New Zealand Food Safety
Haumaru Kai Aotearoa

Risk Profile: Chemical Forms of Contaminant Elements (Species)

Part 1: Arsenic

Part 2: Chromium, Selenium and Vanadium

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Scientific Interpretative Summary

This SIS is prepared by New Zealand Food Safety (NZFS) risk assessors to provide context to the following report for MPI risk managers and external readers

Risk profile: chemical forms of contaminant elements (species).

Chemical elements in the environment, in food and in the human body can often exist in a number of chemical forms dependant on their oxidation state and the chemical groups they are bound too. These different chemical forms, or species, can dictate how the element behaves and how toxic or nutritious it might be when ingested.

Chemical speciation has conventionally been regarded as highly important in the dietary risks associated with mercury, with the organic form methylmercury, presenting a greater risk to health, than the inorganic or elemental form. However, the importance of speciation for other contaminant and nutrient elements is gaining greater recognition.

As testing for the species an element may be present in is more complex than establishing the presence of the element it is important for New Zealand Food Safety to understand the risks associated with species of different elements to prioritise any survey work.

The two parts of this risk profile focus on different chemical elements where speciation may influence the toxicity or nutritional status.

Part 1: Arsenic

ESR report: FW15032

This Risk Profile evaluate the potential risk of the chemical species of arsenic that can occur in food, in particular inorganic arsenic (iAs) which is generally regarded as the most toxic form. Prevalence of iAs differs between foods ranging from less than 1% in fish to nearly 100% in some plant based foods.

Analysis for total arsenic has been the focus of all New Zealand Total Diet Studies prior to 2016 and hence the risk from inorganic arsenic had to be estimated. While conservative estimates did not identify the New Zealand population as having a high dietary exposure to inorganic arsenic this risk profile identified the absence of an accurate exposure assessment for iAs as an important data gap.

Accredited analytical methodology to determine inorganic arsenic is available in New Zealand. As a result, the recommendations of this risk profile were used to inform the addition of inorganic arsenic testing in the 2016 New Zealand Total Diet Study.

Part 2: Chromium, Selenium and Vanadium

ESR report: FW15032

Chromium, selenium and vanadium are trace components of the diet. Trivalent chromium is considered non-toxic and potentially essential for nutrition, while hexavalent chromium is listed by the International Agency for Research on Cancer (IARC) as carcinogenic to humans. The report summarises the data available and identifies there is limited evidence for hexavalent chromium being a risk of exposure through food.

Selenium is an essential trace element, although it can also be toxic in excess. Selenomethionine is the predominant dietary source of selenium and may have nutritional advantages over other forms of selenium as it can be stored by the body. Associations are identified between higher selenium status and reduced disease risk, however the evidence for selenium supplementation to reduce disease risk is not supported. Adverse effect from

selenium deficiency have not been identified in New Zealand. Testing for selenium in the 2016 New Zealand Total Diet Study identified levels are adequate for nutritional needs.

Vanadium has not yet been established as an essential trace element in the diet. While some forms of vanadium, such as vanadium pentoxide, can be a concern for health, there is no evidence dietary vanadium is a health risk. There is insufficient data available to identify the impact of vanadium speciation on dietary exposure risks.

RISK PROFILE: CHEMICAL FORMS OF CONTAMINANT ELEMENTS (SPECIES). PART 1: ARSENIC

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CONTENTS

LIST OF TABLES

LIST OF FIGURES

EXECUTIVE SUMMARY

The purpose of a Risk Profile is to provide contextual and background information relevant to a food/hazard combination so that risk managers can make decisions and, if necessary, take further action. Risk Profiles include elements of a qualitative risk assessment, as well as providing information relevant to risk management. Risk profiling may result in a range of activities, such as immediate risk management action, a decision to conduct a quantitative risk assessment, or a programme to gather more data. Risk Profiles also provide information for ranking of food safety issues.

This Risk Profile addresses the chemical species of arsenic that can occur in food, in particular inorganic arsenic (iAs). Arsenic is ubiquitous in foods, although concentrations differ markedly between food types. Consequently, this risk profile will not focus on specific foods, apart from increased emphasis on foods known to be significant contributors to dietary exposure. The proportion of total arsenic (tAs) present as iAs varies from less than 1% in finfish to almost 100% in some samples of plant-based foods.

It is well accepted that iAs is carcinogenic, causing cancer of the skin, lung and urinary bladder, albeit there is increasing evidence that disease mainly results from chronic exposure to high levels of arsenic.

The toxic mode of action of iAs is not known, but it is believed that it involves several mechanisms, including oxidative stress, non-mutagenic genotoxicity and epigenetic changes. Molecular evidence suggests that there may be a threshold to the toxic effects of iAs, including carcinogenic effects. In addition, several recent meta-analyses and systematic reviews have questioned whether associations between iAs exposure and some disease states are significant at iAs water concentrations less than 100 μg/L.

While tAs has been determined in a wide range of foods consumed by New Zealanders, little direct evidence is available for the iAs content of the New Zealand food supply. However, dietary exposure estimates, derived by application of conservative assumptions to tAs data, do not suggest that New Zealanders have particularly high dietary exposure to iAs (0.2-0.5 μg/kg bw/day), when compared to other international estimates of dietary exposure. However, as has been noted in international assessments of iAs (EFSA, 2009; JECFA, 2011), dietary exposures are similar to benchmark doses, indicating negligible margins of exposure. Consequently, the possibility of a risk to some consumers due to iAs at normal dietary exposure levels cannot be excluded.

Determination of iAs can be successfully achieved by hyphenated, as well as simpler non-chromatographic, methods. One New Zealand laboratory (Cawthron Institute) is currently accredited for iAs analysis. For determination of individual arsenic species in foods a hyphenated technique, linking chromatographic or electrophoretic separation with sensitive arsenic determination would be required. Such techniques are not currently offered by any New Zealand analytical providers

There are relatively few risk management options available for control of iAs exposure in New Zealand. Maximum limits and Imported Food Requirements are in place to control exposure to highly contaminated seafood, cereals and hijiki seaweed.

While it is unlikely to change the profile of risks associated with arsenic species in New Zealand foods, information on the species composition and content of New Zealand foods

would establish whether they are similar to the composition and content of arsenic species in foods reported in the literature.

It is known that some drinking water supplies in the central North Island of New Zealand contain arsenic at concentrations up to 100 μg/L. With the exception of a study demonstrating very high tAs concentrations in watercress from the Waikato River, it is not currently known whether other foods from this area are similarly high in arsenic. If so, consideration of region specific risk assessments may be warranted.

While information is unlikely to come from New Zealand, further information informing the potential for a dose threshold for the adverse health effects due to arsenic is required. Information on the toxicity of organoarsenic species, and the mechanism by which arsenic species exert their toxicity would assist risk assessment.

1. INTRODUCTION

The purpose of a Risk Profile is to provide information relevant to a food/hazard combination or combinations so that risk managers can make better informed decisions and, if necessary, take further action. Risk Profiles are part of the Risk Management Framework (RMF)[1] approach taken by the Ministry for Primary Industries (MPI) Food Safety. The Framework consists of a 4-step process:

- Preliminary risk management activities
- Identification and selection of risk management options
- Implementation of control measures, and
- Monitoring and review

This initial step in the RMF, Preliminary Risk Management Activities, includes a number of tasks:

- Identification of food safety issues
- Risk profiling
- Establishing broad risk management goals
- Deciding on the need for a risk assessment
- If needed, setting risk assessment policy and commissioning of the risk assessment
- Considering the results of the risk assessment
- Ranking and prioritisation of the food safety issue for risk management action.

Risk profiling may be used directly by risk managers to guide identification and selection of risk management options, for example where:

- Rapid action is needed;
- There is sufficient scientific information for action;
- Embarking on a risk assessment is impractical.

1.1 HAZARDS AND RISK MANAGEMENT QUESTIONS

This Risk Profile addresses the chemical species of arsenic that can occur in food, in particular inorganic arsenic (iAs). It should be noted that arsenic is ubiquitous in foods, although concentrations may differ markedly between food types. Consequently, this risk profile will not focus on specific foods, apart from increased emphasis to foods known to be significant contributors to dietary exposure.

The current risk profile does not address issues related to arsenic contamination of animal feed or animal health issues, except where these are relevant to human health.

The sections in this Risk Profile are organised as much as possible as they would be for a conventional qualitative risk assessment.

Hazard identification, including:

- A description of the chemical(s).

[1] http://www.foodsafety.govt.nz/elibrary/industry/RMF_full_document_-_11604_NZFSA_Risk_Management_Framework_3.1.pdf accessed 22 July 2015

Hazard characterisation, including:

- A description of the adverse health effects caused by the chemical.
- Dose-response information for the chemical in humans, where available.

Exposure assessment, including:

- Data on the occurrence of the hazard in the New Zealand food supply.
- Data on the consumption of relevant foods by New Zealanders, where appropriate.
- Qualitative estimate of exposure to the chemical (if possible).
- Overseas data relevant to dietary exposure to the chemical.

Risk characterisation:

- Information on the number of cases of adverse health effects resulting from exposure to the chemical with particular reference to the identified food (based on surveillance data) or the risk associated with exposure (based on comparison of the estimated exposure with exposure standards).
- Qualitative estimate of risk, including categorisation of the level of risk associated with the chemical in the food.

Risk management information

- A description of relevant food safety controls.
- Information about risk management options.

Conclusions and recommendations for further action

1.2 MAIN INFORMATION SOURCES

Information on the toxicology of and exposure to arsenic has been reviewed or otherwise considered by a number of groups. These assessments were major resources for the current project. Sources included:

- JECFA (the Joint FAO/WHO Expert Committee on Food Additives). Assessment reports were accessed at: http://www.inchem.org/
- EFSA (European Food Safety Authority). Opinions were accessed at: http://www.efsa.europa.eu/
- IARC (the International Agency for Cancer Research). Monographs were accessed from ESR's standing collection or at: http://monographs.iarc.fr/ENG/Monographs/PDFs/index.php. Summaries can be accessed at: http://www.inchem.org/

More recent and additional information than that included in these resources was located by general searching of the World Wide Web (internet) and use of specific citation databases, including:

- PubMed. Accessed at: http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?DB=pubmed
- Web of Science. Accessed at: http://apps.webofknowledge.com/

Due to the existence of several recent international reviews of arsenic species (ATSDR, 2007; EFSA, 2009; IARC, 2012; JECFA, 2011), searches of the literature were focussed predominantly on the very recent literature (2010 onwards).

2. HAZARD IDENTIFICATION

Arsenic is a metalloid occurring widely in the earth's crust, with an average abundance of approximately 5 mg/kg (IARC, 2012). Arsenic is the 20th most abundant element in the earth's crust and is present in more than 200 mineral species.

2.1 STRUCTURE AND NOMENCLATURE

Arsenic can exist in four oxidation states; -3, 0, +3 and +5 (IARC, 2012). However, under normal environmental conditions the +5 oxidation state is the most stable and the majority of arsenic species in organisms and in food contain arsenic in the +5 oxidation state (EFSA, 2009). From a biological and toxicological perspective the most important structural distinction is that arsenic may occur in both inorganic and organic forms, of which the inorganic forms are the more toxic (EFSA, 2009; IARC, 2012; JECFA, 2011). The arsenic content of a material, without reference to species forms, is referred to as total arsenic (tAs). The arsenic-containing compounds found in water, foods and biological samples are summarised in Table 1.

Table 1. Arsenic compounds found in water, foods and biological samples

Chemical Name	Synonyms	Abbreviation	CAS No.
Inorganic			
Arsenate		As^{v} or As^{5+}	
Arsenite		As^{III} or As^{3+}	
Organic			
Methylarsonic acid	Monomethylarsonic acid, methylarsonate	MMA^{v}	124-58-3
Dimethylarsinic acid	Dimethylarsinite, cacodylic acid	DMA^{v}	75-60-5
Methylarsonous acid	Monomethylarsonous acid	MMA^{III}	63869-12-5
Dimethylarsinous acid		DMA^{III}	55094-22-9
Arsenobetaine		AB	64436-13-1
Arsenocholine		AC	39895-81-3
Trimethylarsine oxide		TMAO	4964-14-1
Tetramethylarsonium ion		TMA^{+}	27742-38-7
Dimethylarsinylethanol		DMAE	
Trimethylarsoniopropionate		TMAP	
Dimethylarsinylribosides	Oxo-arsenosugars		
Dimethylmonothioarsinic acid		$DMMTA^{v}$	
Dimethyldithioarsinic acid		$DMDTA^{v}$	

CAS No.: Chemical Abstract Service Registry Number

2.2 OCCURRENCE

2.2.1 Inorganic arsenic

Low levels of inorganic and organic arsenic are present in most, if not all, foods (IARC, 2012). Levels of inorganic arsenic (iAs) in foods and beverages do not usually exceed 0.1 mg/kg, with mean concentrations generally less than 0.03 mg/kg (JECFA, 2011). There are some exceptions to this generality. Rice often contains 0.1-0.4 mg/kg dry weight of iAs and may contain considerably higher tAs concentrations (EFSA, 2009). The edible marine alga,

hijiki (*Hizikia fusiforme*) can contain iAs at concentrations in excess of 60 mg/kg, while concentrations of iAs of up to 30 mg/kg dry weight have been reported in blue mussels (*Mytilus edulis*) (EFSA, 2009).

For terrestrial foods, iAs constitutes a high, but variable, proportion of tAs, with 30-100% of the arsenic present as iAs. EFSA carried out exposure assessments for iAs by assessing scenarios in which 50, 70 or 100% of the total arsenic (tAs) was present as iAs (EFSA, 2009). Other studies have used proportions as low as 10% to estimate iAs from tAs, but current knowledge suggests that this proportion is unrealistically low.

Arsenic concentrations in groundwater are usually less than 10 μg/L, but can reach concentrations of 5000 μg/L in some places, notably regions of Bangladesh, Taiwan, India and Chile (EFSA, 2009). Virtually all of the arsenic present in water will be iAs. In oxygenated conditions, arsenic will be present as arsenate, but under certain reducing environmental conditions arsenite can be the dominant species.

2.2.2 Organic arsenic

The highest tAs concentrations and the highest concentration of organoarsenic compounds occur in marine organisms. Over 50 organoarsenic compounds have been identified in marine organisms used as human foods (EFSA, 2009).

Arsenic is in the same group of the periodic table as nitrogen and phosphorus. It has been suggested that the high levels of arsenic in marine organisms may be due to an inability of the organisms to distinguish between arsenic oxides and structurally related phosphates, which are actively taken up (EFSA, 2009).

The major form of arsenic in seafood is arsenobetaine (AB) (EFSA, 2009). AB has also been detected in some terrestrial foods, such as mushrooms. Low levels of AB have also been detected in marine algae. Very low concentrations may also be present in freshwater organisms, although higher concentrations can occur in farmed freshwater fish, due to feeding of marine fishmeal (Soeroes *et al.*, 2005a). AB is structurally similar to the osmolyte (substances that maintain osmotic balance under conditions of variable salinity) glycine betaine, and it has been suggested that AB may have a role as an adventitious acquired osmolyte (Clowes and Francesconi, 2004).

Arsenosugars are the major form of arsenic in marine algae (typically 2-50 mg/kg dry weight) (EFSA, 2009). Arsenosugars also constitute a measurable proportion of the arsenic in species such as mussels and oysters that feed on marine algae. Arsenosugars are only present in trace amounts in terrestrial organisms.

Arsenolipids may be present in fish oils at concentrations in the range 4-12 mg As/kg (EFSA, 2009).

TMAP and AC can be found at modest concentration in seafood, while the simple methylated derivatives of arsenic are generally only present at low concentrations (EFSA, 2009).

2.3 ANALYTICAL METHODS

Analytical methods for arsenic species in foods involve three steps:

- Extraction of the arsenic species from the food matrix
- Separation of arsenic species, and
- Detection of arsenic species.

While this section will focus on the analysis of arsenic species, it should be noted that determination of tAs will simply involve wet or dry digestion of the food matrix in combination with one of the detection systems discussed for arsenic species.

2.3.1 Sample extraction for arsenic species

The extractants most commonly used are polar solvents, such as methanol, water, methanol/water mixtures, or solutions of nitric acid, tetramethylammonium hydroxide, trifluoracetic acid, phosphoric acid, sodium hydroxide or enzyme mixtures (JECFA, 2011). The chemical conditions used for extraction must be reasonably mild, to maintain the chemical form of the arsenic species (EFSA, 2009). Because extraction efficiency can vary across food types and even across different samples of the same food type, mass balance is an important part of the quality control (JECFA, 2011). This involves comparison of the sum of the determined species with a parallel determination of tAs.

The current focus on iAs has led to the development of extraction protocols with high extraction efficiency for iAs, which may not retain the chemical integrity of other arsenic species (EFSA, 2009; JECFA, 2011). Some of these extractants will also result in interconversion of As^{3+} and As^{5+}, so, although iAs can be reliably determined, the individual components of iAs may differ from their form in the food. For example, trifluoroacetic acid has been used for extraction of iAs from rice, but results in partial reduction of As^{5+} to As^{3+}, while some extraction protocols have included hydrogen peroxide, resulting in oxidation of As^{3+} to As^{5+}.

The EU Institute for Reference Materials and Measurements (IRMM) has included iAs in several interlaboratory comparisons under the International Measurement Evaluation Programme (IMEP). IMEP-107 involved analysis of rice for tAs and iAs (de la Calle *et al.*, 2011b). Seven certifying laboratories all used extraction methods based on acid (nitric, hydrochloric or trifluoroacetic) extraction for iAs. Two laboratories included hydrogen peroxide, while a further two included a reducing agent (bromic acid and hydrazine sulphate). The latter reducing methods included separation of the arsenic species into chloroform, then back extraction in dilute hydrochloric acid. IMEP-116 and IMEP-39 included analysis of mushrooms for tAs and iAS (Cordeiro *et al.*, 2014) and IMEP-112 involved analyses of wheat, spinach and algae in tAs and iAs (de la Calle *et al.*, 2011a), with extraction methods used by certifying laboratories the same as those outlined for rice.

In a US-based interlaboratory proficiency trial, analysing rice, kelp and apple juice for arsenic species, 26 of 41 participating laboratories used nitric acid extraction (Briscoe *et al.*, 2015). Comparison of a wide range of potential extractants concluded that the highest extraction efficiencies and the least impact on the identity of the species extracted was achieved with 0.15 M nitric acid containing silver (to remove potential for iodide to reduce As^{5+} to As^{3+}) or 0.05 M perchloric acid (Narukawa *et al.*, 2014). Dilute nitric or hydrochloric acids were shown to have the best extraction efficiencies for determination of iAs from seafood matrices (Pétursdóttir *et al.*, 2014).

2.3.2 Separation of arsenic species

The nature of the food being analysed dictates the range of arsenic species present and the complexity of the separation required. High-performance liquid chromatography (HPLC) is the separation technique that has been most widely used for arsenic species analysis (JECFA, 2011), with ion exchange or ion-pairing conditions most commonly used (EFSA, 2009). The most generally occurring species, As^{3+}, As^{5+}, MMA and DMA, can be satisfactorily separated by anion exchange under isocratic conditions, while the wider range of species present in seafood can be separated by a combination of anion and cation exchange columns (JECFA, 2011).

Some of the newly discovered arsenic species require different chromatographic conditions. For example, arsenolipids are separated by reversed-phase HPLC (EFSA, 2009). Gas-chromatography (Yuan *et al.*, 2014) and capillary electrophoresis (Matusiewicz and Ślachciński, 2012; Qu *et al.*, 2015) have also occasionally been used to separate arsenic species.

As for extraction, an important quality assurance procedure is the determination of chromatographic column efficiency; the proportion of the arsenic applied to the column that can be accounted for by the species separated (and quantified).

A review of separation conditions used in a number of studies has been published (Sadee *et al.*, 2015).

2.3.3 Detection of arsenic species

Detection and quantification of arsenic species are usually performed by atomic absorption spectrometry (AAS), atomic fluorescence spectrometry (AFS) or inductively-coupled plasma-mass spectrometry (ICP-MS) (EFSA, 2009; JECFA, 2011). The sensitivity of AAS and AFS is substantially increased by derivatisation of arsenic through a process called hydride generation (HG). Inorganic arsenic species, MMA and DMA all form volatile arsines[2] in reaction with sodium tetrahydroborate in acidic conditions. However, for organoarsenic species it is often necessary to chemical treat samples before they will form hydrides (EFSA, 2009). HG-AAS or HG-AFS can be interfaced directly with HPLC for determination of arsine-forming arsenic species (Anawar, 2012).

ICP-MS is becoming the method of choice for arsenic speciation analysis, as the HPLC can be directly coupled to the ICP-MS, without the need for intermediate chemical treatment. Such techniques are known as hyphenated techniques (Sadee *et al.*, 2015). In the IMPE-107 study, 6 of the 7 certifying laboratories used ICP-MS detection, while the seventh used HG-AAS (de la Calle *et al.*, 2011b). In a US-based interlaboratory proficiency trial, analysing rice, kelp and apple juice, 37 of 41 participating laboratories used ICP-MS detection (Briscoe *et al.*, 2015).

HPLC with MS or MS/MS detection is very useful for the identification of arsenic species, but cannot reliably provide quantification of the species detected unless individual calibration for all species is included (EFSA, 2009; Serpe *et al.*, 2013).

[2] Inorganic forms of arsenic are converted to AsH_3 (arsine), MMA is converted to CH_3AsH_2 (monomethylarsine) and DMA is converted to $(CH_3)_2AsH$ (dimethylarsine)

2.3.4 Methods for total inorganic arsenic

Other methods have been developed that do not allow determination of the different arsenic species that may be present in food, but allow iAs to be separated from organic arsenic for subsequent quantification by techniques such as HG-AAS or ICP-MS.

Some recent studies have proposed a method without chromatography for the determination of iAs, based on the greater propensity for iAs species to form hydrides and partition into the gas phase than organoarsenic species (Chen *et al.*, 2014; Musil *et al.*, 2014).

Extraction of rice with 1 M nitric acid followed by treatment with 0.1% (w/v) EDTA was reported to result in quantitative extraction of iAs for subsequent determination by electrothermal AAS (Pasias *et al.*, 2013). As^{3+} was determined following microwave-assisted extraction of rice with 1 M nitric acid and EDTA (5% w/v) treatment by complexation with ammonium pyrrolidinedithiocarbamate (APDC) and separation into methyl-isobutyl ketone (MIBK). As^{5+} was determined by difference.

Microwave assisted extraction into dilute hydrochloric acid, containing hydrogen peroxide, results in oxidation of As^{3+} to As^{5+}, which can then be selectively eluted from a strong anion exchange solid-phase extraction (SPE) cartridge (Chen and Chen, 2014; Rasmussen *et al.*, 2012). In these studies, subsequent quantification of arsenic was carried out by HG-AFS and HG-AAS.

The reduction-chloroform extraction-dilute acid back-extraction method mentioned in the extract section has also been used for chromatography-free determination of iAs, in combination with ICP-MS (Chung *et al.*, 2013; Fontcuberta *et al.*, 2011).

A colourimetric method for determination of iAs has been outlined (Gürkan *et al.*, 2015). Inorganic arsenic was extracted in acidic hydrogen peroxide, converting all iAs to As^{5+}. Arsenic was then concentrated using cloud-point extraction, a micelle-mediated process. Arsenic was determined as the As^{5+} complex with acridine orange and tartaric acid by absorbance at 494 nm.

A wide range of voltammetric techniques have been developed for the determination of iAs (Liu and Huang, 2014). However, there is little evidence that these techniques have been applied to the determination of iAs in foods.

2.3.5 Arsenic species testing in New Zealand

Only one New Zealand laboratory (Cawthron Institute) is accredited for inorganic arsenic analysis.[3] Cawthron use the reduction-chloroform extraction-dilute acid back-extraction method mentioned above and originally developed by Munoz *et al*. (1999), with ICP-MS quantification (Geoff Miles, Cawthron Institute, personal communication). Cawthron participated in the IMEP-107 interlaboratory study on tAs and iAs in rice (de la Calle *et al.*, 2011b; de la Calle *et al.*, 2012) and a more recent interlaboratory study of arsenic species in rice, kelp and apple juice, run by Brooks Rand Laboratories (Briscoe *et al.*, 2015). This extraction method was used by two of the six certifying laboratories for the IMEP programme.

[3] http://www.ianz.govt.nz/directory/ Accessed 28 July 2015

2.3.6 Comparison of method performance

While no quantitative comparison of different arsenic speciation methods has been performed, inter-laboratory comparison studies have consistently noted that there is no clustering of results for iAs on the basis of methodology (Baer *et al.*, 2011; Cordeiro *et al.*, 2014; de la Calle *et al.*, 2012). It was concluded that analysis for iAs was not method dependent.

For determination of individual arsenic species a hyphenated technique, linking chromatographic or electrophoretic separation with sensitive arsenic determination would be required. Such techniques are not currently offered by any New Zealand analytical providers. Determination of iAs can be successfully achieved by hyphenated, as well as simpler non-chromatographic, methods. There is no evidence to suggest that there is a 'preferred method' for iAs analysis and method performance data would be a better guide to the suitability of a method-provider combination.

3. HAZARD CHARACTERISATION: ADVERSE HEALTH EFFECTS

3.1 CONDITIONS

While trivalent arsenic and trivalent arsenic compounds are generally considered to be more toxic than their pentavalent equivalents, the co-occurrence of both forms and their interconversion under a range of environmental and physiological conditions means they are generally considered collectively (EFSA, 2009). Inorganic arsenic is acutely toxic with a minimum lethal dose of approximately 2 mg/kg bw (ATSDR, 2007). However, acute arsenic poisoning is usually associated with accidental, suicidal, homicidal, or medicinal ingestion of arsenic-containing powders or solutions. Rodent LD_{50} for arsenate and arsenite have been reported in the range 15 to 175 mg/kg bw (ATSDR, 2007). No information has been reported on human deaths following ingestion of organic arsenic species (ATSDR, 2007; EFSA, 2009), but LD_{50} for MMA, DMA and roxarsone[4] have been reported in the range 102 to 3184 mg As/kg bw (ATSDR, 2007). AB and TMAO have been reported to be virtual non-toxic following acute administration, with LD_{50} greater than 10,000 mg/kg bw (JECFA, 2011).

