to cadmium are unlikely to be of any toxicological concern for consumers.

References

Ahokas, R. and Dilts, P. (1979). Cadmium uptake by the rat embryo as a function of gestational age. *Am. J. Obstet. Gynecol*. **135:** 219-222.

Aoshima, K. and Kasuya, M. (1991). Preliminary study on serum levels of 1,25-dihydroxyvitamin D and 25-hydroxyvitamin D in cadmium induced renal dysfunction. *Toxicol. Lett.* **57:** 91-99.

Choudhuri, S., Liu, W.L., Berman, N.E. and Klaassen, C.D. (1996). Cadmium accumulation and metallothionein expression in the brain of mice at different tages of development. *Toxicol. Lett* **84:** 127-133.

COT (2003). Statement on twelve metals and other elements in the 2000 Total Diet Study. COT Statement 2003/07. Annual Report 2003 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Foods, Consumer Products and the Environment.

Blakley, B.R. 1985. The effects of cadmium chloride on the immune response in mice. *Can. J. Comp. Med*. **49:**104-108.

Flanagan, p., McLellan, J., Haist, J., Cherian, M., Chaimerlain, M. and Valberg, L. (1978). Increased dietary cadmium absorption in mice and human subjects with iron defficiency. *Gastroenterology*, **74:** 841-846.

Friberg, L., Elinder, C., Kjellestrom, T. and Nordberg, G. (1985). *Cadmium and Health, A toxicological and Epidemiological Appraisal,* Vol. I, *Exposure, Dose Metabolism,* Cleveland, OH, CRC Press.

IARC (1993). Cadmium and Cadmium compounds, IARC Summary and Evaluation, Volume 58.

Koller, L.D., Exon, J.H. and Roan, J.G. (1975). Antibody suppression by cadmium. *Arch. Environ. Health* **30:** 598.

Kotsonis, F.N. and Klaassen C.D. (1978). The relationship of metallothionein to the toxicity of cadmium after prolonged administration to rats. *Toxicol. Appl. Pharmacol.* **46:** 39-54.

Liu, Y., Liu, J., Habeebu, S. and Klaassen, C. (1999). Metallothionein protects against the nephrotoxicity produced by chronic CdMT exposure. *Toxicol. Sci.* **50:** 221-227.

Miles, A.T., Hawksworth, G.M., Beattie, J.H. and Rodilla, V. (2000). Induction. Regulation, Degradation and Biological significance of mammalian metallothioneins. *Crit. Rev. Biochem. Mol. Biol.* **35:** 35-75.

Nogawa, H. and Nishijo, M. (1999). Dose-response relationship and mortality in inhabitants of cadmium-polluted areas. In Nogawa, K., Kurachi, M. and Kasuya, M., eds, *Advances in the Prevention of Environmental Cadmium Pollution and Countermeasures,* Kanazawa, Eiko Laboratory, pp. 110-115.

Stowe, H.D., Wilson, M. and Goyer, R.A. (1972). Clinical and morphological effects of oral cadmium toxicity in rabbits. *Arch. Pathol*. **94:** 389-405.

USAF. (1990). Cadmium. In: Installation Restoration Program Toxicology Guide, Vol. 5. Harry G. Armstrong Aerospace Medical Research Laboratory, Wright Patterson AFB, OH.

US EPA (1994). Integrated Risk Information System (IRIS): Cadmium. US Environmental Protection Agency National Center for Environmental Assessment.

WHO (1989) Safety evaluation of certain food additives and contaminants. WHO Food Additives Series 24, Joint FAO/WHO Expert Committee on Food Additives.

WHO (1992) *Cadmium*, Environmental Health Criteria 134, Geneva.

WHO (2003). Safety evaluation of certain food additives and contaminants. WHO Food Additives Series 52. Joint FAO/WHO Expert Committee on Food Additives.

WHO (2006a). Safety evaluation of certain food additives and contaminants. WHO Food Additives Series 55. Joint FAO/WHO Expert Committee on Food Additives.

WHO (2006b). Guidelines for Drinking-Water Quality. Volume 1 Recommendations. First addendum to the third edition. World Health Organization, Geneva.

7. Chromium (Cr)

The key sources used in this summary were the EVM report (EVM, 2003) and Environmental Health Criteria 61 (WHO, 1988).

Chemistry and occurrence

Chromium has been shown to potentiate insulin activity and therefore has a role in carbohydrate, lipid and protein metabolism.

The transition element chromium (Cr) belongs to Group VI of the Periodic Table. Chromium can exist in any oxidation state from –2 to +6; however, oxidation states other than 0, +2, +3 and +6 are uncommon.

In biological materials chromium is most stable in the oxidation state +3, as chromium compounds with oxidation states below +3 are reducing, and those above +3, are oxidising. Chromium (III) is soluble in acidic solutions, readily forming hexahedral complexes with appropriate ligands, such as oxalate and sulphate ions. At the oxidation state of +6, chromium forms chromates and dichromates, which have strong oxidising potential.

Almost all of the sources of chromium in the earth's crust are in the trivalent state, naturally occurring chromium compounds in the hexavalent state are rare. Hexavalent chromium compounds are thus man-made products (WHO, 1988). Therefore the predominant form of chromium in food is the trivalent form. Trivalent chromium is also an essential element, forming a complex with vitamin B_{12}.

Absorption and Elimination

Intestinal absorption of trivalent chromium is low in both humans and animals, varying between approximately 0.5 and 2.0% depending on dietary intake, with higher intakes resulting in proportionally less absorption (Kumpulainen *et al.,* 1983; Anderson and Kozlovsky, 1985; Offenbacher *et al.,* 1986; Felter and Dourson, 1997).

Absorbed trivalent chromium does not enter blood cells. A portion is incorporated into glucose tolerance factors, and the remainder is bound to transferrin and transported to the liver. In contrast, hexavalent chromium does penetrate red blood cells and is reduced to trivalent chromium, which binds haemoglobin. Excess hexavalent chromium is absorbed into the kidneys, spleen, liver, lungs and bone.

Ingested chromium remains largely unabsorbed and is excreted in the faeces. Assimilated chromium is mainly excreted via urine, with only small amounts being lost in perspiration and bile (EVM, 2003).

Toxicity in animals

The available data are limited, but oral exposure to chromium (III) and chromium (VI) compounds have resulted in gastrointestinal, hepatic, renal, immunological, neurological, developmental and reproductive effects.

Chromium (III) compounds are less toxic than chromium (VI) compounds, with chronic intakes of up to 750 mg/kg body weight/day not being associated with any adverse effects. Doses of 14 mg/kg body weight/day chromium (VI) for 3 weeks resulted in decreased body weights. No treatment related effects were observed when chromium picolinate was administered in the diet to rats and mice for 13 weeks (Rhodes *et al.*, 2005).

Both Chromium (III) and (VI) have been reported to reduce fertility, fetal weight, and crown length and increase post-implantation losses in mice.

There was no clear evidence of carcinogenicity when chromium has been tested via the oral route.

In general, *in vitro* mutagenicity tests have yielded positive results for hexavalent chromium, and negative results for trivalent. However, two studies *in vitro* have shown that chromium picolinate (a form of trivalent chromium) may cause DNA damage. The authors of one study (Stearns *et al.,* 1995) proposed that the picolinate moiety was responsible for the activity. However, the authors of the second study (Speetjens *et al*., 1999) proposed a mechanism involving reduction to chromium II and production of hydroxyl radicals. Chromium nicotinate and chromium III chloride were also investigated in one of these studies and did not damage DNA. The significance of these observations is unclear. Of the limited number of in vivo studies available all have given negative results (COM, 2004).

Toxicity in humans

The oxidation state of chromium is a critical factor, determining not only the route dependent bioavailability, but also its toxicity.

Chronic exposure to hexavalent chromium has been reported to induce renal failure, anaemia, haemolysis and liver failure at doses of between 0.6 - 2.4 mg/day for between 6 weeks and 5 months. The few reports that exist describing chronic exposure to trivalent chromium describe similar effects. Where follow up was reported, these were reversible and had returned to normal parameters in one year. Other randomised controlled trials have shown no adverse effects in individuals given chromium at doses of up to 1mg per day for up to 63 weeks.

A limited retrospective mortality study, conducted on a population which resided in a polluted area near an alloy plant that smelted chromium, and where contamination of water supplies had occurred (20 mg Cr VI/L), found

increased incidences of lung and stomach cancer (Zhang and Li, 1987). However, the study population would have been exposed to chromium by all routes, i.e. air, water, food and soil and all other contaminants that were in the environment.

Hexavalent chromium is carcinogenic via the inhalation route; epidemiological studies have found an association between exposure to chromium (VI) and lung cancer (WHO, 1988). IARC (1990) classified hexavalent chromium, on the basis of combined results of epidemiological studies (exposure to chromium by inhalation) and carcinogenicity studies in experimental animals, into Group 1 'carcinogenic to humans' and trivalent chromium into Group 3 'not classifiable as to its carcinogenicity in humans'.

Previous Evaluations

The US Environmental Protection Agency derived oral reference doses for chromium (III) and chromium (VI) in 1998 (US EPA, 1998). The reference dose for chromium (III) is 1.5 mg/kg body weight/day based on a NOAEL of 1468 mg/kg body weight/day in a chronic rat feeding study with chromic oxide. An uncertainty factor of 100 (to allow for inter- and intra-species variation) and a modifying factor of 10 (to allow for database deficiencies) were applied to the NOAEL.

In 2003, the SCF concluded that there were insufficient data available to derive a tolerable upper intake level for chromium (EFSA, 2006).

The Expert Group on Vitamins and Minerals reviewed the toxicity of trivalent chromium in its report on safe upper levels for vitamins and minerals (EVM, 2003). The Group was unable to establish a safe upper level for chromium due to inadequacies in the database, but recommended that, for guidance purposes only, a total daily intake of 0.15 mg/kg body weight/day trivalent chromium would be expected to be without adverse effects. This was based on a study in which chromium picolinate and chromium chloride were added to the diets of rats, providing a daily intake of 15 mg/kg body weight/day, for 24 weeks without adverse effects (Anderson *et al.,* 1997). An uncertainty factor of 100 was applied to this NOAEL to account for inter-species and intra-species variability. This guidance level of 0.15mg/kg body weight/day is based on the most current information available and is considered the most relevant.

Previous COT evaluations

The COT reviewed the toxicity of chromium compounds in food in 1995. They concluded, "the levels of chromium in the UK diet (average and 97.5th percentile intake, 0.25 and 0.38 mg/kg body weight/day respectively) are not a cause for toxicological concern" (MAFF 1998). This assessment was based on the conclusion of the joint MAFF/Department of Health Working Group on dietary supplements and health foods that chromium would have undesirable effects above a dose of 1-2 g/day (MAFF/DH, 1991).

In 2003, the COT evaluated chromium in food when the results of the 2000 TDS were considered (COT, 2003). The estimated population dietary exposure to chromium from the 2000 TDS was 0.046 mg/day, approximately 0.77 µg/kg body weight/day for a 60 kg adult. The Committee concluded that current dietary exposures to chromium are unlikely to be of any toxicological concern for consumers.

In 2004 the COM reviewed the available information on the mutagenicity of trivalent chromium and specifically on chromium picolinate, a form of chromium used in some dietary supplements (COM, 2004). The Committee concluded that overall the balance of the data suggested that chromium picolinate should be regarded as not being mutagenic *in vitro*. The available *in vivo* tests in mammals with chromium picolinate were negative (COM, 2004).

References

Anderson, R.A. and Kozlovsky, A.S. (1985) Chromium intake, absorption and excretion of subjects consuming self-selected diets. *American Journal of Clinical Nutrition*, **41**, 1177 – 1183.

Anderson, R.A. *et al* (1997) Lack of toxicity of chromium chloride and chromium picolinate in rats. *Journal of the American College of Nutrition*, **16**, No.3, 273 – 279.

COT (2003). Statement on twelve metals and other elements in the 2000 Total Diet Study. COT Statement 2003/07. Annual Report 2003 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Foods, Consumer Products and the Environment.

COM (2004). Statement on the mutagenicity of trivalent chromium and chromium picolinate. COM/04/S3. Annual Report 2004 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Foods, Consumer Products and the Environment.

EFSA (2006) Tolerable Upper Intake Levels for Vitamins and Minerals. February 2006

EVM (2003). Safe upper levels for vitamins and minerals. Report of the Expert Group on Vitamins and Minerals.

Felter, S.P. and Dourson, M.L. (1997) Hexavalent chromium-contaminated soils: options for risk assessment and risk management. *Regulatory Toxicology and Pharmacology*, **25**, 43-59.

IARC (1990). Monographs on the Evaluation of Carcinogenic Risks to Humans: Chromium, Nickel and Welding, Volume 49. IARC, Lyon, France.

Kumpulainen, J. *et al* (1983) Determination of chromium in human milk, serum and urine by electrothermal atomic absorption spectrometry without preliminary ashing. *Sci. Tot. Environ.*, **31**, 71 – 81.

Ministry of Agriculture Fisheries and Food (1998). Cadmium, Mercury and other metals in food. Food Surveillance Paper 53.

Ministry of Agriculture Fisheries and Food (MAFF) and Department of Health (1991). Report of the Working Group on Dietary Supplements and Health Foods.

Offenbacher, E.G. *et al* (1986) Rapid enzymatic pretreatment of samples before determining chromium in serum and plasma. *Clinical Chemistry*, **32**, 1383 – 1386.

Rhodes, M.C., Hebert, C.D., Herbert, R.A., Morinello, E.J., Roycroft, J.H., Travlos, G.S. and Abdo, K.M. (2005). Absence of toxic effects in F344/N rats and B6C3F1 mice following subchronic administration of chromium picolinate monohydrate. The National Toxicology Programme. *Food and Chemical Toxicology*, **43**(1), 21-29.

Speetjens, J.K., Collins, R.A., Vincent, J.B., Woski, S.A. (1999). The nutritional supplement chromium (III) tris(picolinate) cleaves DNA. *Chem. Res. Toxicol.* **12:** 483-487.

Stearns, D.M., Wise, J.P., Patierno, S.R., Wetterhahn, K.E. (1995) Chromium (III) picolinate produces chromosome damage in Chinese hamster ovary cells. *FASEB Journal*, **9**, 1643 – 1649.

US EPA (1998). Integrated Risk Information System (IRIS): Chromium. US Environmental Protection Agency National Center for Environmental Assessment.

WHO (1988). Environmental Health Criteria 61: Chromium.

Zhang, J.D. and Li, S. (1997). Cancer mortality in a Chinese population exposed to hexavalent chromium in water. *J. Occup. Environ. Med.* **39(4):** 315-9.

8. Copper (Cu)

The key sources for this summary were the EVM report (EVM, 2003), the JECFA reviews of copper (WHO, 1974 and 1982) and Environmental Health Criteria 200 (WHO, 1999).

Chemistry and occurrence

Copper occurs in nature mainly in the form of its oxide, Cu_2O and sometimes as $CuCl_2.H_2O$ which, in the presence of humidity and oxygen, is oxidised to the basic copper (II) chloride, Cu(OH)Cl.

Copper is an essential trace element in both animals and humans. It plays a vital role in a number of critical enzyme systems and is closely linked with normal haematopoiesis and cellular metabolism.

The mean daily dietary intake of copper in adults ranges between 0.9 and 2.2 mg. A majority of studies have found intakes to be at the lower end of this range. The variation reflects different dietary habits as well as different agricultural and food processing practices used worldwide. In some cases, drinking-water may make a substantial additional contribution to the total daily intake of copper, particularly in households where corrosive waters have stood in copper pipes (WHO, 1999).

Absorption and elimination

Absorption of ingested copper in man ranges from 25 to 60% and has been shown to vary with diet and physiological status, for example pregnancy, which causes an increase in copper absorption and a decrease in biliary secretion. Copper absorption may be reduced by other metals, such as zinc or cadmium, and by organic materials, such as ascorbic acid (WHO, 1982). Age also affects absorption of copper, for example human infants are apparently unable to absorb copper to the same extent as adults, and are often found to be in negative copper balance (EVM, 2003). Conversely, there is also evidence that homeostatic mechanisms preventing absorption of excess copper (billiary excretion) may not be fully developed in children (Danks, 1991). Evidence from animal studies has shown that suckling rats absorb more copper following ingestion compared to weaning rats (WHO, 1982).

Faecal excretion following biliary secretion is the main route of elimination, with only minor amounts being excreted in the urine.

Toxicity in animals

In animals, copper salts have moderate acute toxicity, with soluble salts being

more toxic than insoluble ones. The data are limited but rats appear to be more tolerant to acute copper toxicity than other laboratory species. In sub-chronic repeat dose toxicity tests, copper salts are associated with effects such as gastro-intestinal irritation and liver and kidney toxicity. Herbert *et al.* (1993) reported a NOAEL for copper sulphate of 16 mg/kg body weight/day in a 13-week study in F344 rats. Above this NOAEL, effects were seen in the liver, kidneys and fore-stomach.

Copper salts have been reported to have adverse effects on reproductive and developmental parameters in animal studies. Effects on the testes, seminal vesicles, uterus and ovaries have been reported at doses of <27 mg/kg body weight/day copper, though this is not consistent (WHO, 1999). Reduced fetal weight, size and viability and delayed ossification have been reported in the offspring of rats exposed to greater than 65 mg/kg body weight/day copper (Haddad *et al.,* 1991).

There are few adequate data on chronic toxicity of copper salts. There is no evidence of direct carcinogenicity but different copper concentrations appear to have a modifying effect on tumours initiated by other agents (WHO, 1982). However, high levels of copper accumulate in the liver of the Long Evans Cinnamon rat and this is associated with a high incidence of hepatocellular carcinoma (Masuda *et al.,* 1992).

Toxicity in humans

Few data are available on chronic copper toxicity in humans.

Indian childhood cirrhosis (ICC) is a fatal disorder associated with accumulation of massive levels of copper in the liver. Although ICC has been attributed to boiling and storing milk in copper and brass vessels, elevating copper content, there also appears to be an element of genetic predisposition in many cases of ICC. Isolated cases of idiopathic copper toxicosis (ICT), identical in nature to ICC, have also been reported in non-Indian communities in the US and Europe.

Wilson's disease is an autosomal recessive inherited disorder of copper metabolism, which normally manifests in late childhood. There is a failure of normal copper excretion into the bile and of incorporation into caeruloplasmin. As a result, copper accumulates and causes toxicity, primarily in the liver and brain. Clinical manifestations may include liver disease, and neurological and psychiatric disturbances.

Increased copper levels in adults with untreated Wilson's disease, or in children who have recovered from ICC, have been associated with a possible increased incidence of hepatoma. High copper levels have also been cited as a possible risk factor for heart disease.

In the normal population, there is no evidence that elevated copper intake is associated with cancer incidence.

Previous Evaluations

In 1982 JECFA set a provisional maximum tolerable daily intake (PMTDI) of 0.05 to 0.5 mg/kg body weight/day for copper (WHO, 1982). JECFA considered that nutritional data relating to background exposure to copper from the diet indicated that the level of copper in food met the nutritional requirements (2-3 mg/day). However, they recognised that this level of intake was likely to be significantly exceeded by sections of the population, particularly in arid areas where there may be a high intake of water containing high levels of copper. There is no information that indicates that such populations are adversely affected and in addition, JECFA did not consider copper to be a cumulative toxic hazard for man, except for individuals with Wilson's disease. On this basis the previous tentative evaluation of a maximum daily load of 0.5 mg/kg body weight (WHO 1974) was reaffirmed as a provisional value for a maximum tolerable intake of 0.5 mg/kg body weight/day from all sources.

The EVM recommended a safe upper level of 0.16 mg/kg body weight/day for copper (equivalent to 10 mg/day in a 60 kg adult) (EVM, 2003). This was based on a NOAEL of 16 mg/kg body weight/day from a 13-week study in rats (Hebert *et al*., 1993) and the application of uncertainty factors of 10 to account for inter-species variability and 10 for intra-species variability. The EVM noted that the safe upper level was consistent with small-scale human studies that had indicated that intakes of up to 10 mg/day were without adverse effects. This safe upper level is based on the most current information available and is considered the most relevant.

The SCF derived a tolerable upper intake level of 5mg Cu/day (or 0.08mg/kg bw/day for a 60 kg adult) based on a small supplementation study by Pratt et al, 1985 with adults consuming 10mg copper per day for 12 weeks. An uncertainty factor of two was applied to the NOAEL from this study (EFSA, 2006).

In 1993 the WHO recommended a drinking water guideline value for copper of 2 mg/L (WHO, 1993). This value was confirmed in the most recent edition of the Guidelines (WHO, 2006). The guideline value was set to be protective against acute gastrointestinal effects and to provide an adequate margin of safety in populations with normal copper homeostasis. For adults with normal copper homeostasis, the guideline value should permit consumption of 2 or 3 litres of water per day, use of a nutritional supplement and copper from foods without exceeding the tolerable upper intake levels of 10 mg/day or eliciting an adverse gastrointestinal response (WHO, 2006).

Copper has been included in the rolling revision of the WHO Guidelines for Drinking Water Quality and an updated factsheet is due to be published in the second addendum to the third edition (WHO, 2008). Recent studies in rabbits

have suggested a link between copper in drinking water at levels below the current guideline of 2 mg/L and Alzheimer's disease although it has not been established whether rabbits are a good model for this disease (WHO, 2008).

Children may be at increased risk of copper toxicity due to immature biliary excretion of copper. However, infants are apparently unable to absorb copper to the same extent as adults and are often found to be in negative copper balance. There is also evidence that copper is accumulated in the third trimester of pregnancy without apparent adverse effect, suggesting that neonates may be resistant to high levels of hepatic copper (EVM, 2003). WHO are currently reviewing the drinking water guideline value for copper with regard to toxicity in the preparation of formula for bottle-fed infants (WHO, 2008).

Previous COT evaluations

In 1998 when considering the levels of lead, arsenic and other metals in food (MAFF, 1998), the COT concluded that "the animal toxicity data on copper are inadequate, but there is no evidence that the dietary intake of copper in the UK (1.4 mg/day) is harmful to human health".

In 2003, the COT evaluated copper in food when the results of the 2000 TDS were considered (COT, 2003). The estimated population dietary exposure to copper from the 2000 TDS was 1.3 mg/day, approximately 21.6 µg/kg body weight/day for a 60 kg adult. The Committee concluded that current dietary exposures to copper are unlikely to be of any toxicological concern for consumers.

References

COT (2003). Statement on twelve metals and other elements in the 2000 Total Diet Study. COT Statement 2003/07. Annual Report 2003 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Foods, Consumer Products and the Environment.

Danks, D.M. (1991). European Journal of Paediatrics, **150**: 142-148.

EVM (2003). Safe upper levels for vitamins and minerals. Report of the Expert Group on Vitamins and Minerals.

EFSA (2006) Tolerable Upper Intake Levels for Vitamins and Minerals. February 2006

Haddad, D.S., Al-Alousi, L.A. and Kantarjian, A.H. (1991). The effect of copper loading on pregnant rats and their offspring. Funct. Dev. Morphol. **1**: 17-22.

Hebert, C.D., Elwell, M.R., Travlos, G.S., Fitz, C.J. and Bucher, J.R. (1993). Subchronic toxicity of cupric sulfate administered in drinking water and feed to rats and mice. Fundam. Appl. Toxicol. **21**: 461-475.

Masuda, R., Yoshida, M.C., Sasaki, M., Dempo, K. and Mori, M. (1992). High susceptibility to hepatocellular carcinoma development in LEC rats with hereditary hepatitis. Jpn. J. Cancer Res. **79**: 828-835.

MAFF (1998). Lead, Arsenic and other metals in Food. Food Surveillance Paper No. **52**. Ministry of Agriculture, Fisheries and Food. The Stationery Office, London.

WHO (1974). Toxicological evaluation of some food additives including anticaking agents, antimicrobials, antioxidants, emulsifiers and thickening agents: Cupric sulfate, WHO Food Additives Series No. 5 .

WHO (1982). Toxicological evaluation of certain food additives. WHO Food Additives Series, No. 17. Copper.

WHO (1999). Environmental Health Criteria 200: Copper. World Health Organization, Geneva, pp. 100-129.

WHO (1993). World Health Organisation Guidelines for Drinking Water Quality, Volume 1 p42-43, Geneva

WHO (2006). Guidelines for Drinking-Water Quality. Volume 1 Recommendations. First addendum to the third edition. World Health Organization, Geneva.

WHO (2008). Plan of work for the rolling revision of the WHO Guidelines for Drinking Water Quality. World Health Organization website: http://www.who.int/water_sanitation_health/gdwqrevision/en/index.html. Accessed July 2008.

9. Germanium (Ge)

The key source for this summary was the EVM report (EVM, 2003).

Chemistry and occurrence

Germanium is a non-metallic element which can exit in valence states of 2 and 4. It is not considered to be an essential trace element for the human diet.

In foods, organic germanium is present in beans, tomato juice, oysters, tuna and garlic. Germanium food supplements (mainly inorganic germanium) were voluntarily withdrawn by industry in the UK due to toxicity but can be obtained by mail order over the internet. Germanium is used for its semiconducting properties in diodes and transistors; as a component of glass of high refractive index; in some alloys and acid-lead batteries; and as a catalyst.

Dietary intake is the principal source of exposure in the general population. Reviews usually quote an estimated daily intake of about 1.5 mg in the US, on the basis of a 1967 paper (Schroeder and Balassa, 1967), but a lower estimate of 367±159 µg/day was derived for the UK from the 1966/7 samples of diet obtained by MAFF for pesticide surveillance (Hamilton and Minski, 1972).

Absorption and elimination

Inorganic germanium compounds are absorbed rapidly and nearly completely from the gastrointestinal tract. Organic compounds such as carboxyethyl germanium sesquioxide (also known as Ge-132, propagermanium, or SK-818) are less well absorbed (about 30% in rats and humans). Following low doses, single or repeated, the tissue distribution is even. In contrast, repeated high doses of inorganic germanium compounds result in preferential accumulation in the kidneys, and in other tissues including intestine and spleen.

Germanium is distributed throughout the body tissues, particularly the kidneys and thyroid glands. The biological half-life of germanium compounds is typically relatively short (several days in rats, and several hours in mice). Excretion of absorbed germanium is largely through the kidneys (Beliles, 1994; Furst, 1987; and Tao and Bolger, 1997).