Laboratory animals appear to be substantially less susceptible to the toxic effects of iAs than humans and most information on the adverse health effects of arsenic exposure comes from human epidemiological investigations. These investigations have usually assessed arsenic exposure in terms of the concentration of arsenic in the water supply and, therefore, relate to iAs. Studies have focussed on five regions (south-west and north-east Taiwan, northern Chile, the Cordoba region of Argentina, Bangladesh and the West Bengal region of India) with particularly high water arsenic concentrations (IARC, 2012).

IARC concluded there was sufficient evidence of a causal relationship between ingestion of iAs and cancer of the lung, urinary bladder and skin (IARC, 2012). Skin cancer was considered to be primarily squamous cell carcinoma (non-melanoma skin cancer). Associations with kidney, liver and prostate cancer were considered to be suggestive, but evidence fell short of establishing a causative relationship.

Skin lesions, including hyperpigmentation and hyperkeratosis, are sensitive indicators of chronic iAs arsenic exposure (EFSA, 2009). Significant associations between skin alterations and risks of skin cancer have also been identified.

Effects on foetal development (increased risk of spontaneous abortion, stillbirth, preterm birth and neonatal death, birth defects, lower birth weight, lower head or chest circumference), child health and development (neurobehavioural deficits, central nervous system disorders), neurotoxicity (peripheral neuropathy, central nervous system toxicity), cardiovascular disease, and abnormal glucose metabolism and diabetes have all been associated with water iAs levels in regions with high water arsenic (>100 μg/L). However, these associations have not been demonstrated in regions with lower water arsenic concentrations.

No human toxicity data are available for organoarsenic compounds.

[4] Roxarsone is an organoarsenic compound that may be added to poultry feed as a coccidiostat. Roxarsone has been voluntarily withdrawn from use in the USA and is not registered for use in the EU or New Zealand.

3.2 TOXICOLOGICAL ASSESSMENT

Five reasonably recent toxicological assessments of arsenic have been carried out, with particular focus on iAs (ATSDR, 2007; EFSA, 2009; IARC, 2012; JECFA, 2011; US Environmental Protection Agency (USEPA), 2010). It should be noted that the USEPA assessment is still in draft form and has not been summarised here.

3.2.1 IARC

IARC concluded that there was:

- Sufficient evidence that iAs compounds cause cancer of the lung, urinary bladder and skin. A positive association was noted to have been observed between exposure to arsenic and iAs compounds and cancer of the kidney, liver, and prostate.
- Sufficient evidence in experimental animals for the carcinogenicity of iAs compounds.

Based on these conclusions, arsenic and iAs compounds were classified as Group 1 (carcinogenic to humans). DMA^{v} and MMA^{v} were classified as possibly carcinogenic to humans (Group 2B), while arsenobetaine and other organic arsenic compounds are not metabolised in humans and are not classifiable as to their carcinogenicity to humans (Group 3).

3.2.2 EFSA

EFSA identified a range of values for the 95 % lower confidence limit of the benchmark dose of 1% extra risk ($BMDL_{01}$), instead of a single reference point, for use in the risk characterisation for iAs. The $BMDL_{01}$ values for the relevant health endpoints, skin lesions, cancers of the skin, urinary bladder and lung, ranged from 0.3 to 8 µg/kg bw/day. The estimated dietary exposures for European populations to iAs for average and high level consumers are within the range of the $BMDL_{01}$ values identified, and therefore there is little or no margin of exposure (MOE), and the possibility of a risk to some consumers cannot be excluded. Exposure estimates for potentially highly exposed population sub-groups (high consumers of rice or algae-based products) were also within the range of the $BMDL_{01}$ values identified.

EFSA noted that the available data on mean and median urinary arsenic in European populations, without specific high level exposure to arsenic, are in the region of 5 to 6 µg/L. This concentration range is close to, or below, the concentrations in the reference populations in the epidemiological studies providing the basis for the $BMDL_{01}$ values. However, data on European sub-groups with high dietary inorganic arsenic exposure were not available.

Because of the lack of data on the toxicity and food concentrations of arsenosugars, arsenolipids, MMA^{v} and DMA^{v} it was not possible for the risks associated with exposure to these compounds to be characterised.

EFSA recommended that:

- Dietary exposure to inorganic arsenic should be reduced.
- Speciation data for different food commodities should be produced to allow refinement of risk assessment.
- The suitability of arsenic speciation methods needs to be established for a range of food samples and/or arsenic species.
- There is a need for robust validated analytical methods for determining inorganic arsenic in a range of food items.

- Certified reference materials, especially for inorganic arsenic in products such as water, rice and seafood, are required.
- Future epidemiological studies should incorporate better characterisation of exposure to inorganic arsenic, including food sources.
- There is a need for more information on critical age periods of arsenic exposure, in particular in early life. Studies should include effects later in life of early life arsenic exposure.
- There is a need for improved understanding of the human metabolism of organoarsenicals in foods (arsenosugars, arsenolipids, etc.) and the human health implications.

3.2.3 JECFA

JECFA based their risk characterisation on an iAs BMDL for a 0.5% increased incidence of lung cancer. The $BMDL_{0.5}$ was determined by using a range of assumptions to estimate exposure from drinking-water and food with differing concentrations of iAs. The $BMDL_{0.5}$ was estimated to be 3.0 µg/kg bw/day (2.0–7.0 µg/kg bw/day based on the range of estimated total dietary exposure). It should be noted that this range is within the EFSA range of values for their $BMDL_{01}$. The JECFA Committee noted that the previously derived Provisional Tolerable Weekly Intake (PTWI) of 15 µg/kg bw (2.1 µg/kg bw/day) is in the region of the $BMDL_{0.5}$ and therefore was no longer appropriate, and withdrew the previous PTWI.

Mean dietary exposure to iAs in the USA and various European and Asian countries was reported to be in the range 0.1 to 3.0 µg/kg bw/day. Drinking water was a major contributor to iAs dietary exposures and, depending on the concentration, can also be an important source of arsenic in food through food preparation and possibly through irrigation of crops. The proportion of total exposure to iAs due to food consumption relative to the proportion from water intake increases as the concentration of iAs in the water decreases.

JECFA recommended that:

- There is a need for validated methods for selective extraction and determination of iAs in food matrices and for certified reference materials for iAs.
- There is a need for improved data on occurrence of different arsenic species in, and their bioavailability from, different foods as consumed in order to improve the estimates of dietary and systemic exposure.
- Further information on the toxicity of arsenic species found in food is also required.
- Future epidemiological studies of the health impacts of arsenic should incorporate appropriate measures of total exposure to iAs, including from food and from water used in cooking and processing of food.
- Epidemiological studies should not only focus on relative risks, but also analyse and report the data such that they are suitable for estimating exposure levels associated with additional (lifetime) risks, so as to make their results usable for quantitative risk assessment.

It is worth noting that there is a high level of agreement between the recommendations made by EFSA and JECFA.

3.2.4 Agency for Toxic Substances and Disease Registry (ATSDR)

ATSDR have derived estimates of exposure posing minimal risks to human (MRLs). An MRL is defined as an estimate of daily human exposure to a substance that is likely to be without an appreciable risk of adverse effects (non-carcinogenic) over a specified duration of exposure.

For iAs, ATSDR derived:

- an MRL of 5 μg/kg bw/day for acute-duration (14 days or less) oral exposure, based on human poisoning case studies
- an MRL of 0.3 μg/kg bw/day for chronic-duration (365 days or more) oral exposure, based on a NOAEL for development of skin lesions in a Taiwanese study.

For organoarsenic compounds, ATSDR derived:

- an MRL of 100 μg/kg bw/day for intermediate-duration (15-364 days) oral exposure to MMA, based on a benchmark dose (BMD) for a 10% increase in the incidence of diarrhoea in animal studies
- an MRL of 10 μg/kg bw/day for chronic-duration (365 days or longer) oral exposure to MMA, based on a BMD for a 10% increase in the incidence of progressive glomerulonephropathy in animal studies
- an MRL of 20 μg/kg bw/day for chronic-duration (365 days or longer) oral exposure to DMA, based on a BMD for a 10% increase in the incidence of vacuolisation of the urothelium in the urinary bladder in animal studies.

No MRLs were derived for roxarsone, due to a lack of appropriate toxicological data.

3.3 PROPOSED MECHANISMS OF CARCINOGENICITY

Several modes of action have been proposed to explain the carcinogenicity of iAs (ATSDR, 2007; EFSA, 2009; JECFA, 2011). It is likely that multiple mechanisms may be involved and that, at least, some of them may also have relevance for non-cancer endpoints. Proposed modes of action include:

- Oxidative stress, through the production of reactive oxygen species
- Genotoxicity. While iAs is only weakly mutagenic, it appears to be capable of inducing DNA damage, such as strand break
- Altered growth factors, leading the cellular proliferation and promotion of carcinogenesis
- Modification of expression of genes involved in cell growth and defence (epigenetic mechanisms)
- Alteration of binding of nuclear transcription factors.

In vitro studies were considered with epidemiological evidence to elucidate the mode of action of arsenic with respect to bladder cancer (Gentry *et al.*, 2014). Studies in cell lines exhibited a transition in gene expression with increasing exposure to arsenic from an adaptive response to frank toxicity. At low arsenic doses changes in gene expression related to preinflammatory responses and delay of apoptosis (Gentry *et al.*, 2010). At intermediate arsenic doses gene expression changes were observed related to oxidative stress, proteotoxicity, inflammation, proliferative signalling and induction of apoptosis, while at high arsenic doses expression changes in genes associated with apoptosis dominate. This suggests that linear extrapolation from high to low doses may not be appropriate for bladder carcinogenicity due to arsenic and an effect threshold was proposed. It was also noted that this finding is consistent with meta-analyses that have failed to establish a relationship between water arsenic and bladder cancer at water arsenic concentrations below 100 μg/L (Mink *et al.*, 2008).

3.4 CARCINOGENIC POTENCY OF INORGANIC ARSENIC

The US Environmental Protection Agency (USEPA) has derived several estimates of carcinogenic potency for arsenic over the last 30 years. The potency is usually expressed as the cancer slope factor or unit risk and is the slope of the low dose relationship derived between arsenic exposure and cancer incidence. The first of these was a cancer slope factor

of 1.5 (mg/kg bw/day)$^{-1}$ for skin cancer, based on data from a Taiwanese study (US Environmental Protection Agency (USEPA), 2010). USEPA amended this cancer slope factor to 3.67 (mg/kg bw/day)$^{-1}$ in 2006 (US Environmental Protection Agency (USEPA), 2006). Subsequent assessments have focussed on internal cancers (lung and bladder) and the current draft USEPA assessment has proposed a cancer slope factor of 25.7 (mg/kg bw/day)$^{-1}$ for lung and bladder cancer combined (US Environmental Protection Agency (USEPA), 2010).

In an assessment of the global burden of disease from foodborne arsenic, cancer slope factors, also known as cancer potency factors or unit risk factors, were consolidated for the three main cancer types causally associated with arsenic exposure (Oberoi *et al.*, 2014). Slope factors are summarised in Table 2.

Table 2. Cancer slope factors for lung, bladder and skin cancers due to inorganic arsenic exposure

Cancer type	Slope factor (increased population risk per μg iAs/day)	
	Males	Females
Lung	0.0000137	0.0000194
Skin	0.000015	0.000015
Bladder	0.0000127	0.0000198

The slope or cancer potency factors are quite similar across the different cancer types. For comparison, assuming a 70 kg body weight, the USEPA cancer slope factor would equate to a value of 0.000367 (μg/day)$^{-1}$.

3.5 METABOLITES AND THEIR RELATIVE TOXICITY

Arsenate (As^{V}) is rapidly reduced to the more reactive arsenite (As^{III}), mainly by the enzyme glutathione reductase (EFSA, 2009). In mammals, arsenite undergoes oxidative methylation in the liver, catalysed by the enzyme arsenic-methyltransferase, to give methylarsonate (MMA^{V}). MMA^{V} is then reduced to MMA^{III} by the enzyme glutathione-S-tranferase ω1. Formation of MMA^{III} facilitates further oxidative methylation to give dimethylarsinate (DMA^{V}). Arsenobetaine is not metabolised in humans and excreted unchanged. Arsenosugars and arsenolipids appear to be completely metabolised to DMA^{V}, the same end product as inorganic arsenic (EFSA, 2009).

In humans, arsenic is mainly excreted in urine, with a typical excretion profile made up of 10-30% iAs, 10-20% MMA^{V} and 60-70% DMA^{V} (EFSA, 2009). However, the profile of arsenic metabolites excreted can vary considerably from person to person and this is believed to reflect variations in methylation efficiency. It has been suggested that methylation efficiency may be a risk factor for carcinogenesis, with higher proportions of MMA in urine associated with increased risk of lung and bladder cancers (Melak *et al.*, 2014).

4. EXPOSURE ASSESSMENT

4.1 ARSENIC IN THE NEW ZEALAND FOOD SUPPLY

4.1.1 Inorganic arsenic

Little information is available on the inorganic arsenic content of New Zealand foods. In 2004-2005, seven samples of imported hijiki, an edible marine alga (called 'seaweed' in the report), were analysed for inorganic arsenic (ESR Report number CFC0990375). Samples were extracted by a method to separate inorganic and organic forms of arsenic, with the inorganic fraction subsequently analysed for total arsenic. Inorganic arsenic concentrations in the range 60-90 mg/kg were reported. Three samples of other seaweed types were analysed and found to contain <0.1 mg/kg inorganic arsenic.

4.1.2 Total arsenic

The total arsenic content of more than 100 different foods available in New Zealand are periodically determined as part of the New Zealand Total Diet Study (NZTDS). Table 3 summarises information from the most recent completed NZTDS (2009) (Vannoort and Thomson, 2011).

Table 3. Total arsenic in New Zealand foods

Food/food group	Number of samples	Number of samples <LOD	Arsenic concentration (µg/kg)	
			Mean	Range
Fruits and vegetables, included canned, excluding dried	296	252 (85%)	0.7-2.4	<2-9.4
Raisins/sultanas	8	0	24.3	17.0-32.0
Mushrooms	8	0	195	56.3-389
Cereal products, excluding rice, bran cereal and wheat biscuit cereals	112	55 (49%)	5.4-8.2	<2-24.5
Rice	8	0	33.4	14.4-49.7
Bran cereal	8	0	29.6	17.0-51.4
Wheat biscuit cereal	8	0	29.1	18.5-38.5
Dairy products	80	74 (93%)	0.4-3.5	<1-10.1
Meat, excluding bacon	72	22 (31%)	4.0-4.6	<2-11.5
Bacon	8	0	55.9	3.5-111
Fish fingers	8	0	1020	500-1880
Fish in batter	8	0	2660	313-5810
Fish, canned	8	0	623	324-1130
Fish, fresh	8	0	3990	2070-6310
Mussels	8	0	2220	1690-3420
Oysters	8	0	2390	1630-3220
Yeast extract	6	0	142	119-165

Source: 2009 New Zealand Total Diet Study (Vannoort and Thomson, 2011)

As previously observed, total arsenic concentrations in fish and shellfish are at least two orders of magnitude greater than most other foods. Not all foods are summarised here, but those not summarised all have concentrations of total arsenic of 10 μg/kg or less.

A study carried out on the Waikato River found concentrations of tAs in watercress (*Lepidium sativum*) leaves of up to 103 mg/kg, fresh weight (103,000 μg/kg; mean 29,000 μg/kg, *n* = 27) (Robinson *et al.*, 2003). River water samples were found to contain tAs in the range 1 to 150 μg/L. The concentrations of tAs found in watercress in this study were approximately 10,000 times the concentrations found in fruits and vegetables in the NZTDS. While arsenic was not speciated in this study, the watercress is clearly accumulating arsenic from the river water and the majority of the arsenic is likely to be iAs. The elevated tAs levels in the Waikato River are considered to be a consequence of geothermal activity in the region. This study demonstrates the potential for local conditions to substantially impact the arsenic content of the food supply.

A later study in the same region analysed a range of mainly non-edible aquatic and terrestrial plant species for tAs (Robinson *et al.*, 2006). Similar, 'hyperaccumulation' of arsenic in aquatic plant species was noted, but the investigators found that arsenic was not taken up to any extent by terrestrial plants. It should be noted that the limit of detection for tAs analyses in this and the earlier study (Robinson *et al.*, 2003) were quite high (500 μg/kg).

4.1.3 Arsenic in New Zealand drinking water

While this Risk Profile is concerned with speciated forms of arsenic in the food supply, most of the epidemiological information on arsenic relates to arsenic in drinking water.

Results have been summarised of a survey of arsenic in water from 342 New Zealand community drinking water zones (Davies *et al.*, 2001). The sample set was biased to zones where there was some expectation of arsenic being present (central plateau and Waikato), due to geothermal activity. Arsenic was detected at concentrations >5 μg/L in 70 drinking water zones providing drinking water to 284,720 people, while arsenic concentrations >10 μg/L were detected in 28 zones providing water 21,284 people. While actual arsenic concentration data were not provided it was noted that the maximum concentration was 100 time higher than the limit of detection. The limit of detection was 1 μg/L, suggesting that the maximum concentration observed was about 100 μg/L.

A further survey of arsenic in New Zealand coastal groundwaters (*n* = 45) detected arsenic in 9 samples, with a maximum concentration of 4 μg/kg (Nokes and Ritchie, 2002).

4.2 ARSENIC IN THE AUSTRALIAN FOOD SUPPLY

4.2.1 Inorganic arsenic

Studies of iAs in Australian foods have mainly focussed on seafoods, due to the higher concentrations of tAs in these foods. Results of recent surveys are summarised in Table 4.

Table 4. Inorganic arsenic in foods in Australia

Food	Number of samples	Number of samples <LOD	Inorganic arsenic concentration (μg/kg)		Reference
			Mean[a]	Range	
Dried seaweed					(FSANZ, 2013)
- Wakame	4	0	180	160-200	
- Kombu	4	0	220	160-330	
- Hijiki	1	0	7800	-	
- *Sargassum fusiforme*	1	0	320	-	
- Nori	4	0	110	90-160	
- Other	2	1	60-85	<50-120	
- Sea vegetable	1	0	100	-	
Seaweed-containing foods					
- Miso soup (dry)	1	1	0-50	-	
- Seasoning sauce	3	3	0-50	-	
- Seaweed chips	3	1	83-100	<50-140	
- Desserts	3	3	0-50	-	
- Japanese tea (dry)	1	1	0-50	-	
Fish fillets, battered	10	10	0-50	-	(FSANZ, 2011)
Fish portions, frozen	4	4	0-50	-	
Prawns	8	8	0-50	-	
Tuna, canned in brine	4	4	0-50	-	
Seaweed	48	5	1060-1070 (279-285)[b]	<50-38,000 (<50-750)	(New South Wales Food Authority, 2010)
Fish	10	10	0-50	-	
Apple juice, from Australia and New Zealand	96	76[c]	3.0-3.2	<2.5-11.3	(FSANZ, 2014)
Rice					(Rahman *et al.*, 2014)
- Australian grown	4	0	199	165-276	
- Imported	7	0	124	19-271	

LOD: Limit of detection

[a] Where the results included results below the LOD, lower bound (results below the LOD set to zero) and upper bound (results below the LOD set to the LOD) estimates of the mean were calculated

[b] The sample set contained one seaweed with very high concentrations of inorganic arsenic (38,000 μg/kg). The figures in parentheses are the upper and lower bound mean calculated, excluding this sample

[c] Samples were initially analysed for total arsenic. Only the 34 samples that contained detectable total arsenic were analysed for inorganic arsenic

It should be noted that the survey of inorganic arsenic in apple juice determined As^{3+} and As^{5+} separately (FSANZ, 2014). Inorganic arsenic was exclusively present as As^{5+}.
In the rice study of Rahman *et al*. (2014) the average proportion of iAs to tAs in Australian-grown rice was 69% (range 58 to 94%).

4.3 OVERSEAS CONTEXT

While inorganic arsenic may be present in any food, international activities have tended to focus on a reasonably narrow range of foods. The rapid evolution of analytical methods for arsenic species has resulted in a proliferation of publications on the arsenic species content of foods in recent years. In the following sections studies have only been summarised that have been published since 2010, except where earlier studies provide unique or particularly important information. It should be noted that a number of reviews have been conducted, summarising earlier studies.

4.3.1 Plant foods

Rice

Inorganic arsenic in rice has received considerable recent attention, due to the being a major staple food in many countries and containing elevated concentrations of arsenic, mainly present as iAs. Table 5 summarises study information on the iAs content of rice.

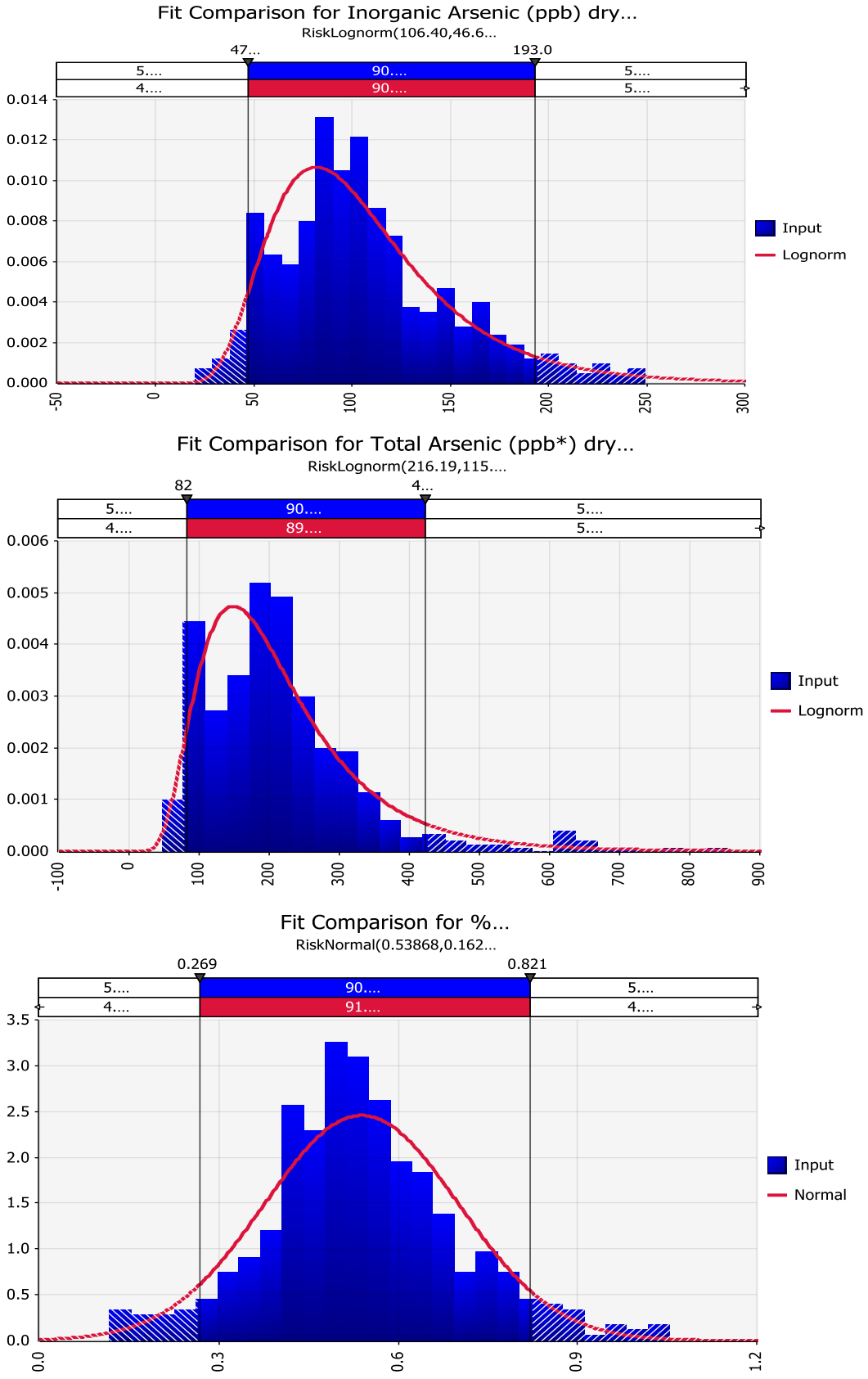
A meta-analysis of studies providing information on the tAs and iAs content of foods has been conducted (Lynch *et al.*, 2014). For rice (*n* = 2000), the mean tAs concentration across all studies considered was 200 μg/kg (range <3-1800 μg/kg). The mean iAs concentration was 130 μg/kg (range <1.6-300 μg/kg), mostly as As^{3+}, giving an average proportion of iAs of 65%. DMA was the other main species reported to be present in rice.

Rahman and Hasegawa (2011) summarised studies carried out between 1998 and 2009 and found mean tAs concentrations in rice in the range 20 to 930 μg/kg, mean iAs concentrations in the range 30 to 510 μg/kg and mean iAs to tAs proportions in the range 35 to 100%. For studies where the proportion of iAs to tAs was determined, the overall unweighted average proportion was 61%.

Analysis of data from Australia, China, Japan, Mercosur, the EU and the USA (*n* = 1243) by the Codex Committee on Contaminants in Foods (CCCF) fitted the available iAs concentration data to a continuous distribution with a mean of 139 μg/kg and a standard deviation of 65 μg/kg (CCCF, 2012). While CCCF did not fit a parametric distribution to these data, the distribution curve depicted is slightly right skewed and likely to conform to a distribution such as the lognormal.

Some studies have suggested that rice from the USA contains comparatively high concentrations of tAs, but a comparatively low proportion of tAs as iAs (30-40%) (Adomako *et al.*, 2011; Meharg *et al.*, 2009). However, the United States Food and Drug Administration (FDA) have analysed almost 500 rice samples for concentrations of tAs and iAs and found the proportion of iAs to be similar to the results of studies conducted in other countries (US Food and Drug Administration, 2013). Results of this programme are summarised in Table 5. The availability of the full FDA dataset allowed further analysis of the distribution of data. The distribution of tAs and iAs concentrations in rice can be adequately modelled as lognormal distributions, while the ratio of iAs to tAs can be modelled as a normal distribution, as would be expected under the Central Limit Theorem (Figure 1).

Figure 1. Distribution of inorganic and total arsenic and their ratio in rice tested in the USA



The body of information on arsenic in rice appears to be sufficiently consistent to draw several general conclusions:

- The concentration of iAs in rice averages about 100 μg/kg and only occasionally exceeds 200 μg/kg
- While the proportion of tAs in rice present as iAs is highly variable, overall it appears that about 60% of arsenic is present as inorganic species
- There is some evidence that the proportion of iAs may be greater in brown rice than white rice, suggesting that there may be a greater concentration of iAs in the bran of rice
- The dominant inorganic species of arsenic in rice is usually As^{3+}
- The dominant organic species of arsenic in rice is DMA

New Zealand imports rice predominantly from Australia and Thailand (10-18 thousand tonnes per annum from each country over the period 2012-2014), with the USA, India and Pakistan also being significant sources for imported rice.[5] However, it should be noted that the mean tAs concentration for rice determined in the 2009 NZTDS was very low (33.4 μg/kg). Even after consideration of the diluting effect of water absorbed during the cooking of the rice (about 60% of the raw rice weight), this concentration does not seem consistent with published information on the iAs content of Australian and Thai rice.

[5] http://www.stats.govt.nz/infoshare/ Accessed 16 July 2015

Table 5. Summary information on inorganic arsenic in rice

Country	Samples	Number of samples	Arsenic, total	Arsenic, inorganic	Percent inorganic arsenic	Main species found	Reference
Argentina	Rice, various	59	380 (80-1290)	100 (20-280)	26	DMA	(Farías *et al.*, 2015)
Asia, South-east	White rice					As^{3+}	(Nookabkaew *et al.*, 2013)
	- South Asia	6	69 (25-125)		85 (71-98)		
	- Malaysia	3	103 (88-123)		69 (66-71)		
	- Vietnam	10	136 (93-204)		67 (51-78)		
	- Thailand	79	139 (23-304)		63 (43-96)		
	Brown rice						
	- Thailand	14	239 (118-343)		54 (41-84)		
	- Vietnam	2	299 (254-345)		73 (68-78)		
	Red rice – Thailand	27	211 (77-375)		63 (40-96)		
	White sticky rice – Thailand	38	93 (46-166)		64 (44-89)		
	Black sticky rice – Thailand	6	124 (31-313)		66 (44-104)		
Brazil	White	23	223 (109-376)	112 (45-193)	50 (23-77)	As^{3+}, As^{5+}, DMA	(Batista *et al.*, 2011)
	Parboiled white	12	215 (108-367)	130 (86-187)	60 (48-80)		
	Brown	3	348 (271-428)	188 (185-190)	54 (43-70)		
	Parboiled brown	6	266 (226-316)	156 (132-174)	59 (50-68)		
China, Guangdong Province	Rice (not further specified)	260		44.1		As^{3+}	(Lin *et al.*, 2015)
China, Hong Kong	White, cooked		46	22	48 (38-57)		(Chung *et al.*, 2013)
	Brown, cooked		67	43	64 (58-79)		
China, Taiwan/Japan	Rice , various	121	125 (3-412)		75	As^{3+}, DMA	(Huang *et al.*, 2012)
Czech Republic	Rice	10	150 (36-286)	93 (25-171)	62 (57-85)		(Raber *et al.*, 2012)
Denmark	White rice	11		89 (30-160)			(Rasmussen *et al.*, 2013)
	Brown rice	10		163 (80-400)			
	Black rice	5		158 (80-310)			
	Red rice	2		370 (130-600)			
	Parboiled rice	1		160			
	Rice crackers	7		337 (190-570)			
Finland	Long grain rice	8	253 (110-650)	158 (90-280)	62 (34-110)		(Rintala *et al.*, 2014)
Ghana	Rice originating from					As^{5+}, DMA[a]	(Adomako *et al.*, 2011)
	- Ghana	7	110	91	83		
	- Thailand	7	150	101	67		
	- USA	6	220	92	42		

Country	Samples	Number of samples	Arsenic, total	Arsenic, inorganic	Percent inorganic arsenic	Main species found	Reference
Greece	Rice	15	167 (42-271)	93 (<30-147)	64 (28-91)	As^{3+}	(Pasias *et al.*, 2013)
India, West Bengal, Nadia district	Rice, not further specified	50	130		74		(Mondal and Polya, 2008)
Iran	White, aromatic and non-organic rice	15	121 (51-222)	82 (40-135)	70 (40-89)	As^{3+}	(Cano-Lamadrid *et al.*, 2015)
Italy	Rice, various	101	211 (70-460)	94 (1-200)	45 (0.4-96)		(Sommella *et al.*, 2013)
Japan	Polished white rice flour	20	149 (83-322)	102 (56-197)	71 (58-83)	As^{3+}	(Narukawa *et al.*, 2012)
Spain	White rice Brown rice	17 6	220 360	110 190	50 53		(Fontcuberta *et al.*, 2011)
Thailand	Brown rice Jasmine rice Sticky rice White rice, polished Jasmine rice, polished Sticky rice, polished Brown rice, bran Jasmine rice, bran Sticky rice, bran	30 30 30 30 30 30 15 15 14	194 (129-258) 186 (117-246) 198 (116-300) 125 (88-220) 124 (86-165) 136 (87-262) 894 (566-1395) 832 (599-1163) 963 (623-1430)	124 (75-193) 120 (73-174) 131 (78-188) 68 (45-106) 38 (42-101) 76 (44-156) 633 (375-919) 599 (447-824) 673 (436-1071)	64 (49-81) 64 (46-80) 67 (52-88) 55 (38-75) 55 (35-73) 56 (43-74) 71 (63-85) 72 (64-83) 70 (58-82)		(Ruangwises *et al.*, 2012)
Turkey	Rice, not further specified	50	202	160	80 (42-97)	As^{3+}	(Sofuoglu *et al.*, 2014)
USA	All rice Basmati Brown Instant Jasmine Parboiled White, long grain White, medium grain White, short grain Other	486 53 99 14 13 39 149 91 23 6	216 (47-854) 134 (47-526) 271 (57-854) 136 (90-244) 150 (63-225) 217 (91-362) 243 (74-776) 208 (54-717) 123 (81-180) 157 (112-227)	106 (20-249) 78 (20-200) 160 (34-249) 59 (31-134) 87 (34-151) 114 (71-191) 103 (49-196) 81 (39-174) 79 (52-102) 124 (88-161)	54 (12-106) 65 (20-104) 64 (20-104) 43 (24-68) 58 (49-76) 54 (43-78) 45 (24-71) 49 (12-76) 65 (42-90) 81 (70-106)		(US Food and Drug Administration, 2013)
USA	Rice, various	17		83 (45-235)			(Chen and Chen, 2014)

[a] The analytical technique used added hydrogen peroxide, which would have oxidised any As^{3+} present to As^{5+}

Other cereals

A meta-analysis consolidated information on tAs and iAs in 'grains, flours and breads' (Lynch *et al.*, 2014). The mean tAs concentration was 16 μg/kg (*n* = 161, range 4.6 to 290 μg/kg), while the mean iAs concentration was 9.2 μg/kg (*n* = 37, range <3 to 96 μg/kg), giving a mean ratio of iAs to tAs of 58%.

Results from the first Hong Kong Total Diet study reported low concentrations of tAs (mean 16 μg/kg, range ND to 79 μg/kg) and iAs (mean 8 μg/kg, range ND to 46 μg/kg) in cereals and their products (Chung *et al.*, 2013). This will presumably include rice and rice products, as well as other cereals. The mean proportion of iAs was 50%.

Arsenic speciation was examined in cereal products (*n* = 30; bread, biscuits, breakfast cereals, flour, snacks, pasta and infant cereals) purchased in Barcelona, Spain (Llorente-Mirandes *et al.*, 2014b). Concentrations of tAs (maximum 35.6 μg/kg) and iAs (maximum 26.0 μg/kg) were generally low, with the proportion of arsenic present as iAs being in the range 73 to >100%.

Analysis of a single wheat sample from Italy found 165 μg/kg of tAs and 152 μg/kg of iAs, giving an overall proportion of iAs of 92% (Raber *et al.*, 2012).

In a further Italian study, four composite samples, one each of wheat grain, bread, flour and pasta were analysed for arsenic species (D'Amato *et al.*, 2011). Concentrations of tAs were in the range 8.6 to 29.8 μg/kg, with the percentage of arsenic present as iAs in the range 50 to 100%. It is interesting to note that the dominant iAs species found in the samples was As^{3+}, This contrasts to the study of Raber *et al*. (2012), in which iAs was found to be solely in the As^{5+} form. However, the extraction solution used by Raber *et al*. included hydrogen peroxide, which would have resulted in oxidation of As^{3+} to As^{5+}.

In a study of household exposure to arsenic in the Turkish city of Izmir, bulgur, a product prepared from wheat, samples (*n* = 50) were analysed for arsenic species (Sofuoglu *et al.*, 2014). The mean tAs concentration was 21 μg/kg (range ND to 75 μg/kg), with virtually all of the arsenic present as iAs, predominantly As^{3+}.

Concentrations of tAs found in New Zealand cereal products are generally less than 30 μg/kg, except for products with a high bran content (Vannoort and Thomson, 2011).

Mushrooms

Arsenic species were determined in fresh shiitake mushroom (*Lentinula edodes*, *n* = 5) and products prepared from shiitake mushrooms (*n* = 9) (Llorente-Mirandes *et al.*, 2014a). Concentrations of tAs were in the range 110-1440 μg/kg dry weight, with iAs concentrations in the range 100 to 1380 μg/kg. The mean proportion of iAs to tAs was 84% (range 53 to 100%). Wong *et al.* (2013) reported lower concentrations of iAs in dried shiitake mushrooms (36 to 53 μg/kg).

Fresh shiitake mushrooms were used as the test material for an inter-laboratory proficiency test under the International Measurement Evaluation Program (IMEP) concluded by the Institute for Reference Materials and Measurement (IRMM) (Cordeiro *et al.*, 2014). The mean tAs and iAs concentrations determined by the reference laboratories were 646 and 321 μg/kg, respectively (50% iAs).

A study was conducted comparing the tAs and iAs concentrations in Chinese wild mushrooms (*Agaricus balieimurrill, Leucopaxillus giganteus, Pleurotas eryngii* and *Legista nuda*) and Spanish cultivated mushrooms (*Pluerotas eryngii* and *Agaricus bisporus*) (Gonzálvez *et al.*, 2009). It should be noted that the white form of *Agaricus bisporus* is the button mushroom, commonly cultivated in New Zealand. Concentrations of tAs were in the range 130 to 1400 μg/kg, with the lowest concentrations in cultivated *Agaricus bisporus* and the highest concentrations in wild *Leucopaxillus giganteus*. Concentrations of iAs were in the range 140 to 888 μg/kg. The proportion of iAs to tAs ranged from 57 to >100%.

A Canadian study of edible mushrooms determined arsenic species in 73 samples from 46 fungal species (Nearing *et al.*, 2014). The arsenic species composition of different fungal species was highly variable. Inorganic arsenic was the dominant species (up to 70% of total) in mushrooms of the families Cantharellaceae and Russulaceae, while AB was the dominant species in the families Lycoperdaceae and Agaricaceae. DMA was the dominant species in mushrooms from the other families examined (Amaniticeae, Hymenogastraceae and Suillaceae). Inorganic arsenic dominated in most types of store-bought mushrooms, except *Agaricus* spp., in which AB dominated.

These results are consistent with an earlier study on arsenic speciation in *Agaricus bisporus* (Soeroes *et al.*, 2005b). *Agaricus bisporus* grown on untreated compost contained 500 μg/kg tAs on a dry weight basis (50 μg/kg fresh weight, assuming 90% water content). AB was the main arsenic species present. However, when mushrooms were grown on a substrate containing 1000 mg/kg of As^{5+}, the tAs content was 22,800 μg/kg dry weight, with the majority of the arsenic present in inorganic forms.

In the 2009 NZTDS, mushrooms were found to have a mean tAs concentration of 195 μg/kg (Vannoort and Thomson, 2011). Mushrooms sampled for the 2009 NZTDS were all described as 'white button mushrooms' and are almost certainly all *Agaricus bisporus*.

Fruit juices

A meta-analysis consolidated information on tAs and iAs in apple juices and apple drinks (Lynch *et al.*, 2014). The mean tAs concentration was 8.1 μg/kg (*n* = 114, range 1.3 to 42 μg/kg), while the mean iAs concentration was 5.8 μg/kg (*n* = 105, range <2.5 to 88 μg/kg), giving a mean ratio of iAs to tAs of 72%.

Total arsenic was detected (LOD = 0.005 μg/kg) in 2 of 4 apple juice samples at concentrations of 1.7 and 1.8 μg/kg (Kutscher *et al.*, 2012). Inorganic arsenic species were found to account for 71 and 56% of tAs. DMA and MMA were also detected in one sample.

The first Hong Kong Total Diet Study included analysis of fruit and vegetable juice (Chung *et al.*, 2013). A mean tAs concentration of 10 μg/kg and a mean iAs of 5 μg/kg were reported, with a mean ratio of iAs to tAs of 52% (range 0 to 60%).

Analysis of fruit juices and fruit drinks (*n* = 96) found a mean tAs concentration of 4.2 μg/L (range ND to 24.4 μg/L) and a mean iAs concentration of 3.6 μg/L (range ND to 23.9 μg/L) (Wang *et al.*, 2015). This equates to a mean iAs to tAs proportion of 85%. Mean tAs and iAs concentrations were higher in grape and pear juices and drinks than in apple and other, but the ratio of iAs to tAs was quite consistent across juice/drink types. MMA and DMA were present at lower concentration and, on average, were present at similar levels to one another (approximately 7-8% of tAs each).

Carrington *et al*. (2013) reported results of monitoring of apple juice over the period 2005 to 2011, conducted by the US Food and Drug Administration. Analysis of 247 samples of apple juice gave results in the range ND to 45 μg/L, with a mean iAs concentration of 5.2 μg/L. The mean was calculated by assigning a value of 0.5 μg/L to samples of apple juice where iAs was not detected.

For comparison, the 2009 NZTDS detected tAs in all samples of 'apple-based juices', with concentrations in the range 2 to 8 μg/kg (mean 4.1 μg/kg) (Vannoort and Thomson, 2011). Assuming 85% iAs, this equates to a mean iAs concentration of 3.5 μg/kg.

Seaweed

Arsenic species were determined in five species of edible seaweed purchased in the Netherlands; Hijiki (*Hizikia fusiforme*), Kombu (*Laminaria japonica*), Laminaria (*Laminaria* spp.), Nori (*Porphyra* spp.) and Wakame (*Undaria pinnatifida*) (Brandon *et al.*, 2014). Seaweed species analysed had tAs concentrations in the range 1400 to 19,000 μg/kg, but differed substantially in the arsenic species present. Inorganic arsenic was only detected in Hijiki, where it accounted for 54% of tAs (10,000 μg/kg). The predominant species across all five seaweed types was DMA, accounting for 18 to 84% of tAs, depending on the species. Arsenosugars accounted for 54-58% of tAs in the *Laminaria* seaweeds (Kombu and Laminaria).

A Korean study determined tAs and iAs in samples of sea mustard (Wakame, *Undaria pinnatifida*), laver (*Porphyra tenera*), green laver (*Monostroma* and *Enteromorpha* spp.) and sea tangle (Kombu, *Laminaria japonica*) (Cui *et al.*, 2013). Concentrations of tAs ranged from 3620 (green laver) to 52,100 μg/kg (sea mustard). Inorganic arsenic was not detected in green laver, but accounted for 3.8 to 5.5% of tAs in other seaweed types. Maximum concentrations of iAs were found in sea tangle (2900 μg/kg).

A further Korean study analysed seaweed samples (*n* = 198) for tAs and arsenic species (Khan *et al.*, 2015). Samples included laver (*n* = 53), sea tangle (*n* = 45), sea mustard (*n* = 58), Hijiki (*n* = 27) and gulf weed (*Sargassum fulvellum, n* = 15). Mean tAs concentrations for the different species were in the range 1840 to 6480 μg/kg, with the highest concentrations found in gulf weed. Inorganic arsenic was only detected in Hijiki (mean 2350 μg/kg) and gulf weed (mean 5347 μg/kg), equating to mean proportions of iAs to tAs of 52 and 83%, respectively.

Laver (*Porphyra haitanensis*) from the Fujian region of south-east China were found to contain tAs concentrations in the range 30,000 to 61,500 μg/kg (Yang *et al.*, 2012). However, iAs accounted for no more than 0.9% of tAs in any sample (*n* = 18). This is consistent with the study of Khan *et al*. (2015) summarised above.

A Chilean study examined 14 seaweed species from the classes Rhodophyceae (red algae), Phaeophyceae (brown algae) and Chlorophyceae (green algae) (Díaz *et al.*, 2012). Concentrations of tAs were in the range 3100 to 68,000 μg/kg, while iAs concentrations were in the range 150 to 1700 μg/kg. The ratio of iAs to tAs was in the range 0.6 to 12.9%. The proportion of iAs in the different seaweed classes was Chlorophyceae > Rhodophyceae > Phaeophyceae. However, it should be noted that only one green algae species was analysed.

Concentrations of tAs in seaweeds from the Thermaikos Gulf, Greece were in the range 1400 to 55,000 μg/kg (Pell *et al.*, 2013). The highest concentration of iAs was in a brown algae, *Cystoseira barbata* (27,000 μg/kg, 49% of tAs). Red and green algae analysed

contained only negligible amounts of iAs. This is completely at odds with the results of the Chilean study summarised above.

Further evidence of the potential for seaweed of the family Sargassaceae to accumulate appreciable amounts of iAs is provided by a study from an environment described as 'pristine', in the Solomon Islands (Grinham *et al.*, 2014). Of the flora and fauna examined, the highest concentration of iAs was found in a brown alga (*Sargassum* sp.), containing 4830 μg/kg iAs, mainly in the form of As^{5+}. This equated to 56% of tAs (8.68 μg/kg). While higher tAs concentrations were reported for lobster and midnight snapper, the arsenic in these biota was substantially (>85%) in the form of AB.

Accumulation of iAs in seaweed appears to be quite species specific. Hijiki consistently has a high proportion of tAs present in inorganic forms. There is also some evidence to suggest that seaweed of the family Sargassaceae may contain a high proportion of iAs (Khan *et al.*, 2015; Pell *et al.*, 2013).

Other foods

Inorganic arsenic was determined in a range of root and tuber vegetables and in plantains, mainly originating in the Americas (Chen *et al.*, 2015). Concentrations of iAs were generally modest (0.9 to 14.1 μg/kg wet weight). The highest iAs concentrations were seen in plantains.

Arsenic species were determined in white and red wine samples (Escudero *et al.*, 2013). Mainly inorganic species were found, with As^{3+} concentrations in the range 1.1 to 5.6 μg/L and 1.6 to 6.5 μg/L in white and red wines, respectively, while As^{5+} concentrations were in the range 2.2 to 10.9 μg/L and 2.5 to 11.2 μg/L. Methylated metabolites were detected at maximum concentrations of less than 1 μg/L in either wine type.

A German study found As^{5+} concentrations in the range ND to 3.13 μg/L and As^{3+} in the range ND to 18.9 μg/L in a range of wine types, including rice wine (Huang *et al.*, 2011). It should be noted that this pattern of arsenic species is the reverse of that reported by Escudero *et al*. (2013). Beer contained higher relative amounts of As^{5+}, with maximum concentrations up to 9.5 μg/L. Based on median values, iAs accounted for 58-98% of tAs in wine and beer samples.

In the 2009 NZTDS, tAs was detected in all wine samples, with a pooled mean concentration across red and white wine types of 4.3 μg/L (Vannoort and Thomson, 2011).

4.3.2 Foods of animal origin

Seafood

It has been conservatively estimated that approximately 10% of the arsenic in seafood is in inorganic forms. Given the high concentrations of tAs that may be present in seafood, this suggests that seafoods have the potential to be major contributors to dietary iAs exposure.

A meta-analysis of concentrations of arsenic species in foods derived a mean iAs concentration for fish of 45 μg/kg (*n* = 1374) and a mean tAs concentration of 5000 μg/kg (*n* = 1920), giving a mean proportion of iAs of 0.9% (Lynch *et al.*, 2014). However, it should be noted that the same meta-analysis derived a mean As^{3+} concentration for fish of 86 μg/kg (*n* = 334) and a mean As^{5+} concentration of 52 mg/kg (*n* = 267), suggesting a much higher iAs content. Seafood was presented as a separate food category to fish and included mollusca

and crustacea, with a mean iAs concentration of 130 μg/kg (*n* = 835) and a mean tAs concentration of 10000 μg/kg (*n* = 2417) to give a proportion of iAs of 1.3%. As for fish, the mean concentrations for As^{3+} (150 μg/kg, *n* =1109) and As^{5+} (84 μg/kg, *n* = 814) suggest a higher proportion of iAs in seafood than that derived from measurements of iAs.

A French study determined arsenic species in fish (*n* = 30 species) and shellfish and crustacea (*n* = 17 species) (Sirot *et al.*, 2009). Amongst the fish species, tAs was in the range 710 to 34,300 μg/kg fresh weight, while the ratio of iAs to tAs was in the range 0.1 to 3.1% (mean 0.7%). Amongst the shellfish and crustacea species, tAs was in the range 2420 to 42,300 μg/kg fresh weight, with the ratio of iAs to tAs in the range 0.1 to 6.7% (mean 1.9%).

A study of economically important fish species from Norwegian waters (halibut, cod, herring, mackerel, tusk and saithe) found tAs concentrations in the range 10 to 110,000 μg/kg (Julshamn *et al.*, 2012). However, iAs was not detectable in most samples (*n* = 986), with limits of quantification in the range 2 to 4 μg/kg. Overall, the results of this study suggest that the proportion of tAs present as iAs in fish is considerably less than 1%.

A study of fish species from Izmir Bay, Turkey (*n* = 854) found iAs concentrations as high as 2170 μg/kg and iAs/tAs as high as 11.8% (Kucuksezgin *et al.*, 2014). The average proportion of iAs, across all samples, was 3.4%.

A French study examined arsenic speciation in composite samples of fish, shellfish and crustacea from the French Total Diet Study (Leufroy *et al.*, 2011). In fish samples, tAs concentrations were in the range 2200 to 10,700 μg/kg (dry weight basis), with iAs accounting for 0.5 to 1.9% of tAs (mean 1.2%). For shellfish (mussels, oysters, scallops) and crustacea (shrimps), tAs concentrations were in the range 1840 to 34,100 μg/kg (dry weight basis), with iAs accounting for 0.4 to 16% of tAs (mean 4.6%).

A Belgian study investigated speciation of arsenic in a range of marine and freshwater fish, shellfish and crustacea (Ruttens *et al.*, 2012). Freshwater species (tAs range 30 to 1700 μg/kg wet weight) accumulated considerable less arsenic than marine species (tAs range 630 to 25,100 μg/kg). Inorganic arsenic species were rarely detected in species from either source, at limits of detection in the range 1 to 2 μg/kg. While the available information does not allow calculation of iAs/tAs ratios, it appears these ratios are well below 1% for most species examined. The highest concentrations of iAs were detected in shellfish and crustacea.

A further study in Belgium analysed tAs and 'toxic arsenic' in 19 species of North Sea fish (*n* = 179) (Baeyens *et al.*, 2009). Toxic arsenic was defined as the sum of iAs, MMA and DMA. The mean tAs concentration across all fish samples was 12,830 μg/kg, with an average of 1.0% present as toxic arsenic. For 4 species of mollusca and crustacea (*n* = 38) the mean tAs concentration was 21,570 μg/kg, with on average 0.9% present as toxic arsenic.

Fish and seafood samples from Spain and Brazil contained tAs in the range 1000 to 33,800 μg/kg dry weight (Zmozinski *et al.*, 2015). Inorganic arsenic was not detected in any fish sample at a limit of detection of 6 μg/kg. Inorganic arsenic was detected in all seafood (shellfish and crustacea) samples at concentrations in the range 33 to 350 μg/kg. The maximum ratio of iAs to tAs for any seafood sample was 3.6%.

Analysis of mussels (*n* = 13) found a mean tAs concentration of 4730 μg/kg (range 1380 to 12790 μg/kg), with a mean iAs concentration of 70 μg/kg (range ND to 160 μg/kg) (Serpe *et*

al., 2013). The mean proportion of tAs as iAs was 1.5% (range 0.7 to 10.1%). On average AB accounted for 81% of tAs, while DMA accounted for 13% of tAs.

Canned tuna samples (*n* = 24) from Germany were found to contain tAs in the range 720 to 5000 μg/kg (Raber *et al.*, 2012). Only low concentrations of iAs were detected (<2 to 18 μg/kg). The proportion of tAs present as iAs was less than 0.5% in most samples, with a maximum proportion of 1.2%.

Fish-based infant foods (*n* = 6) were found to contain tAs in the range 108 to 275 μg/kg, but no iAs was detected in any sample, although the limit of detection was rather high (50 μg/kg) (López-García *et al.*, 2011).

In summary, the available information on iAs in seafood suggests that the previously used assumption of iAs as 10% of tAs is extremely conservative and most studies suggest a figure of 1% may still be conservative for fish. It appears that a higher proportion of the tAs in shellfish (and possibly crustacea) is present in inorganic forms. However, it still appears that, on average, no more than 5% of tAs is present as iAs.

Poultry meat

A meta-analysis consolidated information on tAs and iAs in chicken and chicken products (Lynch *et al.*, 2014). The mean tAs concentration was 8.9 μg/kg (*n* = 278, range <1.7 to 160 μg/kg), while the mean iAs concentration was 1.4 μg/kg (*n* = 273, range <0.4 to 40 μg/kg), giving a mean ratio of iAs to tAs of 16%.