Toxicity in animals

Acute oral LD50s are reported to range from 83 mg Ge/kg body weight (triethyl germanium acetate in rats; Furst, 1987) to 870 mg Ge/kg body weight (GeO_2 in rats and mice; Lewis, 199) and 5000 mg Ge/kg body weight (Ge-132 in male rats; Schroeder and Hamilton and Minski, 1972; Balassa, 1967; Lewis, 1996; and Tao and Bolger, 1997).

Several studies have demonstrated that ingestion of inorganic germanium compounds (usually germanium dioxode, GeO_2) at high doses for prolonged

periods can affect longevity, growth, kidneys, liver, central and peripheral nervous systems, skeletal and cardiac muscle, and haematopoiesis (EVM, 2003). Many of the studies have concentrated on nephrotoxicity, in the light of clinical reports. Organic compounds are less toxic but produce weight loss and decreased red blood cell counts (EVM, 2003). The mechanisms of germanium-induced target organ toxicity are uncertain.

The nephrotoxicity of germanium typically affects the thick ascending limb of the loop of Henle and the distal convoluted tubule. There is severe degeneration of mitochondria, with vacuolar degeneration. The glomeruli are spared, but their capillaries may be dilated. The concentration of germanium in the kidney is usually greatly elevated. This pathology has been produced in rats, mice, rabbits, guinea pigs, monkeys, and humans. The lowest effective oral dose identified in experimental animals is about 0.35 mg Ge/kg body weight/day, in poorly-described lifetime studies in rats and mice, published in the late 1960s (Kanisawa and Schroeder, 1967, 1969; and Schroeder *et al.*, 1968). In these studies, animals on a diet 'low in many trace elements' were given drinking-water containing sodium germanate to provide 5ppm Ge (although there is some ambiguity about this, and the concentration may be as germanate ion). In Long Evans rats, nephrotoxicity and hepatotoxicity (fatty degeneration) were seen, with reduced weight gain in males, and reduced lifespan in both sexes. In white Swiss mice, weight gain and survival were reduced, but no changes in kidney or liver were reported. In another study in rats (Taylor, 1991), 50 ppm Ge (as GeO_2) in the diet for 6 weeks (i.e. about 2.5 mg Ge/kg body weight/day) produced renal pathological changes with apparently normal renal function. The other studies all generated nephrotoxicity with higher doses and longer periods of dosing. None of the studies established a NOAEL.

Other adverse effects seen in the studies of oral dosing with inorganic germanium include mitochondrial swelling and fibre atrophy in skeletal and cardiac muscles, peripheral neuropathy (segmented demyelination and remyelination, without axonal degeneration) and anaemia, at doses of about 9 mg Ge/kg body weight/day and above. None of the studies is adequate to establish NOAELs for these effects.

Several studies of oral dosing of rats with the organic germanium compound Ge-132 have not detected evidence of nephrotoxicity, even at doses as high as 320 mg Ge/kg body weight/day for 1 year (Nakagawa *et al.*, 1990), and 1720 mg Ge/kg body weight/day for 3 months (Kanda *et al.*, 1990). In contrast, one study found renal tubular changes at 300 mg Ge/kg body weight/day for 6 months, in male but not female rats (Anger *et al.*, 1992); while another reported renal pathological changes with normal renal function after only about 2.5 mg Ge/kg body weight/day for 6 weeks (Taylor, 1991).

Few data are available on reproductive toxicity (EVM, 2003).

Sodium germinate was not carcinogenic in rats. Mutagenicity data are limited, but negative results were obtained with the germanium compounds tested (EVM, 2003).

Toxicity in humans

Optimistic extrapolation from laboratory data resulted in the popular use, in the 1970s and 1980s, of inorganic and organic germanium preparations as dietary supplements with supposed beneficial effects in relation to a range of conditions including immunodeficiency, cancer, heart disease, arthritis, mental illness and so forth (Goodman, 1988). Subsequently, cases of renal failure were reported in users of germanium supplements (Tao and Bolger, 1997). Initial symptoms include anorexia, weight loss, fatigue and muscle weakness. This is followed by renal dysfunction and failure, which can be fatal. Fatal renal failure in humans has occurred following cumulative doses of >20 g germanium. Where patients have survived, renal function has not returned to normal. The pathological findings of tubular damage were identical to those seen in experimental animals. Tao and Bolger (1997) suggested that the lowest germanium dose associated with clinical toxicity is about 0.7 mg/kg body weight/day. The subject of one case report (Asaka, 1995) of nephrotoxicity and neurotoxicity was a 63-year old woman who died after taking about 0.6 mg inorganic Ge/kg body weight/day for 6 years.

Previous evaluations

Germanium has not been considered by JECFA, IARC or IPCS. In 1989, following reports of nephrotoxicity in chronic consumers of germanium dietary supplements (in the range of 50-250 mg daily for 4-18 months), DH advised the public that it was not safe to take any preparation containing germanium, and asked suppliers to withdraw all germanium products from retail sale immediately. The COT endorsed this action. At that time, the normal dietary intake of germanium was believed to be about 1 mg/day.

The EVM reviewed germanium in 2003 concluding that, germanium is a cumulative toxin causing serious, and potentially fatal, adverse effects on the kidneys (EVM, 2003). Naturally occurring germanium present in food does not appear to be associated with any adverse effects, but there were insufficient data to define a NOAEL for chronic exposure at levels in excess of this. Therefore, given the cumulative nature of germanium toxicity, there were insufficient data to set a safe upper level for any amount of germanium in excess of that provided by the diet (EVM, 2003).

Previous COT evaluations

In 1998, the COT evaluated germanium in food when the results of the 1994 TDS were considered (COT, 1998). The estimated population dietary exposure to germanium from the 1994 TDS was 0.004 mg/day, approximately 0.07 µg/kg body weight/day for a 60 kg adult. The Committee concluded that the estimated dietary intakes of germanium in adults did not give cause for concern. The estimated intakes from the dietary supplements sampled, which did not include germanium supplements, were very low and did not give cause for concern. However, DH advice to the public that it was not safe to take any preparation containing germanium (i.e. germanium supplements) remained appropriate (COT, 1998).

References

Anger F, Anger JP, Guillou L and Papillon A (1992). Appl Organometallic Chem **6,** 267-272.

COT (1998). Statement on the results of multielement surveys in various items of the diet. COT Statement 1998. Annual Report 1998 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Food, Consumer Products and the Environment.

Beliles RP (1994). Germanium in Clayton GD and Clayton FE (eds). Patty's Industrial Hygiene and Toxicology, Vol II, Part C, pp 2013-2020.

EVM (2003). Safe upper levels for vitamins and minerals. Report of the Expert Group on Vitamins and Minerals.

Furst A (1987). Toxicology and Industrial Health **3,** 167-204.

Goodman S (1988). Germanium, the health and life enhancer. Thorsons Publishing Group, Wellingborough.

Hamilton EI and Minski MJ (1972/1973). Sci. Total Environ. **1,** 375-394.

Kanda K, Ikawa E, Tagawa Y et al (1990). Clin Rep **24,** 46-89.

Kanisawa M and Schroeder HA (1967). Cancer Res **27,** 1192-1195.

Kanisawa M and Schroeder HA (1969) Cancer Res **29,** 892-895.

Lewis RJ Sr (1996). Sax's Dangerous Properties of Industrial Materials, 9th edition. Vol II, p 1727.

Nakagawa H, Segawa M, Kanore M et al (1990). Clin Rep **24,** 144-177.

Tao S-H and Bolger PM (1997). Regul. Toxicol. Pharmacol. **25,** 211-219.

Schroeder HA and Balassa JJ (1967). J. Chronic Dis. **20,** 211-224.

Schroeder HA, Kanisawa M, Frost DV and Mitchener M (1968). J Nutr **96,** 37-45.

Taylor A (1991). Clin Chem **37,** 985.

10. Indium (In)

Indium has not been the subject of any international review by WHO or other relevant bodies (e.g. EFSA, EPA etc).

Chemistry and occurrence

Indium is used in the electronics industry for production of semiconductors and photovoltaic cells; in medicine, for scanning of organs and treatment of tumours, and; in alloys (including some dental alloys), solders and bearings.

Indium was included in a MAFF multi-element survey of samples of crops and cows' milk from locations around industrial sites. Estimated daily maximum intakes from crops and milk for adults in this survey were 0.06 µg and 0.8 µg, respectively.

No information is available on the forms of indium found in foods.

Absorption and elimination

Gastrointestinal absorption of indium salts is poor. Only from 0.2 to 0.7% was absorbed in dogs (Beliles, 1994), mice (Valberg *et al.*, 1981), and rats (Zheng *et al.*, 1994). Once in the circulation, indium ions are uniformly distributed among tissues. Urinary excretion was small, and it appeared that faecal excretion was the major route for absorbed indium. Multiple oral dosing did not increase the total body burden of indium, suggesting that it is unlikely to accumulate (Zheng *e al.*, 1994).

Toxicity in animals

The lowest lethal oral dose of indium oxide reported (in the mouse) is 10 g/kg body weight, corresponding to an indium dose of 8300 mg/kg body weight. The lowest lethal oral dose of indium sulphate reported (in the rat) is 1200 mg/kg body weight, corresponding to an indium dose of 530 mg/kg body weight (Lewis, 1996).

There are few data on the oral toxicity of indium and its compounds. One review states, without references, that indium oxide at 8 % in the diet of rats for 3 months had no discernible effect on growth, mortality or tissue morphology, but that indium trichloride caused a slight growth depression at 2.4 % and a marked depression at 4 %(Beliles, 1994). Such results would imply a NOAEL of about 3300 mg In/kg body weight/day in the first study, but a LOAEL (for slight growth depression) of about 620 mg In/kg body weight/day in the second study.

In a lifetime study, the growth rate of mice receiving indium trichloride in drinking-water at 5 ppm indium, was reduced (Schroeder and Mitchener, 1971). There was no evidence of carcinogenicity. This would suggest a LOAEL of about 250 µg/kg body weight/day.

No data are available on the reproductive or developmental toxicity of indium following oral dosing. Limited studies indicate that indium is not mutagenic or carcinogenic.

Toxicology in humans

There appear to be no reports of human toxicity from oral indium or its compounds.

Previous COT evaluations

Data from the 1979 Total Diet Study were evaluated by the COT (MAFF, 1985). The mean daily dietary intake of indium was estimated as between 5 and 27 µg, with intakes ranging from 0 to 30 µg/day. Intakes from water and air were believed to be negligible. The COT considered that the level of indium in food was not a cause for concern (MAFF, 1985).

In 1998 the COT evaluated the results from a multi-element survey of crops and cows' milk from locations around industrial sites (COT, 1998). The COT concluded that the intakes of indium in adults are very low, and the data indicate that the upper bound estimates of dietary intakes in the 1979 Total Diet Study were probably significantly inflated by the relatively high limit of detection. The sparse data on the oral toxicity of indium did not suggest that the estimated intakes give any cause for concern (COT, 1998).

References

Beliles RP (1994) Indium in Clayton GD and Clayton FE (ed). Patty's Industrial Hygiene and Toxicology, Vol II, Part C, pp 2032-2038.

COT (1998). Statement on the results of multielement surveys in various items of the diet. COT Statement 1998. Annual Report 1998 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Food, Consumer Products and the Environment.

Lewis RJ Sr (1996). Sax's Dangerous Properties of Industrial Materials, 9th edition. Vol III, p 1919.

MAFF (1985). Food Surveillance Paper No 15. London, HMSO.

Schroeder HA and Mitchener M (1971). J Nutr **101**, 1431-1438.

Valberg LS et al (1981). Clinical and Investigative Medicine **4**(2), 103-108.

Zheng W et al (1994). J Toxicol Environ Health **43**, 483-494.

11. Lead (Pb)

The key sources of information for this summary were the JECFA evaluations of lead (WHO, 1987 and 2000), Environmental Health Criteria 165 (WHO, 1995) and the ATSDR toxicological profile (ATSDR, 2007).

Chemistry and occurrence

Lead occurs naturally as a sulphide in galena but is also found naturally in soil as a sulphate, carbonate or oxide. It is a soft, malleable metal with a melting point of 327.5 ^{o}C. Elemental lead reacts with hot boiling acids and is attacked by pure water. The solubility of lead salts in water varies from insoluble to soluble depending on the type of salt (IARC, 1980).

Lead is persistent in water and soil. Most of the lead in environmental media is of anthropogenic sources. Human exposure is mainly to inorganic lead and occurs primarily through diet, air, drinking water, and ingestion of dirt and paint chips (ATSDR, 2007).

Absorption and elimination

The efficiency of lead absorption depends on the route of exposure, age, and nutritional status. Adult humans absorb about 10-15% of ingested lead, whereas children may absorb up to 50% (WHO, 2000).

Lead absorbed into the body is distributed to three major compartments: blood, soft tissue, and bone. The largest compartment is the bone, which contains about 95% of the total body lead burden in adults and about 73% in children. The half-life of bone lead is more than 20 years. In rats during pregnancy and lactation, lead in the bones is mobilised and transferred to more available compartments of the maternal circulation and therefore increasing the exposure of the fetus. An increase in the calcium intake during pregnancy appears to cause an increase in this mobilisation, enhancing the efflux of lead from the bone. It is possible that this also occurs in humans (Vega *et al.,* 2002).

The concentration of blood lead changes rapidly with exposure and its half-life of only 25-36 days is considerably shorter than that of bone lead. The soft tissues that take up lead are liver, kidneys, brain, and muscle. Lead is excreted primarily in the urine following conjugation with glutathione (WHO, 2000). Due to the short half-life of lead in the blood and soft tissues, blood lead concentration is only useful as an indication of recent lead exposure, whereas bone lead levels are a better indication of lifetime exposure.

Toxicity in animals

Lead is a chronic or cumulative poison. Health effects are generally not seen after a single dose. The lowest observed lethal doses in animals range from 300 to 4000 mg/kg body weight given in multiple administrations. The wide range is attributable to the differences in the absorption of the different lead compounds used and the differences in exposure method (ASTDR, 2007).

In laboratory animals lead has been shown to have effects on bone and the immune system as well as haem synthesis, neurological and behavioural effects, renal effects, cardiovascular effects, and effects on the reproductive system. Impaired learning ability has been reported in rats with blood lead concentrations of 15-20 µg/dL and in non-human primates with blood levels of <15 µg/dL. Visual and auditory impairments have been reported in experimental animals. Renal toxicity in rats appeared to occur at concentrations exceeding 60 µg/dL, while cardiovascular effects have been noted at 40 µg/dL (WHO, 2000).

Lead-induced gene mutations in cultures of mammalian cells have only been observed at concentrations toxic to the cells (WHO, 2000).

Lead arsenate and lead phosphate have been found to produce an increase in renal tumour frequency in rodents at doses >10 mg/kg body weight, which also caused renal toxicity (WHO, 2000).

Toxicity in humans

Children are more sensitive to lead exposure than adults. Irreversible brain damage has been reported at blood lead levels greater than or equal to 100 µg/dL in adults, whereas deaths have been reported at blood lead levels as low as 80 µg/dL in children. Children who survive these high levels of exposure suffer permanent and severe mental retardation. Young children are vulnerable to the effects of lead because they absorb a higher percentage of ingested lead and are more susceptible to the neurotoxicity, which may result in deficits in Intelligence Quotient (IQ) (ATSDR, 2007).

Reduced birth weight and gestational age have been associated with exposure to lead *in utero* (Dietrich *et al*., 1987), but conflicting results have been obtained (Graziano *et al*., 1990; ATSDR, 2007). The most important and best-documented effect of lead at the concentrations most commonly encountered outside occupational settings is on the neurobehavioural development of children of mothers exposed to lead. Studies have shown an association between blood lead concentrations and reduced IQ in children exposed pre- and post-natally (WHO, 2000). No threshold for intellectual deficits has been identified but there is evidence of an association at blood lead concentrations below 10 µg/dL (WHO, 2000).

Neuropsychological impairment and cognitive (IQ) deficits are sensitive indicators of lead exposure, particularly *in utero* exposure; both have been the subject of cross-sectional and longitudinal studies in children. The developing nervous system is especially susceptible to lead toxicity because lead interferes with brain development and neurobehavioural deficits in children are associated with blood lead concentrations <10 µg/dL (ATSDR, 2007). However, the lack of a demonstrated convincing biological mechanism which could explain the dose-effect relationship at lower blood lead levels remains uncertain (ATSDR, 2007; Bellinger, 2004). There therefore remains no identified threshold. Meta-analyses conducted on cross-sectional studies or a combination of cross-sectional and prospective studies suggest that an IQ decline of 1-5 points is associated with an increase in blood lead concentration of 10 µg/dL (ATSDR, 2007). Surveys of blood lead concentrations have indicated reductions in mean blood lead concentrations since the late 1970s (ATSDR, 2007; Koller *et al.*, 2004; WHO, 2000; WHO, 2007). Current mean levels in children of developed countries are in the region of 3 µg/dL (Koller *et al.*, 2004). This reduction has been attributed to the reduction in the use of lead in petrol and as a result of programmes to reduce exposure.

Exposure to lead has been associated with increased incidences of hypertension and cardiovascular disease (Pirkle *et al*., 1985; ATSDR, 2007). The relationship between blood lead and blood pressure appears to be greatest at concentrations below 50 µg/dL, and increases in blood lead concentration beyond this level do not appear to correlate with greater hypertension. The causal relationship between lead and hypertension is not clear, however the elevation may be secondary to effects on other organ systems such as the kidney or the haematopoetic system.

In a recent evaluation, IARC classified inorganic lead compounds as ‘probably carcinogenic to humans’ (Group 2A) and organic lead compounds as ‘not classifiable as to their carcinogenicity to humans’ (Group 3) (IARC, 2006).

Previous evaluations

JECFA set a PTWI for lead of 25 µg/kg body weight in 1986 based on evidence that a mean daily intake of 3-4 µg/kg body weight of lead by infants and children was not associated with an increase in blood lead levels (WHO, 1987). The JECFA again evaluated lead in 1993 when the Committee estimated what blood lead level the PTWI would lead to. As this was below levels known to be associated with intellectual deficits in children at the time, the PTWI of 25 µg/kg body weight for infants and children was re-confirmed and extended to all age groups (WHO, 1993). The review of the health effects of lead in 1993 was based on an assessment of lead that had been performed by an International Programme on Chemical Safety Task Group which was subsequently published (WHO, 1995). In 2000, the JECFA assessed the risk of dietary exposure of infants and children, with special emphasis on the most critical effect, which was considered to be impaired neurobehavioural development. The PTWI was not re-considered (WHO, 2000).

Based on the JECFA PTWI, WHO set a drinking water guideline value of 0.01 mg/L, assuming a 5 kg infant drinking 0.75 litres/day and allocating 50% of the PTWI to drinking water. This guideline value was first set in 1993 and was recently confirmed in the latest edition of the Guidelines for Drinking Water Quality (WHO, 2006).

The US EPA reviewed lead in 2004 concluding that it was inappropriate to develop reference values for lead (US EPA, 2004). This conclusion was reached given the difficulty in accounting for pre-existing body burdens, the bioaccumulation of lead, the variation in body burdens, and the continued apparent lack of a threshold effect.

EFSA have recently issued a call for new data on the safety of lead and intend to update the current knowledge on exposure from food and beverages including drinking water.

Previous COT evaluations

The COT reviewed the toxicity of lead in 1995 and stated that 'we welcome the decline in lead levels compared with earlier surveys. It is encouraging that measures taken to reduce lead contamination of food and the environment appear to be reducing the average dietary intake of lead in the UK, although we recognise that changes in the preparation and analysis of the total diet may be responsible for part of this apparent decline. Since it is not possible to identify, from epidemiological studies, a threshold for the association between exposure to lead and decrements in IQ, efforts should continue to reduce lead exposure from all sources' (MAFF, 1998).

In 2003, the COT considered the levels of lead in infant foods following a multi-element survey of infant foods and formulae (COT, 2003a). They concluded that since it is not possible to identify a threshold for the association between lead exposure and decrements in intelligence quotient, efforts should continue to reduce lead exposure from all sources. They were reassured that there has been an apparent decline in lead exposure since the previous survey of infant food and formulae.

Also in 2003, the COT evaluated lead in food when the results of the 2000 TDS were considered (COT, 2003b). The estimated population dietary exposure to lead from the 2000 TDS was 0.0074 mg/day, approximately 0.12 µg/kg body weight/day for a 60 kg adult. The Committee noted that estimates of total exposure to lead, including that from the diet, did not exceed the PTWI. The Committee concluded that current dietary exposures to lead were unlikely to result in adverse effects, but that efforts should continue to reduce exposure to lead from all sources (COT, 2003b).

References

ATSDR (2007). *Toxicological Profile for Lead.* Agency for Toxic Substances and Disease Registry. Atlanta, GA: Department of Health and Human Services.

Bellinger, D.C. (2004). Lead. *Paediatrics*, **113** (supp 4), 1016-1022.

COT (2003a). Statement on a survey of metals in infant foods (2003-02). Committee on Toxicity of chemicals in foods, consumer products and the environment). Available at: http://www.food.gov.uk/science/ouradvisors/toxicity/statements/cotstatements2003/statementmetals.

COT (2003b). Statement on twelve metals and other elements in the 2000 Total Diet Study. COT Statement 2003/07. Annual Report 2003 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Foods, Consumer Products and the Environment.

Dietrich, K.N., Krafft, K.M., Bornschein, R.L., Hammond, P.B., Berger, O., Succop, P.A. & Bier, M. (1987). Low-level fetal lead exposure effect on neurobehavioral development in early infancy. *Pediatrics*, **80**: 721-730.

Graziano, J.H., Popova, D., Factor-Litvak, P., Shrout, P., Kline, J., Murphy, M.J., Zhao, Y., Mehmeti, A., Ahmedi, X., Rajovic, B., Zvicer, Z., Nenezic, D.U., Lolacono, N.J. & Stein, Z. (1990). Determinants of elevated blood lead during pregnancy in a population surrounding a lead smelter in Kosovo, Yugoslavia. *Environ. Health Perspectives*, **89**: 95-100.

IARC (1980). *Lead and lead compounds, Vol. 23*. IARC Monographs on the Evaluation of the Carcinogenic Risk of Chemicals to Humans, Some Metals and Metallic Compounds. International Agency for Research on Cancer, Lyon, France, pp. 325-415.

IARC (2006). *Inorganic and organic lead compounds,* Volume 87. IARC Monographs on the Evaluation of the Carcinogenic Risk of Chemicals to Humans, International Agency for Research on Cancer, Lyon, France.

Koller, K., Brown, T., Spurgeon, A. and Levy, L. (2004). Recent developments in low-level lead exposure and intellectual impairment in children. *Environmental Health Perspectives,* **112**(9), 987-994.

MAFF (1998). Lead, Arsenic and other metals in Food. *Food Surveillance Paper* No. **52**. Ministry of Agriculture, Fisheries and Food. The Stationery Office, London.

Pirkle, J.L., Schwartz, J., Landis, J.R. & Harlan, W.R. (1985). The relationship between blood lead levels and blood pressure and its cardiovascular risk implications. *Am. J. Epidemiol.*, **121:** 246-258.

US EPA (2004). Integrated Risk Information System (IRIS): Lead and compounds. Environmental Protection Agency National Center for Environmental Assessment.

Vega, M.M., Solorzano, J.C., Salinas, J.V. (2002). The effects of dietary calcium during lactation on lead in bone mobilization: implications for toxicology. *Hum. Exp. Toxicol*. **21(8):** 409-14.

WHO (1987). Safety evaluation of certain food additives and contaminants, WHO Food Additives Series 21. Lead.

WHO (1993). Evaluation of certain food additives and contaminants. forty-first report of the Joint FAO/WHO Expert Committee on Food Additives. WHO Technical Report Series, No. 837, World Health Organization, Geneva.

WHO (1995). *Environmental Health Criteria 165: Inorganic Lead*, Geneva: International Programme on Chemical Safety.

WHO (2000). Safety evaluation of certain food additives and contaminants. WHO Food Additives Series 44: Lead.

WHO (2006). Guidelines for Drinking-Water Quality. Volume 1 Recommendations. First addendum to the third edition. World Health Organization, Geneva.

WHO (2007). Blood lead levels in children. Fact sheet no. 4.5, May 2007. European Environment and Health Information System, World Health Organization.

12. Manganese (Mn)

The key source of information for this summary was the EVM report (EVM 2003).

Chemistry and occurrence

Manganese is an abundant metallic element that can exist in a variety of oxidation states. Mn2+ and Mn3+ are the most biologically important. Within this risk assessment, the word manganese refers to ionic manganese, except when specific manganese compounds are mentioned.

Manganese is present both naturally and as a result of contamination in soils, sediments and water. Manganese is an essential component of a number of enzymes in the body and activates a number of other enzymes. It is therefore an essential component of the human diet.

Manganese is present in foods, particularly green vegetables (2 mg/kg), nuts (14.9 mg/kg), bread (8 mg/kg) and other cereals (6.81 mg/kg). Tea is a rich source of manganese, containing 2.71 mg/kg and is the largest contributor to manganese intake. It is present in several licensed medicines, in combination with other substances, in use for prevention and treatment of nutrient deficiencies and other related conditions. Manganese is also present in a number of multi-vitamin and/or mineral food supplements at levels up to 10 mg/daily intake.

The level of manganese in drinking water ranges from 0.001 to 0.1 mg/L, but is mostly around 0.01 mg/L. Exposure to manganese may also occur through inhalation of airborne particles by miners, smelters and workers in the manufacture of dry batteries.

A method for assessing manganese status has not been defined, because of an inconsistent relationship between tissue measures and external exposures. Measurement of manganese-specific superoxide dismutase (MnSOD) activity and the ratio between manganese-specific superoxide dismutase and zinc copper superoxide dismutase activity has been proposed as a method for assessment, but may be confounded by elevated cytokine levels and disease states which increase MnSOD expression, independent of manganese status.

Absorption and elimination

Absorption takes place in the small intestine via a carrier-mediated mechanism; passive diffusion may also occur. Absorption is generally low but appears to be higher in infants and young animals. Bioavailability of manganese from different food types is variable, but appears to be generally low, due to poor solubility. Manganese and iron compete for absorption sites. Fibre, phytate, calcium, phosphorus and magnesium may also interfere with

manganese absorption. It has been suggested that ethanol may enhance manganese toxicity.