Total arsenic was determined in cooked retail chicken meat samples (*n* = 140), including samples from conventional, antibiotic-free and organic production (Nachman *et al.*, 2013). The geometric mean (GM) concentration of tAs was 3.0 μg/kg, with concentrations lower in antibiotic-free meat than conventional or organic. Species analyses were carried out on samples containing greater than 10 g/kg dry weight (*n* = 78). These samples were found to contain a GM concentration of iAs of 1.1 μg/kg, DMA 3.5 μg/kg and roxarsone 0.6 μg/kg. Raw chicken meat samples (*n* = 116) contain a GM tAs concentration of 2.4 μg/kg, with 65 samples being analysed for arsenic species. GM concentrations of arsenic species in raw chicken were; iAs 0.7 μg/kg, DMA 2.7 μg/kg and roxarsone 0.7 μg/kg.

A speciation study on arsenic in chicken livers, with and without use of roxarsone as a feed additive, suggested higher concentrations of arsenic in this tissue type (Peng *et al.*, 2014). Livers from control birds contained AB (43 μg/kg), As^{3+} (9.3 μg/kg), As^{5+} (4.5 μg/kg), DMA (12 μg/kg) and MMA (1.2 μg/kg), as well as two phenylarsonic acid derivatives and roxarsone (3.1 μg/kg). In roxarsone-treated birds, roxarsone was present in livers at 150 μg/kg, while concentrations of all other species, except AB, were elevated 4 to 18-fold compared to control livers.

4.4 ESTIMATES OF DIETARY EXPOSURE

4.4.1 New Zealand

The 2009 New Zealand Total Diet Study derived estimates of dietary exposure to total arsenic, ranging from 5.5-5.8 μg/kg bw/week[6] for a teenage (11-14 years) girl to 12.8-13.7

[6] The range of exposure estimates relates to the treatment of left-censored ('not detected') data. The lower bound estimate was derived by assigning a value of zero to all left-censored data, while the

μg/kg bw/week for a toddler (1-3 years) (Vannoort and Thomson, 2011). By applying conservative assumptions; that 10% of the arsenic in seafood is iAs and all of the arsenic in other foods is iAs, estimates of iAs exposure were derived. Dietary exposure estimates for iAs ranged from 1.2-1.4 μg/kg bw/week (0.17-0.20 μg/kg bw/day) for a teenage girl to 3.0-3.2 μg/kg bw/week (0.43-0.46 μg/kg bw/day) for a toddler or an infant (6-12 months). The information presented in section 4.3 confirms that the assumptions used to derive these dietary exposure estimates for iAs are highly conservative.

No trends have been apparent in tAs exposures over successive New Zealand Total Diet Studies.

4.4.2 Australia

The 23rd Australian Total Diet Study included analyses of tAs for all foods and iAs for selected foods (seafoods) (FSANZ, 2011). However, estimates of dietary exposure were only derived for tAs, with mean estimates in the range 0.42-1.4 μg/kg bw/day (2.9-9.8 μg/kg bw/week).

Rahman *et al*. (2014) estimated dietary exposure to iAs for average Australians, Asian immigrants and European immigrants from consumption of rice only. Estimates for these three groups were 7.2, 68.4 and 2.5 μg/day, respectively. Assuming a conservative body weight of 60 kg, these exposures equate to 0.12, 1.1 and 0.04 μg/kg bw/day, respectively.

4.4.3 Overseas estimates of dietary exposure

Inorganic arsenic

Table 6 summarises results of overseas studies that have derived estimates of dietary exposure to iAs. Estimates of dietary exposure to tAs have not been included.

Table 6. International estimates of dietary exposure to inorganic arsenic

Country	Population group	Mean (high percentile) exposure, μg/kg bw/day[a]	Main foods contributing	Reference
Belgium	General population	0.097 (0.16)[b,e]	Fish, fruit	(Baeyens *et al.*, 2009)
Brazil	General population	0.16[c]	Only based on rice consumption	(Batista *et al.*, 2011)
Cameroon	Adults	0.23 (0.33)[d]	Beverages	(Gimou *et al.*, 2013)
China, Guangdong province	General population	0.09-0.50[e]	Only based on rice consumption	(Lin *et al.*, 2015)
China, Hong Kong	Adults	0.22 (0.38)	Cereals and cereal products	(Wong *et al.*, 2013)
China, Shandong province	General population Fish farmer Islander	0.097[f] 0.23 0.34	Only based on seafood consumption	(Wu *et al.*, 2014)

upper bound estimate was derived by assigning a value equal to the limit of detection to all left-censored data.

Country	Population group	Mean (high percentile) exposure, µg/kg bw/day[a]	Main foods contributing	Reference
Europe	Infants Toddlers Other children Adolescents Adults Elderly Very elderly	0.24-1.37 (0.54-1.66)[g] 0.32-1.17 (0.61-2.09) 0.20-0.87 (0.36-1.41) 0.12-0.48 (0.23-0.84) 0.11-0.38 (0.18-0.64) 0.09-0.34 (0.14-0.53) 0.09-0.36 (0.16-0.54)		(EFSA, 2014)
Finland	Adults Children	0.36-0.46 0.27-0.67	Only based on rice consumption	(Rintala *et al.*, 2014)
France	Adults Children	0.24-0.28 (0.46-0.51) 0.30-0.39 (0.61-0.77)	Water, beverages	(Arnich *et al.*, 2012)
Iran	General population	0.09	Only based on rice consumption	(Cano-Lamadrid *et al.*, 2015)
International	GEMS/Food cluster diets A B C D E F G H I J K L M	 0.21 (0.36) 0.07 (0.13) 0.22 (0.38) 0.08 (0.13) 0.03 (0.05) 0.03 (0.05) 0.88 (1.51) 0.15 (0.26) 0.09 (0.15) 0.17 (0.30) 0.56 (0.95) 0.89 (1.53) 0.08 (0.14)	Only based on rice consumption	(CCCF, 2012)
Japan	General population - Duplicate diets - Total Japanese diet	 0.11[f] 0.45		(Oguri *et al.*, 2012)
Japan	General population	0.40[f]	Rice, algae (Hijiki)	(Oguri *et al.*, 2014)
Korea	General population	0.19	Fish and shellfish	(Joung *et al.*, 2011)
Spain	Adult Catalan population	0.10-0.31 (0.49-1.1)[f]	Rice	(Fontcuberta *et al.*, 2011)
Spain	Gluten-free diet - Males - Females	 0.47 0.46	Only based on consumption of rice-based gluten-free foods	(Munera-Picazo *et al.*, 2014)
Sub-Saharan Africa	General population	0.041	Only based on root, tuber and plantain consumption	(Chen *et al.*, 2015)
Thailand (Dan Chang district)	Adults, 20-48 years - Duplicate diets (*n*= 180)	1.0 (range 0.34-1.8)		(Ruangwises *et al.*, 2011)
Turkey (Izmir)	Adults	0.15-0.69[h]	Only based on seafood consumption	(Kucuksezgin *et al.*, 2014)
Turkey	Adults (*n* = 50) - from rice - from bulgur	 0.10 (0.26) 0.007 (0.043)		(Sofuoglu *et al.*, 2014)
UK	Toddlers Young people Adults Elderly Vegetarians	0.075-0.25 (0.17-0.40) 0.055-0.16 (0.13-0.29) 0.028-0.093 (0.071-0.17) 0.024-0.079 (0.066-0.17) 0.035-0.10 (0.079-0.16)	Cereals, beverages	(Rose *et al.*, 2010)
USA	General population Children	0.08 (0.15)[i] 0.23 (0.39)	Water, rice, other grains	(Tsuji *et al.*, 2007)

Country	Population group	Mean (high percentile) exposure, μg/kg bw/day[a]	Main foods contributing	Reference
USA	General population 0-<1 year 1-2 years 3-5 years 6-12 years 13-19 years 20-49 years ≥50 years	0.05 (0.19) 0.23 (0.53) 0.10 (0.29) 0.08 (0.21) 0.04 (0.13) 0.03 (0.09) 0.03 (0.11) 0.03 (0.09)	Vegetables, fruit juices and fruits, rice, beer and wine	(Xue *et al.*, 2010)
USA, Arizona	Tap water <10 ppb As Tap water >10 ppb As	0.13-0.20[j] (0.56-0.69) 0.42-0.50 (2.1-22.3)		(Kurzius-Spencer *et al.*, 2014)
USA	Adult, 80 kg	0.0014	Only based on chicken consumption	(Nachman *et al.*, 2013)
Vietnam, Mekong delta	Before 2008 - Males - Females After 2008 - Males - Females	 17 (range 1 to 51) 13 (range 1 to 55) 0.9 (range 0.6 to 1.8) 0.9 (range 0.6 to 1.9)	Water (before 2008) Rice (after 2008)	(Hanh *et al.*, 2011)

[a] High percentile dietary exposure estimates are usually expressed as the 95th percentile, but may occasionally be expressed as the 90th or 97.5th percentile. Where a range is presented for the exposure estimate, this usually refers to different treatments of left-censored analytical data

[b] Exposure estimates are for 'toxic arsenic', which was defined as the sum of inorganic arsenic, MMA and DMA

[c] Recalculated from the original publication using a 70 kg body weight and adding individual estimates for As^{3+} and As^{5+}. Based on consumption of rice only

[d] Calculated from tAs by applied proportions taken from the literature

[e] Exposures were calculated for 13 cities in Guangdong province. The range represents the range of mean exposures across the 13 cities. Exposures have been recalculated from weekly exposure to daily exposures by dividing by 7

[f] Exposures were recalculated from μg/day using a 60 kg body weight

[g] Dietary exposure estimates cover a range of European countries. Ranges of dietary exposure estimates are from the minimum lower bound estimate to the maximum upper bound estimate. Estimates were based on iAs analytical values for seafood and a mixture of iAs and tAs x 0.7 for terrestrial foods

[h] The range represents the range across point estimates for the years 2009 to 2012

[i] Recalculated from the original publication using a 70 kg body weight for adults and a 15 kg body weight for children

[j] For this study the measure of central tendency is a median rather than a mean

While the dietary exposure assessments summarised in Table 6 do not constitute a comprehensive collation of studies that have been carried out, they are representative and suggest that exposures to iAs lie within a reasonably narrow range. All mean estimates of dietary iAs exposure fall within the range 0.024 to 1.37 μg/kg bw/day. Estimates of dietary iAs exposure in New Zealand, based on application of conservative assumptions to tAs data, fall within the range of estimates summarised in Table 6.

Organic arsenic

Few studies have derived exposure estimates for organic arsenic species. Oguri *et al*. (2014) used a market basket approach to determine dietary exposure to 6 organoarsenic species for the general Japanese population. The highest exposure was to AB (73 μg/day), followed by DMA (5.9 μg/day), TMAO (4.6 μg/day), AC (1 μg/day), TMA^+ (0.87 μg/day) and MMA (0.30 μg/day). However, unidentified arsenic species, mainly in algae, have higher levels of dietary exposure (95 μg/day). These unidentified compounds may be arsenosugars. Calculation of dietary exposures based on a 'typical Japanese diet' gave higher estimates of dietary exposure for AB (140 μg/day), DMA (11 μg/day), TMAO (5.9

μg/day) and MMA (3.9 μg/day) (Oguri *et al.*, 2012). However, analysis of duplicate diets for 25 Japanese gave lower geometric mean estimates of dietary exposure for DMA (1.2 μg/day), MMA (0.19 μg/day) and TMAO (0.07 μg/day) (Oguri *et al.*, 2012).

Exposure to MMA and DMA from consumption of rice in Guangdong Province, China was estimated to be 0.035 and 0.051 μg/kg bw/day, respectively (Lin *et al.*, 2015).

A Turkish study estimated mean DMA and MMA exposure from consumption of rice to be 0.024 μg/kg bw/day (95th percentile 0.060 μg/kg bw/day) and 0.002 μg/kg bw/day (95th percentile 0.005 μg/kg bw/day), respectively (Sofuoglu *et al.*, 2014).

4.4.4 Biomarkers of exposure

Arsenic, tAs or certain species, in urine, blood, hair, nails and saliva have been used as biomarkers of inorganic arsenic exposure (EFSA, 2009).

Most forms of arsenic present in food are excreted in urine within a few days (EFSA, 2009). While measurement of tAs in urine can be used as a biomarker of iAs exposure when seafood consumption is negligible, consumption of even small amounts of seafood can substantially increase tAs in urine, due to high concentrations of organic arsenic in these foods (Sirot *et al.*, 2009). Specific measurement of iAs and the methylated metabolites (MMA^v and DMA^v) of iAs provides a more reliable estimate of iAs exposure. DMA^v is the 'iAs and methylated metabolites' species that is most commonly detected and was detectable in 80% of urine samples in population study in the USA (Aylward *et al.*, 2014). However, it should be noted the elevated urinary levels of MMA^v and DMA^v can also result following consumption of seafood, either due to the presence of these species in seafood or due to metabolism of organic arsenic species, such as arsenosugars, to these species (Aylward *et al.*, 2014; Navas-Acien *et al.*, 2011). Consequently, in studies where urinary iAs and metabolites are measured, participants may be asked to refrain from seafood consumption for a period of several days before urine samples are collected (EFSA, 2009; Saoudi *et al.*, 2012) or participants with high seafood consumption may be excluded from further analysis of data (Gilbert-Diamond *et al.*, 2013).

A US study of urinary tAs and DMA found that, after correction for seafood intake and water source, both tAs and DMA concentrations in urine were significantly associated with rice consumption (Wei *et al.*, 2014). It should be noted that this study focused on these arsenic species because they were the most frequently detected species. Inorganic arsenic species were detected in less than 5% of urine samples.

A French cohort study (*n* = 1515) examined dietary risk factors for urinary tAs and iAs plus methylated metabolites (Saoudi *et al.*, 2012). Both urinary measures of arsenic exposure showed dose dependent associations with fish and shellfish consumption, while iAs and methylated metabolites were also associated with wine consumption.

As arsenic is cleared from blood within a few hours, blood arsenic is considered to only be indicative of very recent arsenic exposure and to have limited value as a biomarker of exposure (ATSDR, 2007; EFSA, 2009). However, in situations of chronic exposure to high levels of arsenic, blood arsenic will reach a steady state with the steady state concentration correlated with chronic exposure levels.

Arsenic accumulates in hair and nails, due to the high sulphydryl content of the keratin proteins in these tissue types (ATSDR, 2007; EFSA, 2009). Hair and nail arsenic are considered to be reliable biomarkers of chronic arsenic exposure. In some cases, hair arsenic concentrations may be misleading due to the ability of environmental arsenic to

absorb to the surface of hair (ATSDR, 2007; EFSA, 2009). In such instances, nails are preferred as material for biomarker determination.

A study in Southern Vietnam demonstrated significant correlations (Pearson's, $P<0.05$) between hair arsenic and exposure to iAs from groundwater, rice or rice and groundwater combined (Hanh *et al.*, 2011).

Measurement in iAs in saliva has recently been proposed as a biomarker of exposure (Bhowmick *et al.*, 2014). MMA^{v} and DMA^{v} in saliva were believed to be due to methylation of iAs and to reflect the methylation capacity of the individual. This paper did not discuss the exposure period represented by salivary iAs.

Two urinary biomarkers of DNA damage have been investigated (8-oxo-7,8-dihydro-29-deoxyguanosine (8-oxodG) and N^7-methylguanosine (N^7-MeG)) (Chou *et al.*, 2014). These markers were shown to be significantly correlated with urinary arsenic species. However, although Pearson correlation coefficients were significant, they were still quite modest (maximum of 0.25 between urinary iAs and N^7-MeG).

In a study in Norway involving a wide range of seafood consumption patterns, both urinary and blood tAs were positively associated with seafood consumption (Birgisdottir *et al.*, 2013).

A US study measured tAs and speciated arsenic (iAs and methylated metabolites) in urine and tAs in toenails in adults (n = 904) receiving water from supplies with tAs in the range <3 to 1200 μg/L (Calderon *et al.*, 2013). Increased water arsenic was correlated with all three biomarkers, while recent and higher seafood consumption was associated with urinary tAs, but not urinary speciated arsenic or toenail tAs. Volunteers were fed diets containing cod, salmon or blue mussels for two weeks (Molin *et al.*, 2014). Urinary tAs, AB and DMA increased significantly with seafood consumption in all cases, while urinary iAs and MMA only increased significantly with blue mussel consumption.

No biomarkers of effect have been validated for arsenic exposure (ATSDR, 2007; EFSA, 2009). However, proteomic and genomic analyses on cell lines, animals and humans following arsenic exposure have suggested potential markers for further investigation (Gentry *et al.*, 2010; Hegedus *et al.*, 2008).

5. RISK CHARACTERISATION

5.1 ADVERSE HEALTH EFFECTS IN NEW ZEALAND

5.1.1 Incidence of cancer

Arsenic exposure has been associated with an increased risk of lung, urinary bladder and skin cancer. Registration numbers and rates for these cancers are available.[7] However, information on lung and skin cancers (melanoma) are unlikely to be informative with respect to arsenic due to the confounding influences of smoking and exposure to solar radiation, respectively. It should further be noted that arsenic exposure has been associated with non-melanoma skin cancers (squamous or basal cell carcinomas), while New Zealand information is only available for melanoma. Lung cancer registration rates peaked in New Zealand in the 1980s (age standardised rate of approximately 35 to 40 per 100,000 population) and appear to be following a gradual decline (2005 to 2011; 29 to 31 per 100,000). In contrast melanoma rates have followed a general increase over the last 60 years. Melanoma accounts for approximately 95% of all skin cancer registrations in New Zealand.

Bladder cancer registration rates appeared to peak about 1994 to 2004 at 10.3 to 12.6 per 100,000, but have declined to 4.5 per 100,000 in the last reported year (2011). However, this decrease is largely due to a coding change in 2005, with superficial transitional cell carcinoma of the bladder no longer being coded as an invasive cancer.

According to the Globocan database[8], New Zealand lung cancer incidence rates are moderate by world standards, with more developed countries having higher rates than less developed countries. Melanoma incidence rates in Australasia are more than twice those of any other geographical region. Bladder cancer incidence rates worldwide follow a similar pattern to lung cancer incidence rates, with rates in more developed countries being higher than rates in less developed countries and rates in New Zealand and Australia being moderate.

5.1.2 Risk assessment

The 2009 NZTDS estimated the dietary exposure of New Zealanders to tAs and iAs (Vannoort and Thomson, 2011). However, risk was not further characterised. It was noted that, given the consistency of successive estimates of tAs exposure in New Zealand, exposure to tAs and iAs in New Zealand is likely to already be as low as reasonably achievable (ALARA).

5.2 ADVERSE HEALTH EFFECTS OVERSEAS

5.2.1 Incidence of cancer

According to the Globocan database, the countries with the highest incidence of lung cancer are Hungary (52 per 100,000), Serbia (46 per 100,000) and the Democratic Republic of

[7] http://www.health.govt.nz/nz-health-statistics/health-statistics-and-data-sets/cancer-data-and-stats?mega=Health%20statistics&title=Cancer Accessed 13 July 2015
[8] http://www-dep.iarc.fr/ Accessed 13 July 2015

Korea (44 per 100,000), with lowest incidence rates have been reported for western and central Africa, with mean rates of about 2 per 100,000.

Except for Australia and New Zealand (35 and 35 per 100,000, respectively), the highest melanoma rates are reported for Switzerland (21 per 100,000), the Netherlands (19 per 100,000) and Denmark (19 per 100,000). The regions with the lowest reported incidence of melanoma are southern Asia and Africa (<1 per 100,000).

The highest reported incidence rates for bladder cancer are for Belgium (17.5 per 100,000), Lebanon (16.5 per 100,000) and Malta (15.8 per 100,000), while the lowest incidence rates are reported for African, Asian, Pacific and Central and South American regions (<5 per 100,000).

Cancer incidence rates show no obvious associations with areas of known high arsenic exposure, such as India, Bangladesh and Taiwan.

5.2.2 Epidemiological studies

A number of epidemiological studies have been conducted, and continue to be conducted, examining associations between iAs exposure and various health endpoints. For a summary of earlier epidemiological and toxicological studies on associations between arsenic exposure and cancer see the IARC monograph (IARC, 2012). It should be noted that most of the epidemiological studies only consider exposure to arsenic from drinking water. For this reason, not all recent epidemiological studies have been summarised and those summarised in the following sections should be considered to be representative, but not exhaustive.

Lung or bladder cancer

A prospective cohort study of Japanese men and women aged 45 to 74 years (*n* = 90,378) estimated dietary exposure to tAs and iAs based on a validated food frequency questionnaire covering 138 foods (Sawada *et al.*, 2013). During 11 years of follow-up, 7002 lung cancer cases were identified. The median dietary exposures for the quartiles of exposure to iAs ranged from 36.5 to 107.6 μg/day (0.61 to 1.8 μg/kg bw/day for a 60 kg body weight). Significant hazard ratios were only seen for lung cancer across quartiles of tAs exposure in men, but not women. For iAs exposure, significant hazard ratios were seen for lung and kidney cancer in men, but only at the highest exposure quartile. No significant associations were seen in women. Positive associations were strengthened when the analysis only considered currently smoking men.

Associations between arsenic exposure and cancer mortality were examined in a cohort of American Indians (*n* = 3932), aged 45 to 74 years (Garcia-Esquinas *et al.*, 2013). Inorganic arsenic exposure was assessed as the sum of urinary iAs and methylated metabolites. Significant differences in cancer mortality between iAs exposures at the 20th and 80th percentiles were observed for lung, prostate and pancreatic cancer.

A case-control study in Chile matched lung cancer (*n* = 94) and bladder cancer (*n* = 117) cases to controls (*n* = 347) (Melak *et al.*, 2014). Parameters of arsenic exposure were assessed from analysis of urine samples. Significantly elevated odds ratios for both cancer types were found with an increasing proportion of MMA in urine. This was interpreted as evidence that lower arsenic methylation capacity was a risk factor for arsenic-related cancers. Individuals with greater methylation capacity would be expected to metabolise the majority of ingested iAs to DMA. Urinary iAs was not associated with significantly elevated odds ratios for either lung or bladder cancer. In fact, higher urinary iAs concentrations appear to be protective for bladder cancer.

A recent case-control study conducted in Chile reported significant associations between lung cancer and arsenic exposure at drinking water arsenic concentrations <100 μg/L (Steinmaus *et al.*, 2014). The final groups of cases (*n* = 92) and controls (*n* = 288) were individuals definitely known to have never been exposed to water supplies with arsenic content >100 μg/L. Statistically significant odds ratios were found for the highest tertile of water arsenic for highest single year (>63.6 μg/L) and highest 5-year average (>55.8 μg/L), but not highest lifetime average (>37.2 μg/L). As this study is novel in proposing cancer causation at relatively low water arsenic concentrations it has attracted considerable comment and some criticism, due to methodological aspects.

A meta-analysis of epidemiological studies of bladder cancer in moderate to low water arsenic regions (<100-200 μg/L) was conducted (Tsuji *et al.*, 2014a). The summary relative risk estimate (SRRE) from the nine studies included was not significantly elevated (1.07, 95%CI 0.95-1.21). It has been suggested that arsenic may act as a co-carcinogen, potentiating other carcinogens. The SRRE was elevated, but still not significant, when only 'ever smoked' cases were considered.
A re-analysis of lung and bladder cancer information from the Blackfoot disease endemic region arrived at similar conclusions (Lamm *et al.*, 2014). When only data from villages with water arsenic concentrations of <100 μg/L were considered associations went from being positive to negative, but not significant. The authors suggested this represents an inflection in the dose-response relationship at water arsenic concentrations of about 100-200 μg/L.

Skin cancer

A case-control study (470 cases, 447 controls) was conducted to examine associations between arsenic exposure, as assessed by urinary arsenic species, and squamous cell carcinoma (SCC), a non-melanoma skin cancer (Gilbert-Diamond *et al.*, 2013). Participants who reported seafood consumption within two days before urine collection were excluded from the study. Urinary MMA, DMA and the sum of iAs and methylated metabolites, but not urinary iAs, were all significantly associated with SCC incidence. The study was conducted in a region with detectable, but moderate water arsenic concentrations (<2 μg/L). Ratios of the arsenic species were not associated with SCC, suggesting that the methylation capacity of cases is not a determining factor in carcinogenesis.

The Arsenic Health Risk Assessment and Molecular Epidemiology (ASHRAM) study, a case–control study, was conducted in areas of Hungary, Romania, and Slovakia with reported presence of iAs in groundwater (Leonardi *et al.*, 2012). Consecutively diagnosed cases of basal cell carcinoma (BCC) of the skin (*n* = 529) were histologically confirmed; controls (*n* = 540) were general surgery, orthopaedic, and trauma patients who were frequency matched to cases by age, sex, and area of residence. Exposure was assessed in terms of water supply iAs. The adjusted odds ratio per 10 μg/L increase in average lifetime water iAs concentration was 1.18 (95% CI 1.08-1.28). The estimated effect of iAs on cancer was stronger in participants with urinary markers indicating incomplete metabolism of iAs, that is, a higher percentage of MMA in urine or a lower percentage of DMA.

Cardiovascular disease

A case-cohort study was conducted in south-central Colorado (James *et al.*, 2015). The study included 555 participants, with 96 coronary heart disease (CHD) events between 1984 and 1998. Long-term arsenic exposures were determined from structured interviews and data on water supply arsenic concentrations. After correction for known CHD risk factors, a positive association was found between time-weighted average (TWA) inorganic arsenic

exposure from water and CHD risk (hazard ratio = 1.38, 95% CI 1.09-1.78). The risk increased monotonically with increasing TWA inorganic arsenic exposure from water, relative to <20 μg/L. Hazard ratios (HR) were significantly elevated for water supply TWA iAs concentrations of 30-45 μg/L (HR = 2.2, 95% CI 1.2-4.0) and 45-88 μg/L (HR = 3.1, 95% CI 1.1-9.1).

A case-control study was conducted in the Inner Mongolia province of China (Wade *et al.*, 2015). Cardiovascular disease cases (*n* = 298) and age and gender frequency matched controls, with no history of arsenic-related disease (*n* = 275) were recruited from a large hospital. Water from the individuals' primary water source and toenail clippings were collected for iAs analysis. Water iAs concentrations were modest (mean = 8.9 μg/L). A significant odds ratio was found for a 10 μg/L increase in water arsenic (1.18, 95%CI 1.03-1.38). The adjusted odds ratio for water supplies over 40 μg/L compared to those less than 10 μg/L was 4.05 (95%CI 1.1-14.99).

A further case-control study in Inner Mongolia recruited cases with hypertension (*n* = 126) and non-hypertense controls (*n* = 386) (Li *et al.*, 2015). Urinary levels of iAs and methylated metabolites were determined for all participants. Urinary concentrations of iAs, MMA, DMA and tAs were significantly higher in cases than controls. %MMA was significantly higher and %DMA significantly lower in cases, with the associated secondary methylation index (SMI) significantly lower in cases. However, odds ratios were not significantly elevated, with the exception of the highest category of %MMA, but all odds ratios related to SMI were significantly lower. Stronger associations were found when indicators of lower methylation capacity were considered jointly with increased age, higher BMI or smoking status. However, it should be noted that the cases and controls were significantly different with respect to these variables.

Two recent systematic reviews of epidemiological studies conducted in the USA, Taiwan, Bangladesh and China have concluded that there is no conclusive evidence for an association between cardiovascular disease and arsenic exposure at water arsenic concentrations less than 100 μg/L (Moon *et al.*, 2012; Tsuji *et al.*, 2014b). Moon *et al*. (2012) conducted a meta-analysis and derived statistically significant pooled relative risk ratios for cardiovascular disease (1.32, 95%CI 1.05-1.67), coronary heart disease (1.89, 95%CI 1.33-2.69) and peripheral arterial disease (2.17, 95%CI 1.47-3.20), but not stroke (1.08, 95%CI 0.98-1.19) and high drinking water arsenic (>50 μg/L). Tsuji *et al*. (2014) concluded that the literature supported a water arsenic concentration of 100 μg/L as a toxicological no observed adverse effect level (NOAEL). This water arsenic level was used to derived a NOAEL dose for iAs of 9 μg/kg bw/day and applied an uncertainty factor of up to three to define a chronic reference dose (RfD) for cardiovascular disease of 3-9 μg/kg bw/day.

Diabetes

A US study recruited diabetes-free participants, aged 45-75 years (*n* = 1694) and followed for a cumulative 11,263.2 person years (Kuo *et al.*, 2015). The proportions of iAs, MMA and DMA in urine were determined. Diabetes developed in 396 participants. Higher diabetes incidence was associated with lower %MMA and higher %DMA in urine. The authors concluded that arsenic metabolism, but not arsenic exposure was associated with diabetes incidence.

American Indians from Arizona were recruited into a study of diabetes development (Kim *et al.*, 2013). Study participants were tested every two years between 1965 and 2007. Cases (*n* = 150) were defined as participants who were greater than 25 years of age and non-diabetic between 1982 and 1989, but developed diabetes before 2007. Each case was matched to a control who remained non-diabetic at the conclusion of the study. Arsenic species were

determined in urine samples from cases and controls. Odds ratios for development of diabetes and various urinary arsenic parameters were all insignificant. However, *post hoc* analysis, comparing diabetes occurrence across quartiles of tAs and iAs exposure found a significant two-fold higher risk of diabetes in the upper three quartiles of iAs exposure compared to the first quartile.

In a cohort study (*n* = 374) in Chihuahua, Mexico, arsenic exposure was assessed in terms of iAs, MMA or DMA in exfoliated urothelial cells (EUCs) (Currier *et al.*, 2014). Associations between arsenic species and diabetes were determined by logistic and linear regression. Significant odds ratios were found for As^{3+} and MMA^{III} in EUCs and diabetes.

A cross-sectional study, involving 1004 men and women, was carried out in Bangladesh (Islam *et al.*, 2012). Arsenic exposure was determined in terms of household drinking water arsenic concentrations. The prevalence of type 2 diabetes in the cohort was 9% (95% CI 7-11%). After adjusting for known diabetes risk factors, multiple logistic regression showed an increased risk of type 2 diabetes associated with water arsenic concentrations >50 μg/L. Those in the highest quartile of water arsenic (>262 μg/L) had almost double the risk of type 2 diabetes.

In 2011, the US National Toxicology Program (NTP) convened a workshop that considered the evidence for a causative relationship between arsenic exposure and diabetes (Maull *et al.*, 2012). The workshop concluded that existing human data provides "limited to sufficient support for an association between arsenic and diabetes in populations with relatively high exposure levels (≥ 150 μg arsenic/L in drinking water). The evidence is insufficient to conclude that arsenic is associated with diabetes in lower exposure (< 150 μg arsenic/L drinking water), although recent studies with better measures of outcome and exposure support an association. The animal literature as a whole was inconclusive; however, studies using better measures of diabetes-relevant end points support a link between arsenic and diabetes".

Birth outcomes

A birth cohort of 299 pregnant women-newborn pairs was recruited in Taiwan during 2001-2002 (Chou *et al.*, 2014). Arsenic exposure in mothers was assessed by urinary analysis for iAs and methylated metabolites and analysis of two DNA damage biomarkers. The only significant association between arsenic biomarkers and birth characteristics were negative associations between maternal urinary iAs or one of the DNA damage biomarkers and Apgar score at 1 minute. The Apgar score is assigned by a paediatrician or trained nurse based on breathing effort, heart rate, muscle tone, reflexes and skin colour. A significant positive association was also seen between maternal urinary MMA and Apgar score. No associations were found with gestational age, birth weight, birth length, head circumference, chest girth or 5-minute Apgar score.

5.2.3 Risk assessments

International

Meharg *et al*. (2009) used food balance sheet data to estimate rice consumption in 10 countries (Bangladesh, China, Egypt, France, India, Italy, Japan, Spain, Thailand and the USA). Where available, these were combined with country-specific median iAs concentrations for rice to give estimates of dietary exposure to iAs from rice consumption. A cancer slope factor of 3.67 $(mg/kg/day)^{-1}$, derived by the USEPA for skin cancer, was used to calculate cancer risks associated with iAs exposure. Calculated excess cancer rates ranged from 0.7 per 10,000 population (Italy) to 22.1 per 10,000 population (Bangladesh). It

is worth noting that USEPA have subsequently revised their cancer slope factors, basing them on lung and bladder cancer, rather than skin cancer (US Environmental Protection Agency (USEPA), 2010).

Oberoi *et al*. (2014) used GEMS/Food cluster diets food consumption data[9], global literature mean iAs concentration data and cancer slope factors for skin, lung and bladder cancer to derived global estimates of the burden of these cancer due to iAs exposure. The burden of skin cancer due to foodborne arsenic was estimated to be 10,730-110,014 cases, lung cancer 11,844-121,442 cases and bladder cancer 9,129-119,176 cases. Approximately half of the total global burden for all three cancer types was estimated to be due to cases occurring in GEMS/Food cluster G, a group of countries including Pakistan, India, Sri Lanka, China, Vietnam, Cambodia, Laos, Thailand, Malaysia, Singapore and Indonesia.
Chen *et al*. (2015) estimated exposure to iAs of 0.041 μg/kg bw/day in sub-Saharan Africa due to consumption of root and tuber vegetables and plantains. Carcinogenic risk was characterised by two different methods. A margin of exposure[10] of 72 was determined by comparison with the $BMDL_{0.5}$ for lung cancer, determined in the JECFA assessment (JECFA, 2011). While no international standards for acceptable margins of exposure have been agreed, EFSA has proposed that margins should be greater than 10,000 for genotoxic carcinogens (EFSA, 2005). Lifetime cancer risk was also determined, using the USEPA cancer slope factor of 3.67 $(mg/kg/day)^{-1}$ for skin cancer. The mean risk over a 30 year period was assessed by simulation to be 6.3×10^{-5} (6.3 excess cancers per 100,000 population).

China, Shandong Province

A cancer slope factor of 1.5 $(mg/kg\ bw/day)^{-1}$ to calculate excess cancer risks for the general population, fish farmers and island dwellers due to consumption of seafood (Wu *et al.*, 2014). Risk estimates ranged from 1.5×10^{-4} (15 excess cancers per 100,000) for the general population to 5.27×10^{-4} (52.7 excess cancers per 100,000) for island dwellers.

Finland

Risks associated with dietary exposure to iAs by Finnish adults, from consumption of rice, and children, from consumption of rice-based infant foods, were assessed using a margin or exposure approach (Rintala *et al.*, 2014). Exposures were assessed against the lowest $BMDL_{01}$ determined in the EFSA assessment of 0.3 μg/kg bw/day (EFSA, 2009). Margins of exposure were in the range 0.5-1.1.

India

A study in the Nadia district of West Bengal estimated the cancer risk from arsenic in cooked rice, using a USEPA cancer slope factor of 1.5 $(mg/kg/day)^{-1}$ (Mondal and Polya, 2008). Based on a Monte Carlo simulation, the median lifetime risk (maximum 40 years exposure) was 7.62×10^{-4}. The cancer risk from rice consumption was marginally higher than the risk from water consumption in this region. The total risk, from consumption of rice and drinking water, was 1.48×10^{-3} (148 excess cancer per 100,000).

[9] http://www.who.int/nutrition/landscape_analysis/nlis_gem_food/en/ Accessed 15 July 2015
[10] The margin of exposure (MOE) is calculated by dividing a defined point on the dose-response curve (point of departure), such as the benchmark dose (BMD), by estimates of exposure

USA

Estimates of combined lung and bladder cancer cases from consumption of apple juice were derived (Carrington *et al.*, 2013). Estimates were derived for various potential regulatory limits for iAs in apple juice. In other words, the distribution of iAs concentration in apple juice was truncated at various levels, corresponding to potential regulatory limits. At average apple juice consumption and a regulatory limit for iAs of 50 μg/L (essentially no limit) excess cancer cases per million of 67.8-96.6 were estimated, while at a regulatory limit of 10 μg/L these estimates were reduced to 13.1-18.7. Restriction of concentration data to more recently collected data further decreased the estimated number of cancer cases.

The risks associated with iAs in chicken meat (geometric mean concentration 2 μg/kg) were assessed for a putative 80 kg adult, consuming 82.4 g/day of chicken (Nachman *et al.*, 2013). Using an USEPA derived cancer slope factor of 25.7 $(mg/kg\ bw/day)^{-1}$ for lung or bladder cancer, it was estimated that this level of iAs exposure would result in an additional 3.7 cancers per 100,000 population.

6. RISK MANAGEMENT INFORMATION

6.1 RELEVANT FOOD CONTROLS: NEW ZEALAND

6.1.1 Establishment of regulatory limits

Standard 1.4.1 of the Australia New Zealand Food Standards Code specifies a maximum limit (ML) to tAs in cereals of 1000 μg/kg (1 mg/kg) and an ML for iAs in crustacea and fish (2000 μg/kg) and molluscs and seaweed (1000 μg/kg).[11]

6.1.2 Imported Food Requirements (IFR)

There is a current IFR for hijiki seaweed, requiring product imported into New Zealand to contain less than 1 mg/kg (1000 μg/kg) of iAs (Ministry for Primary Industries, 2015).

6.2 RELEVANT FOOD CONTROLS: OVERSEAS

6.2.1 Establishment of regulatory limits

The Codex Committee on Contaminants in Foods (CCCF) first proposed an ML for arsenic (iAs or tAs) in raw rice of 0.3 mg/kg (300 μg/kg) and in polished rice of 0.2 mg/kg (iAs) (CCCF, 2012). In addition, CCCF made several recommendations:

- It is preferable to set MLs specifically for inorganic arsenic rather than total As. However to do this further data needs to be sourced as currently there is insufficient robust occurrence data for inorganic As in raw commodity and processed rice products to set MLs.
- The Committee should ask the Codex Committee on Methods of Analysis and Sampling (CCMAS) to establish the method for determination of inorganic As in rice. The sampling method for contaminants directives (EC 333/2007) should be made available to CCMAS as potential starting point.
- Consideration should be given to the value of developing a Code of Practice which could address factors that influence inorganic arsenic levels in rice and rice products e.g. As content of soil and water, processing and cooking procedures.
- If a ML is set based on the current level of knowledge then it should be set with reference to both total and inorganic As i.e. draft MLs for As in raw rice (brown) would be proposed at 0.3 mg/kg , whether for inorganic As or total As; or 0.2 mg/kg only for inorganic As in polished rice. It might be measured for total As first, and then measured as inorganic As if the total As measurement exceeds 0.3 mg/kg.

CCCF also reviewed current regulatory limits pertaining to arsenic in rice. In addition to the limit for tAs in cereals in Australia and New Zealand, a small number of relevant regulatory limits were identified:

- China; 0.15 mg/kg iAs in rice and rice products
- India; 1.1 mg/kg tAs (uncertainty whether this only applies to rice)
- Mercosur; 0.3 mg/kg tAs in rice
- Singapore; 1 mg/kg tAs
- United Kingdom; 1 mg/kg tAs applicable to all foods

[11] https://www.comlaw.gov.au/Details/F2015C00052 Accessed 16 July 2015

The proposed draft ML for polished rice was forwarded for adoption at the 8th Session of CCCF in the Hague (CCCF, 2014). The draft ML for husked rice (previously referred to as raw rice) was returned to the Electronic Working Group for redrafting. The proposed redrafted ML of 0.35 mg/kg iAs in husked rice (CCCF, 2015c) was forwarded at the 9th Session of CCCF in New Delhi (CCCF, 2015b).

6.2.2 Codes of Practice

A discussion paper on a the possibility of developing a Code of Practice for the prevention and reduction of arsenic contamination in rice was presented to the 7th Session of CCCF in Moscow (CCCF, 2013). The proposed draft Code of Practice was discussed at the 9th Session of CCCF in New Delhi (CCCF, 2015a). The proposed draft Code of Practice includes aspects related to:

- Source directed measures, specifically identification and management of soils and irrigation waters with high arsenic contents, atmospheric emissions and wastewater from industries, materials used in agricultural and livestock production and waste containing arsenic
- Agricultural measures, including farmer education, consideration of use of aerobic or intermittent ponding during rice production and identification of low-arsenic rice cultivars
- Processing and cooking measures, including sharing information on the potential of rice polishing and washing to reduce arsenic content and informing consumers to avoid using highly arsenic contaminated water for cooking rice
- Monitoring, including monitoring of the effectiveness of measures implemented and monitoring soil arsenic in potentially contaminated areas

6.3 INFLUENCE OF FOOD PROCESSING ON ARSENIC SPECIES LEVELS

6.3.1 Grain storage

Storage of rice at 15 or 25ºC for up to 12 months had no appreciable impact on the tAs, iAs or DMA content of the rice (Naito *et al.*, 2015).

6.3.2 Vegetable processing

Arsenic concentrations are much higher in the skin of potatoes, carrots, beetroot and garlic than in the peeled vegetables (JECFA, 2011; Munoz *et al.*, 2002).

6.3.3 Rice processing

White rice is produced by abrading or polishing off the bran layer of brown rice. Polishing of rice decreased both the tAs and iAs concentration of the rice grains (Naito *et al.*, 2015). However, iAs appears to be removed at a greater rate, with the iAs/tAs proportion decreasing with the degree of polishing. No significant changes were seen in the DMA content of rice grains. The resulting bran contained up to five times the tAs concentration of the unpolished rice, with essentially all the arsenic present as iAs. DMA was not detected in rice bran.

Washing of brown rice had little impact on the tAs or iAs content (Naito *et al.*, 2015). However, washing of polished rice reduced the tAs concentration by up to 30% for high arsenic rice (>100 μg/kg) and by up to 95% for low arsenic rice (<50 μg/kg). Decreases in iAs mirrored decreases in tAs, while DMA concentrations were not influenced by washing. It is likely that the washing removed residual bran material adhering to the grain following the polishing process.

6.3.4 Cooking

Both increases and decreases in the arsenic content of food during cooking have been reported. However, it appears that increases are due to weight losses in the food during cooking, while decreases may be due to dilution due to water uptake or solubilisation into the cooking medium (Devesa *et al.*, 2008).

The influence of rice cooking on the arsenic content of the cooked rice is critically dependent on the arsenic content of the cooking water (Rahman and Hasegawa, 2011). A study in West Bengal reported that the mean tAs concentration in cooked rice (170 μg/kg) was higher than that in the raw rice (130 μg/kg) (Mondal and Polya, 2008). The tAs content of cooking water was described as low for this regions, but was still in the range 1.5 to 46 μg/L. In should be noted that the maximum acceptable value (MAV) for tAs in drinking water in New Zealand is 10 μg/L.[12]
Cooking of rice in deionised water resulted in modest decreases in tAs (2-33%) and iAs (0-28%), but no significant change in DMA (Naito *et al.*, 2015). There were negligible or no changes in the tAs and iAs concentrations of brown rice during cooking.

Cooking (175ºC for 30 minutes) of chicken demonstrated that concentrations of the organoarsenic animal therapeutic, roxarsone (20 μg/kg), and an unidentified arsenic species (10 μg/kg) decreased to <2 μg/kg, while the concentration of iAs increased from 11 to 42 μg/kg (Nachman *et al.*, 2013).

6.3.5 Fermentation

There are conflicting reports on the fate of arsenic species during fermentation. Fermentation of fish (sardine, sandfish, squid) to produce fish sauce was reported to result in a change from AB being the dominant species in the raw fish to DMA being the dominant species in the fermented fish sauce (Kato *et al.*, 2004). No change in the total amount of arsenic present was noted.

Analysis of six commercial fish sauces for arsenic species found that AB was the dominant species (84-92%), followed by AC (4.9-7.7%) (Rodriguez *et al.*, 2009). DMA was not detected in any of the fish sauces analysed.

It is plausible that the differences between the findings of these two studies were due to the presence of different fermenting microbial species. In particular, the study of Kato *et al*. (2004) suggests the presence of species with significant capacity to metabolise AB.

[12] http://www.health.govt.nz/system/files/documents/publications/drinking-water-standards-2008-jun14.pdf Accessed 21 July 2015

7. CONCLUSIONS

7.1 DESCRIPTION OF RISKS TO NEW ZEALAND CONSUMERS

It is well accepted that iAs is carcinogenic, causing cancer of the skin, lung and urinary bladder (ATSDR, 2007; EFSA, 2009; IARC, 2012; JECFA, 2011). There is weaker evidence for causation of cancers at other sites and for non-cancer endpoints, such as cardiovascular disease and diabetes. There is increasing evidence that disease mainly results from chronic exposure to high levels of arsenic. Molecular evidence supports this observation and suggests that there may be a threshold to the toxic effects of iAs, including carcinogenic effects. The toxic mode of action of iAs is not known, but it is believed that it involves several mechanisms, including oxidative stress, non-mutagenic genotoxicity and epigenetic changes.

Several recent meta-analyses and systematic reviews have questioned whether associations between iAs exposure and some disease states are significant at iAs water concentrations less than 100 μg/L. The maximum acceptable values (MAVs) for drinking water in New Zealand are based on a consumption level of 2 L/day.[13] Based on a conservative body weight of 60 kg, a water arsenic concentration of 100 μg/L equates to iAs exposure of 3.3 μg/kg bw/day. This level of exposure is near the bottom of the range of $BMDL_{0.5}$ (2-7 μg/kg bw/day) determined by JECFA and within the range of $BMDL_1$ (0.3-8 μg/kg bw/day) determined by EFSA.

While tAs has been determined in a wide range of foods consumed by New Zealanders, little direct evidence is available for the iAs content of the New Zealand food supply. However, dietary exposure estimates, derived by application of conservative assumptions to tAs data, do not suggest that New Zealanders have particularly high dietary exposure to iAs (0.2-0.5 μg/kg bw/day), when compared to other international estimates of dietary exposure. It was noted that, given the consistency of successive estimates of tAs exposure in New Zealand, exposure to tAs and iAs in New Zealand is likely to already be as low as reasonably achievable (ALARA). However, as has been noted in international assessments of iAs (EFSA, 2009; JECFA, 2011), dietary exposures are similar to benchmark doses, indicating negligible margins of exposure.

For determination of individual arsenic species in foods a hyphenated technique, linking chromatographic or electrophoretic separation with sensitive arsenic determination would be required. Such techniques are not currently offered by any New Zealand analytical providers. Determination of iAs can be successfully achieved by hyphenated, as well as simpler non-chromatographic, methods. There is no evidence to suggest that there is a 'preferred method' for iAs analysis and method performance data would be a better guide to the suitability of a method-provider combination. One New Zealand laboratory (Cawthron Institute) is currently accredited for iAs analysis.

7.2 COMMENTARY ON RISK MANAGEMENT OPTIONS

There are relative few risk management options available for control of iAs exposure in New Zealand. MLs and an IFR are in place to control exposure to highly contaminated seafood, cereals and hijiki seaweed.

[13] http://www.health.govt.nz/system/files/documents/publications/drinking-water-standards-2008-jun14.pdf Accessed 24 July 2015

New Zealand does not grow any appreciable amounts of rice, so the risk management options included in the draft Codex Code of Practice are not relevant in the domestic New Zealand context.

7.3 DATA GAPS

While it is unlikely to change the profile of risks associated with arsenic species in New Zealand, information on the arsenic species composition and content of New Zealand foods would establish whether they are similar to the composition and content of arsenic species in foods reported in the literature.

It is known that some drinking water supplies in the central North Island of New Zealand contain arsenic at concentrations approaching 100 μg/L. With the exception of a study demonstrating very high tAs concentrations in watercress from the Waikato River, it is not currently known whether other foods from this area are similarly high in arsenic. If so, consideration of region specific risk assessments may be warranted. To date, the NZTDS has not included potentially high arsenic areas among its sampling sites.

While information is unlikely to come from New Zealand, further information informing the potential for a dose threshold for the adverse health effects due to arsenic is required.

Information on the toxicity of organoarsenic species and the mechanism by which arsenic species exert their toxicity would assist risk assessment.

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RISK PROFILE: CHEMICAL FORMS OF CONTAMINANT ELEMENTS (SPECIES). PART 2: CHROMIUM, SELENIUM AND VANADIUM

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CONTENTS

LIST OF TABLES

LIST OF FIGURES

EXECUTIVE SUMMARY

The purpose of a Risk Profile is to provide contextual and background information relevant to a food/hazard combination so that risk managers can make decisions and, if necessary, take further action. Risk Profiles include elements of a qualitative risk assessment, as well as providing information relevant to risk management. Risk profiling may result in a range of activities, such as immediate risk management action, a decision to conduct a quantitative risk assessment, or a programme to gather more data. Risk Profiles also provide information for ranking of food safety issues.

Chromium, selenium and vanadium are trace components of the diet. For chromium and vanadium there is still conjecture as to whether they are essential trace nutrients or toxic contaminants. While the essentiality of selenium is well established, there is a relatively narrow margin between exposures that satisfy nutritional requirements and exposures that are toxic for this element. All three elements may occur in the diet in different chemical forms, which may impact on their bioavailability and toxicity.

Chromium

Chromium occurs in two main oxidation states; Cr(III) and Cr(VI). The hexavalent form of chromium, Cr(VI), is categorised by the International Agency for Research on Cancer (IARC) as Group 1, carcinogenic to humans. However, this classification mainly relates to occupational exposure by the inhalation exposure route and there is limited evidence of causation of cancer by oral exposure, based on published epidemiological studies to date. Trivalent chromium, Cr(III), is considered to be relatively non-toxic and may be an essential trace element.

There are also some questions over the occurrence of Cr(VI) in food samples, with suggestions that the apparent detection of this chromium species may be an artefact of the analytical methods used. Until further evidence is presented and analytical methods become standardised, evidence for exposure to Cr(VI) from food remains tentative. The concentration of total chromium in foods studied in New Zealand in 1982 and 1987/88, are of comparable levels to international studies, in the parts per billion range.

Selenium

Selenium is an essential trace element in the human diet, due to its inclusion in specific human selenoproteins and particularly seleno-enzymes, where selenium is a component of the enzyme active site. Adverse health effects in humans can result from either selenium deficiency or excessive selenium intake. There is some evidence to suggest a beneficial impact of selenium intakes between nutritional and toxic levels on a number of disease states, including cancer. However, the evidence for such beneficial effects are equivocal. There are reasonably consistent associations between higher selenium status and reduced disease risk. However, the ability of supplementation, in the form of selenite, selenate or selenomethionine (SeMet) or in the form of selenium enriched foods, to reduce disease risk is not supported by the weight of evidence, as measured by meta-analyses.

Despite New Zealand being recognised as a low-selenium environment, supported by studies showing low dietary intake of selenium and low levels of biomarkers, adverse effects due to selenium deficiency have not been identified in New Zealand. There is evidence that the selenium intake and status of New Zealanders has increased in the last 30 years.

SeMet is the main form of selenium present in most plant foods and is almost the sole form in animal products, except for the small amounts of selenocysteine (SeCys) present in

functional selenoproteins. SeMet has some nutritional advantages, as its nonspecific incorporation into body proteins represents a storage form of selenium, in the event of periods of insufficient selenium dietary intake. It is unclear what the toxicological implications are of intake of different selenium species.

Vanadium

Vandium pentoxide (V(V)) has been identified by IARC as a possible carcinogen (Group 2B), on the basis of animal studies, however there is no evidence that vanadium is a risk to humans through food consumption. Adverse effects in humans orally exposed to vanadium have generally been confined to gastrointestinal systems and it is uncertain whether these symptoms are due to a toxicological response or to local irritation of the gastrointestinal tract. While there is suggestive information in some animal species, it has still not been established that vanadium is an essential trace element in humans. Vanadium compounds have been used as supplements by body builders and in clinical trials for people with diabetes mellitus at daily doses three orders of magnitude above international estimates of dietary exposure.

There is insufficient data to comment on the potential impact of vanadium speciation on risks associated with dietary exposure to vanadium.