In the portal blood, manganese may bind to albumin and alpha-2 macroglobulin. A small proportion of manganese is oxidised to Mn3+, and enters the systemic circulation, possibly by binding to transferrin. Manganese accumulates in mitochondria-rich tissues such as liver and pancreas. Manganese also accumulates in the brain, particularly in the globus pallidus, striatum and substantia nigra.

Manganese is excreted largely in the faeces, mostly as a result of biliary excretion, although some direct secretion also occurs. A small amount of manganese is excreted in the urine.

Toxicity in animals

Manganese has low acute toxicity but neurotoxic effects have been observed in animals chronically fed high concentrations of manganese salts in the diet. High doses of manganese have also resulted in anaemia as a result of iron sequestration. Fertility is reduced by high doses of manganese but other reproductive parameters are unaffected.

Other more limited data suggest that adverse effects may occur at even lower intake levels in children. In laboratory animals, adverse effects have been reported following long term exposure to manganese at doses greater than 50-200 mg/kg bw/day. Detailed neurological examinations were performed in only one study in mice which detected effects at ≤130 mg/kg bw/day.

Manganese has been tested for carcinogenicity and for genotoxicity in bioassays in rats and mice. No clear evidence of carcinogenicity has been found in either species. Some positive *in vitro* mutagenicity tests have been reported, although results are generally mixed. Studies in mice have indicated that manganese is not mutagenic *in vivo*.

Toxicity in humans

Occupational exposure, for example in manganese miners and smelters, to high levels of inhaled manganese has been associated with manganism, a neurotoxic condition similar to Parkinson's disease. Drinking water contaminated with manganese has also been associated with neurological and behavioural effects. There is an association between manganese accumulation and liver disease but this may be due to impaired biliary excretion caused by the liver disease rather than manganese toxicity. Effects on the immune system have been reported.

Supplementation trials of human volunteers with 15 or 9 mg manganese/day for 124 days and 'many months' respectively have not reported any adverse effects, but it is unclear whether information on such effects was specifically sought.

Several hypotheses have been proposed to explain the neurotoxicity of manganese, including irreversible oxidation of dopamine, via the reduction of Mn3+ to Mn2+ or interference with calcium. Other hypotheses include inhibition of mitochondrial respiration, decreased glutathione peroxidase and oxidant damage.

Manganese is a known neurotoxin at high occupational levels of inhalation exposure. However, it has also been suggested that exposure from lower levels in drinking water may result in more subtle neurological effects in human populations. Neurological effects have been reported at estimated intakes of 3.6-4.6 mg manganese from water, though effects were not found at comparable intakes in other studies.

Anaemic individuals may be vulnerable to the toxic effects of manganese due to the increased absorption that occurs in states of iron deficiency. Groups with impaired biliary clearance, such as patients with liver disease or older people, may also be susceptible to manganese accumulation and toxicity. It has also been reported that ethanol and long-term use of anti-psychotic drugs increases the susceptibility of humans to manganese toxicity.

A retrospective study of three cohorts who were exposed to water containing 0.0036-0.015, 0.08-0.25 or 1.8-2.3 mg/L manganese was carried out. The subjects were all aged over 50 years and had been exposed to manganese for >10 years. No estimates of the actual amount of water consumed or the manganese content of the diet were provided, although the 3 cohorts were considered to be comparable. Assuming 2L/day water consumption, intakes for the three groups can be estimated to be 0.0072-0.03, 0.16-0.5 and 3.6-4.6 mg manganese plus dietary manganese. There was no difference in blood manganese levels in subjects from the 3 areas, but hair manganese was higher in subjects in the area with the highest level of manganese. It has been noted (US DHHS, 1997) that hair manganese is a useful measure of exposed versus unexposed populations but that it is of limited use in assessing individual exposure. The authors suggested that this could be because blood levels are highly variable and depend on how recent intake is, and thus could mask the influence of manganese stored in body tissues. A history was taken and a physical examination conducted by a neurologist who was unaware of the manganese exposure status. Neurological signs and symptoms were scored and were elevated in subjects from the highest manganese area. The symptoms scored ranged from depression, fatigue and hallucinations to tremor and impaired reflexes. Some of the symptoms were relatively subjective (e.g. fatigue) and it is uncertain whether one or two unusual ‘outlier’ individuals with a number of symptoms and/or signs may have produced the observed elevation in the high intake group. The authors noted that an effect was apparent at exposure levels which were negative in occupational studies. The authors attributed this finding to the age of the subjects, making them more susceptible to neuronal loss with ageing. The study was limited by inconsistent evidence of significant excess exposure and the subjective ascertainment of clinical effects (Kondakis et al., 1989).

A retrospective study of two cohorts exposed to water containing <0.05 mg/L or >0.3 mg/L (range 0.3-2.16 mg/L) manganese was carried out. The subjects

in the two groups were aged 41-84 (mean = 57.5) years and 41-86 (mean 56.9) years. Long duration of exposure (10-40 years) to manganese had taken place. No estimates of the actual amount of water consumed or of the manganese content of the diet were provided by the authors, but the cohorts were considered to be comparable. Assuming 2 L/day water consumption, manganese intakes of 0.1 and 4.3 mg from water can be estimated. A neurologist blinded to the group status of the participants conducted the examination. No significant differences in blood manganese levels or neurological scores were found between the two groups. The authors noted the conflict with the findings of Kondakis *et al*., which they attributed to the greater age of the participants in that study. This study was also limited by the lack of exposure data (Vieregge et al., 1995*)*.

A major limitation of both studies (Vieregge et al., 1995, Kondakis et al., 1989) is the failure to provide water consumption or dietary manganese intake data.

It has generally been considered that manganese uptake from oral exposure is subject to homeostatic control. However, there is also a suggestion of less severe neurotoxic effects at lower levels of exposure from consuming drinking water containing higher levels of manganese. The reported symptoms include muscle pain, fatigue, tremor, memory problems and impaired reflexes. Although other human data are available as noted above, it is not possible to draw conclusions from these due to problems with co-exposure, study design or lack of data.

Update since the EVM report

Since the EVM report was published in 2003, a number of new studies on manganese have been published. Whilst it is known that high level acute exposure to manganese can cause manganism, a syndrome resembling Parkinson's disease, there are an increasing number of studies suggesting that low level longer term exposure may also cause neurotoxicity manifesting itself as sub-clinical and sub-functional declines in the performance of specialised neuropsychological tests but data on the levels necessary to cause this are unclear (Martin, 2006). There is also evidence to suggest that many of the existing models for neurotoxicity may be of limited relevance for manganese because of differences of neurotoxicity observed between species (Gwiazda et al, 2007).

Previous evaluations

In 1996, the US Environmental Protection Agency set a reference dose for manganese of 1.4 mg/kg body weight/day using dietary information from a range of population groups (US EPA, 1996).

In 2000, the SCF concluded that there was insufficient evidence available to set an Upper Limit for manganese in the diet (EFSA, 2006).

In 2003, the UK Expert Group on Vitamins and Minerals concluded that the data were insufficient to establish a Safe Upper Level for manganese.

For guidance purposes, the EVM concluded that it was reasonable to assume that in the general population, a supplemental intake of up to 4 mg manganese/day in addition to the diet would be unlikely to produce adverse effects (equivalent to 0.07 mg/kg body weight for a 60 kg adult) based on the NOAEL from the Vieregge study. No uncertainty factor was required as the NOAEL was based on a large epidemiological study. Using the NOAEL from the Kondakis study, it could be assumed that up to 0.5 mg/day manganese (equivalent to 0.008 mg/kg body weight for a 60 kg adult) in addition to the diet would not result in adverse effects in older people. Assuming a dietary intake of 8.2 mg, acceptable total manganese intakes can be estimated to be 12.2 mg/day in the general population (equivalent to 0.2 mg/kg body weight in a 60 kg adult) and 8.7 mg/day (equivalent to 0.15 mg/kg body weight in a 60 kg adult) for older people.

Some population groups may be exposed to high levels of manganese as a result of tea consumption. The significance of this level of exposure is uncertain (EVM, 2003).

In 2004 the WHO looked at the safety of manganese in drinking water. They derived a NOAEL using the upper range manganese intake value of 11 mg/day from dietary studies. The WHO calculated a guideline value using this upper range value. A TDI of 0.06 mg/kg of body weight was calculated by dividing the NOAEL of 11 mg/day by an uncertainty factor of 3 (to allow for the possible increased bioavailability of manganese from water) and an adult body weight of 60 kg. The guideline value of 0.4 mg/litre was then derived from the TDI by assuming an allocation of 20% of the TDI to drinking-water (because manganese is an essential trace element) and consumption of 2 litres of drinking-water per day by a 60-kg adult.

In their assessment, the WHO stated the following:

"…manganese body burden in infants may be influenced by the fact that the biliary excretion system, which is the primary route of manganese excretion, is not completely developed in human infants (Lönnerdal, 1994). Dörner et al. (1989) reported high retention of manganese in infants ingesting both human milk and cow's milk formulas. Studies in rats have demonstrated that young animals absorb significantly more manganese in the gut than do mature animals (Lönnerdal et al., 1987). Also, animal studies have shown that manganese crosses the blood–brain barrier in neonates at a rate 4 times higher than that in adults (Mena, 1974). The relevance of these studies to humans is unknown, however, and few direct absorption data for manganese in human infants are available. Evidence exists, however, to indicate that infants are less well protected than adults against manganese overload.

The manganese contents of erythrocytes in infants up to the age of 6 weeks are higher by about 7–9% than those in adults (Hatano *et al.*, 1985). Collipp *et al.* (1983) reported manganese levels in hair that increased significantly from birth (0.19 µg/g) to 6 weeks (0.865 µg/g) and 4 months (0.685 µg/g) of age in infants given formula, whereas infants given breast milk exhibited no significant increase (0.330 µg/g at 4 months). This study also reported that the average manganese level in hair in children exhibiting learning disabilities was

significantly increased (0.434 µg/g) compared with that in children who exhibited normal learning ability (0.268 µg/g). It should be noted that the Collipp et al. (1983) study did not indicate that the increased manganese level in hair was from ingested manganese."

All of the above evaluations make the assumption that commonly occurring dietary exposure to manganese does not result in adverse effects.

Previous COT evaluations

The COT looked at manganese in 2003 in the light of the results of the 2000 Total Diet Survey. The COT concluded that there was insufficient information to determine whether or not there were toxicological risks associated with dietary exposure to manganese at the levels found in the UK diet and no available safety guideline. The Committee noted that the population exposures had not increased since 1983 and that there was no basis for assuming that the current dietary exposure to manganese is a concern for health.

References

COMA (1998). Nutrition and Bone Health: with particular reference to calcium and vitamin D. Report of the Subgroup on Bone Health, Working Group on the Nutritional Status of the Population, Committee on Medical Aspects of Food and Nutrition Policy. The Stationery Office, London.

EVM (2003). Safe upper levels for vitamins and minerals. Report of the Expert Group on Vitamins and Minerals. Food Standards Agency.

EFSA (2006) Tolerable Upper Intake Levels for Vitamins and Minerals. February 2006

Gwiazda, R., Lucchini R. & Smith, D. (2007) Adequacy and consistency of animal studies to evaluate the neurotoxicity of chronic low-level manganese exposure to humans. J Toxicol Environ Health A. 70 (7) 594-605

Kondakis, X. G., Makris, N., Leotsinidis, M., Prinou, M., Papapetropoulos, T. (1989) Possible health effects of high manganese concentration in drinking water. *Archives of Environmental Health* 44, 175-178

Martin, C.J. (2006) Manganese neurotoxicity: connecting the dots along the continuum of dysfunction. Neurotoxicity 27 (3) 347-9

US DHSS Department of Health and Human Services, Agency for Toxic Substances and Disease Registry (1997) Draft toxicological profile for manganese update.

US EPA (1996) Integrated Risk Information System. Found at: http://www.epa.gov/ncea/iris/subst/0373.htm

US Department of Health and Human Services, Agency for Toxic Substances and Disease Registry (1997). Draft toxicological profile for manganese update.

Vieregge, P., Heinzow, B. Korf, G., Teichert, H.F., Schleifenbaum, P., Mosinger, H.U. (1995) Long term exposure to manganese in rural well water has no neurological effects. *Canadian Journal of Neurological Sciences* 22, 286-289.

WHO (2004) Manganese in Drinking-water – Background document for development of WHO guidelines for drinking water quality. Available at: http://www.who.int/water_sanitation_health/dwq/chemicals/manganese.pdf

13. Mercury (Hg)

The key sources of information for this summary were the JECFA monographs on mercury (WHO, 1972 and 2000) and Environmental Health Criteria 1 (WHO, 1976).

Chemistry and occurrence

Mercury exists in multiple forms and in various oxidation states. Mercury exists in three oxidation states: Hg^{o} (elemental mercury), Hg_2^{2+} (mercurous mercury), and Hg^{2+} (mercuric mercury). The properties and chemical behaviour of mercury strongly depend on its oxidation state and its chemical form. Mercurous and mercuric mercury form numerous inorganic and organic chemical compounds. Organic forms of mercury, especially methylmercury, $CH_3Hg(II)X$, where "X" is a ligand, typically Cl^- or OH^-, are the most toxic forms. Airborne mercury is primarily inorganic mercury. It is used in a wide variety of products and processes. In the environment, mercury may undergo transformations among its various forms and among its oxidation states.

Oral exposure to mercury can occur through dental amalgam fillings (elemental and inorganic mercury), or through the consumption of food contaminated with mercury (inorganic mercury or methylmercury). Exposure via food is the largest source of exposure, with methylmercury in fish being responsible for most of this. This toxicity summary will concentrate on exposure to inorganic mercury compounds through food. Toxicity information on methylmercury is contained in the COT statement on methylmercury in fish (COT, 2003a).

Absorption and elimination

Mercurous salts are poorly absorbed (<0.10%) following oral exposure (Friberg and Nordberg, 1973). Absorption of mercuric chloride by adult mice was reported to be only 1 to 2% (Clarkson, 1971) but 1-week-old mice absorbed 38% of the orally administered compound. Gastrointestinal absorption of inorganic salts of mercury from food has been reported to be <15% for mice and about 7% for humans (Goyer, 1991).

Following oral exposure to inorganic mercury salts the largest concentration of mercury can be found in the kidneys.

Animal data indicate that all forms of mercury cross the placenta and that mercury levels may be 2-fold greater in the fetus than in the mother, with fetal red blood cells containing mercury levels 30% higher than maternal red blood cells (Goyer, 1991).

The urine and faeces are the primary routes for the excretion of inorganic mercury by humans (ATSDR, 1989) with the faeces accounting mainly for any

unabsorbed mercury. Following brief exposure of humans to inorganic mercury, urinary excretion accounts for 13% of the total body burden, whereas this value increases to 58% for long-term exposure. For inorganic mercury, the urinary levels do not parallel blood levels (ATSDR, 1989). The biological half-life for inorganic mercury salts is about 40 days (Goyer, 1991).

Toxicity in animals

Oral exposure to inorganic mercury has produced neurological, immunological, and systemic effects in rodents exposed for periods of 1 to 11 weeks. The NOAEL for these studies was 0.42 mg/kg body weight/day, and the LOAEL was 0.8 mg/kg body weight/day (ATSDR, 1989).

Evidence for a systemic autoimmune response involving the kidneys was reported by Bernaudin *et al*. (1981) in rats given mercuric chloride (3 mg/kg body weight/week) orally for up to 60 days. Druet *et al*. (1978) noted renal immunologic insufficiencies in Brown Norway rats given subcutaneous injections of mercuric chloride (1 mg/kg body weight) for 8 to 12 weeks. Andres (1984) also reported autoimmune glomerulonephritis in brown Norway rats administered mercuric chloride (3 mg/kg body weight) by gavage twice weekly for 60 days.

Only limited information is available regarding the developmental toxicity of inorganic mercury. Gale (1974) reported an increase in fetal resorptions in hamsters receiving a single oral dose of mercuric chloride (31.4 mg Hg/kg body weight). This study identified a NOAEL of 15.7 mg Hg/kg body weight/day based on the absence of developmental toxicity.

Mercuric chloride was tested for carcinogenicity in mice and rats by oral gavage. In mice, a few renal adenomas and adenocarcinomas occurred in males only. In rats, a few renal adenomas occurred in females; there was a dose-related increase in the incidence of squamous-cell papilloma of the forestomach in males, and a few papillomas were seen in females. Dose-related hyperplasia of the forestomach was seen in both males and females.

Toxicity in humans

Generally, any form of mercury in high acute doses may cause tissue damage resulting from the ability of mercury to denature proteins, thereby disrupting cellular processes (WHO, 1976). However, oral exposure to mercury metal is usually without serious effects.

Ingestion of inorganic salts of mercury such as mercuric chloride may cause gastrointestinal disorders including pain, vomiting, diarrhoea and haemorrhage, and renal failure resulting in death. Additional effects of acute mercury poisoning include shock and cardiovascular collapse (WHO, 1976). Acute lethal doses in humans range from 1 to 4 g (10 to 42 mg Hg/kg body weight for a 70 kg adult) for inorganic mercuric salts (ATSDR, 1989).

Mercurous compounds are less corrosive and less toxic than mercuric salts, presumably because they are less soluble. A hypersensitivity reaction to mercurous chloride (calomel), a teething powder used in children has been reported (Matheson *et al.,* 1980; Goyer, 1991). The reaction is hypothesised to be a hypersensitivity reaction since the effects are independent of the dose, but appears to be limited to mercurous chloride.

Chronic oral exposure to mercury or mercury compounds may affect the central nervous system, gastrointestinal tract, and the kidneys; the renal effect, in part, involves an immunologically-mediated response (ATSDR, 1989). Davis *et al.* (1974) reported dementia, colitis, and renal failure in two women chronically (6 and 25 years) ingesting a mercurous chloride-containing laxative. However, little information is available regarding the toxicity of inorganic mercury following chronic oral exposure.

In 1993 IARC concluded that metallic mercury and inorganic mercury compounds were 'not classifiable as to their carcinogenicity to humans' (Group 3).

Previous evaluations

In 1978, JECFA established a PTWI of 0.005mg/kg body weight for total mercury. More recently JECFA have lowered the PTWI for methylmercury and they noted the need to review the PTWI for total mercury.

WHO derived a guideline value for inorganic mercury in drinking water of 0.006 mg/L in 2003, this was re-affirmed in 2006 (WHO, 2006). The guideline value was based on a NOAEL of 0.23 mg/kg body weight/day for kidney effects in a 26-week study in rats. Adjustments were made for 5 days/week dosing and an uncertainty factor of 100 was applied, resulting in a TDI of 2 µg/kg body weight/day. Assuming a 60 kg adult drinking 2 litres of water per today and allocating 10% of the TDI to drinking water, results in the guideline value (0.006 mg/L; WHO, 2006). This TDI of 2 µg/kg body weight/day was also quoted by WHO in the most recent Concise International Chemical Assessment Document (WHO, 2003b).

Previous COT evaluations

In 2003, the COT evaluated mercury in food when the results of the 2000 TDS were considered (COT, 2003c). The estimated population dietary exposure to mercury from the 2000 TDS was 0.0015 mg/day, approximately 0.025 µg/kg body weight/day for a 60 kg adult. The Committee concluded that current dietary exposures to mercury are unlikely to be of any toxicological concern for consumers. The committee noted that with the exception of high-level consumption by children aged 1.5-4.5 years, the estimates of dietary exposure to total mercury for all consumer groups was within the PTWI for methylmercury set by JECFA in 2003. The estimate for high-level consumption by children aged 1.5-4.5 years exceeded the JECFA PTWI for

methylmercury by 17%. It is unlikely that all of the mercury in the diet is in the form of methylmercury. Inorganic mercury is less well absorbed than methylmercury by the oral route, and therefore comparing dietary exposure to total mercury to the PTWI for methylmercury is a worst case scenario. Furthermore, the COT has previously noted that toddlers are likely to be less sensitive to the neurodvelopmental effects of methylmercury than the foetus or infant. Therefore the dietary exposures to mercury do not give rise to toxicological concerns for consumers. The Committee also noted that the population exposures to mercury have decreased since 1976 (0.005mg/day), with the current dietary exposure at its lowest level (0.0015 mg/day) (COT, 2003c).

References

Andres, P. (1984). IgA-IgG disease in the intestine of Brown Norway rats ingesting mercuric chloride. *Clin. Immunol. Immunopathol*. 30: 488-494.

ATSDR (1984). *Toxicological Profile for Mercury*. Agency for Toxic Substances and Disease Registry. ATSDR/U.S. Public Health Service.

Bernaudin, J. F., E. Druet, P. Druet, and R. Masse. (1981). Inhalation or ingestion of organic or inorganic mercurials produces auto-immune disease in rats. *Clin. Immun. Immunopath*. 20: 129-135.

Clarkson, T. W. (1971). Epidemiological and experimental aspects of lead and mercury contamination. *Food Cosmet. Toxicol*. 9: 229-243.

COT (2003a). COT statement on a survey of mercury in fish and shellfish. Committee on Toxicity of chemicals in foods, consumer products and the environment.

COT (2003b). Statement on a survey of metals in infant foods (2003-02). Committee on Toxicity of chemicals in foods, consumer products and the environment.

COT (2003c). Statement on twelve metals and other elements in the 2000 Total Diet Study. COT Statement 2003/07. Annual Report 2003 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Foods, Consumer Products and the Environment.

Davis, L. E., J. R. Wands, S. A. Weiss, et al. (1974). Central nervous intoxication from mercurous chloride laxatives - quantitative, histochemical and ultrastructure studies. *Arch. Neurol*. 30: 428-431.

Druet, P., E. Druet, F. Potdevin, and R. Masse. (1978). Immune type glomerulonephritis induced by $HgCl_2$ in the Brown Norway rat. *Ann. Immunol*. 129C: 777-792.

Friberg, L. and F. Nordberg. (1973). Inorganic mercury--a toxicological and epidemiological appraisal. In: Miller, M.W. and T.W. Clarkson, eds. *Mercury, mercurials and mercaptans*. Charles C. Thomas Co., Springfield, Il. pp. 5-22.

Gale, T. F. (1974). Embryopathic effects of different routes of administration of mercuric acetate on the hamster. *Environ. Res*. 8: 207-213.

Goyer. R. (1991). Toxic effects of metals. In: Amdur, M.O., J.D. Doull and C.D. Klassen, Eds. *Casarett and Doull's Toxicology*. 4th ed. Pergamon Press, New York. pp.623-680.

IARC (1993). International Agency for Research on Cancer (IARC) - Summaries & Evaluations: Mercury and Mercury Compounds. Vol. 58.

Matheson, D. S., T. W. Clarkson, and E. Gelfand. (1980). Mercury toxicity (acrodynia) induced by long-term injection of gamma globulin. *J. Pediatr*. 97: 153-155.

WHO (1972). Evaluation of Mercury, Lead, Cadmium and the food additives Amaranth, Diethylpyrocarbonate and Octyl Gallate. FAO Nutrition Meetings Report Series, No. 51A: WHO Food Additives Series No. 4.

WHO. (1976). *Environmental health criteria. 1. Mercury*. United Nations Environment Programme and World Health Organization, Geneva, 131 pp.

WHO (2000). Safety Evaluation of Certain Food Additives and Contaminants. Methylmercury. WHO Food Additives Series 44.

WHO (2003a). Safety Evaluation of Certain Food Additives and Contaminants. Methylmercury. Summary and conclusions of the 61st JECFA meeting. ftp://ftp.fao.org/es/esn/jecfa/jecfa61sc.pdf

WHO (2003b). Concise International Chemical Assessment Document (CICAD) 50: Elemental mercury and inorganic mercury compounds: human health aspects. World Health Organization, Geneva.

WHO (2007). Safety Evaluation of Certain Food Additives and Contaminants. Methylmercury. WHO Food Additives Series 58. World Health Organization.

WHO (2006). Guidelines for Drinking-Water Quality. Volume 1 Recommendations. First addendum to the third edition. World Health Organization, Geneva.

14. Molybdenum (Mo)

The key source of information for this summary was the EVM report.

Chemistry and occurrence

The transition element molybdenum exists in five oxidation states (II – VI), the predominant states are Mo (IV) and Mo (VI).

Molybdenum does not exist naturally in the metallic state, but occurs in association with other elements. The predominant form of molybdenum occurring in soil and natural waters is the molybdate anion, $MoO4^{-2}$.

Molybdenum is an essential component in the diet as it has a role in a number of important enzymes.

Generally, foodstuffs from above ground plant material, such as legumes, leafy vegetables and cauliflower, contain relatively high concentrations of molybdenum compared with food from tubers or animals. Few data are available on the variability of molybdenum content of individual plant species, but it is affected by the soil molybdenum concentration and pH, with molybdenum uptake increasing with soil pH. Foodstuffs from plants grown in alkaline or neutral soils with high molybdenum concentrations can be expected to have the highest molybdenum levels.

Molybdenum is available in food supplements at levels up to 0.33 mg and licensed medicines. The latter are used (maximum daily dose 0.25 mg) to treat patients with malabsorption states and conditions leading to hypoproteinaemia and in perioperative nutritional support.

Most natural waters contain low levels of molybdenum. The WHO recommends a maximum level of molybdenum in drinking water of 0.07 mg/L and notes that concentrations of molybdenum in drinking water are typically less than 0.01 mg/L. However, in areas near mining sites, molybdenum concentrations up to 0.2 mg/L have been reported (WHO, 1993).

It is recognised that molybdenum interacts with copper and sulphates in living organisms, but the mechanism of this interaction has not been elucidated. The presence of dietary copper and sulphate affects the amount of molybdenum absorbed and retained by the body.

Absorption and elimination

Absorption of molybdenum varies over a wide range (25 – 93%). Reports suggest that soluble molybdenum compounds are readily absorbed, whilst insoluble compounds are not.

Molybdenum occurs in low concentrations in all the fluids and tissues of the body. The greatest amounts are found in the kidney, liver, small intestine and adrenals. It is found largely as molybdoenzymes. In plasma, molybdenum is bound specifically to alpha2-macroglobulin.

Molybdenum is primarily excreted in the urine and experimental data from humans suggest that up to 80% of the absorbed amount is excreted via urine. Normally only small amounts are excreted in the faeces with exceptions in certain gastrointestinal disorders.

Toxicity in animals

Although non-ruminant animals will develop symptoms of toxicity when fed high molybdenum diets, ruminants are much more sensitive. Thus the toxicity seen in these species cannot be related to the effects that would be expected in man. The toxicity is primarily expressed as a copper deficiency and the ambient sulphate level has a marked effect on the interaction between copper and molybdenum.

Molybdenum toxicity in animals is commonly referred to as molybdenosis or teart. In appearance it is similar to the disease of copper deficiency. Signs of molybdenum toxicity in animals include anaemia, anorexia, profound diarrhoea, joint abnormalities, osteoporosis, hair discoloration, reduced sexual activity and death.

There are no data on the carcinogenicity of molybdenum. No data are available on the genotoxicity of molybdenum compounds in vitro, although there is limited evidence of genotoxicity from in vivo tests, the reliability of which is questionable.

No data on mechanisms of toxicity or dose response characterisation have been identified.

Molybdenum (as the trivalent form molybdenite, and as the hexavalent forms molybdenum trioxide, calcium molybdate and ammonium molybdate) was administered in the diet to rats at levels of 10 – 500 mg molybdenum/animal/day, for periods up to 232 days. No signs of toxicity were observed in the groups ingesting molybdenite. In the groups receiving hexavalent molybdenum, signs of toxicity were observed at all dose levels and included loss of appetite, weight loss, a tendency to become quiet and listless and premature death. One hundred percent mortality was recorded in the groups receiving >100 mg hexavalent molybdenum/animal/day. Limited experimental details were provided (Fairhall et al., 1945).

Hexavalent molybdenum (sodium molybdate) was administered in the diet to groups of 2-5 weanling or mature rabbits at levels of 140, 500, 1000, 2000 and 4000 mg/kg diet (4, 15, 30, 60 or 120 mg/kg bw/day – estimated) for up to 12 weeks (average survival being 3 – 54 days in the higher dose groups). In the highest dose group 100% mortality was observed and 80% mortality in the group receiving 60 mg/kg bw/day. Anaemia, anorexia, loss of weight, alopecia, dermatosis and effects on the skeletal system were noted in animals receiving 30 mg/kg bw/day and above. Limited experimental details were provided. Haemoglobin levels appear to decrease by >50% within a few weeks of treatment. It is uncertain whether this is a specific effect or related to factors such as haemodilution. Data for the 500 ppm (4 and 15 mg/kg bw)

group are not provided so it is uncertain whether the decrease would have also have been apparent at this level (Arrington & Davies, 1953).

Sodium molybdate (hexavalent molybdenum) was administered in drinking water (0.0025 mg/kg bw/day) and diet (0.07 mg/kg body weight/day) in lifetime studies over three generations of mice. In the first and second generations, 15/238 and 7/242 premature deaths, respectively, were observed, compared to 0/209 and 6/248 in the control populations. In the third generation increased numbers of premature deaths, maternal deaths, failure to breed, runting and production of single sex or dead litters were observed (Schroeder & Mitchener, 1971).

Toxicity in humans

Few data are available on human toxicity following ingestion. Acute intakes of more than 100 mg/kg have been shown to produce signs of toxicity, which include diarrhoea, anaemia and high levels of uric acid in the blood. Elevated uric acid levels, which are associated with the onset of gout, are hypothesised to be caused by stimulation of xanthine oxidase by high molybdenum intake. Occupational exposure, by inhalation, to molybdenum containing dusts has been associated with pneumoconiosis. No vulnerable groups or genetic susceptibilities have been identified.

In a retrospective study of environmental exposure to increased molybdenum (form unclear), medical reports revealed a prevalence of gout in 31% and 18% of the exposed and control populations, respectively. Intake was calculated to be 10 – 15 mg molybdenum per day in the exposed population compared to 1 – 2 mg/day in the control area. It is possible that the 1-2 mg intake produced a low level of response. Symptoms included arthralgia in the hand, feet and knee joints, accompanied by elevated blood and urine molybdenum levels (Kovalsky *et al.*, 1961).

Previous evaluations

In 1993, the US Environmental Protection Agency set a reference dose for molybdenum of 5 micrograms/kg bw/day using a cross-sectional epidemiology study on two population groups from Armenia (US EPA, 1993).

In 2000, the SCF established a tolerable upper intake level (UL) of 0.01mg/kg bw/day based on a 9 week study in Sprague-Dawley rats by Fungwe *et al*. (1990). This study was used because of its satisfactory design with full histopathology, adequate number of animals, demonstration of a clear dose – response relationship and clear toxicological endpoints. The NOAEL of the study was 0.9mg/kg bw/day for reproductive toxicity and uncertainty factors of 100 were incorporated into the UL.

The UK Expert Group on Vitamins and Minerals concluded that there were insufficient data from human or animal studies to establish a Safe Upper Level for molybdenum. They concluded that the molybdenum intake from the UK diet (estimated maximum intake 0.23 mg/day) was not expected to present any risk to health, but there were insufficient data on the safety of

molybdenum intakes in excess of those naturally occurring in the diet to be able to provide further guidance (EVM, 2003).

In 2003, the WHO looked at the safety of molybdenum in drinking water. They concluded that no data were available on the carcinogenicity of molybdenum by the oral route. They derived a NOAEL from a 2-year study of humans exposed via drinking-water of 0.2 mg/litre, but they had some reservations over the quality of this study. Although an uncertainty factor of 10 would normally be applied to reflect intraspecies variation, it was recognized that molybdenum is an essential element, and a factor of 3 was therefore considered to be adequate.

This calculation gave a guideline value of 0.07 mg/litre (rounded figure), which was in the same range as that derived on the basis of the results of toxicological studies in animals and is compatible with the essential daily requirement for molybdenum.

No assessments have been carried out by EFSA or the COT.

References

Arrington, L.R. and Davies, G.K. (1953) Molybdenum toxicity in the rabbit. Journal of Nutrition 51, 295-304.

EFSA (2006) Tolerable Upper Intake Levels for Vitamins and Minerals. February 2006

EVM (2003). Safe upper levels for vitamins and minerals. Report of the Expert Group on Vitamins and Minerals. Food Standards Agency.

Fairhall, L.T., Dunn, R.C., Sharpless, N.E., Pritchard, E.A. (1945) The toxicity of molybdenum. US Public Health Service. Public Health Bulletin No. 293, 1-36.

Kovalsky, V.V., Yarovaya, G.A., Shmavonyan, D.M. (1961) The change in purine metabolism of humans and animals under the conditions of molybdenum biogeochemical provinces. Zhurnal Obshchei Biologii 22, 179-191 (Full translation of original Russian paper obtained).

Schroeder, H.A and Mitchener, M. (1971) Toxic effects of trace elements on the reproduction of mice and rats. Archives of Environmental Health 23, 102-106.

US EPA (2003) Intergrated Risk Information System. Available at http://www.epa.gov/ncea/iris/subst/0425.htm

WHO (1993) Guidelines for drinking water quality. Second edition. World Health Organisation, Geneva.

WHO (2003) Molybdenum in Drinking-water – Background document for development of WHO guidelines for drinking water quality. Available at: http://www.who.int/water_sanitation_health/dwq/chemicals/molybdenum.pdf

15. Nickel (Ni)

The key sources of information for this summary were Environmental Health Criteria 108 (WHO, 1991) and the EVM report (EVM, 2003).

Chemistry and occurrence

Nickel exists in various mineral forms and may be found throughout the environment including rivers, lakes, oceans, soil, air, drinking water, plants, and animals. Soil and sediment are the primary compartments for nickel, but mobilisation may occur depending on physico-chemical characteristics of the soil (ATSDR, 1988; USAF, 1990). Nickel is used in a wide variety of metallurgical processes such as electroplating and alloy production as well as in nickel-cadmium batteries. Some evidence suggests that nickel may be an essential trace element for mammals (Goyer, 1991).

Nickel concentrations in food are usually below 0.5 mg/kg fresh weight. Cocoa, soybeans, some dried legumes, various nuts, and oatmeal contain high concentrations of nickel. Daily intake of nickel from food will vary widely, because of different dietary habits, and can range from 100 to 800 µg/day.

In humans, nickel influences iron absorption and metabolism and maybe an essential component of the haematopoietic process. It is primarily found in nuts, legumes, chocolate, canned foods and grain products and is absorbed from the soil where it is often found in ferromagnesium minerals.

Absorption and elimination

Pulmonary absorption is the major route of concern for nickel-induced toxicity and this mainly involves occupational exposure. Although nickel is poorly absorbed from the gastrointestinal tract, dietary exposure and exposure via drinking water provides most of the intake of nickel and nickel compounds for the general population (Coogan *et al.,* 1989; Goyer, 1991). Gastrointestinal absorption of nickel depends on the composition of the diet with the presence of food reducing the amount of nickel absorbed. Approximately 27% of nickel in water is absorbed, whereas less than 1% of nickel in food is absorbed (WHO, 1991).

Following absorption, nickel is transported in the blood, principally bound to albumin. Faecal nickel exceeds urinary nickel by approximately one to two orders of magnitude, but is mainly unabsorbed. The primary excretory route for absorbed nickel is via the kidneys in the form of low-molecular-weight complexes, mainly with histidine. The elimination half-life for ingested nickel into urine is ~28 $\pm$ 9 hours.

Toxicity in animals

The acute toxicity of nickel-containing compounds is strongly dependent on solubility. Prolonged exposure to various nickel compounds, in the diet, by gavage, or in drinking water, has resulted in decreases in body weight gain, increases in relative heart weight and plasma alkaline phosphatase, altered haematology, changes in relative liver weight and in liver and heart cytochrome c oxidase, histologic lesions in the lung and altered glomerular and myeloid function. Increases in arterial blood pressure and vascular resistance of the kidneys have been seen with doses of 5 mg/kg body weight/day (WHO, 1991, 1993).

Oral exposure of pregnant rats to soluble nickel is associated with increased perinatal mortality; dietary exposure (≥ 12.5 mg/kg body weight/day) of dams and their mates increased stillbirths in F_1 generation pups, however these doses also caused maternal toxicity (decreased organ weights) (Ambrose *et al.,* 1976). A NOAEL of 5 mg/kg body weight/day was identified based on decreased body and organ weights.

A No Observed Adverse Effect Level (NOAEL) of 5 mg/kg body weight/day has been identified, based on decreased organ to body weight ratios in a two year feeding study in rats (Ambrose *et al.,* 1976). Survival at the end of this study was poor, particularly in the controls, leading to questions about the sensitivity of the study, however a 90-day gavage study with nickel chloride in rats also produced a NOAEL of 5 mg/kg body weight/day.

Smith *et al*. (1993) reported an increased incidence of perinatal mortality when nickel chloride was given to female rats 11 weeks prior to mating and during two successive gestation and lactation periods. This occurred at the lowest dose of 1.3 mg Ni/kg body weight/day in the second litter.

Nickel compounds have been shown to be genotoxic in cultured mammalian cells. There is evidence to suggest that nickel and poorly soluble nickel compounds are carcinogenic by inhalation and by various routes of injection. Although the data are limited, the evidence does not suggest that nickel or nickel compounds are carcinogenic when administered by the oral route (IARC, 1990).

Toxicity in humans

There is limited information on the acute and chronic effects of nickel exposure via the oral route. In acute poisoning incidents where individuals ingested large amounts of nickel sulphate or chloride in drinking water, nausea, vomiting, diarrhoea, giddiness, headache, coughing, shortness of breath and evidence of nephrotoxicity were seen (Sunderman *et al*., 1989).

Much of the acute and chronic toxicity information comes from poisoning incidents and epidemiological studies focussing on occupational exposure to

nickel compounds via the inhalation route. Nickel compounds have been associated with an increase in the incidence of cancer (lungs and nasal passages), leading the IARC to classify nickel compounds as 'carcinogenic to humans' (Group 1), and metallic nickel as 'possibly carcinogenic to humans' (Group 2B) (IARC, 1990). However there is insufficient evidence regarding the carcinogenicity of nickel and its compounds via the oral route.

The main toxic effect associated with nickel is skin sensitisation. Nickel and its water-soluble salts are potent skin sensitisers. Approximately 7-10 % of the population is affected by nickel allergic dermatitis (Schollhammer *et al.*, 1994), with a 7-14:1 apparent predominance in women. There is no clear threshold for aggravation of nickel sensitisation, but it appears possible at the levels of nickel found in food. Oral intakes as low as 0.49 mg nickel could trigger symptoms in pre-sensitised individuals, particularly in the fasting state (Nielsen *et al.*, 1990). Although certain sensitised subjects will be aware that exposure to nickel may be responsible for their dermatological symptoms, other subjects may only be aware that they are unable to tolerate certain jewellery. They also may not be aware that their dermal symptoms could be aggravated by food. Despite the potential for aggravation of nickel sensitivity, there are no data to support the conclusion that that oral exposure to nickel can initiate nickel sensitivity.

It has been reported that that oral exposure to nickel at an early age might serve to inhibit subsequent development of allergic hypersensitivity to nickel (Van Hoogstraten *et al.,* 1991*)*.

Previous evaluations

The US EPA used the NOAEL of 5 mg/kg body weight/day from the study of Ambrose *et al.* (1976) to derive an oral reference dose of 20 µg/kg body weight/day (US EPA, 1996). This NOAEL in which decreased organ to body weight ratios were reported at 50 mg/kg bw/day from a rat 2-year feeding study. An uncertainty factor of 300 (100 for inter- and intra-species variation and 3 to allow for inadequacies in the reproductive toxicity database) was applied, resulting in a reference dose (rounded) of 20 µg/kg body weight/day (US EPA, 1996).

In 2005, the EFSA Scientific Panel on Dietetic Products, Nutrition and Allergies concluded that there were insufficient data available to derive a tolerable upper intake level for nickel (EFSA, 2006).

The Expert Group on Vitamins and Minerals could not recommend a safe upper level or guidance level for supplemental intake of nickel because of the high prevalence of nickel sensitivity, the fact that many individuals may be unaware that they are sensitised, and that certain sensitised individuals may not be aware that food could aggravate their dermal symptoms (EVM, 2003). However, using a LOAEL of 1.3 mg/kg body weight/day from Smith *et al.* (1993) and applying an uncertainty factor of 300 (100 for inter- and intra-

species variation and 3 for the use of a LOAEL), they indicated that a total nickel intake of 4.3 µg/kg body weight/day would not be expected to have effects in non-sensitised individuals (EVM, 2003).

In order to establish a guideline for drinking water quality the WHO (2003) derived a TDI for nickel of 12 µg/kg body weight, based on a LOAEL established after oral intakes in fasted patients. The dose of 12 µg/kg body weight/day was considered to be an acute LOAEL. Absorption from drinking water on an empty stomach is 10-40 fold higher than absorption from food. Assuming a 60 kg adult, drinking 2 litres of water per day and allocating 20% of the TDI to drinking water, results in a guideline value of 0.07 mg/L (WHO, 2003). This guideline value was re-affirmed in the latest edition of the Drinking Water Guidelines (WHO, 2006).

Nickel has been included in the rolling revision of the WHO Guidelines for Drinking Water Quality and an updated factsheet is due to be published in the second addendum to the third edition (WHO, 2008). Nickel has been included in the plan of work of the rolling revision because of a new study that reported significant addition of nickel to drinking water through household appliances such as kettles; as well as expected results from a reproductive study.

Previous COT evaluations

As part of a MAFF food surveillance exercise, the COT concluded that the levels of nickel present in food in 1991 resulted in an average intake of 170 µg/day. These levels were not considered to be of concern other than for individuals sensitised to nickel (MAFF, 1998). In 2003 the COT evaluated nickel in food when the results of the 2000 TDS were considered (COT, 2003b). The estimated population dietary exposure to nickel from the 2000 TDS was 0.13 mg/day, approximately 2.2 µg/kg body weight/day for a 60 kg adult. The Committee concluded that current dietary exposures to nickel were unlikely to be of any toxicological concern for consumers (COT, 2003b).

In 2003, the COT reviewed a survey of metals in infant formula carried out by the Food Standards Agency. The committee concluded that the estimated intakes were lower than for the previous survey. The worst case intakes (based on manufacturers feeding guidelines) for 7-12 month old infants (normal diet) and for 4-12 month old infants (soya diet) exceeded the WHO TDI of 5µg /kg bw/day by up to 68%. Taking into account that this was likely to be an over estimate of exposure due to the use of upper bound concentrations and worst case scenario consumption, this exceedance of the TDI was considered unlikely to be of significance. Ingestion of nickel may exacerbate contact dermatitis/ eczema in pre-sensitised individuals. Infants ar less likely than adults to be sensitised to nickel and are therefore not to be considered a susceptible sub-group. Overall, the dietary exposures were not considered to be of concern (COT, 2003c).

References

Ambrose, A.M., Larson, P.S., Borzelleca, J.F., Hennigar Jr, G.R. (1976). *J Food Sci. and Technol*. **13**: 181-187.

ATSDR (1988). *Toxicological Profile for Nickel*, ATSDR/U.S. Public Health Service, ATSDR/TP-88/19. Agency for Toxic Substances and Disease Registry

Coogan, T.P., Latta, D.M., Snow, E.T., and Costa, M. (1989). Toxicity and carcinogenicity of nickel compounds, In: *Critical Reviews in Toxicology*, Vol 19. McClellan, R.O., ed., CRC Press, Boca Raton, FL. pp. 341–384.

COT, (2003a). Opinion on Nickel Leaching from Kettle Elements into Boiled Water. (TOX/2003/11).

COT (2003b). Statement on twelve metals and other elements in the 2000 Total Diet Study. COT Statement 2003/07. Annual Report 2003 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Foods, Consumer Products and the Environment.

COT (2003c). Statement on a survey of metals in infant food. COT Statement 2003/02. Annual Report 2003 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Foods, Consumer Products and the Environment.

COT (2007). Nickel leaching from kettle elements into boiled water. Committees on Toxicity, Mutagenicity, Carcinogenicity of chemicals in Food, Consumer Products and the Environment. Annual Report 2007.

EFSA (2006) Tolerable Upper Intake Levels for Vitamins and Minerals. February 2006

EVM (2003). Safe upper levels for vitamins and minerals. Report of the Expert Group on Vitamins and Minerals.

IARC (1990). IARC Monograghs on the evaluation of carcinogenic risks to humans: Chromium, Nickel and Welding. Volume 49.

Goyer, R. (1991). Toxic effects of metals, In: *Casarett and Doull's Toxicology*, 4th ed. Amdur, M.O., J.D. Doull and C.D. Klaassen, eds., Pergamon Press, New York. pp.623–680.

MAFF (1998). Lead Arsenic and other metals in food. *Food Surveillance Information Sheet No. 52*. Ministry of Agriculture Fisheries and Food.

Nielsen, G.D., Jepsen, L.V., Jorgensen, P.J. *et al.* (1990). *British Journal Dermatology,* **122**: 299-308.

Schollhammer M, Guillet MH, Guillet G (1994). *Ann. Dermatol. Venereol.* **121:** 338-345.

Smith, M.K., George, E.L., Stober, J.A., Feng, H.A. and Kimmel, G.L. (1993). Perinatal toxicity associated with nickel chloride exposure. *Environmental Research*, **61:** 200-211.

Sunderman Jr, F.W., Hopfer, S.M., Sweeney, K.R. *et al.* (1989). Proc. Soc. Exp. Biol. Med. **191**: 5-11.

USAF (1990). Nickel, In: *Installation Restoration Program Toxicology Guide*, Vol. 5. Harry G. Armstrong Aerospace Medical Research Laboratory, Wright Patterson AFB, OH.

US EPA (1996). Integrated Risk Information System (IRIS): Nickel, soluble salts. Environmental Protection Agency National Center for Environmental Assessment.

Van Hoogstraten *et al.,* (1991). Reduced frequency of nickel allergy upon oral nickel contact at an early age. *Clinical and Experimental Immunology* **85:** 441-445.

WHO (1991). International Programme on Chemical Safety. Environmental Health Criteria 108: Nickel.

WHO (1993). Guidelines for drinking-water quality. World Health Organisation. Geneva.

WHO (2003). Guidelines for Drinking-Water Quality. Volume 1 Recommendations. Third edition. World Health Organization, Geneva.

WHO (2006). Guidelines for Drinking-Water Quality. Volume 1 Recommendations. First addendum to the third edition. World Health Organization, Geneva.

WHO (2008). Plan of work for the rolling revision of the WHO Guidelines for Drinking Water Quality. World Health Organization website: http://www.who.int/water_sanitation_health/gdwqrevision/en/index.html. Accessed July 2008.

16. Palladium (Pd)

The key source of information for this summary was Environmental Health Criteria 226 (WHO, 2002).

Chemistry and occurrence

Palladium is one of the six platinum-group metals in Group VIII of the periodic table. It is a soft, white, ductile metallic element, of molecular weight 106.4. It is present in the Earth's crust at very low levels, i.e. about 0.01 ppm. It is the second most widely mined and used of the platinoid elements (after platinum) and is used industrially in catalysts, in the electrical and chemical industries, in jewellery and in dental alloys. Palladium forms di- and tri-valent salts, which are of varying solubility in water. Unlike platinum, palladium coordination complexes have not been demonstrated to be present in biological systems (Goering, 1992).

Palladium is present naturally in foods such as, nuts, bread, offal and fish. However, the chemical form(s) in which palladium occurs in food is unknown. The average human dietary intake of palladium is up to 2 µg/day (WHO, 2002).

As part of the total diet study from 1994, the levels of platinum group metals were analysed in the average UK diet. Concentrations of the platinum group elements (i.e. ruthenium, rhodium, iridium, palladium and platinum) were very low with few food groups containing concentrations of these elements above their LODs. Those food groups which contained detectable concentrations of the platinum group elements were nuts (0.003 mg/kg palladium, 0.004 mg/kg rhodium); and oils and fats, offal, and bread which all had palladium concentrations of 0.002 mg/kg[1,4].

Dietary intake estimates of the platinum group elements for average, mean and upper range (97.5 percentile) consumers were very low.

Absorption and elimination

Following a single oral dose of ^{103}Pd (as $PdCl_2$) in rats, most was eliminated rapidly in the faeces, with less than 0.5% absorbed and subsequently eliminated in the urine. Higher retention and absorption was seen following intratracheal, inhalation and intravenous exposure. Twenty-four hours after peroral dosing, detectable quantities of ^{103}Pd were found only in the kidney and liver. In comparison, 24 hours after intravenous dosing, ^{103}Pd was found in all major tissues, with the highest concentrations in the kidney, spleen, liver, adrenals, lung and bone, respectively. No significant amounts of ^{103}Pd were found in any tissues when the rats were sacrificed 104 days after peroral dosing (Moore et al., 1975).

From a single study in rats using $^{103}PdCl_2$ injected intravenously on day 16 of pregnancy, it appears that Pd does not readily move across the placental barrier in the rat (Moore et al., 1975).

Studies with palladium chloride and sodium tetrachloropalladate showed elimination via the faeces and urine following intravenous dosing. Urinary excretion rates ranged 6.4-76% in rats and rabbits, and elimination in the faeces ranged from trace amounts to 13% of the administered dose. However, following oral administration of palladium chloride, >95% of palladium is eliminated in the faeces due to non-absorption.

Toxicity in animals

The acute oral toxicity of $PdCl_2$ in male Sprague-Dawley rats has been determined as 287 mg/kg body weight (equivalent to 172 mg Pd/kg body weight) (Holbrook et al., 1975).

The acute oral toxicity of PdO in male Sprague-Dawley rats was determined to be >425 mg/kg body weight (equivalent to > 385 mg Pd/kg body weight) and that of $PdSO_4$ to be >744 mg/kg body weight (equivalent to >390 mg Pd/kg body weight) (Holbrook et al., 1975).

Short term exposure to various palladium compounds in rodents results mainly in changes to biochemical parameters and clinical signs.

In a limited, early study, mice were given 5 mg/L Pd^{2+} in the drinking water from weaning until death. The authors reported that mice exposed to Pd had an increased frequency of amyloidois in the kidney, liver, spleen and heart but no further details are provided (Schroeder and Michener, 1971, reported in Hildebrand et al., 1996).

Decreases have been reported in the levels and/or activity of several microsomal enzymes following oral administration of unspecified amounts of $PdCl_2$ or $Pd(NO_3)_2$ to rats or mice, but the data are very poor. Intravenous injection of soluble Pd salts in rats have been shown to induce cardiac arrhythmias and a fall in blood pressure. The effective dose of $PdCl_2$ was >0.4 mg/kg body weight (studies reviewed by Hildebrand et al., 1996).

A 28-day gavage study in rats reported a NOAEL of 1.5 mg tetra-ammine palladium hydrogen carbonate/kg body weight/day (equivalent to 0.54 mg palladium/kg body weight/day) (Johnson Matthey, 1997b). Animals were dosed with 1.5, 5 or 150 mg/kg body weight/day and treatment-related abnormalities confined to histopathological changes were observed in the top two dose groups. Although, the authors considered 1.5 mg/kg body weight/day to be the NOAEL, significant increases in absolute brain and ovary weights were observed in females of this dose group.

There are insufficient data on the reproductive and developmental effects of palladium and its compounds. Tumours have been associated with palladium exposure via the drinking water (5 mg/L) in mice. However, the tumours observed were concomitant with an increased longevity which may explain the increased tumour rate.

$PdCl_2$ is negative in the Ames test with and without metabolic activation (Mortelmans *et al.*, 1986). A number of divalent Pd salts - $PdCl_2$, K_2PdCl_4,

$Pd(NH_3)_2J_2$, $Pd(NH_3)_4Cl_2$ and transpalladium ($Pd(NH_3)Cl_2$) were tested for genotoxicity in an *in vitro* micronucleus test using human lymphocytes and the bacterial SOS chromotest (an unvalidated mutagenicity test). All results were negative (Gebel *et al.,* 1997).

Toxicity in humans

Human exposure to palladium in the occupational context is largely via the inhalation and dermal routes. The Hazardous Substances Data Bank reports that "despite the wide use of palladium, especially the handling of materials in laboratories and in photography, no occupational poisoning has been shown". There is no occupational exposure standard for palladium in the UK or USA.