1. INTRODUCTION

The purpose of a Risk Profile is to provide information relevant to a food/hazard combination(s) so that risk managers can make more informed decisions and, if necessary, take further action. Risk Profiles are part of the Risk Management Framework (RMF)[1] approach taken by the Ministry for Primary Industries (MPI) Food Safety. The Framework consists of a 4-step process:

- Preliminary risk management activities
- Identification and selection of risk management options
- Implementation of control measures, and
- Monitoring and review

This initial step in the RMF, Preliminary Risk Management Activities, includes a number of tasks:

- Identification of food safety issues
- Risk profiling
- Establishing broad risk management goals
- Deciding on the need for a risk assessment
- If needed, setting risk assessment policy and commissioning of the risk assessment
- Considering the results of the risk assessment
- Ranking and prioritisation of the food safety issue for risk management action.

Risk profiling may be used directly by risk managers to guide identification and selection of risk management options, for example where:

- Rapid action is needed;
- There is sufficient scientific information for action;
- Embarking on a risk assessment is impractical.

1.1 HAZARDS AND RISK MANAGEMENT QUESTIONS

This Risk Profile addresses the chemical species of chromium, selenium and vanadium that can occur in food. It should be noted that these elements are widespread in the biosphere and will be present, at some level in all foods, although concentrations may differ markedly between food types. Consequently, this Risk Profile will not focus on specific foods, apart from increased emphasis on foods known to be significant contributors to dietary exposure. In the context of this Risk Profile, chemical species are different oxidation states of the same element (e.g. Cr(III) and Cr(VI)) or different stable molecules incorporating the element (e.g. selenomethionine, selenocysteine).

The current Risk Profile does not address issues related to chromium, selenium or vanadium in animal feed or associated animal health issues, except where these are relevant to human health.

The sections in this Risk Profile are organised as much as possible as they would be for a conventional qualitative risk assessment.

Hazard identification, including:

[1] http://www.foodsafety.govt.nz/elibrary/industry/RMF_full_document_-_11604_NZFSA_Risk_Management_Framework_3.1.pdf accessed 22 July 2015

- A description of the chemical(s).

Hazard characterisation, including:
- A description of the adverse health effects caused by the chemical.
- Dose-response information for the chemical in humans, where available.

Exposure assessment, including:

- Data on the occurrence of the hazard in the New Zealand food supply.
- Data on the consumption of relevant foods by New Zealanders, where appropriate.
- Qualitative estimate of exposure to the chemical (if possible).
- Overseas data relevant to dietary exposure to the chemical.

Risk characterisation:
- Information on the number of cases of adverse health effects resulting from exposure to the chemical with particular reference to the identified food (based on surveillance data) or the risk associated with exposure (based on comparison of the estimated exposure with exposure standards).
- Qualitative estimate of risk, including categorisation of the level of risk associated with the chemical in the food.

Risk management information
- A description of relevant food safety controls.
- Information about risk management options.

Conclusions and recommendations for further action

1.2 MAIN INFORMATION SOURCES

Information on the toxicology of and/or exposure to chromium, selenium and vanadium has been reviewed or otherwise considered by a number of groups. These assessments were major resources for the current project. Sources included, but were not limited to:

- JECFA (the Joint FAO/WHO Expert Committee on Food Additives). Assessment reports were accessed at: http://www.inchem.org/
- EFSA (European Food Safety Authority). Opinions were accessed at: http://www.efsa.europa.eu/
- IARC (the International Agency for Cancer Research). Monographs were accessed from ESR's standing collection or at: http://monographs.iarc.fr/ENG/Monographs/PDFs/index.php. Summaries can be accessed at: http://www.inchem.org/
- ATSDR (the US Agency for Toxic Substances and Disease Registry). Toxicological profile documents were accessed at: http://www.atsdr.cdc.gov/toxprofiles/index.asp
- USEPA (the US Environmental Protection Agency). Human health and environmental assessments are available through the IRIS (Integrated Risk Information System) portal: https://www.epa.gov/iris

More recent and additional information than that included in these resources was located by general searching of the World Wide Web (internet) and use of specific citation databases, including:

- PubMed. Accessed at: http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?DB=pubmed
- Web of Science. Accessed at: http://apps.webofknowledge.com/

2. HAZARD IDENTIFICATION - CHROMIUM

Chromium is a transition metal ubiquitous in the environment from erosion of chromium-containing rocks. Uses of chromium in industry include stainless steel production for its corrosion resistant properties, leather tanning, wood preservatives and catalysts (EFSA, 2014c). The name chromium comes from the Greek word chroma, meaning coloured, as evident in many brightly coloured chromium-containing compounds such as pigments and dyes (IARC, 1990).

2.1 STRUCTURE AND NOMENCLATURE

Chromium has a wide range of oxidation states from -2 to +6. The most stable and commonly occurring states are trivalent (chromium +3, Cr(III)), a form which is considered an essential element to humans and found in food, and hexavalent (chromium +6, Cr(VI)), which is toxic and a known carcinogen (EFSA, 2014b; c). Therefore, the focus of this chromium section is on Cr(VI). The stable isotopes of chromium are ^{50}Cr, ^{52}Cr, ^{53}Cr and ^{54}Cr with abundances 4.3%, 83.8%, 9.5%, and 2.4%, respectively (EFSA, 2014c).

A list of selected common chromium containing compounds is given in Table 1 (IARC, 1990).

Table 1. Selected chromium compounds, their formula and CAS No.

Compound	Formula	CAS No.	Synonym
Chromium(III) compounds			
Chromic acetate	$Cr(CH_3COO)_3$	1066-30-4	
Chromic chloride	$CrCl_3$	10025-73-7	
Chromic hydroxide	$Cr(OH)_3$	1308-14-1	
Chromium hydroxide sulphate	$Cr(OH)SO_4$	12336-95-7	Chrome sulphate
Chromic nitrate	$Cr(NO_3)_3$	13548-38-4	
Chromite ore	$Cr_2O_3.FeO$	1308-31-2	
Chromium(VI) compounds			
Barium chromate	$BaCrO_4$	10294-40-3	C.I. Pigment Yellow 31
Lead chromate	$PbCrO_4$	7758-97-6	C.I. Pigment Yellow 34
Lead chromate oxide	$PbO.PbCrO_4$	1344-38-3	C.I. Pigment Orange 21
Calcium chromate	$CaCrO_4$	13765-19-0	C.I. Pigment Yellow 33
Potassium chromate	K_2CrO_4	7789-00-6	
Potassium dichromate	$K_2Cr_2O_4$	7778-50-9	

CAS No.: Chemical Abstract Service Registry Number

2.2 OCCURRENCE

Food is the major source of total chromium exposure for the general population accounting for >90% of total exposure. Some overseas studies have shown that drinking water may also be a significant contributor if chromium levels in the water are high. Of the European studies reviewed, EFSA determined the chromium concentration in staple foods was low with the main sources of total chromium being processed meats, whole grain products, pulses and

spices. Smaller amounts were found in dairy products, fruit, and vegetables (EFSA, 2014c). In Australian and New Zealand studies across a range of foods in the diet, moderate contributors to total chromium in the diet were processed meats (ESR/MoH, 1994; FSANZ, 2011).

Copper, chromium and arsenic (CCA) is a pesticide used for the preservation of timber in outdoor settings. Uses include decking, garden furniture, landscaping, fencing, playground equipment, and agricultural posts. CCA is available in a number of different formulations with typical ratios of copper, chromium and arsenic of 23-25%, 38-45%, and 30-37%, respectively. Chromium is present as Cr(VI), including compounds such as chromium trioxide and sodium dichromate. Copper is included in the formulation to control fungal growth, while arsenic controls insect populations, such as borer. Cr(VI) is reduced by the timber's organic matter to Cr(III). Cr(III) is insoluble resulting in the fixation of copper and arsenic in the wood. A report by Environmental Risk Management Authority New Zealand (ERMA, now the Environmental Protection Authority) concluded that, while the bioavailability of chromium from CCA-treated wood in soil requires further research, any uptake by plants would be confined to the roots (including root vegetables). The effects of exposure to chromium from CCA-treated wood may differ from the individual effects of exposure to copper, chromium and arsenic in isolation (ERMA, 2003). The Agency for Toxic Substances and Disease Registry (ATSDR) advises CCA-treated timber should not be burnt, or used as mulch and wood chips.[2] The United States Environmental Protection Agency (USEPA) advises against the treated timber being used for raised vegetable beds.[3] There is not enough evidence to conclude whether CCA-treated timber would cause adverse health effects to the public if treated timber were to be used in domestic and commercial food production (within close proximity to food production), or burning the timber for the smoking foods.

2.2.1 Hexavalent chromium (Cr(VI))

Traditionally chromium analysis has concentrated on total chromium levels without separation and quantitation of the different relevant species, Cr(III) and Cr(VI). In recent years through the development of new technologies the scientific community has slowly advanced into determining Cr(VI) levels in food and if present, exposure to humans.

Cr(VI) has been reported in foods at low levels, albeit in a limited number of studies. Such studies include the works of Ambushe *et al.* (2009); Figueiredo *et al.* (2007); Lameiras *et al.* (1998); Mahmud *et al.* (2011); Mandiwana *et al.* (2011); Paleologos *et al.* (1998); Soares *et al.* (2000); Soares *et al.* (2010); Svancara *et al.* (2004); Vieira *et al.* (2014).

A study in Slovenia has presented findings that show Cr(VI) is not present in foods of terrestrial (plant) origin and predicts that Cr(VI) does not exist in foods of animal origin (Novotnik *et al.*, 2013). This is anomalous compared to other studies.

The bioconcentration factor (BCF) for Cr(VI) was studied in freshwater fish. BCF was determined to be low at approximately 1. This is compared to a BCF of approximately 100 for total chromium, resulting from the reduction of Cr(VI) to Cr(III) in organisms such as freshwater fish (IPCS, 2013). There is no current evidence that chromium bioaccumulates up the terrestrial food chain (EFSA, 2014c).

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[2] http://www.atsdr.cdc.gov/CCA-Treated_Wood_Factsheet.pdf Accessed 4 May 2016.
[3] https://www.epa.gov/ingredients-used-pesticide-products/chromated-copper-arsenate-cca Accessed 4 May 2016.

2.2.2 Trivalent chromium (Cr(III))

Similarly to Cr(VI) analysis, most studies of chromium do not include details on speciation, or separated Cr(III). In many reports, it has been presumed that total chromium concentration is present in the form of Cr(III).

2.3 ANALYTICAL METHODS

There are few published analytical methods for chromium speciation in food and over recent years few advances in limits of detection and specificity have occurred, restricting those methods for use as research tools. Validation techniques across the methods employed are varied with no consistency in extraction techniques. The EFSA noted that no standardised methods were available for Cr(VI) determination in foods (EFSA, 2014c).

The majority of methods for the analysis of chromium species in food employ a two-step process:

- Extraction and separation of the species from the food matrix
- Detection of the chromium species

While this section will focus on the analysis of Cr(VI), it should be noted that determination of total chromium will simply involve dry or wet digestion of the food matrix in combination with the detection systems discussed for Cr(VI).

2.3.1 Sample extraction and separation specifically for Cr(VI)

Published extraction techniques for Cr(VI) broadly fall into two categories of aqueous extractions. The first type of extraction is a simple aqueous alkaline extraction followed by direct detection (Mandiwana *et al.*, 2011; Novotnik *et al.*, 2013; Soares *et al.*, 2010). Cr(VI) is soluble and therefore amenable to alkaline conditions, whereas Cr(III) is sparingly soluble and is strongly absorbed to organic and mineral surfaces (Izbicki *et al.*, 2015; Kotas and Stasicka, 2000). In addition to an alkaline extraction, Vacchina *et al.* (2015) ultra-filtered the alkaline extraction solution from the samples with a 10kDa molecular weight cut-off filter prior to analysis.

The second type of extraction employs an acidic extraction with alizarin, used to bind Cr(III) to prevent oxidation, followed by Chromabond solid phase extraction (SPE) for specific Cr(VI) retention and subsequent elution with nitric acid (Ambushe *et al.*, 2009; Lameiras *et al.*, 1998; Soares *et al.*, 2000; Vieira *et al.*, 2014).

Cr(VI) detection is then by the instrument of choice of the researcher, the instruments of which are described in the section below (2.3.2).

2.3.2 Sample extraction and detection for all chromium species

The selective extraction of Cr(VI) in foods has typically involved extraction of total chromium using a harsh acid digestion, with microwave assisted digestion and/or hydrogen peroxide. Such methods are called “off-line” methods, for the individual determination of chromium species after sample pre-treatment. It is then possible to quantitate Cr(III) if desired through the derivation of the difference between total chromium concentration (described above) and Cr(VI) concentration (EFSA, 2014c).

Two recent studies use high-performance liquid chromatography (HPLC) as a separation technique between Cr(III) and Cr(VI) coupled to their analytical instrument of choice (hyphenated) enabling simultaneous detection of Cr(III) and Cr(VI), otherwise known as "on-line methods" (Novotnik *et al.*, 2013; Vacchina *et al.*, 2015). On-line coupling attempts to extract all the original oxidation states of the chromium species, followed by separation and quantification of Cr(III) and Cr(VI). Other species of chromium between its oxidation states of -2 to +6 are thermodynamically highly unstable and are therefore not detected. This is common practice in the analysis and quantification of Cr(VI) in water, soil and environment samples, for example, the analysis of Cr(VI) drinking water by EPA Standard Method 218.7 utilises HPLC and post-column derivatisation with UV-Visible spectroscopic detection.[4]

Traditional atomic absorption spectrometry (AAS) or electrothermal atomic absorption spectrometry (ETAAS) is commonly used for the detection and quantification of Cr(VI), with limits of detection at low ppb levels. More recently the use of inductively coupled plasma-mass spectrometry (ICP-MS) has seen an increase in the sensitivity of metal ions detection, and in some case a 10-fold increase in Cr(VI) sensitivity compared to AAS techniques (Ambushe *et al.*, 2009; Novotnik *et al.*, 2013). ICP-MS detection is also more amenable to hyphenation with chromatographic techniques, used for separation of ions prior to detection.

One published paper details an alkaline chromium extraction followed by detection using an electrochemical technique, differential pulse cathodic stripping voltammetry (DPCSV) to report Cr(VI) concentration (Svancara *et al.*, 2004). This utilises the reduction properties of chromium to study the conversion of Cr(VI) to Cr(III). The author notes a certified reference material for tea was available and using their DPCSV method gained a recovery of 80%. The study repeated the sample analysis by inductively coupled plasma-atomic emission spectroscopy (ICP-AES). It is unclear based on the extraction techniques and dialogue in the paper whether the author is reporting results for total chromium or Cr(VI). During sample preparation, the extraction solution was heated to ensure the quantitative conversion of Cr(III) to Cr(VI), therefore culminating in all chromium in the extraction solution now present as Cr(VI). DPCSV analysis for Cr(VI) was then conducted at this point. It could be presumed that the quantitative conversion of Cr(III) to Cr(VI) was subtracted from the final Cr(VI) DPCSV analysis, to give Cr(VI) concentration in the starting sample, although it is uncertain whether this is the case. Given the ambiguity, the method is possibly detecting and reporting total chromium.

Speciation of chromium was also studied using flow injection analysis (FIA) with fluorescence detection (Paleologos *et al.*, 1998). Cr(VI) present in the sample is used to selectively oxidise the non-fluorescing reagent 2-(α-pyridyl)thioquinaldinamide (PTQA), generating an intensely fluorescent product.

2.3.3 Chromium species testing in New Zealand

To our knowledge, chromium speciation in food has not been studied in New Zealand, while a few analytical laboratory providers have accredited methods for hexavalent chromium in leather and water.[5] Overseas, accredited methods for Cr(VI) are common for water, soil, leather and environmental samples, but not food.

[4] https://clu-in.org/download/contaminantfocus/chromium/method-218-7.pdf Accessed 5 April 2016
[5] http://www.ianz.govt.nz/directory/ Accessed 18 February 2016

2.3.4 Comparison of method performance

The published studies to date all note that a certified reference material or proficiency programme for Cr(VI) was not available at the time of analysis, therefore no inter-laboratory comparison studies with method accuracy and proficiency have been conducted. It is unknown whether the analysis of Cr(VI) is method dependent. Without this knowledge the published data should remain tentative until the suitability of methods has been reviewed.

3. HAZARD CHARACTERISATION: ADVERSE HEALTH EFFECTS - CHROMIUM

3.1 CONDITIONS

Trivalent chromium, Cr(III), has previously been regarded as an essential trace element with benefits postulated to include efficacy of insulin, and glucose and lipid metabolism (Anderson, 1989). The role of Cr(III) in the body and its classification as an essential element was reviewed by EFSA, who concluded that beneficial effects of Cr(III) intake were not observed in healthy populations, and therefore there is no evidence to suggest chromium is essential in the body. From the review it was determined that formulating an Adequate Intake for chromium was not appropriate (EFSA, 2014b).

The International Agency for Research and Cancer (IARC) defines Cr(III) as Group 3, not classifiable with respect to its carcinogenicity in animals and humans (EFSA, 2014b; IARC, 1990). In contrast, hexavalent chromium, Cr(VI) is a strong oxidiser and classified as a Group 1, carcinogenic to humans (IARC, 1990).

Acute oral toxicity in humans was studied after intentional or accidental poisoning at high doses of Cr(VI). Sources of Cr(VI) were chromic acid, potassium chromate, and ammonium dichromate. Clinical effects of the high dose poisoning in humans included haematological hepatic and renal injury. Respiratory and gastrointestinal lesions were also observed. Lethal doses of Cr(VI) were reported to range from 4 mg/kg bw to 360 mg/kg bw (EFSA, 2014c). The wide range in lethal dose concentrations is not particularly informative without further investigation.

Laboratory rats were found to have lethal oral acute toxicity (LD_{50} mg/kg bw) to Cr(VI) from potassium dichromate of 16.9 (F) and 26.2 (M), sodium chromate 13 (F) and 28 (M), and calcium chromate 108 (F) and 249 (M), respectively (EFSA, 2014c; Gad, 1989; Vernot *et al.*, 1977).[6] Experimental oral exposure studies in animals have linked exposure to Cr(VI) compounds to gastrointestinal cancers (EFSA, 2014c).

3.1.1 Deficiency and excess

Case studies documenting Cr(III) supplementation in total parental nutrition patients have provided inconclusive evidence as to the essentiality of the element in humans (EFSA, 2014b), leading to the conclusion that chromium is probably not essential. To date, no evidence exists of adverse effects related to high intake of Cr(III) (up to 1 mg/day). Deficiency and excess Cr(III) intake is therefore unlikely to be of concern to the general population (EFSA, 2014c).

3.2 TOXICOLOGICAL/NUTRITIONAL ASSESSMENT

A number of recent toxicological reports for chromium species have been published, although there has been little focus on exposure to chromium from foods (ATSDR, 2012b;

[6] F: female, M: male

EFSA, 2014c; IARC, 1990; 2012; National Health and Medical Research Council/Ministry of Health, 2006; USEPA, 1998; 2010)

3.2.1 International Agency for Research on Cancer (IARC)

IARC concluded that:

- Among workers in chromate production plants, almost all studies showed an increased risk for lung cancer (Luippold *et al.*, 2005).
- Chromate pigment production workers showed an elevated risk of lung cancer.
- There is positive correlating evidence between Cr(VI) exposure and occurrence of nasal and nasal sinus cancers.
- There is little evidence that exposure to Cr(VI) causes stomach or other cancers.
- Absorption fractions from human volunteers for Cr(III) and Cr(VI) were 0.13% and 6.9%, respectively.

Based on the evidence it was determined that Cr(VI) compounds are carcinogenic to humans (Group 1), while Cr(III) were defined as Group 3 not classifiable.

3.2.2 European Food Safety Authority (EFSA)

For a substance that is both genotoxic and carcinogenic, it is recommended the margin of exposure (MOE) approach is adopted to characterise the risks from neoplastic effects of Cr(VI). The lower 95% confidence limit for a benchmark response at 10% extra risk ($BMDL_{10}$) for Cr(VI) was defined as 1.0 mg/kg bw/day for the combined incidence of adenomas and carcinomas in the small intestine of mice, as a toxicological point of departure. For carcinoma only, at all sites, a $BMDL_{10}$ of 3.8 mg/kg bw/day was determined.

For substances that are both genotoxic and carcinogenic, an MOE of 10,000 or higher is considered to be of low concern for public health (based on $BMDL_{10}$ animal data) (EFSA, 2005). In the EFSA assessment, for the consumption of drinking water containing Cr(VI), all MOE values were >10,000 for all age groups, except infants where the maximum upper bound (UB) exposure estimate equated to a MOE of 6,300. At the 95th percentile exposure, MOE values below 10,000 were observed at maximum UB exposure estimates for infants (3,100), toddlers (4,200) and children (6,600).

EFSA summarised from recently published speciation work that food is a reducing medium for Cr(VI) to Cr(III) and it is unlikely Cr(VI) would be present in food. Therefore oxidation of Cr(III) species to Cr(VI) would also be unlikely. However it was also assumed in the report by EFSA that all chromium in drinking water was Cr(VI), consequently water added during food preparation (coffee, tea, infant formula, instant soup etc.) will contain Cr(VI). This could lead to a two-fold increase in exposure to Cr(VI), but the report was unable to derive a MOE due to a lack of existing reliable data quantifying Cr(VI) in foods.

For non-cancer effects, an MOE of 100 or higher is considered to be of low concern for public health. Non-cancer effects from Cr(VI) exposure via drinking water (non-neoplastic lesions and haematological effects) where EFSA derived separate $BMDL_{10}$ values had MOEs values of >100.

The EFSA report summarises human environmental exposure in a few retrospective observational studies. The studies were of weak epidemiological design, and in particular there was a lack of recognition of the confounding contribution of Cr(VI) from smoking to human health in the populations studied. Of the studies documented, there was a variety of health end points assessed, including leukaemia, lung cancer, stomach cancer and liver

cancer. Approximately half of the studies determined no significant health effects due to Cr(VI) exposure.

EFSA recommended that:

- Further data should be generated to specifically measure the content of Cr(III) and Cr(VI) in food, using sensitive analytical methods.
- Further data are needed to estimate percentage reduction of Cr(VI) to Cr(III) in the gastrointestinal tract for relevant human exposure.

3.2.3 Agency for Toxic Substances and Disease Registry (ATSDR)

Studies have shown less-water soluble chromium compounds have a longer retention time than more soluble forms in the pulmonary tract. While gastrointestinal absorption of chromium is estimated to be <10% of ingested dose, soluble chromium has greater absorption potential than insoluble chromium. In addition, soluble Cr(VI) has greater absorption than soluble Cr(III). Absorbed chromium is primarily excreted in urine.

Cr(VI) is reduced in the stomach to Cr(III), lowering the absorbed dose from ingested Cr(VI). The reduction process of Cr(VI) to Cr(III) produces reactive intermediates Cr(V) and Cr(IV). Cr(VI) compounds rapidly (seconds to minutes) enter cells by facilitated diffusion while Cr(III) enters cells more slowly (days) by passive diffusion.

With regard to public health, Cr(VI) compounds are of greater concern. Chromium induces many types of DNA lesions, for example, chromium-DNA complexes, DNA adducts, and DNA-protein crosslinks. Evidence also suggests unscheduled DNA synthesis and chromosome aberrations take place. These are potential markers of genotoxicity or cancer.

Minimum risk levels (MRL) are based on non-cancer effects only, to identify the target organ(s) of effect or the most sensitive health effect(s). The MRL is set below levels where the most sensitive individuals can experience a chemical-induced adverse health effect.

Intermediate[7] oral exposure from the study of rats derived an MRL for Cr(VI) of 0.005 mg Cr(VI)/kg/day. No human studies for the effects of intermediate-duration were identified. For chronic[4] oral exposure to Cr(VI), a MRL from the study of mice was determined to be 0.0009 mg Cr(VI)/kg/day for non-cancer adverse health effects. Critical observations for the derivation of chronic exposure in mice included non-neoplastic lesions of the duodenum from exposure to Cr(VI) in drinking water. Intermediate- and chronic- oral duration MRLs for Cr(VI) include uncertainty factors of 10 for the extrapolation from animals to humans, and an additional factor of 10 for human variability. Available animal studies have yet to conclusively determine toxicological end points, therefore the data available is inadequate to derive acute-duration oral MRL for Cr(VI).

MRLs for oral exposure to Cr(III) over acute-, intermediate-, and chronic-durations could not be derived. ATSDR commented the studies of adverse health effects of Cr(III) exposure to humans and animals were inadequate for establishing exposure concentrations.

The EPA and FDA have established a maximum contaminant level of 0.1 mg/L for total chromium in drinking water and total chromium in bottled drinking water, respectively.[8]

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[7] ATSDR defines acute duration 1 - 14 days, intermediate duration 15 - 364 days, and chronic duration >365 days.

[8] http://www.atsdr.cdc.gov/toxfaqs/tfacts7.pdf Accessed 26 February 2016

3.2.4 United States Environmental Protection Agency (USEPA)

A toxicological review of Cr(VI) was conducted by the USEPA in 1998 and subsequently updated in 2010 (available as draft). Very limited data were identified regarding oral exposure to Cr(VI) either from environmental sources or accidental poisoning. The data presented were of very weak epidemiological design (for example, case reports or cross-sectional studies with no control population). Adverse health end-points varied considerably ranging from leukaemia, oral ulcers, indigestion, abdominal pain, and diarrhoea, to kidney failure. USEPA collated data on the lethal dose for Cr(VI) in humans, and found it to range by two-orders of magnitude (approximately 4-350 mg/kg bw), therefore this information currently remains inconclusive.

In the previous assessment conducted by USEPA in 1998, an oral reference dose (RfD) of 0.003 mg/kg bw/day Cr(VI) was derived. This resulted from a no-observed-adverse-effect-level (NOAEL) of 2.5 mg/kg bw/day from a study of rats receiving Cr(VI) spiked drinking water, and an uncertainty factor of 300 (10 for interspecies extrapolation, 10 variability within species, 3 to compensate for the less than lifetime exposure of the study) (USEPA, 1998). It should be noted that the NOAEL was the highest dose used in the study (i.e. no adverse effects were seen at any dose).