In dentistry, palladium is a very common component of dental casting alloys of all types, and its use has increased in recent decades in response to the increased cost of gold. No satisfactory studies have been carried out to investigate palladium release from the alloys *in vivo.* A number of studies investigating palladium release *in vitro* after incubation with artificial saliva solution indicate that palladium is not released in measurable quantities. Nevertheless, a few case reports have appeared in the literature of palladium allergy due to exposure via dental alloys. In addition, a number of studies of the frequency of palladium allergy have been reported, in which the allergic response is usually assessed by means of a patch test using $PdCl_2$. Some of these studies have been on selected populations e.g. populations employed in platinum refineries or with a history of other allergy. The frequency of palladium sensitivity appears to range from 2% to 18%. The overwhelming proportion of individuals sensitive to palladium are also sensitive to nickel (Wataha and Hanks, 1996).

Previous evaluations

Due to the lack of suitable data the WHO considered it not possible to make a quantitative risk assessment and therefore, could not derive a health based guidance value (WHO, 2002). With regards to dental health they concluded that protection of the public may be achieved either by limiting the use of certain alloys or using alloys with minimal release of palladium. They also considered that both dentists and patients should be informed about the composition of dental alloys and the possible sensitisation effects of palladium (WHO, 2002).

Previous COT evaluations

In 1998, the COT evaluated palladium in food when the results of the 1994 TDS were considered (COT, 1998). The estimated population dietary exposure to palladium from the 1994 TDS was 0.001 mg/day, approximately 0.017 µg/kg body weight/day for a 60 kg adult. The Committee concluded that from the available data, there is was reason to believe that current intakes of palladium from the diet pose a risk to health. However, the toxicological database on palladium metal and its compounds were extremely limited (COT, 1998).

References

COT (1998). Statement on the results of multielement surveys in various items of the diet. COT Statement 1998. Annual Report 1998 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Food, Consumer Products and the Environment.

Gebel T, Lantsch H, PleBow K and Dunkelberg H (1997). Genotoxicity of platinum and palladium compounds in human and bacterial cells. *Mutation Research* **389**, 183-190.

Goering PL (1992) Platinum and related metals: palladium, iridium, osmium, rhodium and ruthenium. *In* Hazardous Materials Toxicology, Clinical Principles of Environmental Health, ed Sullivan and Krieger, Williams and Wilkins, Baltimore, US.

Hildebrand HF, Floquet I, Lefevre A and Veron C (1996). Biological and hepatotoxic effects of palladium. An overview on experimental investigations and personal studies. *Int J Risk & Safety in Medicine* **8**, 149-167.

Holbrook DJ, Washington ME, Leake HB and Brubaker PE (1975). Studies on the evaluation of the toxicity of variuos salts of lead, manganese, platinum and palladium. *Envir Health Pers* **10**, 95-101.

Johnson Matthey (1997). Tetraammine palladium hydrogen carbonate: Twenty-eight day repeated dose oral (gavage) toxicity study in the rat. Hertfordshire, Johnson Matthey plc (SPL Project No. 036/084; unpublished report reported in WHO, 2002, Health Criteria 226: Palladium. International Programme on Chemical Safety. World Health Organization, Geneva.)

MAFF (1998) Food Survey Information Sheet No 149 Dietary intakes of metals and other elements.

Moore W, Hysell D, Campbell K and Stara J (1975). Preliminary studies on the toxicity and metabolism of palladium and platinum. *Envir Health Persp* **10**, 63-71.

Mortelmans K, Haworth S, Lawlor T, Speck W, Tainer B and Zeigler E (1986). Salmonella mutagenicity tests. 2. Results from the testing of 270 chemicals. *Environ Mutagen* **8 (Supp 7**), 1-119.

Wataha JC and Hanks CT (1996). Biological effects of palladium and risk of using palladium in dental casting alloys. *J of Oral Rehabilitation* **23**, 309-320.

WHO (2002). Environmental Health Criteria 226: Palladium. International Programme on Chemical Safety. World Health Organization, Geneva.

17. Platinum (Pt)

The key sources of information for this summary were statements from the COT from 1996 (platinum based fuel catalysts) and 1998 (multi-element surveys on certain dietary components), the WHO Environmental Health Criteria (No 125, WHO, 1991) and the Criteria Document for an Occupational Exposure Limit from the UK Health and Safety Executive. (EH65/24)

Toxicity evaluations on platinum have made distinctions between the toxicity of platinum and is salts on one hand and the toxicity of platinum complexes which can have medicinal (i.e. cis-platinum) or industrial applications (fuel catalysts). Platinum complexes have mutagenic, teratogenic and carcinogenic activities, while soluble platinum salts are relatively less toxic. Insoluble salts and platinum metal are considered to be of relatively low systemic toxicity.

Chemistry and occurrence

Platinum is one of the platinum group metals, along with palladium, rhodium, ruthenium, iridium and osmium, and occurs in Group VIII of the periodic table which also includes iron, nickel and cobalt. These elements have a tendency to form complex salts, although platinum naturally also forms some simple salts.

The average concentration of platinum in the earth's crust ranges between 0.001 and 0.005 ppm. It is found in its metallic form and in a number of minerals such as sperrylite (PtAs) cooperite (Pt, Pd)S and braggite (Pt, Pd, Ni)S.

Platinum escapes into the environment from various industrial sources and is also emitted from vehicle exhausts, mainly in the metallic form. Airborne concentrations from a single engine are calculated to be 0.005 $\mu g/m^3$ for platinum and 0.00008 $\mu g/m^3$ for soluble salts of platinum (COT, 1996). The precise form of platinum compounds in the diet is unknown.

As part of the total diet study from 1994, the levels of platinum group metals were analysed in the average UK diet. Concentrations of the platinum group elements (i.e. ruthenium, rhodium, iridium, palladium and platinum) were very low with few food groups containing concentrations of these elements above their LODs. Those food groups which contained detectable concentrations of the platinum group elements were nuts (0.003 mg/kg palladium, 0.004 mg/kg rhodium); and oils and fats, offal, and bread which all had palladium concentrations of 0.002 mg/kg[1,4].

Dietary intake estimates of the platinum group elements for average, mean and upper range (97.5 percentile) consumers were very low.

Absorption and elimination

Platinum and its salts are poorly absorbed following exposure by inhalation, and absorption via the gastro-intestinal tract is likewise very limited. Following

absorption, platinum metal and salts are preferentially taken up by the kidney, with significant uptake also by the liver and the spleen. Uptake by the brain is negligible. Platinum salts appear to cross the placenta and enter the foetus in small amounts, and also distribute to the suckling rodent via the maternal milk. Following uptake most of the administered metal or salt is eliminated via the faeces with a small amount in the urine (HSE, 1996).

The retention of a single oral dose of platinum (amount not clearly specified) was measured in adult and suckling rats in a briefly reported study (Moore *et al.*, 1975 a, b). After dosing, animals were placed in metabolism cages for 24-hour collection of urine and faeces. Immediately after dosing and at intervals of up to 14 days, whole-body radioactivity was measured. Results indicated that gastro-intestinal absorption was probably extremely low, with over 99% of the administered dose eliminated in the faeces within 3 days.

As part of a large study, rats were given a single intragastric dose of $Pt(SO_4)_2$ at 382 mg Pt/kg body weight. After two weeks, the highest levels were found in the kidney (16 ppm), with detectable levels in the liver and spleen (range 1-4 ppm). Rats were also give soluble Pt^{4+} salts in the drinking water ($PtC1_4$ or $Pt(SO_4)_2$) at a concentration of 319 ppm Pt for 8-9 days. High tissue concentrations were found in the kidney (4.5-5 ppm), followed by the liver (0.8-2.2 ppm) (Holbrook *et al.,* 1975).

Few data are available on the toxicokinetics of platinum and its compounds in humans, other than those relating to the use of compounds such as cisplatin (*cis*-$[PtC1_2(NH_3)_2]$), in clinical oncology, where only parenteral routes of administration are involved.

Toxicity in animals

The acute toxicity of platinum salts is low and depends on the chemical species of platinum involved, with soluble platinum salts more toxic than insoluble salts. The following platinum salts are in approximate order of acute oral toxicity: $Na_2PtCl_6 > K_2PtCl_6 > (NH_4)_2PtCl_6 > (NH_4)_2\ PtCl_4 > Pt(SO_4)_2 > Na_2\ Pt(OH)_6$, all with a range of LD_{50} values from 10 mg/kg body weight to 1137 mg/kg body weight (HSE, 1996).

Rats were administered platinum salts in the drinking water using a variety of concentrations for periods of up to 30 days (Holbrook *et al.*, 1975). A 30-day exposure to a saturated solution of $PtCl_2$, containing only trace amounts of platinum (14 ppb) led to no changes in body weight gain. Similarly, a 30-day exposure to $PtC1_4$ in drinking water at a concentration of 105 ppm led to no changes in body weight gain, organ weights or any biochemical changes. In rats given 317 ppm of $PtCl_4$ or $Pt(SO_4)_24H_2O$ in the drinking water, body weight gain, food and fluid consumption all went down by 20%, relative to control values during the first week of exposure. However, these parameters returned to control levels in the following 3 weeks. At the end of a 30-day exposure to 317 ppm $PtCl_4$ in the drinking water, there were no changes in organ weights, or hepatic biochemical changes. Assuming a body weight of 250 g and a daily water intake of around 50 mL, the NOAEL of 105 ppm from

this study equates to 21 mg $PtCl_4$/kg body weight/day or 13 mg Pt/kg body weight/day.

The genotoxic potential of platinum salts has not been systematically investigated. From limited data it appears that some soluble platinum salts are mutagenic *in vitro,* although this potential has not been demonstrated *in vivo*.

Regarding *in vitro* studies, $Pt(NH_3)_4$, $(NH_4)_2PtCl_6$ and $PtCl_4$ have all been found to be mutagenic to bacteria. $Pt(SO_4)_2$ induced an increase in forward mutations at the HGPRT locus in mammalian cells, but $K_2PtCl_4$and K_2PtCl_6 were inactive in this assay. $Pt(NH_3)_4Cl_2$ and K_2PtCl_4 were not clastogenic to the bone marrow cells of mice or Chinese hamster (HSE, 1996).

No carcinogencity data are available for soluble forms of platinum. There is however, sufficient evidence for the carcinogenicity of cisplatin in animals.

In a reproductive study, pregnant mice were given single intragastric doses of $Pt(SO_4)_2$ at 200 mg Pt/kg body weight on days 7 or 12 of gestation or postpartum (D'Agostino *et al.*, 1984). Other dams were given single subcutaneous injections of Na_2PtCl_6 on day 7 or 12 of gestation. Control groups were similarly treated with either intragastric sulphuric acid or subcutaneous administration of phosphate-buffered saline. The only findings were that pups with pre-, but not those with post-natal exposure to $Pt(SO_4)_2$ showed a reduction in body weight up to day 45 post-partum compared with low-pH exposed control groups. Also, there was said to be reduction in the activity level of pups from dams given Na_2PtCl_6 on gestation day 12 but not in those dosed on day 7. However, as no information on maternal toxicity was given in this report no conclusions could be drawn (HSE, 1996).

Toxicity in humans

The most significant health effect from exposure to soluble platinum salts is sensitisation. Halogenated platinum salts are highly allergenic in humans, producing a type 1 (IgE mediated) response. There is evidence that platinum salts of relatively low molecular mass act as haptens and combine with serum proteins to form complete antigens. Reactive platinum species with good leaving groups (e.g. chloride ligands) are more likely to bind covalently to nucleophilic groups on protein molecules and are therefore more likely to elicit allergic responses. Both dermal and respiratory sensitisation has been observed in occupationally exposed groups. There are no studies of sensitisation by the oral route. Signs and symptoms in affected individuals include scaly erythematous dermatitis on the hands, face and neck; conjunctivitis and rhinitis; wheezing, breathlessness and cyanosis. Some individuals show extreme sensitivity and have even developed acute anaphylactic responses (including profound drop in blood pressure, intense bronchospasm and generalised urticaria) as a result of skin testing with dilute solutions of platinum chloride salts (HSE, 1996).

It was noted in one study that in workers with known hypersensitivity to $[PtCl_6]^{2-}$ and $[PtCl_4]^{2-}$ salts, positive dermal reactions were obtained on skin prick testing with platinum halide salts, but not with neutral complexes or salts

in which the platinum species is not directly associated with a halide ligand. This indicates a lack of cross-reactivity for the antibody response to platinum halide salts with other platinum complexes. These observations, together with the consistently negative results obtained in animal studies with $PtSO_4$, provide suggestive, but not conclusive evidence, that it is only the platinum halide (chiefly chloride) salts which pose a sensitisation hazard (HSE, 1996).

A number of studies have shown that allergic sensitivity to platinum halide salts is retained even at 5 years after cessation of exposure (as indicated by skin prick testing). However, one study found that skin sensitivity to platinum salts declined on cessation of exposure. This may be attributed to the policy of ensuring the rapid removal from occupational exposure of those individuals showing positive skin test results at regular monitoring (HSE, 1996).

Previous evaluations

WHO reviewed platinum in 1991, although the review concentrated mainly on the inhalation risks from platinum emitted from vehicle exhausts and on dermal effects. Limited attention was directed towards the oral toxicity of platinum and its compounds (WHO, 1991).

A WHO task group on metals met in 1994 to revise the Air Quality Guidelines for Europe (WHO, 1994). The group concluded that on the basis of observations from work-related exposure, the lowest adverse effect level for soluble platinum was equal to or below 0.05 µg Pt/m^3 (derived from an occupational study of refinery workers in Germany in which symptoms of asthma, rhinitis, dermatitis and conjunctivitis were recorded). The group considered that (apart from a very small number of individuals sensitised to platinum who may develop adverse effects at much lower levels than 0.05 $\mu g/m^3$), adverse effects arising in the general population would be unlikely at levels of 0.05 $\mu g/m^{3.}$ This would equate to an individual inhalation exposure of 1µg Pt/day (assuming 20 m^3 of air inhaled daily).

A review by the Health and Safety Executive (HSE, 1996) evaluated the toxicity of platinum metal and soluble platinum salts (specifically excluding cis-platinum) with a view to setting occupational exposure limits for workplace exposure. In view of the available data on the possible sensitising potential of platinum salts, it was not possible to draw conclusions as to whether or not a threshold for sensitisation could exist, and a pragmatic limit, based on good practice in industry, was recommended (0.002 mg/m^3, 8-hour time weighted average). Assuming 10 m^3 of air inhaled over a working shift, this limit would equate to a daily intake of some 20 µg of platinum.

There are currently no relevant safety guidelines for oral exposure to platinum.

Previous COT evaluations

The COT has previously looked at organometallic platinum compounds in the context of their use as diesel fuel catalysts (COT, 1996). The Committee considered the proposed usage and the projected emissions and noted that, if

the majority of the emissions were in the form of the metal, there would be no risk to health; and that the platinum emissions from the catalyst were unlikely to be in an allergenic form (COT, 1996).

In 1998, the COT evaluated platinum in food when the results of the 1994 TDS were considered (COT, 1998). The estimated population dietary exposure to platinum from the 1994 TDS was 0.0002 mg/day, approximately 0.003 µg/kg body weight/day for a 60 kg adult. The highest concentrations of platinum were detected in oils and fats (2 µg/kg) (MAFF, 1997). The Committee concluded that there was no evidence that current dietary intakes of platinum in the UK diet may be harmful to health (COT, 1998). An earlier dietary evaluation (Hamilton and Minski, 1972/3) estimated a total platinum intake of less than 1µg/day. There was no information for intake from dietary supplements or from particular food sources.

References

COT (1996). Platinum-based fuel catalyst for diesel fuel. Annual Report 1996 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Food, Consumer Products and the Environment.

COT (1998). Statement on the results of multielement surveys in various items of the diet. COT Statement 1998. Annual Report 1998 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Food, Consumer Products and the Environment.

D'Agostino RB, Lown BA, Morganti JB et al (1984). Effects on the development of offspring of female mice exposure to platinum sulphate. J Toxiol Environ Health 13 p879-891.

Hamilton EL and Minski MJ (1972/3). Abundance of the chemical elements in main diet and possible relations with environmental factors. Sci Tot Env 1 p375-394.

Holbrook DJ, Washington ME, Leake HB and Brubaker PE (1975). Studies on the evaluation of the toxicity of various salts of lead, manganese, platinum and palladium. Env Health Perspect 10 p95-101.

HSE (1996). Criteria document for an occupational exposure limit. HSE Document EH65/24, HSE Books.

MAFF (1998) Food Survey Information Sheet No 149 Dietary intakes of metals and other elements.

MAFF (1997). 1994 Total diet study for England and Wales.

Moore WJV,Hysell D; Crocker W and Stara JF (1975a). Biological fate of a single administration of ^{191}Pt in salts following different routes of exposure. Env Res 9 p152-158.

Moore WJV, Hysell D, Hall L, Campbell K and Stara J (1975b). Preliminary studies on the toxicity and metabolism of palladium and platinum. Environ Health Perspect 10 p63-71.

WHO (1991). Platinum, Environmental Health Criteria 125. WHO, Geneva.

WHO (1994). WHO Task Group on metals for revision of Air Quality Standards. (Unpublished Report).

18. Rhodium (Rh)

There have been no published reviews of rhodium toxicity carried out by regulatory authorities or by international organisations (e.g. WHO, EFSA). There are insufficient experimental and human toxicological data to be able to make an appraisal of the toxicity of rhodium and its compounds. Chlorinated compounds of rhodium show similarities with related platinum compounds in terms of mutagenic and cytotoxic activity and there is some evidence of cross-sensitivity to allergenic effects. Rhodium compounds however, would appear to be less potent than their platinum counterparts.

Chemistry and occurrence

Rhodium, along with platinum, palladium, ruthenium, iridium and osmium is a member of Group VIII of the Periodic Table. This group is commonly known as the platinoids or platinum group metals (PGM). The platinoids form an important group of commercial metal alloys - platinum-rhodium alloy is used as a catalyst and in glass manufacture. It forms salts with valence status +2, +3, +4, - the chloride, nitrate sulphate and soluble hexachloro complexes are trivalent. In biological media rhodium forms stable compounds with a co-ordination number of 6 (Goering, 1992).

The uses of rhodium are distributed among the chemical (38%), electrical (25%), jewellery (17%) and glass (12%) industries. Carbonyl complexes of rhodium catalyse the hydrogenation of olefins and acetylene. Other rhodium catalysts are involved in the control of NO emissions for combustion engines and industrial stacks. Rhodium is also used in small amounts in some metallic dental materials in order to increase their hardness (Goering, 1992).

As part of the total diet study from 1994, the levels of platinum group metals were analysed in the average UK diet. Concentrations of the platinum group elements (i.e. ruthenium, rhodium, iridium, palladium and platinum) were very low with few food groups containing concentrations of these elements above their LODs. Those food groups which contained detectable concentrations of the platinum group elements were nuts (0.003 mg/kg palladium, 0.004 mg/kg rhodium); and oils and fats, offal, and bread which all had palladium concentrations of 0.002 mg/kg[1,4].

Dietary intake estimates of the platinum group elements for average, mean and upper range (97.5 percentile) consumers were very low.

Absorption and elimination

Oral uptake of the platinoids is very low. Only the intravenous and inhalation routes have shown significant uptake (Goering, 1992).

Whole-body retention half-times of rhodium were 1750 and 3230 days for a $^{106}RhO_2$ mixture (particle size 0.07 to 0.12 µm), as estimated from single exposure by inhalation in beagle dogs. After 3.25 years following the single exposure, 82 to 85 percent of the total body burden still remained in the lung,

with the remainder translocated predominantly in the lymph nodes (Beliles, 1994).

Excretion of rhodium (as for platinum and ruthenium) following intravenous injection is mainly in the urine: 20-45% of the dose is excreted in 24 hours, and 80% in 1 week. Orally administered platinoids are excreted primarily in the faeces without significant absorption (Goering, 1992).

Toxicity in animals

Moderately low acute toxicities have been reported for rhodium; the intravenous LD_{50} of $RhCl_3$ for rat and rabbit being 198 and 215 mg/kg body weight, respectively. All rat deaths occurred within 48 hr post-injection. After 100 days no histological lesions in selected organs were found among the survivors. For the rabbits, all deaths occurred within 12 hours, and after 30 days, no histological lesions attributable to rhodium were visible (Beliles, 1994).

The mouse LD_{50} (route not specified) of the triaminetrichloro Rh (III) complex was reported as 225 mg/kg body weight, a value indicating about seven times greater toxicity than that of a comparable iridium complex (Beliles, 1994).

A slight suppression in growth in Charles River CD mice was observed following lifetime administration of 5 ppm rhodium in drinking water - equivalent to less than 1 mg Rh/kg body weight/day (Schroeder, 1973).

Studies in mice showed a slightly higher malignancy rate (28.8% with malignant tumours versus 13.8% in controls) after the addition of 5 ppm rhodium (ie <1mg Rh/kg body weight/day) to their drinking water. There is insufficient reporting in the published article in terms of pathological findings and experimental detail to be able to make an evaluation of the carcinogenicity of rhodium from this study (Schroeder and Mitchener, 1971).

Positive results in the Rec assay with *Bacillus subtilis* (H17 and M45 strains) were reported for rhodium trichloride ($RhCl_3$) and nitrate ($RhNO_3$) at a concentration of 0.005 M. Spot mutation induction tests with rhodium trichloride in *E coli* WP2 (base change mutations) were also positive as were findings in *Salmonella typhimurium* strain TA98 although negative results were observed in TA100, TA1535, TA1537 and TA1538. A dose response relationship for $RhCl_3$ was also demonstrated in *E coli.* The authors considered these rhodium compounds (along with chloro-platinum compounds; $PtCl_4$ $NH_4)_2PtCl_6$) as potent mutagens (Kanematsu *et al*., 1980). Other Rh(III) and Rh(I) salts have also been shown to be mutagenic in bacterial systems (Warren *et al.,* 1981; Aresta *et al.,* 1985).

Toxicity in humans

The ability of rhodium salts to cross-react with the principal sensitising agent ammonium hexachloroplatinate was investigated in refinery workers. Selected subjects were screened by skin-prick test, specific RAST, RAST inhibition, and primate PCA tests. These showed - but only in platinum-sensitive

subjects - a low prevalence of skin and RAST sensitivity to the other platinum group metals (e.g. palladium, ruthenium etc.) and limited evidence of hapten-specific cross-reactivity (Beliles, 1994).

There are no data in the literature relating to the acute or chronic health effects of rhodium or its compounds in man. Despite industrial usage of rhodium compounds, effects upon the respiratory system and skin, similar to those produced by platinum compounds have not been observed. A single unpublished observation (quoted in ACGIH,1981) refers to nasal irritation in a plant where the concentration of rhodium salt was in the order of 0.002 mg Rh/m^3. A long-term exposure limit of 0.001 mg Rh/m^3 has been set for soluble salts of rhodium by the ACGIH and this has also been taken as the occupational exposure limit in the UK (HSE, 1997). Workplace inhalation intake at this limit corresponds to a theoretical intake by inhalation of 10 µg Rh/day (assuming inhalation of 10 m^3 over a daily working shift).

Previous COT evaluations

In 1998, the COT evaluated rhodium in food when the results of the 1994 TDS were considered (COT, 1998). The estimated population dietary exposure to rhodium from the 1994 TDS was 0.0003 mg/day, approximately 0.005 µg/kg body weight/day for a 60 kg adult. The Committee concluded that despite a lack of information on the effects of low doses of rhodium upon man, the level of rhodium in the diet did not indicate a cause for concern (COT, 1998).

References

ACGIH (1981). Documentation on the Threshold Limit Values 4 ed p358-359. American Congress of Governmental Industrial Hygienists.

Aresta M Treglia S Collucia M *et al* (1985). Mutagenic activity of transition metal complexes: Relation structure - mutagenic and anti-bacterial activity for some Pd(II) Pt(II) and Rh(I) complexes. In: Merian E *et al* eds Carcinogenic and Mutagenic Metal Compounds. New York. Gordon and Breach p453-456.

Beliles RP (1994). The Metals - Platinum Group Metals. In: Patty's Industrial Hygiene and Toxicology 4th edition. Ed Clayton GD and Clayton FE Vol IIC, p2183-2201.

COT (1998). Statement on the results of multielement surveys in various items of the diet. COT Statement 1998. Annual Report 1998 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Food, Consumer Products and the Environment.

Goering PL (1992). Hazardous Material Toxicology. Clinical Principles of Environmental Health. Eds Sullivan JB Jr and Krieger G D. Chapter 82: Platinum and Related Metals p874-881.

HSE (1997). Occupational Exposure Limits 1997. Guidance Booklet EH40/97, HSE Books.

Kanematsu N, Hara M and Kada T (1980). Rec Assay and Mutagenicity Studies in Metal Compounds. Mut Res 77 p109-116.

MAFF (1998) Food Survey Information Sheet No 149 Dietary intakes of metals and other elements.

Schroeder HA (1973). Recondite toxicity of trace elements. In: Essays in Toxicology Vol 4. Ed Hayes WJ Jun. Academic Press New York p105-199.

Schroeder and Mitchener M (1971). Scandium, chromium (VI), gallium, yttrium, rhodium, palladium, indium in mice. Effects on growth and life span. J Nutr. 101 p1431-1437.

Warren G Abbott E Schultz P *et al* (1981). Mutagenicity of a series of hexacoordinate rhodium (III) compounds. Mut Res 88 p165-173.

19. Ruthenium (Ru)

There have been no published reviews of ruthenium toxicity carried out by regulatory authorities or by international organisations (e.g. WHO, EFSA). There are very limited experimental toxicological data for ruthenium and its compounds.

In comparison to other platinum group metals the compounds of ruthenium would appear to be less toxic than equivalent compounds/complexes of platinum.

Chemistry and occurrence

Ruthenium, along with platinum, palladium, rhodium, iridium and osmium is a member of Group VIII of the Periodic Table. The group is commonly known as the platinoids or platinum group metals (PGM). Ruthenium occurs as the natural mineral laurite (RuS_2) and constitutes about 0.4% of the earth's crust. Two-thirds of ruthenium is used by the electrical industry and most of the remainder by chemical industry. It is a potent hardener for platinum, and ruthenium alloys are used in electrical contacts. Occupational exposure to ruthenium is possible (Beliles, 1994).

Ruthenium can exhibit valence states ranging from +2 to +8, but +3 is the most common. $Ru(OH)_2$, $RuCl_4$ and RuO_2 are stable and water soluble, but generally the trivalent salts are not soluble. There is no information on the forms of ruthenium found in foods.