USEPA reviewed information on the carcinogenicity of Cr(VI) following oral exposure, but were unable to derive a quantitative risk estimate (unit risk or carcinogenic potency).

3.2.5 National Health and Medical Research Centre (NHMRC)

NHMRC derives nutrient reference values (NRVs) for Australia and New Zealand. NHMRC considered that chromium has a role in the potentiation of insulin. There is a lack of data across all gender-age cohorts for estimated average requirements (EAR) to be set from New Zealand, Australian and worldwide chromium dietary information.

Adequate intakes (AI) for total chromium were determined to be 0.2 µg/day to 5.5 µg/day for infants, 11 µg/day to 35 µg/day for children and adolescents, and 25 µg/day to 45 µg/day for adults. However, upper limits (UL) of intake for chromium have not been defined due to limited data from toxicological studies of chromium in food (National Health and Medical Research Council/Ministry of Health, 2006).

3.3 PROPOSED MECHANISMS OF CARCINOGENICITY

Cr(VI) has been shown to be carcinogenic in animal experiments via oral administration. Increasing rates of squamous epithelium tumours in the oral cavity of rats were reported, along with epithelial tissue in the small intestines of mice (EFSA, 2014c). Studies in bacterial and mammalian cells determined that Cr(VI) compounds are likely to be genotoxic, while intraperitoneal administration of Cr(VI) leads to genotoxicity, indicating that the reductive capacity of the gastrointestinal tract was important (EFSA, 2014c).

While Cr(VI) is efficiently reduced to Cr(III) in the gastrointestinal tract, it is possible that even at low doses, a small percentage of Cr(VI) is not reduced (EFSA, 2014c). Cellular uptake of Cr(VI) is likely to be the first stage of Cr(VI)-induced carcinogenesis. Cr(VI) is similar in structure to tetrahedral sulphate and phosphate anions, and therefore can readily pass into cells via non-specific sulphate and phosphate anion transport channels (Bridges

and Zalups, 2005).[9] Cr(III), and Cr(VI) reduced to Cr(III) before entering cells, is not tetrahedral in structure and does not readily pass into cells. Intracellular reduction of Cr(VI) takes place following intermediate reduction to thermodynamically unstable Cr(V) and Cr(IV) and finally stable Cr(III) (USEPA, 2010).

DNA damage and mutagenicity occurs via the reduced species of Cr(V) and Cr(IV). They have been shown to be reactive towards DNA, leading to DNA strand breaks, chromium-DNA adducts, chromosomal aberrations, and genomic instability. If inadequately repaired, mutations can occur (USEPA, 2010). Oxidative stress takes place with the formation of reactive intermediates (EFSA, 2014c).

3.4 CARCINOGENIC POTENCY OF CHROMIUM

USEPA has derived estimates of cancer potency to Cr(VI) via inhalation (where data is reasonably comprehensive), but not for oral exposure. There is a lack of consistency in the limited data from oral exposure concerning whether Cr(VI) is carcinogenic by the oral exposure route.

[9] As cited by USEPA. (2010) Toxicological review of hexavalent chromium (CAS No. 18540-29-9) in support of summary information on the integrated risk information system (IRIS) EPA/635/R-10/004A Draft. Accessed at: https://cfpub.epa.gov/ncea/iris_drafts/recordisplay.cfm?deid=221433. Accessed: 1 March 2016.

4. EXPOSURE ASSESSMENT - CHROMIUM

4.1 CHROMIUM IN THE NEW ZEALAND FOOD SUPPLY

4.1.1 Total chromium

Total chromium was studied as part of the 1987/88 New Zealand Total Diet Survey in 105 foods. The majority of foods reported results were less than the limit of detection (<LOD) at 0.02 mg/kg, while bacon had the highest mean concentration of chromium (0.15 mg/kg). The next highest chromium concentrations were in beef steak mince and mixed confectionary, both with a concentration of 0.05 mg/kg (ESR/MoH, 1994).[10] Chromium content in foods have not been studied since 1987/88.

4.2 CHROMIUM IN THE NEW ZEALAND WATER SUPPLY

Chromium, whether total or Cr(VI) species, has yet to be analysed for in the New Zealand drinking-water supply, although the New Zealand drinking water standards contain a provisional maximum acceptable value (MAV) of 0.05 mg/L total chromium (Ministry of Health, 2008).

4.3 CHROMIUM IN THE AUSTRALIAN FOOD SUPPLY

4.3.1 Total chromium

Total chromium was studied in the 23rd Australian Total Diet Study across 92 foods and beverages (FSANZ, 2011).

Mango was determined to have the highest maximum concentration of 1.6 mg/kg and the highest mean concentration of 0.39 mg/kg, in comparison with the other foods studied, but it was noted that three out of the four samples had concentrations below the limit of reporting (LOR), i.e. the high concentration was caused by a single, presumably anomalous, result.[11]

Sliced ham, chocolate milk, hamburger, bacon and lean minced beef were also determined to have high maximum concentrations of total chromium, with concentrations of 0.35, 0.27, 0.26, 0.23, and 0.22 mg/kg, respectively. It should be noted that processed meats, in particular bacon, were also found to have high chromium concentrations in the 1987/88 New Zealand Total Diet Survey.

The highest mean concentration of total chromium was found in processed meats, with concentrations of 0.24 mg/kg and 0.15 mg/kg for sliced ham and bacon, respectively. It is uncertain whether the elevated levels of chromium of processed meats are related to the curing process or contamination during the slicing process.

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[10] Concentrations given in Appendix 8 of report assumed to be mean concentrations ESR/MoH. (1994) 1987/88 New Zealand Total Diet Survey Report. ESR Client Report FW9411. Wellington: Institute of Environmental Science and Research.

[11] For calculation of mean concentrations, not detected results were assigned the value of half limit of reporting.

Moderately high concentrations were found in grain-containing products (0.07-0.2 mg/kg). Fresh fruit and vegetables mostly had low mean concentrations (<0.03 mg/kg), whereas processed fruit and vegetables tending to have high concentrations. For all foods the minimum concentration of total chromium was less than the limit of reporting (<LOR). The LOR for chromium in the study was 0.0005 to 0.025 mg/kg.

Speciation data has not been identified in Australian studies to date. Some studies outside of Australia and New Zealand report chromium speciation (see Section 4.4).

4.4 OVERSEAS CONTEXT

Information on the chromium content of food is largely restricted to total chromium data, with few studies reporting speciated results. Known Cr(VI) data in food is summarised in the next section.

EFSA (2014b) found the food groups with the highest average chromium occurrence values (after data clean-up, *n* = 4647) were: products for special nutritional use; herbs, spices and condiments; sugar and confectionary; vegetables and vegetable products (including fungi); and animal and vegetable fats and oils. Processed meats were not high contributors to chromium content.

4.4.1 Hexavalent chromium (Cr(VI)) in plant foods

Bread

Chromium speciation was studied in one hundred and fifty-two bread samples in northern Portugal, where 76 samples were white bread and 76 samples wholemeal bread (Soares *et al.*, 2010). It is worth noting that results in the study are given on a dry weight basis, but the temperature/time parameters employed (30-35º C for 1 hour) may not have completely dried the samples. The mean Cr(VI) concentrations (dry weight basis) were 5.65 μg/kg (range <5.60 to 18.80 μg/kg) and 6.82 μg/kg (range <5.60 to 19.70 μg/kg) for white bread and wholemeal bread, respectively. For the purpose of calculating mean values, analytical results below the limit of quantification (LOQ) were assigned a value of half the limit of detection (0.85 μg/kg). The differences in mean Cr(VI) concentrations between white and wholemeal bread were not statistically significant. Cr(VI) in the bread samples was found to represent approximately 10 to 20% of total chromium. It is worth noting that the LOQ for the Cr(VI) method (5.60 μg/kg) is very similar to the mean values determined.

Mushrooms

Total chromium and hexavalent chromium were determined in wild mushrooms (*n* = 34) from two regions in Portugal (Figueiredo *et al.*, 2007). Mushrooms were collected from regions in different locations that were classified as non-contaminated or contaminated (proximity to nearby industrial processes). Of the 34 mushroom samples, 26 were edible varieties from 10 different species, 7 were non-edible varieties from 7 different species and 1 was poisonous. The cap of the mushroom and stalk were analysed separately. Concentrations of total chromium in the cap were in the range 0.02 to 13.84 mg/kg dry weight, with Cr(VI) concentrations in the range <0.0085 to 0.580 mg/kg dry weight. Concentrations of total chromium in the stalk were in the range 0.04 to 6.50 mg/kg dry weight, with Cr(VI) concentrations in the range <0.0085 to 0.81 mg/kg dry weight. There was no significant difference between Cr(VI) concentrations between the caps and the stalks. No results were given as a breakdown for the edible, non-edible and poisonous species groupings. Underlying soil samples, with environmental detritus removed, were taken and analysed for total chromium and Cr(VI) in order to derive bioconcentration factor (BCF) values. BCF

geometric means ranged from 0.004 to 0.024 and 0.093 to 0.187, for total chromium and Cr(VI), respectively. All BCFs were <1 indicating chromium does not bioaccumulate in the mushroom species analysed. It should be noted that the BCF for Cr(VI) was 1 order of magnitude greater than total chromium. The percentage of total chromium as Cr(VI) in the samples ranged from approximately 4 to 40%.

Juice

Tomato juice (*n* = 5) obtained from fresh tomatoes from a local grocery store were analysed for Cr(VI) in a Greek study (Paleologos *et al.*, 1998). Cr(VI) was not detected in tomato juice, with a relatively high LOD of 0.05 mg/kg.

Alcoholic beverages

Total chromium and Cr(VI) were determined in Portuguese and imported beers (*n* = 70), from nine different beer styles (Vieira *et al.*, 2014). Seven of the beer styles (fruit beer, Dunkel/Tmavý, Schwarzbier, premium lager, amber lager, Heller bock, and canned pale lager) reported Cr(VI) of <LOQ (1.61 µg/L). Pale lager style beer, presumed to be bottled beer, had a mean Cr(VI) of 0.94 µg/L (range <LOQ to 9.9 µg/L), while low-alcohol style beer had a mean Cr(VI) concentration of 2.51 µg/L (range <LOQ to 13.0 µg/L). Cr(VI) concentration in pale lager and low-alcohol represented 0.29%, and 0.58% of total chromium, respectively. Results lower than the LOQ were assigned the value of limit of detection (0.68 µg/L) for the purpose of calculating mean values. Different beer styles contained different chromium content, suggestions for why this might be the case were not provided.

Teas

A study of total chromium, water soluble Cr(VI) and total Cr(VI) in tea varieties originating from Sri Lanka, China, Taiwan and of unknown origins was conducted (Mandiwana *et al.*, 2011). For total Cr(VI), black tea had a mean concentration of 1.07 mg/kg (*n* = 11, range 0.03 to 3.15 mg/kg), oolong tea mean of 0.12 mg/kg (*n* = 2, range 0.03 to 0.21 mg/kg), green tea mean of 0.09 mg/kg (*n* = 5, range 0.03 to 0.14 mg/kg), pu-erh tea[12] mean of 0.075 mg/kg (*n* = 2, range 0.05 to 0.1 mg/kg), and herbal tea mean of <LOD (0.02 mg/kg). The concentration of water soluble Cr(VI) in infused black tea, as consumed, was determined to be 17.5 µg/L, equating to 3.5 µg per cup of tea (200 mL). Water soluble Cr(VI) made up approximately 2 to 20% of total chromium in tea samples, and up to 100% of total Cr(VI).

Two commercially available black tea samples were analysed for Cr(VI) in a study in the Czech Republic (Svancara *et al.*, 2004), with mean concentrations of 1.0 mg/kg and 2.4 mg/kg, respectively, by DPCSV methods. For comparison with DPCSV results, the samples were also analysed by ICP-AES with results of 0.87 mg/kg and 2.12 mg/kg, respectively. As discussed in the methodology section (**Error! Reference source not found.**) of this report, he techniques used in the study may be reporting total chromium instead of Cr(VI). The results should be viewed with caution.

[12] Pu-erh tea; fermented dark aged tea from China

4.4.2 Hexavalent chromium (Cr(VI)) in foods of animal origin

Milk

A South African study of pasteurised cows' milk of different supermarket brands (*n* = 8) analysed in triplicate found concentrations of Cr(VI) in the range 0.61 to 1.44 µg/L. The milk was found to contain 33.2 to 57.1 µg/L of total chromium, with a ratio of Cr(VI) to total chromium of 1.3 to 3.3% (Ambushe *et al.*, 2009).

A study of UHT milk samples (*n* = 60) in Portugal found Cr(VI) concentrations made up approximately 15 to 40% of total chromium (Lameiras *et al.*, 1998). Four different types of UHT milk were available representing four different fat-content categories. [13] Skim milk had a mean Cr(VI) of <0.15 µg/L (range <0.15-0.15 µg/L), simple half-fat milk had a mean Cr(VI) of 0.48 µg/L (range 0.15-0.74 µg/L), supplemented half-fat milk had a mean Cr(VI) of 0.40 µg/L (range 0.24-0.60 µg/L), and whole milk had a mean Cr(VI) of 0.68 µg/L (range 0.20-1.20 µg/L). The basis for the correlation of fat content to Cr(VI) concentration was not further studied.

The concentration of Cr(VI) was determined in powdered milk infant formula (*n* = 20) representing the most available brands in Portugal (Soares *et al.*, 2000). Prior to analysis, the powders were reconstituted in water (1 g sample with 15 mL deionised water), with results reported as Cr(VI) in the initial powders. Amongst the infant formula samples[14] (*n* = 7), Cr(VI) had a range of <10 to 75 µg/kg (mean 24 µg/kg); for follow-up milk samples[15] (*n* = 5), Cr(VI) had a range of <10 to 26 µg/kg (mean 12 µg/kg); and dietetic milk samples[16] (*n* = 8), Cr(VI) had a range of <10 to 75 µg/kg (mean 33 µg/kg).

Poultry meat

Cr(VI) was determined in raw chicken body parts, locally sourced in Pakistan (*n* = unknown) (Mahmud *et al.*, 2011). The young chicks were fed a diet containing a small amount of treated leather for protein nourishment. Leather is obtained from processed animal hide by a tanning process using potassium chromate and potassium dichromate, both of which are sources of Cr(VI). There is reasonable scope for Cr(VI) migration into the chicken parts, and of particular concern are the parts typically available for human consumption. Body parts of the chickens studied were sternum, leg, arm, gizzard, neck, heart and liver, with the results given inTable 2. The paper uses the terms sternum and arm, while for the purposes of the current report they are presumed to mean breast meat and wing, respectively. Standard deviations for intra-body parts were low and typically <0.331, apart from the heart showing considerable variability (standard deviation 1.32). It is not evident from the paper that the methodology used would adequately perform chromium speciation followed by AAS detection. The acid digestion employed in the study is typically used in the scientific literature for determination of total chromium. Mahmud *et al.* (2011) may have assumed all of the Cr(VI) ingested by the chicken remained as Cr(VI) in the animal, therefore the results should be interpreted with caution.

[13] Typical fat content of milk: skim milk <0.5%, half-fat milk 1.5-2%, and whole milk 3-3.5%.
[14] Infant formula samples defined as 'milk prepared for newborn infants (0-4 months) in physiological situations'.
[15] Follow-up milk samples defined as 'milk preparations for feeding infants older than 4 months in physiological situations'.
[16] Dietetic milk samples defined as 'preparations manufactured for infants in pathological situations'.

Table 2. Hexavalent chromium concentration in chicken body parts

Chicken body part	Range (mg/kg)	Mean (mg/kg)
Breast meat (Sternum)	0.670-0.800	0.734
Leg	1.200-1.330	1.266
Wing (Arm)	0.200-0.270	0.233
Gizzard	0.800-1.070	0.933
Neck	0.330-0.600	0.489
Heart	0.300-2.670	1.144
Liver	0.330-0.610	0.415

From Mahmud *et al.* (2011)

4.4.3 Is hexavalent chromium (Cr(VI)) present in foods?

The chromium speciation experiments in bread by Soares *et al.* (2010), and tea by Mandiwana *et al.* (2011) were repeated by Novotnik *et al.* (2013). It was concluded that detection of Cr(VI) in the bread and tea were artefacts of experimental conditions and Cr(VI) could not be present in these food types. The study by Novotnik *et al.* (2013) was carried out by HPLC-ICP-MS using stable isotope spikes to determine speciation in the samples. The stable isotopes ^{50}Cr(VI) and ^{53}Cr(III) were spiked into samples and the differing masses used for identification of the different chromium species by mass spectrometry. Each isotope has its own elution time from the HPLC column, and these peaks were used as references to the chromium peaks obtained from the samples.

The authors concluded that tea itself, containing natural antioxidants, would reduce Cr(VI) to Cr(III). The authors described how the ^{50}Cr(VI) spike was reduced to ^{50}Cr(III) in a neutral organic form, but no elution peak at Cr(III) retention time is observed. It was concluded that the ^{50}Cr(III) was absorbed strongly to the chromatography column resin. No further evidence of this was given nor further investigation using different columns to overcome this issue.

Likewise bread samples by their very nature could not contain Cr(VI) because of the presence of organic matter. Despite the samples being spiked with stable isotopes to study the effect of redox reactions, both in food samples and blank samples, there is no reference to spike recoveries. Although the authors claim that the method had been validated, no validation data were provided for the extraction method and instrumentation conditions.

Visually, from chromatogram figures in the paper, the recovery for ^{50}Cr(VI) in both tea and bread samples is poor compared to ^{53}Cr(III). The authors do not appear to have studied the effect of ionisation suppression or food matrix effects as an explanation of very poor to non-existent ^{50}Cr stable isotope recoveries.

It is the opinion of the authors of the current report that the conclusions of the study (Novotnik *et al.*, 2013) should be treated with caution until further substantiated evidence is published.

Current scientific literature does not provide enough information to conclude whether Cr(VI) is present in foods or Cr(VI) is an artificial species of extraction techniques from oxidation of Cr(III). Additional information requires more vigorous studies to confirm chromium species present in food, with an emphasis on method development with robust method validation.

4.4.4 Hexavalent chromium (Cr(VI)) in drinking water

Where drinking water is sourced from groundwater supplies, Cr(VI) is rapidly reduced to Cr(III) in the groundwater due to anaerobic conditions. Any chromium then present in water is likely to be deposited in sediments, rather than remain in water systems (IPCS, 2009).

An EFSA assessment assumed that chromium in drinking water was entirely Cr(VI), to reflect a worst case scenario. Concentration data gathered by EFSA from the analysis of Cr(VI) in drinking water found that the mean Cr(VI) to total chromium ratio in drinking water was 0.97. Tap water is often treated with oxidisers for increase potability, therefore the oxidation of Cr(III) to Cr(VI) would be promoted, and Cr(VI) in drinking water conserved.

In a study of water from public-supply wells across the state of California, almost all dissolved chromium was present as Cr(VI), the ratio of Cr(VI) to total chromium was 0.9. The state has a maximum contaminant level (MCL) for Cr(VI) of 10 µg/L. Of the wells sampled (*n* = 918), 31% had concentrations >LOR at 1 µg/L, while 4% of sampled wells exceed the MCL. It was noted that Cr(VI) was most abundant in aquifers with chromium-containing source rock and alkaline, oxic[17] groundwater, especially alluvial aquifers. Many factors influence Cr(VI) concentration in waters including geologic, hydrologic and geochemical conditions. For example, waters with an alkaline pH of ≥8 contained increasing concentrations of Cr(VI) (Izbicki *et al.*, 2015).

An Italian study of tap waters and commercial mineral waters compared the ratio of Cr(VI) to total chromium concentrations, with results of 0.82 and 0.57 respectively. In mineral waters, the persistence of both Cr(III) and Cr(VI) species was suggested to occur because the waters remained untreated, and geological and biochemical features in the deep underground aquifers supported Cr(III) existence. In tap waters Cr(VI) concentrations ranged from 0.1-10.88 µg/L and in commercial mineral waters Cr(VI) concentrations ranged from 0.25-3.4 µg/L. The study had an LOD of 0.1 µg/L (Catalani *et al.*, 2015).

Chromium concentration data from tap waters, municipal waters and bottle waters of numerous countries was collated in a chromium speciation review study. The ratio of Cr(VI) to total chromium was variable in the range 0.6 to 0.96 (Markiewicz *et al.*, 2015).

4.5 ESTIMATES OF DIETARY EXPOSURE

4.5.1 New Zealand

A study of total chromium in foods was conducted as part of the 1982 New Zealand Total Diet Survey (Pickston *et al.*, 1985). Table 3 summarises the results of the study and the derived estimates of dietary exposure. Vegetables had the highest contribution of total chromium to the diet (24%) followed by dairy products (17%) and cereal-based foods (16%). A total daily intake of 0.058 mg/day of total chromium was determined, equating to a derived dietary exposure of 0.97 µg/kg bw/day, based on a conservative body weight of 60 kg.

[17] Oxic groundwater: oxygen-present groundwater

Table 3. Total chromium dietary exposure estimates from the 1982 NZTDS

Food group	Daily intake, μg/day	Derived exposure estimate, μg/kg bw/day[a]	Contribution of food group to total exposure estimate, %
Cereal-based foods	9	0.15	16
Meat, fish, eggs	8	0.13	14
Dairy products	10	0.17	17
Fats and oils	<1	<0.02	-
Fruit	4	0.07	7
Vegetables	14	0.23	24
Sweet foods and buts	8	0.13	14
Instant foods	<1	<0.02	-
Drinks	5	0.08	9
Total	**58**	**0.97**	

[a] Calculated using a conservative body weight of 60 kg

In the 1987/88 New Zealand Total Diet Survey, for females and young males the total daily chromium intake was 36 μg/day and 54 μg/day, respectively. This equated to 0.6 μg/kg bw/day and 0.9 μg/kg bw/day, for females and young males respectively, based on a conservative body weight of 60 kg. Whole milk was found to be the highest contributor of total chromium in the diet representing 32% of chromium exposure in young males and 62% in young children.

4.5.2 Australia

The 23rd Australian Total Diet Study (ATDS) included the analysis of total chromium for all foods and for the purposes of the study was considered a nutrient element, rather than a contaminant element (FSANZ, 2011). Major contributors to chromium exposure were milks and creams, and breads (white, wholemeal, multigrain and rye). The mean intake of total chromium for males and females aged 17 to 29 years were 146 μg/day and 99 μg/day, respectively. ATDS estimates body weights for these age-gender cohorts as 77 kg for males and 64 kg for females. Exposure to total chromium equates to 1.9 μg/kg bw/day and 1.5 μg/kg bw/day, respectively.

4.5.3 Overseas estimates of dietary exposure

In the 2006 UK Total Diet Study, sugars and preserves were major contributors to chromium in the diet at 16%, followed by miscellaneous cereals, potatoes and beverages (13%, 12%, and 13%, respectively) due to their high consumption levels. Total exposure to chromium was calculated to be 29 μg/day, equating to 0.5 μg/kg bw/day based on a conservative body weight of 60 kg (Rose *et al.*, 2010).

Further overseas dietary exposure estimates are presented in Table 4.

Table 4. Summary information on total chromium in foods from selected overseas studies

Country	Samples	Number of samples	Total chromium, mean, µg/kg	Estimated dietary intake, µg/day		Estimated dietary intake 95th percentile, µg/day		Reference
Cameroon[a]	Cereals and cereal products	7	279	58		130		Gimou *et al.* (2013)
	Tubers and starches	11	94	23		62		
	Fruits, vegetables and oilseeds	14	170	24		68		
	Oils and fats	4	779	23		61		
	Beef, poultry and eggs	4	157	2.5		9.7		
	Fish	6	460	46		140		
	Milk and milk products	4	129	1.1		5.1		
	Beverages	8	30	34		73		
	Sugar and cocoa products	2	500	3.8		16		
	Condiments, salt and flavourings	4	1140	7.8		34		
	Food eaten outside	-	90	8.3		35		
	All food	**64**		**230**		**430**		
France				*Children 3–14 years*	*Adults 15 + years*	*Children 3–14 years*	*Adults 15 + years*	Leblanc *et al.* (2005)
	Bread, rusk	12	160	6.2	12	19	34	
	Breakfast cereals	10	130	2.1	0.51	8	4	
	Pasta	2	40	1.4	1.3	3.8	3.8	
	Rice and semolina	6	60	1.1	1.0	3.2	3.6	
	Miscellaneous cereals	4	10	0.02	0.01	0.11	0	
	Viennese bread, buns	12	160	3.3	2.0	12	11	
	Biscuits	14	140	3.9	1.3	16	6.3	
	Cakes	12	70	1.4	1.5	6.6	5.9	
	Milk	16	20	2.5	1.4	5.4	4.2	
	Ultra-fresh dairy products	30	30	1.1	1.2	3.8	5	
	Cheeses	16	104	1.5	3.1	5.2	9.3	
	Eggs and egg products	30	50	0.77	1.2	2.4	4.0	
	Butter	2	70	0.35	0.54	1.3	2.0	
	Oils	2	40					
	Margarine	2	60					
	Vegetal oils			0.01	0.05	0	0.16	
	Meat	44	50	2.4	2.6	5.9	6.4	
	Poultry and game	24	30	0.76	1.1	2.4	3.7	
	Offals	18	100	0.09	0.19	0.86	1.5	
	Delicatessen	32	170	2.5	3.0	8.0	9.7	
	Fish	62	80	0.94	1.0	3.1	3.8	
	Shellfish	18	90	0.1	0.23	0.79	1.6	

Country	Samples	Number of samples	Total chromium, mean, µg/kg	Estimated dietary intake, µg/day		Estimated dietary intake 95th percentile, µg/day		Reference
	Vegetables (exc. potatoes)	198	50	3.7	6.1	9.7	16	
	Starchy vegetables	26	50	3.5	3.4	8.4	8.3	
	Pulses	16	80	0.67	0.95	3.7	5.5	
	Fruits	79	10	0.63	1.03	1.8	3.1	
	Nuts and oilseeds	22	60	0.03	0.08	0.22	0.51	
	Ices cream	2	100	0.74	0.52	3.5	2.6	
	Chocolate	4	340	1.5	0.8	6.6	4.6	
	Sugars, confectionery	18	120	3.2	3.0	10	8	
	Drinking waters	12	10	2.2	2.7	5.8	7.9	
	Non-alcoholic beverages	46	50	8.0	4.0	25	18	
	Alcoholic beverages	22	20	0.02	1.9	0	8.8	
	Coffee	12	10	0.13	2.8	0.86	8.2	
	Hot beverages	4	20	0.51	0.91	1.6	4.7	
	Pizzas, salt cakes, quiche	6	70	0.31	0.49	1.6	2.3	
	Sandwiches	12	80	0.65	0.58	3.2	3.6	
	Soups	38	50	1.3	2.6	5.5	10	
	Mixed dishes	75	90	5.4	5.9	16	20	
	Salads	4	140	0.29	0.59	2.1	4.1	
	Dessert	12	90	2.5	1.7	11	8.8	
	Stewed fruit, compote	8	30	0.21	0.22	1	1.3	
	Condiments, sauces	12	120	0.35	0.44	1.5	2.0	
	Substitute meals	2	60					
	All Foods			**68**	**77**	**110**	**120**	
France[b]	Whole diet (breakfast, lunch)	50	102 (41-179)	150				Noel *et al.* (2003)
Spain[c]	Meat	15		23				Bocio *et al.* (2005)
	Fish and seafood	15		65				
	All foods			**88.4**				
Spain (Catalonia)[d]	Whole diet	600		280				Domingo *et al.* (2012)
USA[e]	Not specified	10	82.5	<39				Iyengar *et al.* (2000)

[a] Concentration and dietary exposure data are upper bound estimates, from not detected analytical results assigned a value equal to the limit of detection
[b] Dietary exposure was determined from duplicate diet samples. Breakfast and lunch meals were collected from restaurants and institutions. Daily exposure was taken to be the sum of breakfast plus two-times lunch items
[c] Estimated dietary intake, µg/day based on adult male of 70 kg body weight
[d] Duplicate diets
[e] Estimated dietary intake, µg/day based on adult males 25-30 years

4.5.4 Dietary exposure to chromium species

With the limited available data on Cr(VI) across just a few foods, Cr(VI) in foods as a percentage of total chromium is difficult to estimate. No estimates of dietary exposure to chromium species have been derived.