As part of the total diet study from 1994, the levels of platinum group metals were analysed in the average UK diet. Concentrations of the platinum group elements (i.e. ruthenium, rhodium, iridium, palladium and platinum) were very low with few food groups containing concentrations of these elements above their LODs. Those food groups which contained detectable concentrations of the platinum group elements were nuts (0.003 mg/kg palladium, 0.004 mg/kg rhodium); and oils and fats, offal, and bread which all had palladium concentrations of 0.002 mg/kg[1,4].

Dietary intake estimates of the platinum group elements for average, mean and upper range (97.5 percentile) consumers were very low.

Absorption and elimination

Oral absorption of the platinoids is very low. Absorption can occur after intravenous injection (but not generally via other parenteral routes). About 20-45% of an intravenous dose of ruthenium is excreted within 24 hours, and 80% is excreted in a week. Orally administered platinoids are excreted in the faeces (Goering, 1992).

A metabolic study of the simple salt $^{106}RuCl_3$ has also been made by Furchner *et al.* (1971) in rats, mice, and dogs by the oral, intraperitoneal, and intravenous routes. From plots of whole-body retention of ^{106}Ru from gavage and intraperitoneal injection in mice and rats, metabolic patterns remarkably

similar to those of the other platinum-group metals were found, with very uniform values between the two species. Retention from oral administration was a fraction of that from intraperitoneal injection. The amounts of ^{106}Ru absorbed were so small as to preclude activity measurements beyond 23 days in rats and 33 days in mice. Across species, the cumulative 3-day faecal excretion differed by less than 4 percent from an estimated gastrointestinal absorption of 3.5 percent.

The distribution pattern of $^{103}RuCl_3$, $RuCl_4$, and nitrosyl ruthenium ingested in tracer amounts by humans to reflect the movement from the stomach to the large intestine was determined by Yamagata *et al.* (1989). As in animals, the absorption of ^{103}Ru "was very small". Even after 2 and 4 days, peaks in the distribution patterns were still present in the large intestine. Comparison of the excretion of Ru^{3+}, Ru^{4+}, and the chloro complexes of nitrosyl Ru^{3+} revealed that, although only 0.5 percent of the dose of Ru^{3+} and Ru^{4+} was eliminated in the urine in 24 hours, this was almost three times the amount of nitrosyl Ru^{3+} eliminated. The remainder was excreted in the faeces in each instance. This was a demonstration of the slower hydrolytic conversion of the nitrosyl complex to insoluble, less absorbable compounds in the gut than that of the simple salts.

Toxicity in animals

Minimal acute toxicity data on ruthenium exist. One report of a mouse LD_{50} (route not specified) of 132 mg/kg body weight for a Ru^{3+} amine complex indicates a toxicity about 11 times greater than that of the corresponding Ir^{3+} complex.

Oral LD_{50} values for a range of ruthenium compounds are as follows:

Ruthenium Chloride hydroxide	Rat	1250 mg/kg bw
Ruthenium Chloride hydroxide	Mouse	463 mg/kg bw
Ruthenium Chloride hydroxide	Guinea Pig	210 mg/kg bw
Ruthenium Oxide (RuO_2)	Rat	4580 mg/kg bw
Ruthenium Oxide (RuO_2)	Mouse	5570 mg/kg bw

Ruthenium (III) ion has been shown to bind to nucleic acid bases and to nucleosides (Tselepi-Kalouli and Katsaros, 1988).

In mutagenicity screening tests with *Bacillus subtilis* rec assay with H17 and M45 strains, ruthenium trichloride produced a negative response. Positive responses were obtained for chloroplatinum salts and for chlororhodium salts. (Kanematsu *et al.*, 1980; Nishioka, 1975)

Chloro-aminoruthenium complexes, possible candidates for chemotherapeutic use, have been shown to induce mutations in the Ames *Salmonella* test system in strains TA98 (frame shift mutations) and TA100 (base pair mutations). In contrast cis-platin (cis-$Cl_2(NH_3)_2$ Pt) induced only base pair

mutations (TA100), although potency was 100 times greater than for ruthenium complexes. Ruthenium complexes also induced `SOS' functions in the *Bacillus subtilis* Comptest (Yasbin *et al.*, 1980).

The ruthenium complex, $RuCl_2$ $(DMSO)_2$ (4-nitroimidazole), which has hypoxic radiosensitising properties was investigated for genotoxic activity in *in vitro* induction of chromosome abberations (chromatid breaks and chromatid exchanges) in Chinese hamster ovary cells. A dose dependent increase in the frequencies of metaphases with chromatid abberations was observed: addition of S9 mix did not alter the clastogenic activity. The mutagenic activity was much less than that of cis-platin (Chan *et al.*, 1986).

Ruthenium (II) tris 4,4 substituted bipyridine complexes with potential medical applications as glucose biosensor components were reported to be mutagenic in the Ames test, (*Salmonella* strains, TA98 and TA100). Metabolic activation with S9 mix did not significantly alter mutagenic response (Gasiorowski *et al.*, 1995).

Toxicity in humans

There are no data on the human toxicity of ruthenium compounds although there is evidence of some clinical usage.

Following the success of cis platin (cis-$Cl_2(NH_3)_2$ Pt) as a anti-tumour agent, ruthenium compounds have been evaluated as potential chemotherapeutic candidates that may offer some advantages over platinum. Cis-$Cl_2(DMSO)_4Ru(II)$, while showing less activity than cis-platin, was also significantly less toxic. Aminoruthenium (III) complexes, and their adducts with DNA bases have been investigated for potential clinical usage. As a consequence, systematic studies of mutagenic properties of these compounds have been conducted (Yasbin *et al.*, 1980). Laboratory work has also shown that adenine and cytosine are capable of forming stable bonds with ruthenium through their exocyclic nitrogen, and this may provide a mechanism for the observed bacterial mutagenicity (Clarke, 1980).

Previous COT evaluations

In 1998, the COT evaluated ruthenium in food when the results of the 1994 TDS were considered (COT, 1998). The estimated population dietary exposure to ruthenium from the 1994 TDS was 0.004 mg/day, approximately 0.067 µg/kg body weight/day for a670 kg adult. The intake of ruthenium was higher than the total for the other four Platinum Group Metals investigated in the diet by MAFF (MAFF, 1997). The Committee concluded that levels of ruthenium in the UK diet were not a cause of concern, although there were insufficient data upon which a full evaluation could be made (COT, 1998).

References

Beliles RP (1994). The Metals - Platinum Group Metals. In: Patty's Industrial Hygiene and Toxicology 4th edition. Ed Clayton GD and Clayton FE Vol IIC, p2183-2201.

Chan PKL, Skov KA, James BR and Farrell NP (1986). Chem-Biol Interactions 59 p247

COT (1998). Statement on the results of multielement surveys in various items of the diet. COT Statement 1998. Annual Report 1998 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Food, Consumer Products and the Environment.

Clarke MJ (1980). Mutagenicity and Modes of Metal Binding to Nucleic Acids. Inor Chem 19 p1103-1104.

Furchner JE *et al* (1971). Health Physics 21 p355.

Gasiorowski K, Szyba K, Urban J *et al* (1995). Mutagenic activity of groups VIII metal-organic complexes in the Ames test: evaluation of potential glucose biosensor components. Bio Metals 8 p257-262.

Goering PL (1992). Hazardous Material Toxicology. Clinical Principles of Environmental Health. Eds Sullivan JB Jr and Krieger G D. Chapter 82: Platinum and Related Metals p874-881.

Kanematsu N Hara M and Kada T 1980. Rec Assay and Mutagenicity Studies on Metal Compounds. Mut Res 77 p109-116.

MAFF (1998) Food Survey Information Sheet No 149 Dietary intakes of metals and other elements.

MAFF (1997). 1994 Total Diet Study.

Nishioka N (1975). Mutagenic activities of metal compounds in bacteria. Mut Res 31 p185-189.

Sax I (1996). Sax's Dangerous Properties of Industrial Materials 9th ed

Lewis RS Snr Ed. Volume II.

Tselepi-Kalouli E and Katsaros N (1988). Ruthenium (III) ion complexes with nucleic acid bases and nucleosides. J Inorg Biochem 34 p63-74.

Yamagata N *et al* (1989). Health Phys 16 p159.

Yasbin RE, Mathews CR and Clarke MJ (1980). Mutagenic and Toxic Effects of Ruthenium. Chem-Biol Interactions 31 p355-365.

20. Selenium (Se)

The key source of information for this summary was the EVM review of selenium (EVM, 2003).

Chemistry and occurrence

Selenium is a group VI element and has both metallic and non metallic properties. It has an atomic weight of 78.96. It can exist in four oxidation states (-2, 1, +2, +6) and forms chemical compounds analogous to those of sulphur. The salts of selenous acid (H_2SeO_3) and selenic acid (H_2SeO_4) are selenites (Se^{4+}) and selenates (Se^{6+}) respectively.

Selenium is an essential trace element, which is necessary for the functioning of the enzyme glutathione peroxidase, which protects against intracellular oxidative damage. Other selenoproteins exist and the element may also be involved in thyroxine metabolism.

Selenium occurs in a number of foodstuffs. In the UK, the highest mean levels are found in nuts, offal, fish, eggs and poultry (MAFF, 1997a, 1997b). Most of the selenium in food is thought to be present as the amino acid derivatives selenomethionine or selenocysteine (COMA, 1991), however, knowledge of the other selenium compounds present in food is incomplete (Nordic Project Group, 1995).

Absorption and elimination

Absorption and thus bioavailability can be affected by the physical or chemical form of the selenium compound or the dosing regimen. In general, the degree of selenium absorption is independent of the exposure but in some instances, absorption can be greater where selenium deficiency exists. It is thought that 55-60% of the selenium in food is absorbed following ingestion. Human volunteer studies suggest that there is greater absorption of selenate and selenomethionine than selenite.

Absorbed selenium is rapidly distributed and does not accumulate in any specific organs although the concentration is higher in the liver and kidney. The pattern of distribution is similar for both organic and non-organic selenium. Tissue and blood levels reflect dietary intakes. Selenium from sodium selenite and selenate is found in the highest concentrations in the liver and kidneys of both humans and animals following oral exposure.

Selenite is reduced to selenide. Methylation of selenide leads to the formation of methylated selenium derivatives that are excreted in the breath (resulting in a garlic-like smell) and the urine. Urine is the major route of excretion for selenium however some excretion also occurs via the faeces (Thomson and Stewart, 1976).

Toxicity in animals

The toxicity of different selenium compounds varies depending on the nature of the compound, particularly its solubility. For example, selenite, selenate and selenomethionine are more toxic than the insoluble selenium sulphide and disulphide. Similarly, elemental selenium is less toxic than sodium selenite or selenate as a result of its lower solubility; an oral LD50 of 6700 mg/kg body weight has been reported for elemental selenium in Swiss Webster mice (Cummins and Kimura, 1971). In animals, acute toxicity is characterised by central nervous system toxicity and degenerative changes in the liver.

Adverse effects on growth rates, the kidneys and reproductive parameters have been reported in rats and mice dosed with selenium compounds chronically and sub-chronically. Domestic animals develop a condition known as blind staggers, involving impaired vision and eventual respiratory failure. Some adverse reproductive effects have also been reported in rats and mice fed selenium compounds. Seleno-methionine did not have adverse reproductive effects in macaques at doses of up to 300 μg/kg body weight/day where dose-related maternal toxicity was observed (anorexia, vomiting, and a significant reduction in body weight increased with increasing duration of selenium exposure) (Tarantal *et al.,* 1991).

The results of genotoxicity studies on selenium compounds are conflicting and are reviewed by Shamberger (1985). Selenite and selenate are weakly mutagenic in Salmonella strain TA 100. Unscheduled DNA synthesis, sister chromatid exchange and chromosomal aberrations have been observed *in vitro* in the presence of glutathione. In an *in vivo* study in hamster bone marrow, an increase in sister chromatid exchange was observed at doses of selenite which were toxic. It has been suggested (Nordic Project Group, 1995) that glutathione is required for the production of reactive oxygen metabolites, and that this may be a concentration dependent effect. Conflicting results on the co-mutagenic effects of selenium have also been reported.

Toxicity in humans

The acute effects of selenious acid have been summarised by Mack (1990). Patients suffer from hypersalivation, repetitive copius emesis (which may contain blood), diarrhoea and garlic odour to the breath. Burns and erosions may occur in the mouth and upper gastro-intestinal tract. Extreme restlessness, muscle spasms, tachycardia, pulmonary oedema and toxic cardiomyopathy may also be experienced. In some patients, rather than hypertension, a state of severe shock develops, possibly as a result of decreased contractility secondary to the cardiomyopathy as well as lowered peripheral vascular resistance. Stupor, respiratory depression and death can occur several hours post ingestion.

In China where endemic selenosis (selenium poisoning) occurs due to the high selenium content of the soil, symptoms such as brittle and pigmentless hair, skin lesions, pathological changes to the nails and neurological disturbances are observed. In a study by Yang *et al*. (1989a and b), conducted in an area of China where dietary selenium exposure is high, selenium intakes were correlated with blood levels to determine the intakes at which marginal selenium toxicity occur. This was at a total intake of 0.91 mg/day selenium.

There are some discrepancies in the NOAELs described by Yang and colleagues in that the range of intakes in high selenium areas that was not associated with selenosis were given as 0.24-1.51 mg/day whereas a LOAEL of 0.91 mg selenium/day was determined in a later study. This may have resulted from the way in which the dietary intakes were calculated but it has also been argued that subjects may have become sensitised to selenium as a result of earlier outbreaks of selenosis. A follow up study (Yang and Zhou, 1994) indicated that the symptoms of selenosis could be reversed if selenium intake was reduced.

The World Cancer Research Fund/American Institute for Cancer Research has recently concluded that selenium probably protects against prostate cancer and that there is limited evidence to suggest that selenium may protect against lung, colorectal and stomach cancers (WCRF/AICR, 2007).

Previous evaluations

In 1996 The World Health Organisation considered the most desirable level and the upper limit for safety of selenium in the diet (WHO 1996). The population *normative* intakes [§] for selenium were defined for a number of age groups. The *normative* intakes for 0-0.25, 0.25-0.5 and 0.5-0.1 year olds were 1.2, 1.29 and 1.33 μg/kg body weight/day, respectively. The upper limit of the safer range for selenium intake, 400 μg/day, was determined for adults only based on epidemiological data. The data were insufficient to determine upper limits for each age range and so it is unclear how this limit would apply to infants.

In 2000, the SCF used the study by Yang et al mentioned above (Yang *et al* 1989a and 1989b) to set a Tolerable Upper Intake Level of 300μg Se/day or 5μg Se/kg body weight/day for a 60kg adult.

The Expert Group on Vitamins and Minerals, in its report on vitamins and minerals, recommended a safe upper level for total selenium intake of 0.45 mg/day (equivalent to 0.007 mg/kg body weight/day in a 60 kg adult) (EVM, 2003). The safe upper level was based on a LOAEL of 0.91 mg/day, derived from an epidemiological study in which signs of selenosis (prolonged

[§] This refers to the level of intake that serves to maintain a level of tissue storage or other reserve that is judged by the Expert Consultation to be desirable (WHO 1996).

prothrombin time, morphological changes in the nails, increased white blood cell count) were observed in individuals with selenium blood levels of 1.054 to 1.854 mg/L, which was calculated to represent a selenium intake of 0.91 mg/day. An uncertainty factor of 3 was applied to extrapolate from the LOAEL to a NOAEL. The US EPA also used this study to derive an oral reference dose for selenium of 0.005 mg/kg body weight/day (US EPA, 1991).

The World Health Organization also derived a health-based drinking water guideline value of 0.01 mg/L for selenium (WHO, 1993), which was re-affirmed in the latest edition (WHO, 2006). This guideline value was based on an estimated human NOAEL of 4 µg/kg body weight/day from a small clinical study in which subjects with a mean daily intake of 4 µg/kg body weight/day showed no clinical or biochemical signs of selenium toxicity. Assuming a 60 kg adult drinking 2 litres of water per day and allocating 10% of the NOAEL to drinking water, results in the guideline value of 0.01 mg/L (WHO, 2006). Selenium has been included in the rolling revision of the WHO Guidelines for Drinking Water Quality and an updated factsheet is due to be published in the second addendum to the third edition (WHO, 2008). Selenium has been included in the plan of work of the rolling revision because there is a fairly narrow range between doses essential to humans and those associated with adverse effects.

Previous COT evaluations

In 2003 the COT evaluated selenium in food when the results of the 2000 TDS were considered (COT, 2003). The estimated population dietary exposure to selenium from the 2000 TDS was 0.034 mg/day, approximately 0.57 µg/kg body weight/day for a 60 kg adult. The Committee concluded that current dietary exposures to selenium were unlikely to be of any toxicological concern for consumers (COT, 2003).

References

COMA (1991). Dietary Reference Values for Food Energy and Nutrients for the United Kingdom. Report of the panel on Dietary Reference Values of the Committee on Medical Aspects of Food Policy (COMA). *Department of Health RHSS 41.*

COT (2003). Statement on twelve metals and other elements in the 2000 Total Diet Study. COT Statement 2003/07. Annual Report 2003 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Foods, Consumer Products and the Environment.

Cummins, L. M. and Kimura, E. T. (1971). Safety Evaluation of Selenium Sulfide Anti-Dandruff Shampoos. *Toxicology and Applied Pharmacology*, **20:** 89-96.

EVM (2003). Safe upper levels for vitamins and minerals. Report of the Expert Group on Vitamins and Minerals.

EFSA (2006) Tolerable Upper Intake Levels for Vitamins and Minerals. February 2006

IARC (1975). International Agency for Research on Cancer - Summaries & Evaluations Vol: 9 (p. 245). Updated in 1998.

Mack, R. B. (1990). The Fat lady Enters Stage Left. Acute Selenium Poisoning. *NCMJ*, **51:** 636-638.

Ministry of Agriculture Fisheries and Food (1997a). 1994 Total Diet Study: metals and other elements. Food Surveillance Information Sheet no 131.

Ministry of Agriculture Fisheries and Food (1997b). Dietary intake of selenium. Food Surveillance Information Sheet no 126.

Nordic Project Group (1995). Risk Evaluation of Essential Trace Elements - Essential versus toxic levels of intake. Report of a Nordic project group. *Nord 1995*: **18.**

Shamberger, R. J. (1985). The Genotoxicity of Selenium. *Mutation Research*. **154:** 19-48.

Tarantal, A. F. *et al*, (1991). Developmental Toxicity of L- selenomethionine in *Macaca fascicularis. Fundamental and Applied Toxicology,* **16:** 147-160.

Thomson, C. D. and Stewart, R. D. H. (1976). Metabolism of [^{75} Se] selenite in young women. *British Journal of Nutrition,* **32:** 47-57.

US EPA (1991). Integrated Risk Information System (IRIS): Selenium and compounds. Environmental Protection Agency National Center for Environmental Assessment.

World Cancer Research Fund /American Institute for Cancer Research (2007) Food, Nutrition, Physical Activity and the Prevention of Cancer: a Global Perspective. Washington DC: AICR, 2007.

WHO (1996). Trace Elements in human nutrition and health. World Health Organization, Geneva.

WHO (1993). Guidelines for drinking-water quality. World Health Organisation. Geneva.

WHO (2006). Guidelines for Drinking-Water Quality. Volume 1 Recommendations. First addendum to the third edition. World Health Organization, Geneva.

WHO (2008). Plan of work for the rolling revision of the WHO Guidelines for Drinking Water Quality. World Health Organization website: http://www.who.int/water_sanitation_health/gdwqrevision/en/index.html. Accessed July 2008.

Yang, G., et al, (1989a). Studies of Safe Maximal Daily Selenium Intake in a Seleniferous area in China. I. Selenium Intake and Tissue Selenium Levels of the inhabitants. *J. Trace Elemnt. Electrolotes. Health Dis.* **3:** 77-87.

Yang, G., et al, (1989b). Studies of Safe Maximal Daily Selenium Intake in a Seleniferous area in China. II. Relation between Se Intake and the manifestation of Clinical Signs and Certain Biochemical Alterations in Blood and Urine. *J. Trace Elem. Electrolytes. Health Dis.* **3**: 123-130.

Yang, G. and Zhou, R. (1994). Further observations on the human maximum safe dietary selenium intake in a seleniferous area in China. *J. Trace Element. Electrolytes. Health Dis.* **8:** 159-165.

21. Strontium (Sr)

Chemistry and occurrence

There have been no published reviews of strontium toxicity carried out by international organisations (e.g. WHO, EFSA). The US EPA and the UK COT have reviewed the toxicology of strontium and these are the main sources used for the review below.

Strontium occurs in nature chiefly as the minerals celestite ($SrSO_4$) and strontianite ($SrCO_3$), which are widespread in the rocks and waters of the earth's surface averaging 430 ppm in rock and 10 ppm in seawater. Strontium is present in small quantities in most plants (1 to 169 ppm; mean 36 ppm dry weight), but certain plant species growing on strontium rich soils in the UK may contain up to 26,000 ppm (Beliles, 1994).

Strontium is a characteristic divalent alkaline earth metal belonging to Group IIA, along with calcium and barium. As a rule, strontium salts are more water-soluble than those of barium.

Because strontium is ubiquitous in the geosphere in relatively high concentrations (ca. 400 ppm) and is thus present in all living organisms, strontium is omnipresent in human tissues. The average whole-body content of strontium, based on a reference man (a 70-kg American adult) is 320 mg as determined by emission spectroscopy on 400 human subjects from many areas of the world (Tipton & Cook, 1963).

Over 99% of naturally occurring strontium is in the form of its non-radioactive isotopes (strontium 86, 87 and 88). Radioactive strontium-90 (half-life 28.8 years) is a major long-lived fission product from uranium and is found in the fall out from nuclear explosions.

Absorption and elimination

Absorption of strontium from the gastrointestinal tract varies greatly, ranging from 9-63% (average of 38%), based on studies with radioactive strontium-90 (Snyder *et al*., 1975). The bioavailability of strontium was estimated to be 20% in 6 healthy adult males administered 2.5 mmol of strontium chloride (Leeuwenkamp *et al.*, 1990). Deficiency of dietary calcium leads to an increased absorption of strontium (Beliles, 1994).

Ingested strontium is distributed in the body in three compartments: extracellular fluid, soft tissues, and the bone itself (El Solh and Rousselet, 1981). The average adult is estimated to have a body burden of 320 mg strontium, 99% of which is in the bone at an average concentration of 100 ppm (Snyder *et al*., 1975; Beliles, 1994).

In pharmacokinetic studies in six male volunteers, a single oral dose (200mL) of a 2.5 mmol solution of strontium chloride hexahydrate (equivalent to about 1000 mgSr/L or about 3 mgSr/kg body weight) resulted in peak plasma concentrations of 3550 µg/L (Leeuwenkamp *et al.*, 1990).

As opposed to calcium, which is under homeostatic regulation, strontium appears to be passively absorbed (Comar and Wasserman, 1964). However, several factors may affect the bioavailability of ingested strontium, for example, age, species, the form of strontium, and the composition of the diet, especially with regard to phosphorus, vitamin D and calcium levels (US EPA, 1990, 1992). Experiments in animals suggest that the ratio of strontium to calcium in the bone is based on the relative intake of the two elements and the maturity of the intestine and kidney in discriminating between the two cations (Sugihira and Suzuki, 1991).

Toxicity in animals

Toxicity studies with strontium-90 have shown teratogenic, reproductive and carcinogenic effects which are attributable to its radiological rather than its chemical properties.

The capacity of strontium to substitute for calcium is dominant in its mode of toxicity which is characterised by its effects upon bone mineralisation and development. In addition, because of its close relationship with calcium, its interactions with phosphate, magnesium, and potassium must also be noted. Strontium can substitute for calcium in maintaining excitability of nerve and muscle membranes and strontium can activate and enhance transmitter (acetylcholine) release from myoneural junctions at low calcium concentrations. At higher calcium concentrations, strontium inhibits transmitter release. Strontium also substitutes for calcium in the excitation and contraction of skeletal and cardiac muscle, and also substitutes for calcium in the process leading to neurohypophyseal hormone secretion (Beliles, 1994).

The strontium ion is considered to have low oral toxicity. The oral LD_{50} for strontium chloride in the rat is 2250 mg/kg body weight and in the mouse 1874 mg/kg body weight (Sax, 1996). Symptoms of acute toxicity are excessive salivation, vomiting, colic and diarrhoea, and possibly respiratory failure.

Storey (1961) fed young (40-60 g) and adult (200-250 g) female rats diets with adequate calcium (1.6%), phosphorus (0.9%) and vitamin D for 20 days. The dietary levels of strontium (as strontium carbonate) given to both adult and young rats were 0.19, 0.38, 0.75, 1.0 (young rats only), 1.5 and 3.0%. Assuming young rats consume 10% and adult rats consume 5% of their body weight in food per day, these doses correspond to 190, 380, 750, 1000, 1500 and 3000 mg/kg body weight/day for young rats and 95, 190, 375, 750 and 1500 mg/kg body weight/day for adult rats. Rats were examined for changes in bone mineralization and defects in cartilage. They were weighed at the onset and end of the experiment. Young rats were found to be affected more severely at lower dietary strontium levels than were adult rats. In young rats at 0.38% (380 mg/kg body weight/day) the epiphyseal plate was irregular and slightly widened; however, at 0.75% (750 mg/kg body weight/day) this plate was so irregular that measurements were unreliable. Changes observed with the dose of 0.38% and higher were inhibition of calcification, as evidenced by increasing width of epiphyseal cartilage, presence of uncalcified bone matrix and decreased ash weight of bone. In adults, the first obvious bone change

occurred at the 1.5% dietary strontium level (750 mg/kg body weight/day) and included slightly wider than normal epiphyseal cartilage plate and metaphyseal osteoid seams, which were irregularly increased in extent and width. At the 3% strontium level in adult animals (1500 mg/kg body weight/day), the cartilage plate was much larger. For young rats, the dietary level of 0.19% strontium (190 mg/kg body weight/day) was considered to be the NOAEL. For adult rats, the dietary level of 0.75% strontium (375 mg/kg body weight/day) was a NOAEL.

Marie *et al*. (1985) administered stable strontium to weanling male Sprague-Dawley rats. The purpose of this study was to determine the effect of low doses of stable strontium on mineral homeostasis and bone histology. Rats received 0, 0.19, 0.27, 0.34 and 0.40% of $SrC1_2$ in distilled water for 9 weeks. The authors estimated average strontium intakes of 0, 316, 425, 525 and 633 mg/kg body weight/day. Rats in the 0.40% (633 mg/kg body weight/day) dose group showed signs of increased mineralization lag time; excessive osteoid thickness associated with a decline in the rate of calcification, which resulted in slow growth rate; and a decreased double-labelled osteoid surface, which frequently resulted in defective long bone growth. This study identified a NOAEL of 525 mg/kg body weight/day and a LOAEL of 633 mg/kg body weight/day.

Skoryna (1981), as part of wider clinical observations, investigated also the oral toxicity of stable strontium in male adult RVH hooded rats. The rats were fed *ad libitum* a standard laboratory diet and divided into four groups which were administered 0.002, 900, 1900 or 3400 ppm strontium chloride (55% strontium) in their drinking water for 3 years or bred for 3 generations. The experimental doses corresponded to 70, 147 and 263 mg Sr/kg body weight/day. A chronic NOAEL of 263 mg/kg body weight/day was identified from this study.

In a 90 day study, groups of 10 male and 10 female Wistar rats were fed with a semi purified diet containing 75, 300, 1200 or 4800 ppm strontium chloride hexahydrate (approximately 3.8, 15, 60 or 240 mg/kg body weight/day). Adequate levels of Ca (0.85%), Mg, P, and Vitamin D3 were contained in the diet. Thyroid weights were increased in males in the 1200 and 4800 ppm group and histological evidence of increased thyroid activity was noticed in males of the 4800 ppm group. In females, pituitary weights were significantly decreased in the 300 ppm and 4800 ppm groups but not in the 1200 ppm group. Increased strontium concentrations in bone were observed at all levels after 4 weeks but blood calcium levels remained unchanged. The authors, discounted the increase in bone strontium as a toxic event, and considered the NOAEL from these studies to be 300 ppm (calculated to be equivalent to about 15 mg Sr/kg body weight/day; Kroes *et al.,* 1977).

Relatively little information is available regarding the potential for developmental toxicity resulting from exposure to strontium. Pregnant female Wistar rats (3/group) were administered subcutaneous doses of 0, 25, 50, 100 or 200 mg/kg body weight of strontium nitrate (equivalent to 10.3, 20.7, 41.4 or 82.8 mg Sr/kg body weight/day, respectively) during gestational days 9-19 (Landsdown *et al.*, 1972). No effects were seen on the size or body weight of

fetuses, litter sizes or the number of resorption sites. The skeleton and zones of calcification were normal and no histological changes were seen in soft tissues. Although this study reported no teratogenic effects of strontium, the small number of dams exposed and fetuses examined preclude a definite evaluation of the reproductive toxicity of strontium.

In vitro mutagenicity studies with strontium chloride have shown negative results in the rec-assay in *Bacillus subtilis* (Kanematsu *et al.,* 1980) and no effect upon the fidelity of DNA replication (Zakour *et al.,* 1981; Niyogi and Feldman, 1985). However, Ghosh and co-workers (1990) reported that oral administration of large single doses of strontium chloride (from 1/2 to 1/20th of an oral LD_{50} dose) to 6-8 week old mice *in vivo* induced chromosomal aberrations in bone marrow cell metaphase preparations. Increases were seen at the lowest dose investigated (260 mg/kg body weight in male mice and 240 mg/kg body weight in female mice).

Toxicity in humans

In clinical studies to determine calcium and strontium levels in plasma, Skoryna (1981) reported no adverse health effects in 50 patients to whom he administered between 0.1 and 1.5 g/day of strontium gluconate (equivalent to 274 mg Sr/day) for at least 3 months.

There are no epidemiological data concerning the health effects of strontium, although there is a long history of use of strontium clinically in the treatment and prevention of osteoporosis, and relatively high levels of strontium have been given (1700 mg Sr/day) without any clear evidence of toxicity.

Effect of Strontium on the Bone

The toxicity of strontium with respect to effects upon the bone (strontium rickets or rachitic changes) is influenced by the levels of calcium in the diet. In weaning Sprague-Dawley rats, rachitic changes were observed from a diet containing 0.95% strontium (950 mg/kg bodyweight/day) and calcium (0.69%) for 4 weeks (Engfeldt and Hjertquist, 1969). Raising dietary calcium to 1.6% eliminated the rachitic changes.

As their bones are actively growing, young animals are more sensitive than adult animals to excessive strontium intakes (Storey, 1961, 1962). Both young and adult rats of both sexes were provided a diet containing 1.8% strontium as strontium carbonate (doses estimated to be 390 mg/kg body weight/day in young rats and 190 mg/kg body weight/day in adult rats). The exposure continued for up to 7 months with several interim sacrifices. After only 3 weeks of exposure, the young rats exhibited a "rachitic gait" with the most obvious changes occurring in the distal end of the femur and the proximal end of the tibia. The epiphyseal plate was reported to be "grossly widened" and the "metaphysis was a mass of soft white tissue". Conversely, it was 3 months before any change was observed in the adult rats, this being the appearance in the upper tibial metaphysis of fine transverse bands of osteoid trabeculae in which calcified bone or cartilage is absent (Storey, 1962).

In addition to the information available in rats, Marie and Hott (1986) studied the effects of strontium on weanling mice. Eleven male C57BL/6J mice were provided with drinking water containing 0.27% strontium chloride from 21 to 50 days of age (approximately 240 mg Sr/kg body weight/day). Another group of 13 untreated mice served as controls. No significant effects were observed in bone formation parameters except an increase in the osteoid surface and a decrease in the number of osteoclasts involved in bone resorption. No effect was seen on total calcified bone volume.

Patients receiving strontium supplements showed an increase in bone density in areas of lesions due to metastatic cancer, and strontium may have a role in mineralising osteopenic areas and relieving bone pain. Moderate doses of strontium (200-300 mg/day) do not adversely effect either serum calcium levels or bone metabolism as long as a normal dietary intake of calcium is maintained. Higher levels of strontium (1-3% in the diet in experimental animals) are disadvantageous leading to bone changes resulting from a decrease in bone calcium. Strontium may also inhibit resorption of bone by covering endosteal bone surfaces with an osteoid that is resistant to absorption (Skoryna, 1981).

Previous evaluations

The USA Environmental Protection Agency's evaluation of the oral toxicity of strontium (EPA, 1996) derived an oral reference dose for man of 0.6 mg/kg body weight/day (equivalent to 36 mg/day, for a 60 kg adult). As pertinent human data were not available, the reference dose was based on the NOAEL level for strontium carbonate in young rats of 190 mg strontium/kg body weight/day (Storey, 1961). This was the lowest NOAEL derived from this and other oral toxicity studies (Marie *et al.*, 1985; Skoryna, 1981). A total uncertainty factor of 300 (comprising factors of 10 to account for interspecies variation, 3 to account for susceptible groups and 10 to account for data inadequacies) applied to the NOAEL results in the reference dose of 0.6 mg/kg body weight/day. (Note: a 90-day dietary toxicity study of strontium chloride hexahydrate in rats (Kroes *et al.,* 1977) showing a much lower NOAEL of 300 ppm (equivalent to about 15 mgSr/kg body weight/day) is not considered in the EPA evaluation.)

Previous COT evaluations

In 1998, the COT evaluated strontium in food when the results of the 1994 TDS were considered (COT, 1998). The estimated population dietary exposure to strontium from the 1994 TDS was 1.3 mg/day, approximately 21.7 µg/kg body weight/day, for a 60 kg adult. Highest levels of strontium were reported in nuts (8.67 mg/kg). Bread contained 3.7 mg/kg, fish 3.6 mg/kg, green vegetables 1.6 mg/kg and cereals 1.3 mg/kg - other foodstuffs contained values below 1 mg/kg. High-level consumers of marine fish and shellfish might obtain up to 0.78 mg/day from this source alone (MAFF, 1997). Older estimates of dietary strontium intake range from 0.98-2.2 mg/day for adults, with milk providing about one-third of this (Snyder *et al.,* 1975). The Committee concluded that current dietary levels of exposure to strontium were unlikely to be of concern for human health but they acknowledged that intakes

were likely to be an overestimate and that the data available on strontium were limited and did not allow a full assessment of it's safety. (COT, 1998).

References

Beliles RP (1994). The Metals - section on Strontium in Patty's Industrial Hygiene and Toxicology ed Clayton GD and Clayton FE. p 2216-2227.

Comar CL and Wasserman RH (1964). Strontium In: Mineral Metabolism ed Comar CL and Bronner F Vol IIA Academic Press, London, p523-572.

COT (1998). Statement on the results of multielement surveys in various items of the diet. COT Statement 1998. Annual Report 1998 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Food, Consumer Products and the Environment.

El Solh N and Rousselet RH (1981). Effects of stable strontium administration on calcium metabolism with particular reference to low-calcium diet. In: Handbook of Stable Strontium, SC Skoryna, Ed. Plenum Press, New York p. 515-544.

Engfeldt S and Hjertquist SO (1969). The effect of strontium administration on bones and teeth of rats maintained on diets with different calcium contents. Virchows Arch. Abt. A Path. Anat. 346 p330-344.

EPA (1996). US Environmental Protection Agency. Integrated Risk Information System (IRIS) database. Entry for Strontium (updated 9/9/96).

Ghosh S, Talukder G, and Sharma A (1990). Biol Trace Elem. Res. 25 p51-56.

Kanematsu N Hara U and Kara T 1980. Rec Assay and Mutagenicity Studies on Metal Compounds. Mut Res 77 p109-116.

Kroes R, Den Tonkelaar EM, Minderhoud A *et al* (1977). Short -term toxicity of strontium chloride in rats. Toxicology 7 p11-21.

Lansdown ABG, Longland RC and Grasso P (1972.). Reduced foetal calcium without skeletal malformation in rats following high maternal doses of a strontium salt. Experientia 28(5) p558-560.

Leeuwenkamp OR, van der Vijgh WJF, Husken DCP, Lips P and Netelenbos JC. (1990). Human pharmacokinetics of orally administered strontium. Calcif. Tissue Int. 47: 136-141.

MAFF (1997). 1994 Total Diet Study: Metals and Other Elements Food Surveillance Information Sheet No 131

Marie PJ, Garba MT, Hott M and Miravet L (1985). Effect of low doses of stable Sr on bone metabolism in rats. Miner. Electrolyte Metab. 11 p5-13.

Marie PJ and Hott M (1986). Short-term Effects of Fluoride and Strontium on Bone Formation and Resorption in the Mouse. Metab. 35(6) p547-551.

Niyogi SK and Feldman RP (1981). Nucleic Acids Res 9 p2615.

Sax (1996). Sax's Dangerous Properties of Industrial Materials, 9th Ed

Lewis RJ Snr Ed. Volume II.

Skoryna SC and Fuskova M (1981). Effects of Stable Strontium Supplementation. In: Handbook of Stable Strontium, SC Skoryna, Ed. Plenum Press. p 593-613.

Skoryna, SC (1984). Metabolic Aspects of the Pharmacologic use of Trace Elements in Human Subjects with Specific Reference to Stable Strontium. In: Trace Substances in Environmental Health - XVIII, DD Hemphill, Ed. University of Missouri. p. 3-20.

Snyder WS, Cook MJ, Nasset ES, Karhausen LR, Howells GP and Tipton IH, (1975). International Commission on Radiological Protection (IRCP). Report of the Task Group on Reference Man, New York. ICRP Publ. 23.

Storey E (1961). Strontium "rickets" bone calcium and strontium changes. Austral. Ann. Med. 10 p213-222.

Storey E (1962). Intermittent bone changes and multiple cartilage defects in chronic strontium rickets in rats. J Bone Joint Surg. 44B(1) p194-208.

Sugihira N and Suzuki KT (1991). Discrimination between strontium and calcium in suckling rats. Biol Trace Elem Res 29 p1-10.

Tipton IH and Cook MJ (1963). Trace elements in human tissues; II. Adult subjects from the United States. Health Phys. 9, p103.

US EPA (1990). Drinking Water Criteria Document for Stable Strontium. Prepared by the Office of Health and Environmental Assessment, Environmental Criteria and Assessment Office, Cincinnati, OH for the Office of Drinking Water, Washington, DC.

US EPA (1992). Health and Environmental Effects Document for Stable Strontium. Prepared by the Office of Health and Environmental Assessment, Environmental Criteria and Assessment Office, Cincinnati, OH for the Office of Solid Waste and Emergency Response, Washington, DC.

Zakour RA et al (1981). J Can Res Clin Oncol 99 p187.

22. Thallium (Tl)

The key source of information for this summary was Environmental Health Criteria 182 (WHO, 1996).

Chemistry and occurrence

Elemental thallium is a soft and malleable metal with a bluish-white colour. When exposed to humid air or water, thallium is oxidized rapidly on the surface or the hydroxide is formed, respectively. Thallium has two important oxidation states, thallium(I) and thallium(III). Monovalent (thallous) compounds behave like alkali metals, e.g. potassium, whereas the trivalent (thallic) compounds are less basic, resembling aluminium. In contrast to inorganic compounds in which the thallium(I) ion is more stable in aqueous solutions than the thallium(III) ion, the latter is more stable in organic compounds.

Thallium is present in the environment as a result of natural processes and from man-made sources. It is ubiquitous in nature and occurs especially in sulfide ores of various heavy metals, but normally in low concentrations. There are only a few areas with a naturally very high thallium concentration.

Thallium is produced industrially only in small quantities (the worldwide industrial consumption in 1991 was 10-15 tonnes/year). Thallium and its compounds have a wide variety of industrial uses. Its uses as a depilatory agent for humans and as a rodenticide and insecticide are now severely restricted. The main uses are in the electrical and electronic industries and in the production of special glasses. Another important field of application is the use of radioisotopes in medicine for scintigraphy and the diagnosis of melanoma and the use of arylthallium(III) compounds in biochemistry (WHO, 1996).

Food is considered to contain < 1 mg/kg dry weight thallium. In areas not contaminated by thallium, concentrations in air are usually < 1ng/m^3. Mean urinary thallium concentrations in unexposed populations are 0.3 - 0.4 µg/litre (WHO, 1996).

A MAFF survey of vegetable consumption from point sources indicated that 9 µg thallium/day may be ingested (comprising 7 µg/day from a high vegetable intake and 2 µg/day as the mean value from the dietary study; MAFF, 1997b).

Absorption and elimination

Thallium is rapidly absorbed via the gastro-intestinal and respiratory tracts and is also taken up through the skin. It is rapidly distributed to all organs and passes the placental and blood brain barriers in humans. Because of its rapid accumulation in cells, concentrations of thallium in whole blood do not reflect the levels in tissues (WHO, 1996). The total amount of thallium in the body has been estimated to be 100 µg for a 75 kg body weight in an unexposed population (WHO, 1996).

The distribution of thallium in the body in man after either acute poisoning episodes or from chronic intake shows no consistent trend. In acute poisoning episodes, endocrine glands, kidney, liver and intestines showed the highest concentrations (WHO, 1996). Grey matter of the brain has been shown to contain levels 3 times higher than white matter (Cavanagh *et al.,* 1974), and areas of the brain rich in neurons tend to accumulate thallium (Davis *et al.,* 1981).

The excretion of thallium in humans differs substantially from that in laboratory animals, both in the rate of excretion (lower in man) and in the relative contribution to different routes of excretion. In man, renal excretion is much more important than in animals, although data are incomplete. Following therapeutic administration of radioactive thallium (2.3 mg), 11% of the administered dose was excreted in the urine after 72 hours with 0.5% being eliminated through the gastro-intestinal tract (WHO, 1996). According to this study the excreted amounts can be calculated to be: urine, 73%; gastro-intestinal, 3.7%; hair, 19.5%; skin and sweat, 3.7%.

Thallium has a short biological half-life of about 10 days in man. The urinary excretion value can be taken as an indication of the daily absorbed dose of thallium (WHO, 1996).

A population-based study of unexposed healthy subjects living in northern Italy was performed with the aim of determining trace element concentrations, including thallium, in blood, serum or plasma, and urine, in which the collection, handling and analysis of the samples was carried out under rigorous standardised protocols (496 subjects). The mean urinary thallium concentration in this unexposed population was 0.42 ± 0.09 µg/L (range 0.07 - 0.7 µg/L). Other carefully controlled studies in population samples showed similar urinary concentrations, e.g. 0.4 ± 0.2 µg/L and 0.3 ± 0.2 µg/L in rural and urban population samples, respectively, and 0.3 ± 0.14 µg/L in a sample of 149 subjects. This gives credence to a mean value of 0.3 - 0.4 µg/L for urinary thallium concentrations in an unexposed population. In all three studies, involving a total of 686 subjects, the range of urinary thallium concentrations was 0.06 - 1.2 µg/L (WHO, 1996).

By contrast, in a population sample living in the vicinity of thallium emissions into the atmosphere, the mean urinary thallium concentration was 5.2 µg/L ± 8.3 µg/L (range 0.1 - 76.5 µg/L). In a limited study on cement plant workers with thallium exposure, five out of 36 workers showed urinary thallium levels above 5 µg/L (WHO, 1996), but there was no correlation between urinary levels and neurological effects.

The normal daily elimination in humans is estimated to be in the order of 1.64 µg (urine 1.2 µg/day, hair 0.32 µg/day, faeces 0.06 µg/day, skin and sweat 0.06 µg/day) (USEPA, 1980).

Toxicity in animals

In animal studies oral LD_{50} values of thallium (I) and thallium (III) inorganic compounds are generally < 100 mg/kg body weight with an oral LD_{50} of 15

mg/kg body weight being reported for thallium (III) sulphate in the rat and a lowest oral lethal dose of 5.6 mg/kg body weight for thallium (III) oxide in the guinea pig (WHO, 1996).

No lifetime studies of thallium administration have been conducted in laboratory animals. Three studies of intermediate duration by the oral route are described below. A no-observed-effect level of 0.25 mg thallium sulphate/kg body weight/day has been determined from a 90-day gavage study in rats (USEPA, 1986), although the 1996 IPCS evaluation considered that a NOAEL could not be determined from this work and considered that effects were seen at 0.01 mg/kg body weight/day (WHO, 1996). Much lower doses (0.05 - 0.5 µg thallium carbonate/kg body weight/day over 8 months in male rats) have been reported to cause dominant lethal effects on subsequent mating (Zasukhina *et al.,* 1983).

In a study of weanling albino rats fed *ad libitum* for 1 month on a diet containing 2, 10, 50, 100, 500, or 5000 mg thallium (I) acetate per kg diet (5 rats/group), dietary levels of 2 and 10 mg/kg diet caused no effects on growth or survival within the feeding period, whereas the other concentrations resulted in mortalities of 60 to 100% within 10 days. When rats were fed for 15 weeks with 5, 15, 30 or 50 mg thallium acetate/kg diet (5 males and 5 females/group), the two lowest doses did not affect growth of males or females. The 15 and 30 mg/kg doses caused hair loss starting after 2 weeks, and after 15 weeks the rats were almost free of hair. Within the fourth and eighth week and after intakes of 30 mg/kg diet, 80% of males and 60% of females died. At 50 mg/kg all males died within 2 weeks and all females within 8 weeks. No specific pathological alterations were found in any organ. A dose of 30 mg/kg diet resulted in moderate growth depression in males but not in females, while in both sexes increased mortality was observed. For thallium (III) oxide the effective concentrations were similar, and males also reacted more sensitively than females. No specific pathological alterations were found in any organ except the skin, where atrophy of hair follicles and sebaceous glands were seen at both higher dose levels. The exact concentration of thallium ingested by the rats could not be determined but was estimated to be in the range of 1 to 3 mg thallium acetate/kg body weight per day for the diet containing 15 mg thallium acetate/kg food (Downs *et al.*, 1960). Over the 15 week study 5 mg thallium acetate/kg diet could be considered to be a NOAEL, and would equate to 0.25 mg thallium /kg body weight per day.

USEPA (1986) conducted a 90-day study in which male and female Sprague-Dawley rats (20 of each sex per group) were administered aqueous thallium sulphate by gavage at doses of 0.01, 0.05 or 0.25 mg/kg body weight per day. Both untreated controls and vehicle (water) treated controls were included. Clinical observations were recorded daily and neurotoxicological examinations were performed 3 times per week on selected animals. No significant differences were seen in any group for body or organ weights. Moderate changes were reported for blood chemistry parameters. In both males and females, small increases were seen for serum glutamic-oxaloacetic transaminase (SGOT), lactic acid dehydrogenase (LDH) and sodium levels, with statistical significance at many points. At the lowest dose, statistically

significant changes were seen in male rats for all three of these parameters, but only when compared with untreated controls. Higher doses resulted in statistical significance when compared with vehicle controls. Similar patterns were seen in female rats. In reviewing these data (USEPA, 1990) it was considered that the gross pathological finding was alopecia in female rats but there were no histopathological alterations. Based on these results, 0.25 mg thallium sulphate/kg body weight/day was considered as an NOAEL (equivalent to 0.2 mg Tl/kg body weight/day). However, the US EPA commented that confidence in this critical study is rated low because of uncertainties in the results (i.e. vehicle versus untreated control differences) and because other studies show effects at doses slightly higher than the NOAEL.

In a study with 80 female Sprague-Dawley rats, thallium sulphate was given via the drinking water (10 mg thallium/litre) over a period of 40 weeks and with a total intake of about 80 mg thallium/rat, approximating to 1.4 mg Tl/kg body weight/day (Manzo *et al.*, 1983). Starting from day 40, the number of rats showing mild or severe cutaneous disorders increased strongly. After 40 days and a total ingestion of about 10 mg thallium/rat, lethality amounted to 15%, and the surviving rats showed no electrophysiological abnormalities. After 240 to 280 days and the ingestion of about 70 to 80 mg/rat, only an additional 6% of the rats died. About two-thirds of the rats showed electrophysiological effects, reduced motor and sensory action potentials and histopathological changes in the nervous system.

In the review by IPCS (WHO, 1996) no mutagenic effects of thallium were reported in two separate studies in the Ames test (strains TA98, TA100, TA1535, TA1537 and TA1538). Negative results were also obtained in the reverse mutation assay in yeast and negative results were also reported on cell division in yeast and *E. coli* (reviewed in USEPA, 1990).

Positive results were obtained for thallium nitrate in the Rec assay using *Bacillus subtilis* strains H17 and M25. Cytotoxic levels of thallium acetate (1000 µg/mL) caused depression of DNA synthesis in Chinese hamster ovary cells (reviewed in USEPA, 1990).

In a dominant lethal study, increased dominant lethality was observed after exposure of male rats to 0.5 or 0.05 µg/kg body weight/day of thallium carbonate orally (equivalent to 0.43 or 0.043 µg Tl/kg body weight/day) over a period of 8 months before mating. There was a tendency towards an increase in embryonic mortality at these doses. Effects (increased resorptions) were also seen at the low dose of 0.005 µg Tl_2CO_3/kg body weight/day, equivalent to 0.0043 µg Tl /kg body weight/day (Zasukhina *et al.,* 1983)

Leonard and Gerber (1997) published a review of mutagenic, carcinogenic and teratogenic effects of thallium compounds. The authors concluded that while data are sparse, available information did not indicate that thallium could be mutagenic or carcinogenic, although it has teratogenic properties.

The review by Leonard and Gerber (1997) concluded that thallium has some teratogenic properties, especially on cartilage and bone formation, although it

does seem to be more prominent in chicks than in mammals. Effects were noted in chick embryos when administered at doses of 0.4 to 1.2 mg thallium sulphate (approximately 0.32 - 1.0 mg Tl/kg body weight) on days 4 - 11 of incubation, but 3 -6 mg/kg body weight of thallium chloride or thallium acetate (approximately 2.5 – 5 mg Tl/kg body weight) administered during organogenesis to female NMRI mice or Wistar rats produced no increase in malformations. These differences could be due to differences in placental transfer.

Toxicity in humans

The symptomatology of thallium poisoning is usually non specific because of the multi-target organ toxicity involved. Thallium has affinity for sulphydryl groups of mitochondrial membranes and therefore inhibits many enzyme systems. By exchanging with potassium, thallium also interferes with oxidative phosphorylation by inhibition of Na/K ATPase. As it behaves in a similar way to potassium and thus readily enters myocardial cells, thallium 201 is widely utilised for myocardial imaging. Case studies of thallotoxicosis estimate that 1g of soluble thallium salt (about 15 mg/kg body weight) can be considered to be a lethal dose for adults in chronic cases of human poisoning (Beliles, 1994).

The triad of gastroenteritis, polyneuropathy and alopecia is regarded as the classic syndrome of thallium poisoning, but in some cases gastroenteritis and alopecia are not observed. Other symptoms also develop in varying sequences. Both lethal and sublethal doses give rise to most of the symptoms, but these same symptoms vary in intensity and time, probably in a dose-dependent way (WHO, 1996).

Cases of acute intoxication by thallium salts in humans, which always cause severe symptoms, have been reported for single or multiple oral doses of the order of 100 mg or more for adults, i.e. 1.5 to 2 mg/kg body weight (WHO, 1996).

Studies of long-term exposure to thallium resulting in chronic poisoning show considerable variability in symptoms which are in general milder than in cases of acute intoxication. No indication of dose is provided in these reports. Depending on the level of exposure, a relatively long latent period (several weeks) may be followed by few relatively mild symptoms. Peripheral sensory disturbances, mental aberrations, loss of weight and sleeplessness seem to be the most common. In more severe cases, disturbances of vision, pain without marked polyneuritis, and loss of hair were reported. Later, severe polyneuritis may develop, with an inability to walk, blindness and pronounced cachexia. Cardiac disorders include hypertension, irregular pulse and angina-like pain. Renal dysfunction is indicated by albuminuria and haematuria. Other symptoms are gastric anacidity, lack of appetite, loss of weight, endocrine disorders, psychoses and encephalitis. Complete rehabilitation takes months and can be interrupted by relapses, probably caused by remobilisation of thallium from tissue depots (WHO, 1996).

On the basis of acute toxicity values in animals and known lethal doses in man, it appears that humans may be more sensitive than laboratory rodents to the toxic effects of thallium.

The WHO Environmental Health Criteria report on thallium considered that urinary excretion of thallium is the most reliable indicator of human exposure as thallium is well and rapidly absorbed and is rapidly excreted primarily via the urine. They concluded that exposure to thallium causing urinary concentrations below 5 µg/L "are unlikely to cause adverse human health effects" although it is not clear on how this figure was reached and whether it related to acute or chronic exposure (WHO, 1996). This was estimated to correspond to an oral intake of about 11 µg thallium/day in a soluble (i.e. bioavailable) form.

Previous evaluations

Human oral reference doses for thallium salts have been set by the EPA (US EPA 1990). These are based on an NOAEL for thallium sulphate from a 90-day rat study of 0.25 mg/kg body weight/day (equivalent to 0.2 mg Tl/kg body weight/day). Applying uncertainty factors of 10 for extrapolation from subchronic to chronic data, 10 for intraspecies extrapolation, 10 for interspecies variability and 3 to account for a lack of reproductive or chronic data (total uncertainty factor of 3000) gives a human oral reference dose of approximately 0.07 µg Tl/kg body weight/day (or 4µg Tl/day). The IPCS review on the other hand considered adverse effects in this critical study were evident at the lowest dose tested i.e. 0.01 mg thallium sulphate/kg body weight/day, equivalent to about 8 µg Tl/kg body weight/day (WHO, 1996).

The IPCS Task Group (WHO, 1996) considered that exposures causing urinary thallium concentrations below 5 µg/litre are unlikely to cause adverse health effects. In the range of 5 - 500 µg/litre the magnitude of the risk and severity of adverse effects are uncertain, while exposures giving values over 500 µg/litre have been associated with clinical poisoning. The estimated daily oral intake corresponding to a urinary thallium concentration of 5 µg/litre is approximately 11 µg/day or 0.18µg/kg bw/day in an average 60kg adult.

Previous COT evaluations

In 1998, the COT evaluated thallium in food when the results of the 1994 TDS were considered (COT, 1998). The estimated population dietary exposure to thallium from the 1994 TDS was 0.002 mg/day, approximately 0.03 µg/kg body weight/day for a 60 kg adult. Highest levels of 3 µg thallium/kg were found in green vegetables, carcase meat and in offal. Marine fish and shellfish were considered unlikely to contribute more than 0.1 µg/day (MAFF, 1997a). The Committee concluded that there is no evidence that current dietary intake of thallium by the UK population may be harmful to health (COT, 1998).

References

Beliles RP (1994). The Metals - Thallium in Patty's Industrial Hygiene and Toxicology, Volume II, Part C, pp 2234-2249.

Cavanagh J B, Fuller N H, Johnson H R M and Rudge P (1974). The Effects of Thallium Salts with Particular Reference to Nervous System Changes. Q J Med New Ser 43, pp 293 – 319.

COT (1998). Statement on the results of multielement surveys in various items of the diet. COT Statement 1998. Annual Report 1998 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Food, Consumer Products and the Environment.

Davis L E, Standefer J C, Kornfeld M, Abercrombie D M and Butler C (1981). Acute Thallium Poisoning: Toxicological and Morphological Studies of the Nervous System. Ann Neurol 10, pp 38 – 44.

Downs W L, Scott J K, Steadman L J and Maynard E A (1960). Acute and Subacute Toxicity of Thallium Compounds. Am Ind Hyg Assoc J 21, pp 399 – 406.

Leonard A and Gerber G B (1997). Mutagenicity, Carcinogenicity and Teratogenicity of Thallium compounds. Mutation Research 387, pp 47 – 53.

Manzo L, Scebi R, Moglia A, Poggi P, Alfonsi E, Dietra R, Mousty F and Sabbioni E (1983). Long Term Toxicity of Thallium in the Rat. In; Brown S S and Savory J ed. Chemical Toxicology and Clinical Chemistry of Metals. Proc. 2nd Inter. Conf. Montreal, Canada, Academic Press, pp 401 – 405.

MAFF (1997a). 1994 Total Diet Study: Metals and Other Elements. Food Surveillance Information Sheet No 131.

MAFF (1997b). 1997 MAFF Multi element Survey of Foods from around point sources.

MAFF (1997c). 1997 MAFF Multi-element Study of Dietary Supplements.

USEPA (1980). Ambient Water Quality Criteria for Thallium. USEPA-440/5-80-074, Washington.

USEPA (1986). Subchronic (90-day) Toxicity of Thallium (I) Sulfate (CAS No 5446-18-6) In Sprague Dawley Rats: Interim Report.

USEPA (1990). Integrated Risk Information System Database (IRIS). Entries for Thallium sulfate and for other soluble salts of thallium. Washington DC USEPA, Office of Solid Wastes, Washington.

WHO (1996). Thallium - International Programme on Chemical Safety (IPCS). Environmental Health Criteria Document No. 182, WHO Geneva

Zasukhina GD, Vasilyeva IM, Sdirkova NI et al (1983). Mutagenic effect of thallium and mercury salts on rodent cells with different repair activities. Mut Res 124 pp163 – 173.

23. Tin (Sn)

The key sources of information for this summary were the JECFA monographs on tin (WHO, 1988, 2001 and 2006) and the EVM review (EVM, 2003). A further report (WHO, 2005) was also reviewed but provided no additional data.

Chemistry and occurrence

Tin is rarely found as the metallic element in nature and is usually found combined with other substances, most commonly as the dioxide (SnO_2). It has oxidation states of II and IV and the corresponding oxides of these states are amphoteric. Tin also forms organic compounds such as tri-butyl tin, methyltin and dimethyltin. The main form of tin found in food is metallic or inorganic tin, which has leeched from food packaging. Organic forms of tin such as tri-butyl tin are found mainly in shellfish where they accumulate. Tin in food is generally considered to be a contaminant. This summary will focus on the toxicity of inorganic tin.

Tin has not been shown to be an essential element in the diet.

Absorption and elimination

The absorption of tin is low and depends on the oxidation state. For example, following a 24-hour fast, 200-225 g rats were given a single 20 mg tin/kg body weight dose of $Sn(^{+2})$-citrate, -fluoride or –pyrophosphate, or $Sn(^{+4})$-citrate or -fluoride. Changing the anion complement from the citrate to the fluoride did not alter the biological fate of either valency form, while approximately 2.85% and 0.64%, respectively, of the Sn^{+2} and Sn^{+4} were absorbed. Therefore Sn^{2+} appears to be more bioavailable than Sn^{4+} (WHO, 1988).

The tissues and organs that accumulate the highest concentrations of tin are bone, lymph nodes, liver, lung, ovary, testis and kidney. No metabolism of tin has been demonstrated.

The majority of ingested inorganic tin is unabsorbed and is eliminated in the faeces (about 95-99% of ingested dose in animal studies), with the remainder excreted in the urine (WHO, 1988, 2001).

Toxicity in animals

In studies of the acute oral administration of tin to animals, cats have shown a vomiting response similar to that of humans (Cheftel, 1967; Omori *et al.,* 1973).

Most studies of the subchronic toxicity of tin have been carried out in rats, a species that lacks an emetic reflex. Reductions in body weight gain, reduced

appetite and reduced feed conversion efficiency have been reported. These, together with abdominal distension and at autopsy, distended caeca, reddening of the mucosal surface of the stomach and histopathologic changes in the gastrointestinal tract (from the stomach to the ileum), are indicative of gastrointestinal effects in the rat (De Groot *et al*., 1973b). Haematological changes included reduced haematocrit and haemoglobin, which are indicative of anaemia, although no biologically significant changes to leucocytes occurred. These effects may result from disturbance to the pathways of haem biosynthesis and degradation, as tin reduces the activity of the enzyme 5-aminolaevulinate dehydratase and increases haem oxidase (WHO, 1988).

Pathological changes to the pancreas, including atrophy with single-cell necrosis and destruction of complete acini, have been reported in rats fed stannous chloride at 500 mg/kg diet (de Groot, *et al.,* 1973a; Dreef-van der Meulen, *et al.,* 1974). Doses of 0.4 and 0.8% of the diet for 100 days caused severe gastrointestinal irritation and caused a decrease in haematocrit, haemoglobin and serum iron in rats (Fritsch *et al*., 1978). Pathological changes to the liver, spleen, kidneys, adrenals and testicles have also been reported. Various authors have investigated the effects of tin, given orally or parenterally, on calcium balance and bone metabolism in experimental animals. Some changes were detected at low doses of tin. The results of reproductive and developmental toxicity studies were negative (WHO, 1988).

The available data on chronic administration of tin are limited but indicate that doses of up to 100 mg/kg body weight/day as stannous chloride are well-tolerated and do not affect survival (WHO, 1988, 2001).

The results of long-term studies of the carcinogenicity of various tin compounds in animals have been negative. The conformation of DNA is disrupted by *in vitro* exposure to stannous chloride, but it has been suggested that this is a result of the generation of a reactive oxygen species under the conditions used (WHO, 1988, 2001).

Toxicity in humans

The acute toxicity of inorganic tin in humans results from irritation of the mucosa of the gastrointestinal tract, which may lead to vomiting, diarrhoea, anorexia, depression, ataxia, and muscle weakness (WHO, 2001).

Reports of acute poisoning have been associated with high concentrations of tin in drinks or solid foods. Subjects presented with gastrointestinal effects (abdominal cramps, nausea, vomiting, diarrhoea), and headache and chills within 1-2 hours of consuming the implicated foodstuff, and recovered within 1-2 days. However, there is little consistency in the reports in terms of the nature of the foodstuff, or the concentration and chemical form of the tin, all of which may influence human toxicity. From the limited human data available it could be concluded that tin concentrations as low as 150 µg/g in one incident involving canned beverages (Barker & Runte, 1972) and 250 µg/g in other

canned foods may produce acute manifestations of gastric irritation in certain individuals. However, it can also be noted that some canned products containing levels up to 700 µg/g of tin produced no detectable toxic effects (WHO, 1988, 2001).

In 2006, JECFA reviewed a new study that showed tomato juice freshly spiked with tin chloride (≥161 mg/kg) caused gastrointestinal disorders in humans in a concentration-related manner; whereas volunteers receiving tomato soup contaminated with tin (<0.5, 201 and 267 mg/kg) did not experience an increased incidence of adverse effects (Boogaard *et al.*, 2003). This study supports the view that both complexation and adsorption of tin onto solid matter reduce its irritant effect on the gastrointestinal tract (WHO, 2006).

Previous evaluations

JECFA established a PTWI for tin of 14 mg/kg body weight/week (WHO, 2001). This was apparently based on the threshold concentration for acute manifestation of gastric irritancy, 200 mg/kg in food. From this a PMTDI of 2 mg/kg body weight/day was established. JECFA also concluded that tin concentrations as low as 150 mg/kg in canned beverages and 250 mg/kg in other canned foods may produce acute manifestations of gastric irritation in certain individuals. The legal limit for tin in foods (200 mg/kg) may not therefore be sufficiently protective. In 2001 the Scientific Committee on Food (SCF) published its opinion of the Acute Risks Posed by Tin in Canned Foods. In this opinion the SCF agreed with the evaluation made by JECFA.

The EVM was unable to establish a safe upper level for tin in its report (EVM, 2003). The EVM noted that in a subchronic study in rats, intakes of 240 mg tin/day produced pancreatic atrophy, and that anaemia and changes to liver cells had been observed in another study in rats, with a NOAEL of 22-33 mg/kg body weight/day. Applying uncertainty factors of 10 for interspecies variability and 10 for intraspecies variability, the EVM recommended, for guidance purposes only, that a daily intake of 0.22 mg/kg body weight/day (equivalent to 13 mg/day in a 60 kg adult) would not be expected to produce adverse effects in humans (EVM, 2003).

In 2005, the EFSA Scientific Panel on Dietetic Products, Nutrition and Allergy concluded that there were insufficient data available to derive a tolerable upper intake level for tin (EFSA, 2006).

More recently, JECFA evaluated the acute toxicity of tin in humans after consumption of foods containing high concentrations of inorganic tin (WHO, 2006). The Committee concluded that it is the concentration of tin in foods, rather than the dose ingested on a body weight basis, that is critical to the development of gastrointestinal effects. The Committee noted that the basis for this previously established PTWI was unclear and that therefore it would be desirable to reassess the toxicokinetics and effects of inorganic tin after

long-term exposure to dietary doses of inorganic tin at concentrations that did not elicit acute effects (WHO, 2006).

Previous COT evaluations

In 2003 the COT evaluated tin in food when the results of the 2000 TDS were considered (COT, 2003). The estimated population dietary exposure to tin from the 2000 TDS was 1.4 mg/day, approximately 23 µg/kg body weight/day for a 60 kg adult. The Committee concluded that current dietary exposures to tin were unlikely to be of any toxicological concern for consumers (COT, 2003).

References

Barker, W.H. and Runte, V. (1972). Tomato juice-associated gastro-enteritis, Washington and Oregon, 1969. *American Journal of Epidemiology,* **96:** 219-226.

Boogaard, P.J., Boisset, M., Blunden, S., Davies, S., Ong, T.J. and Taverne, J.P. (2003). Comparative assessment of gastrointestinal irritant potency in man of tin (II) chloride and tin migrated from packaging. *Food Chem Toxicol.*, **41**, 1663-1670.

Cheftel, H. (1967) Tin in food. Joint FAO/WHO Food Standards Program: Fourth meeting of the Codex Committee on Food Additives, September 1967. Joint FAO/WHO Food Standards Branch (Codex Alimentarius), FAO, Rome

COT (2003). Statement on twelve metals and other elements in the 2000 Total Diet Study. COT Statement 2003/07. Annual Report 2003 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Foods, Consumer Products and the Environment.

de Groot, A.P., Feron, V.J. & Til, H.P. (1973a) Short-term toxicity studies on some salts and oxides of tin in rats, *Food Cosmet. Toxicol*., **11**, 19–30.

de Groot, A. P., Feron, V. J. and Til, H. P. (1973b). Subacute toxicity of inorganic tin as influenced by dietary levels of iron and copper. *Food and Cosmetics Toxicology*, **11:** 955-962.

Dreef-van der Meulen, H. C., Feron, V. J. and Til, H. P. (1974). Pancreatic atrophy and other pathological changes in rats following the feeding of stannous chloride. *Pathol Eur*, **9**, 185-192.

EFSA (2006) Tolerable Upper Intake Levels for Vitamins and Minerals. February 2006

EVM (2003). Safe upper levels for vitamins and minerals. Report of the Expert Group on Vitamins and Minerals.

Fritsch, P., de Saint Blanquat, G. and Derache, R. (1977). Etude nutritionnelle et toxicologique, chez le rat, d'un contaminant alimentaire: l'etain [Nutritional and toxicological study of rats fed a diet containing tin]. *Toxicology*, **8:** 165-175.

Omori, Y. et al. (1973) Experimental studies on toxicity of tin in canned orange juice, *J. Food Hyg. Soc. Jpn*, **14:** 69-74.

WHO (1988). Safety Evaluation of certain food additives and contaminants. WHO Food Additives Series 24. Joint FAO/WHO Expert Committee on Food Additives.

WHO (2001). Safety Evaluation of certain food additives and contaminants. WHO Food Additives Series No. 46. Joint FAO/WHO Expert Committee on Food Additives.

WHO (2005). Concise International Chemical Assessment Document (CICAD) 65: Tin and inorganic tin compounds. World Health Organization, Geneva.

WHO (2006). Safety Evaluation of certain food additives and contaminants. WHO Food Additives Series No. 55. Joint FAO/WHO Expert Committee on Food Additives.

24. Zinc (Zn)

The key sources for information in this summary were the EVM report (EVM, 2003), the JECFA monologue (WHO, 1982) and Environmental Health Criteria 221 (WHO, 2001).

Chemistry and occurrence

Zinc is an essential element in the nutrition of man, animals and plants. It functions as an integral part of numerous enzymes. Because of its essentiality, zinc is present in all plant and animal tissues. The total body zinc for a 70 kg individual has been estimated to be 2.3 g. Protein foods are important dietary sources of zinc since they contain a large amount of enzymes that form complexes with zinc. Zinc is also present in the diet as inorganic salts such as the sulphate, oxide or gluconate. (WHO, 1973; WHO, 1982).

Absorption and elimination

Zinc, as contained in food and drink, is absorbed through the gut mucosa. Absorption of zinc salts from food is approximately 20-40%, being higher from flesh food but lower from cereals, where phytate content impairs absorption. The absorption of zinc salts depends on their solubility (EVM, 2003). Zinc is absorbed both by passive diffusion and an unknown membrane carrier process, which requires energy. The initial site of absorption is the stomach where absorption occurs within 15 minutes of ingestion; however, the major site is the second portion of duodenum. Absorption also occurs in other segments of both the small and large intestine (Henkin & Aamodt, 1975; Methfessel and Spencer, 1973 a,b).

Many substances can affect the absorption of zinc. Both phytate and soy protein inhibit the formation of the zinc-protein complex and as a result diminish the absorption of zinc (WHO, 1982). Other substances that adversely affect zinc absorption include cottonseed, peanut, safflower, calcium, phosphate, food and zinc itself (WHO, 1982). Iron and copper compete for absorption with zinc. The absorption of zinc has been shown to be enhanced by the presence of histidine, cysteine, methionine and ethylenediamine tetracetic acid. These are thought to act by promoting the formation of the low molecular weight organic zinc complex (WHO, 1982).

The major route of zinc elimination is via the faeces with minor amounts eliminated in the urine, semen and sweat. Administration of large amounts of zinc does not result in elevated tissue levels, since increasing the level of zinc in the diet results in decreased absorption (WHO, 1982).

Studies on retention patterns of intravenously administered ^{65}Zn, by normal human subjects, indicate that there is a 2-component exponential pattern of

retention. One is a rapid component which comprises 20% of the radio-labelled zinc with a biological half-life of 8 days, and the other is a slow component with a biological half-life of 300-500 days (WHO, 1982).

Toxicity in animals

Acute lethality values for various zinc compounds range from 0.25 g/kg body weight for zinc fluoride (guinea pigs) to 2.46 g/kg body weight for zinc acetate dihydrate (rats) (Stokinger, 1981).

Pancreatic lesions were reported in mice ingesting 38 mg Zn/kg body weight/day (as zinc sulphate) in drinking water for 14 months (Aughey *et al*., 1977).

Anaemia has been observed in mice, rats, ferrets, and sheep following oral exposures to zinc oxide, zinc oleate or zinc sulphate (Fox and Jacobs, 1986). The anaemia may have been due to intestinal haemorrhaging, or iron deficiency due to competition for absorption between iron and zinc.

Teratogenic effects have not been observed in animals dosed orally with zinc; however, high oral doses can affect reproduction and fetal growth (Ketcheson *et al.,* 1969; Schlicker and Cox, 1967, 1968). It was reported that these effects might have been associated with a zinc-induced anaemia as indicated by reduced haemoglobin and RBC levels.

Toxicity in humans

Gastrointestinal distress is a common symptom of acute oral exposure to zinc compounds (WHO, 1982), particularly when zinc salts of strong mineral acids are ingested (Stokinger, 1981). Severe toxic effects have also been reported in cases of ingestion of zinc chloride and zinc phosphide (Chobanian, 1981; Mack, 1989). The estimated fatal dose of zinc phosphide is 40 mg/kg body weight.

Consumption of zinc supplements by human volunteers has been reported to cause gastrointestinal effects, including cramping and nausea. This was particularly apparent when the supplements were taken with little or no food. These effects have occurred at doses from 1.57 to 2.24 mg/day (Samman and Roberts, 1987; Henkin *et al*., 1976).

There is limited evidence that the immune system may be affected by excessive intake of zinc. Chandra (1984) reported that zinc sulphate, in doses equivalent to 4.29 mg Zn/kg body weight/day for 6 weeks, impaired the immune system as measured by the mitogenic response of peripheral blood lymphocytes and the chemotactic and phagocytic responses of polymorphonuclear leukocytes.

Chronic oral exposures to zinc (doses of > 2 mg Zn/kg body weight/day for 11 months to 2 years) can result in sideroblastic or hypochromic microcytic anaemia, associated with hypoceruloplasminaemia, hypocupremia, and neutropenia (Prasad *et al*., 1978; Porter *et al*., 1977; Hoffman *et al*., 1988).

Excess levels of dietary zinc interfere with the gastrointestinal absorption of copper, potentially leading to secondary copper deficiency. Signs of this include decreased ESOD (erythrocyte superoxide dismutase) activity, increased LDL cholesterol and decreased HDL cholesterol, decreased glucose clearance and abnormal cardiac function. One of the most sensitive markers of this appears to be ESOD activity. Supplemental doses of 50 mg/day cause significant decreases in the activity of this enzyme (EVM, 2003).

Previous evaluations

In 1982, JECFA (WHO, 1982) proposed a PMTDI of 0.3-1.0 mg/kg body weight/day, corresponding to 18-60 mg/day for a 60 kg adult. This was based on clinical studies, where up to 600 mg of zinc sulphate (equivalent to 200 mg elemental zinc) was administered daily, in divided doses, for periods of several months without any reported adverse effects, including effects on blood counts and serum biochemistry.

The SCF set a tolerable upper intake level for zinc in 2002 of 25mg Zn/day (or 0.4mg/kg bw/day in a 60kg adult) based on a number of small scale studies in humans looking at effects on copper status (EFSA, 2006).

The EVM recommended a safe upper level of 25 mg/day supplemental zinc in its report (EVM, 2003). This was based on human supplementation studies that had indicated supplemental intakes of zinc of 50 mg/day and above were associated with reduced copper status, as indicated by reduced ESOD activity. An uncertainty factor of 2, rather than 3, was applied to extrapolate from a LOAEL to a NOAEL because the effects were considered to be small and inconsistent, resulting in the safe upper level for supplementation of 25 mg/day. This resulted in a total dietary intake of 42 mg/day (equivalent to 0.7 mg/kg body weight/day in a 60 kg adult) that would not be expected to result in any adverse effects (EVM, 2003).

The US EPA derived an oral reference dose for zinc 0.3 mg/kg body weight/day. This was based on an average LOAEL of 0.91 mg/kg body weight/day from four clinical studies, all of which identified similar effects (decreases in erythrocyte copper and ESOD activity) at similar dose levels, in a variety of human subject groups (postmenopausal females, adult males and females). An uncertainty factor of 3 was applied to the LOAEL to allow for variability in susceptibility in human populations (US EPA, 2005).

Previous COT evaluations

In 2003 the COT evaluated zinc in food when the results of the 2000 TDS were considered (COT, 2003). The estimated population dietary exposure to zinc from the 2000 TDS was 8.4 mg/day, approximately 120 µg/kg body weight/day for a 70 kg adult. The Committee concluded that current dietary exposures to zinc are unlikely to be of any toxicological concern for consumers (COT, 2003).

References

Aughey, E., Grant, L., Furman, B.L., Dryden, W.F. (1977). The effects of oral zinc supplementation in the mouse. *J. Comp. Pathol.* **87:** 1-14

Chandra, R.K. (1984). Excessive intake of zinc impairs immune response. *J. Amer. Med. Assoc.* **252:** 1443-1446.

Chobanian, S.J. (1981). Accidental ingestion of liquid zinc chloride: local and systemic effects. *Ann. Emerg. Med.* **10:** 91-93.

COT (2003). Statement on twelve metals and other elements in the 2000 Total Diet Study. COT Statement 2003/07. Annual Report 2003 Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Foods, Consumer Products and the Environment.

EFSA (2006) Tolerable Upper Intake Levels for Vitamins and Minerals. February 2006

EVM (2003). Safe upper levels for vitamins and minerals. Report of the Expert Group on Vitamins and Minerals.

Fox, M.R.S. and Jacobs, R.M. (1986). Zinc: essentiality, function, effects of deficiency, and requirementsy. In: Metal Ions in Biological Systems, H. Sigel, ed. Marcel Dekker, Inc., New York. pp. 214-248.

Henkin, R. and Aamodt, R. (1975). Zinc absorption in acrodermatitis enteropathica and in hypogeusia and hyposmia, *Lancet*, **1**: 1379-1380.

Henkin, R.I., Schechter, P.H., Friedewald, W.T., *et al*. (1976). A double-blind study of the effects of zinc sulfate on taste and smell dysfunction. *Amer. J. Med. Sci.* **272:** 285-299.

Hoffman, H.N., Phyliky, R.L., Fleming, C.R. (1988). Zinc-induced copper deficiency. *Gastroenterology* **94:** 508-512.

Ketcheson, M.R., Barron, G.P., Cox, D.H. (1969). Relationship of maternal dietary zinc during gestation and lactation to development and zinc, iron, and copper content of the postnatal rat. *J. Nutr.* **98:** 303-311.

Mack, R.B. (1989). A hard day's knight, zinc phosphide poisoning. *N.C. Med. J.* **50:** 17-18.

Methfessel, A. and Spencer, H. (1973a) Zinc metabolism in the rat. I. Intestinal absorption of zinc. *J. appl. Physiol*. **34**: 58-62.

Methfessel, A. and Spencer, H. (1973b) Zinc metabolism in the rat. II. Secretion of zinc into intestine. *J. appl. Physiol.* **34:** 63-67.

Porter, K.G., McMaster. D., Elmes, M.E., *et al*. (1977). Anemias and low serum-copper during zinc therapy. *Lancet*. **2:** 744.

Prasad, A.S., Brewer, G.J., Schoomaker, E.B. *et al.* (1978). Hypocupremia induced by zinc therapy in adults. *J. Amer. Med. Assoc.* **240:** 2166-2168.

Samman, S. and Roberts, D.C.K. (1987). The effect of zinc supplements on plasma zinc and copper levels and the reported symptoms in healthy volunteers. *Med. J. Australia* **146:** 246-249.

Schlicker, S.A. and Cox, D.H. (1967). Maternal dietary Zn in excess, fetal development, and Fe and Cu metabolism. *Fed. Amer. Proc.* **26:** 520.

Schlicker, S.A. and Cox, D.H. (1968). Maternal dietary zinc and development; zinc, iron, and copper content of the rat fetus. *J. Nutr.* **95:** 287-294.

Stokinger, H.E. (1981). Zinc. In: Patty's Industrial Hygiene and Toxicology, 3rd rev. ed., vol. 2A. G.D. Clayton and E. Clayton, eds., John Wiley and Sons, New York. pp. 2033-2049.

USEPA (2005). Integrated Risk Information System Database (IRIS). Zinc and compounds. US Environmental Protection Agency, Washington DC.

WHO (1973). Trace elements in human nutrition, WHO Technical Report Series 532, pp 9-15.

WHO (1982). Safety evaluation of certain food additives and contaminants. WHO Food Additives Series 17. Zinc.

WHO (2001). Environmental Health Criteria 221: Zinc. World Health Organization, Geneva.