Where total chromium and Cr(VI) are reported, Cr(VI) as a percentage of total chromium ranges from approximately 0.3% to 40%. No reasonable conclusion can be gained from this, therefore the exposures to total chromium from national total diet studies and other large food studies are not amenable to estimating Cr(VI) in foods. If it was assumed foods were capable of containing Cr(VI), even with high organic contents and on average the foods contained <40% Cr(VI) from total chromium, based on the total diet studies documented above, Cr(VI) exposure would be < 0.8 μg/kg bw/day. However, this estimate is probably too conservative to be useful.

The mean chronic dietary exposure to total chromium ranges from 0.6 μg/kg bw/day (minimum lower bound) to 5.9 μg/kg bw/day maximum upper bound in the EFSA review of chromium in food and water. Foods with the highest content of chromium were water-based foods such as juices and soft drink. The reported analytical results for total chromium were assumed to be Cr(III), due to the immediate reduction of Cr(VI) to Cr(III). EFSA noted that exposure of the general population via the diet is likely to represent the most important contribution to overall chromium exposure. If a small proportion of total chromium in food existed in the Cr(VI) form, it could contribute significantly to Cr(VI) exposure (EFSA, 2014c).

4.5.5 Biomarkers of exposure

Biomarkers of Cr(VI) exposure are used in occupational settings where exposure to Cr(VI) is common and at high levels, for example, leather tanning industries or stainless steel industries. Such exposure is generally through inhalation and dermal contact. Systemic exposure to Cr(VI) can be monitored through red blood cell (RBC) Cr(VI) levels (RBC-Cr), although the taking of blood samples is by nature invasive, requiring skilled personnel. RBC-Cr is suitable as a biomarker because only Cr(VI) is able to cross RBC membranes, the biomarker is therefore species specific, and it persists in the plasma for longer periods of time than Cr(III), being slowly released. Data for the general population using RBC-Cr as a biomarker is currently not available (EFSA, 2014c).

There is a good correlation between chromium exposure by inhalation and the concentration of chromium in urine. Sample collection of urine is much easier than red blood cells and can provide indications of high-level recent occupational exposure by inhalation up to 48 hours following exposure. For a more meaningful exposure assessment, baseline chromium levels should be determined to assess daily increases in chromium in urine (Mutti *et al.*, 1979). Inhalation of chromium by steel alloy welders was reflected in the urinary concentration of chromium, this reflected the exposure to water-soluble Cr(VI). No correlation was observed for water-insoluble Cr(III) (Tola *et al.*, 1977).

It was noted by Gargas *et al.* (1994) that, while increased urinary chromium exposure can be used as a good measure for occupational inhalation exposure, urinary chromium levels are not suitable for assessing incidental exposure to dust and soils (and, presumably, food). The study followed the voluntary ingestion of soil samples for chromium doses up to 200 μg/day for three days. It was concluded that the chromium was not sufficiently bioavailable for biomonitoring of urine, with no difference observed between pre- and post-dosing mean urinary chromium concentrations. (Gargas *et al.*, 1994).

It is possible to use white blood cells (WBC) as biomarkers of chromium exposure as Cr(VI). The exposure of human whole blood to Cr(VI), prior to isolation of WBC, resulted in accumulation of Cr(VI) to a greater extent in WBC than in RBC. In a study of isolated rat WBC the uptake of Cr(VI) was greater than that of human white blood cells, while uptake by human red blood cells was greater than that of rats (Coogan *et al.*, 1991). Similarly to red blood cell studies, the technique is invasive and it is difficult to study large populations to gain meaningful data (USEPA, 2010).

Plasma is another potential biomarker with selectivity towards Cr(III). Plasma studies could be used in conjunction with RBC-Cr data. However, Cr(VI) can only be detected up to 2 hours from exposure and it reduces rapidly in plasma. In addition, this technique is also invasive.

Goulle *et al.* (2012) monitored the concentration of chromium in plasma, red blood cells, and urine for 49 days following an accidental poisoning event. The chromium concentration in the three media decreased from 2088 μg/L to 5 μg/L, 631 μg/L to 129 μg/L, and 3512 mg/L to 10 mg/L, respectively. The half-life of chromium in red blood cells was longer than in plasma.

5. RISK CHARACTERISATION - CHROMIUM

5.1 ADVERSE HEALTH EFFECTS IN NEW ZEALAND

5.1.1 Incidence of cancer

In New Zealand, prostate, colorectal, breast, melanoma of the skin, and lung are the five most prevalent cancers, in descending order.[18]

From overseas occupational studies, Cr(VI) has been classified as carcinogenic for humans. Increased risks of lung, nose and nasal sinus cancers have been documented. There is insufficient evidence to determine if Cr(VI) is carcinogenic following oral exposure from food and water (EFSA, 2014c).

Cancer registration rates in New Zealand are available for lung cancer. However, any contribution of Cr(VI) exposure to lung cancer rates is likely to be minor compared to the known impact of smoking on lung cancer rates, and any potential occupational exposures.[19] Lung cancer registration rates peaked in New Zealand in the 1980s (age standardised rate of approximately 35 to 40 per 100,000 population) and appear to be following a gradual decline (2005 to 2011; 29 to 31 per 100,000).[20]

According to the Globocan database[21], New Zealand lung cancer incidence rates are moderate by world standards, with more developed countries having higher rates than less developed countries.

5.1.2 Risk assessment

The 1982 and 1987/88 New Zealand Total Diet Surveys estimated the dietary exposure of New Zealanders to total chromium (ESR/MoH, 1994; Pickston *et al.*, 1985). More recent studies have not included chromium analysis as exposure in New Zealand is likely to already be as low as reasonably achievable (ALARA).

In the 1982 survey, the intake of chromium for low (8.4 MJ/day), medium (11.3 MJ/day), and high (16.7 MJ/day) energy requirement diets were 58 μg/day, 78 μg/day, and 116 μg/day, respectively. All three intakes were between 50 to 200 μg /day, the range specified as the estimated safe and adequate daily dietary intake (ESADDI) recommendations by the National Research Council, 1989 (Pickston *et al.*, 1985). The ESADDI was the health based exposure guideline used for these surveys.

[18] http://globocan.iarc.fr/Pages/online.aspx Accessed 2 March 2016
[19] http://www.health.govt.nz/nz-health-statistics/health-statistics-and-data-sets/cancer-data-and-stats?mega=Health%20statistics&title=Cancer Accessed 24 February 2016
[20] http://www.health.govt.nz/publication/cancer-new-registrations-and-deaths-2012 Accessed 2 March 2016
[21] http://www-dep.iarc.fr/ Accessed 13 July 2015

5.2 ADVERSE HEALTH EFFECTS OVERSEAS

5.2.1 Incidence of cancer

According to the Globocan database, the countries with the highest incidence of lung cancer are Hungary (52 per 100,000), Serbia (46 per 100,000) and the Democratic Republic of Korea (44 per 100,000), with lowest incidence rates having been reported for western and central Africa, with mean rates of about 2 per 100,000.[22]

5.2.2 Epidemiological studies

Epidemiological studies for chromium and in particular hexavalent chromium are very limited. The relationship between environmental exposure to chromium and cancer outcomes in humans have been summarised in the EFSA report Chromium in food and drinking water, section 7.3.2 (EFSA, 2014c). Representative conclusions are given in the following section, but are not exhaustive.

EFSA observations related to Cr(VI) in humans

The published studies were limited to retrospective observations of cancer after oral exposure to total chromium and/or Cr(VI).

From the limited number of human studies, and subsequent meta-analyses, the data do not provide substantial evidence of an association between oral exposure to total chromium or Cr(VI) and adverse health effects including cancer. Two meta-analyses were cited in the EFSA report, the first was a meta-analysis of 49 epidemiological studies from 1950 to 2005 concluding that chromium was only a weak lung carcinogen and not carcinogenic to other organs, and the second meta-analysis of 32 epidemiological studies from 1950 to 2009 looking at gastrointestinal cancer found no significant association between occupational exposure to Cr(VI) and incidence of gastrointestinal cancers. It should be noted that both meta-analyses were strongly influenced by occupational exposure studies, as opposed to dietary exposure and therefore do not represent exposures to the general population. Due to limitations in the data sets, a dose-response assessment could not be conducted. Likewise, data on the allergenic potential of Cr(VI) via oral ingestion was lacking, with inconclusive assessments.

5.2.3 Risk assessments

International

A risk assessment was conducted by EFSA (2014c) and summarised in section 3.2.2. Exposure to Cr(VI) from consumption of drinking water was determined to be of low public concern after MOEs were calculated. There was insufficient data to determine MOEs for Cr(VI) from consumption of food.

[22] http://globocan.iarc.fr/Pages/summary_table_site_sel.aspx Accessed 1 March 2016

6. RISK MANAGEMENT INFORMATION - CHROMIUM

6.1 RELEVANT FOOD CONTROLS: NEW ZEALAND

6.1.1 Establishment of regulatory limits

No maximum limits for total chromium or Cr(VI) are included in the Australia New Zealand Food Standards Code.

6.1.2 Imported Food Requirements (IFR)

There are currently no IFRs that require products imported into New Zealand to be monitored for chromium.

6.1.3 Establishment of dietary reference values

There is a lack of data across all gender-age cohorts for estimated average (EAR) requirements to be set in New Zealand. Adequate intakes (AI) for total chromium were determined to be 0.2 µg/day to 5.5 µg/day for infants, 11 µg/day to 35 µg/day for children and adolescents, and 25 µg/day to 45 µg/day for adults. Upper limits (UL) of intake for chromium have not been defined due to limited data from toxicological studies of chromium in food (National Health and Medical Research Council/Ministry of Health, 2006).

6.2 RELEVANT FOOD CONTROLS: OVERSEAS

6.2.1 Establishment of regulatory limits

The EU frequently has the most stringent regulatory requirements worldwide and at present there is no EU regulation for maximum levels of chromium in food. A quality standard for total chromium in water for human consumption was determined to be 50 µg/l in the Council Directive 98/83/EC.[23] No standards were set for Cr(VI) (EFSA, 2014c).

The Codex Committee on Contaminants in Foods (CCCF) has not assessed chromium regulations in food.

6.3 INFLUENCE OF FOOD PROCESSING ON CHROMIUM SPECIES LEVELS

6.3.1 Cooking

During the baking and toasting of bread, the change in Cr(VI) content of the flour component and bread itself was studied by graphite furnace atomic absorption spectroscopy (GFAAS). It was determined that Cr(III) did not convert to Cr(VI) and it is likely that the organic content of the flour provides a reducing medium if Cr(VI) was initially present in the bread. The authors stated that the chromium content of the flour and bread was 'nearly the same' as it's starting content. Therefore it could be presumed no change in concentration took place (Kovacs *et al.*, 2007). It should be noted that the study determined Cr(VI) as the water-soluble chromium following dry ashing of samples. It is uncertain whether the oxidation state of chromium would have remained unchanged during dry ashing.

[23] Council Directive 98/83/EC of 3 November 1998 on the quality of water intended for human consumption, OJ L 330, 5.12.98, p. 32-54.

7. CONCLUSIONS - CHROMIUM

7.1 DESCRIPTION OF RISKS TO NEW ZEALAND CONSUMERS

The carcinogenicity of Cr(VI) as defined by IARC is Group 1, carcinogenic to humans, although there is limited evidence of causation of cancer by oral exposure based on published epidemiological studies to date, with evidence for carcinogenicity coming from studies in occupational settings, where exposure was primarily by inhalation. There are also some questions over the detection of Cr(VI) in food samples, with suggestions that the apparent detection of this chromium species may be an artefact of the analytical methods used. Until further evidence is presented and analytical methods become standardised, exposure to Cr(VI) from food remains tentative. The concentration of total chromium in foods studied in New Zealand in 1982 and 1987/88, are of comparable to levels found in international studies in the parts per billion range, albeit the data is now fairly old.

7.2 COMMENTARY ON RISK MANAGEMENT OPTIONS

No specific risk management procedures or models have been identified for chromium.

7.3 DATA GAPS

While the chemistry behind the persistence of Cr(VI) in foods, stipulated to be a reducing medium, is expected to be the same across the world, the agricultural, geological and industrial environment in New Zealand may not be comparable to Europe, where most studies of chromium in foods have been conducted. Chromium in the local environment is likely to affect the levels of chromium in New Zealand grown food. EFSA concluded that more data for Cr(III) and Cr(VI) in food are required. Given that Cr(VI) has the biggest health risks and implications, the determination of this species in food is critical to the derivation of improved dietary risk assessments.

Further new data for total chromium and Cr(VI) in New Zealand food would need to come from a survey of foods, determined to low levels of detection using a hyphenated technique linking chromatography and mass spectrometry for sensitive chromium determination. The greater sensitivities achieved by hyphenated techniques would allow meaningful comparison of total chromium and Cr(VI) with overseas studies. A suitable validated extraction method would be required that crucially maintains the original chromium speciation ratio throughout the process.

8. HAZARD IDENTIFICATION - SELENIUM

Selenium is in group VI of the periodic table, the same group as sulphur (EFSA, 2014a; IPCS, 1987). The chemistry of selenium is very similar to sulphur and selenium forms analogues of the sulphur-containing amino acid (cysteine and methionine).

Selenium and its compounds are used in some photographic devices, gun bluing (a liquid solution used to clean the metal parts of a gun), plastics, paints, anti-dandruff shampoos, vitamin and mineral supplements, fungicides, and certain types of glass (ATSDR, 2003).

8.1 STRUCTURE AND NOMENCLATURE

Selenium can exist in five oxidation states; -2, 0, +2, +4 and +6 (IPCS, 1987). All valence states occur naturally, except the +2 valence state, which is unknown in nature. Selenium also exists in six isotopic forms with ^{80}Se being the most abundant (49.8%), followed by ^{78}Se (23.5%), ^{82}Se (9.1%), ^{76}Se (9.0%), ^{77}Se (7.9%) and ^{74}Se (0.9%). There are no naturally-occurring radio-isotopes of selenium, but one isotope with a half-life of 120 days (^{75}Se) and two short half-life (^{77m}Se, 17.5 seconds and ^{81}Se, 18.6 minutes) isotopes can be generated by neutron activation. The selenium-containing compounds found in water, foods and biological samples are summarised in Table 5.

Table 5. Selenium compounds found in water, foods and biological samples

Chemical Name	Formula	Abbreviation	CAS No.
Inorganic			
Selenate	SeO_4^{2-}	Se(VI)	95788-45-7
Selenite	SeO_3^{2-}	Se(IV)	7783-00-8
Organic			
Selenocysteine	$HOOCCH(NH_2)CH2\text{-}Se\text{-}H$	SeCys	10236-58-5
Selenocystine	$HOOCCH(NH_2)CH_2\text{-}Se\text{-}Se\text{-}CH_2CH(NH_2)COOH$	SeCys2	2897-21-4
Selenomethionine	$HOOCCH(NH_2)CH_2CH_2\text{-}Se\text{-}CH_3$	SeMet	1464-42-2
Selenomethyl-selenocysteine	$HOOCCH(NH_2)CH2\text{-}Se\text{-}CH_3$	SeMeSeCys	26046-90-2
γ-glutamyl-methyl-selenocysteine	$H_2NCH_2CH_2\text{-}CO\text{-}NHCH(COOH)CH_2\text{-}Se\text{-}CH_3$	γ-glutamyl-MeSeCys	
Dimethyl-selenide	$(CH_3)_2Se$	DMSe	593-79-3
Dimethyl-diselenide	$(CH_3)Se–Se(CH_3)$	DMSe2	7101-31-7
Trimethyl-selenonium cation	$(CH_3)_3Se^+$	TMSe+	25930-79-4

CAS No.: Chemical Abstract Service Registry Number

While the list in Table 5 is not comprehensive, it includes the species most commonly found in foods. Selenium will also be present in a wide range of proteins due to incorporation of SeCys and SeMet, in place of the equivalent sulphur-containing amino acids.

8.2 OCCURRENCE

Selenium occurs in most, if not all, foods, although concentrations are usually less than 30 μg/kg fresh weight (EFSA, 2014a). The selenium content of plant foods generally reflects the selenium content of the growing environment (EFSA, 2014a). However, plants may be classified as selenium accumulators or non-accumulators, as some plants (Brazil nut tree, *Brassica* spp., *Allium* spp.) have greater ability to assimilate and accumulate selenium. Selenium accumulators generally have mechanisms to detoxify selenium and will, consequently, contain a wider range of selenium species.

New Zealand soils are low in selenium (Thomson, 2004) and this is reflected in the generally low selenium content in New Zealand-grown fruits, vegetables and grains (Vannoort and Thomson, 2005; Vannoort and Thomson, 2011). This effect of growing conditions is emphasised by comparison of the selenium content of bread from the North Island of New Zealand, mainly produced from Australian wheat, and bread from the South Island, mainly produced from New Zealand wheat. In the 2009 New Zealand Total Diet Study, North Island bread had a mean selenium content of 0.111 mg/kg, compared to 0.026 mg/kg for South Island bread (Vannoort and Thomson, 2011).

The highest selenium concentrations are usually seen in seafood and offal meats, such as liver and kidney from terrestrial animals (IPCS, 1987). Selenium concentrations in foods of animal origin (meat, milk and eggs) will vary depending on the animals' diets. It is interesting to note that in New Zealand, food products from intensively reared animals (chickens and pigs) are higher than equivalent foods from pasture-reared animals (Vannoort and Thomson, 2011).

8.3 ANALYTICAL METHODS

As for other elements, analysis of selenium and its species forms can be considered in terms of three processes; extraction, separation and detection. For total selenium determination the processes of extraction and separation are replaced by a process of matrix destruction and solubilisation.

8.3.1 Sample extraction for selenium species

Extraction procedures for selenium speciation generally involve extraction of total selenium for subsequent separation.

General extraction

There are currently no methods that can reliably extract 100% of the selenium from foods without potentially affecting the species (Fairweather-Tait *et al.*, 2010). General extraction techniques for chemical speciation need to balance maintaining the species in their original form and maximising extraction efficiency. A range of solvents, including tetramethylammonium hydroxide, sodium dodecyl sulphate, hot water and methane sulphonic acid have been used (B'Hymer and Caruso, 2006). The highest extraction efficiencies for any of these solvents were reported with methane sulphonic acid (66-67%).

Extraction can also be enhanced by microwave heat, ultrasonication and/or the use of enzymes, particularly proteolytic enzymes, such as Proteinase K. Such enzymes are able to hydrolyse selenium-containing proteins, while maintaining the SeMet and SeCys moieties. Enzyme-assisted extraction techniques have been reported to extract 60-90% of the total selenium from yeast (B'Hymer and Caruso, 2006), 98% of selenium from broccoli (Šindelářová *et al.*, 2015) and 75% of selenium from mushrooms (Stefánka *et al.*, 2001) and 94-97% from cereals (Tsai and Jiang, 2011; Zhao *et al.*, 2011).

Volatile selenium species can be recovered by headspace analysis, solid phase extraction or solid-phase micro-extraction (B'Hymer and Caruso, 2006).

8.3.2 Separation of selenium species

B'Hymer and Caruso (2006) have reviewed techniques for the separation of selenium species from biological samples. Although HPLC, capillary electrophoresis (CE) (Zhao *et al.*, 2011) and gas chromatography (GC) have all been used, HPLC is by far the most commonly used technique, due to its reproducibility and the fact that, unlike GC, no derivatisation of the analytes is necessary prior to chromatography. Reverse-phase, ion pair, ion exchange, size

exclusion and chiral HPLC have all been used for selenium speciation (B'Hymer and Caruso, 2006; Pedrero and Madrid, 2009).

While a range of non-chromatographic techniques have been used to separate selenium species, it appears that the techniques are typically applied to water or other environmental matrices (Gonzalvez *et al.*, 2009). In such matrices, separation techniques are usually concerned with isolating the inorganic selenium species, selenite and selenate.

8.3.3 Detection of selenium species or total selenium

Inductively-coupled plasma mass-spectrometry (ICP-MS) is used as a general detector for selenium species, with little difference observed in response factors seen between different selenium species (Anan *et al.*, 2015). However, 'softer' mass spectrometric techniques, such as quadrupole time-of-flight mass spectrometry (QTOF-MS) can also be used for direct selenium species detection (Anan *et al.*, 2015). With this technique, species are detected close to their molecular weight, while ICP-MS detects all selenium species as atomic selenium.

Hydride generation atomic absorption spectroscopy (HG-AAS) can also be used in hyphenated techniques or for measurement of total selenium (Matni *et al.*, 1995).

Interferences in the mass spectrometric detection of ^{78}Se and ^{80}Se can occur due to the formation of $^{40}Ar^{38}Ar^{+}$ or $^{40}Ar^{40}Ar^{+}$. However, these interferences can largely be removed by employing a dynamic reaction cell, where the ion stream passes through a gas flow, which removes interfering ions, prior to entering the mass spectrometer (Tsai and Jiang, 2011; Zhao *et al.*, 2011).

8.3.4 Certified reference materials

There is a single certified reference material available for species forms of selenium, a selenium-enriched yeast (Selm-1), certified by the Technical and Advisory Services of the National Research Council Canada.[24] Selm-1 has a certified selenium content of 2031 ± 70 mg/kg and a certified SeMet content of 3190 ± 260 mg/kg. When allowance is made for the difference in molecular weight between selenium and SeMet, these certified values suggest that 63% of the selenium in the enriched yeast is in the form of SeMet.

8.3.5 Selenium species testing in New Zealand

While a number of analytical laboratories in New Zealand are accredited for the analysis of food items for total selenium, usually by ICP-MS, none are accredited for analysis of species forms of selenium.[25] The scientific literature did not identify any New Zealand laboratories testing foods for selenium species.

[24] http://www.nrc-cnrc.gc.ca/eng/solutions/advisory/crm/certificates/selm_1.html Accessed 15 March 2016
[25] http://www.ianz.govt.nz/directory/ Accessed 17 March 2016

9. HAZARD CHARACTERISATION: ADVERSE HEALTH EFFECTS - SELENIUM

9.1 CONDITIONS

Selenium is an essential trace element in the human diet (National Health and Medical Research Council/Ministry of Health, 2006). Adverse health effects in humans can result from either selenium deficiency or excessive selenium intake.

9.1.1 Selenium deficiency

The essentiality of selenium is due to its inclusion in specific human selenoproteins and particularly seleno-enzymes, where selenium is a component of the enzyme active site (Kryukov *et al.*, 2003). The major human selenoproteins and their functions are (EFSA, 2014a):

- Glutathione peroxidase (GPx). A widely distributed family of enzymes involved in the reduction of reactive oxygen species, such as peroxides, to protect various body components from oxidative damage.
- Iodothyronine deiodinase (DIO). A family of enzymes involved in thyroid hormone metabolism.
- Thioredoxin reductase (TR). A family of enzymes with diverse roles including reduction of oxidised thioredoxin, protection against hydrogen peroxide produced by the mitochondrial respiratory chain, and reduction of ribonucleotides to deoxyribonucleotides.
- Selenophosphate synthetase (SPS2). An enzyme involved in synthesis of all selenoproteins.
- Selenoproteins H, N, P, R, S, W. Proteins that are involved in various biochemical functions.

Selenium deficiency results in decreased GPx activity and decreased concentrations of plasma thyroid hormones (EFSA, 2014a). Clinical manifestations of selenium deficiency are poorly defined. However, several conditions have been associated with selenium deficiency:

- Patients on selenium-free total parenteral nutrition have been reported to develop skeletal myopathy and muscle pain/weakness, cardiomyopathy, pseudoalbinism (loss of skin pigment), erythrocyte macrocytosis (enlarged erythrocytes) (Gramm *et al.*, 1995).
- Keshan disease is an endemic cardiomyopathy, occurring mainly in children and young women in low selenium intake regions of China (Ge *et al.*, 1983). The disease is not fully understood, but there is some evidence that it is due to concurrent selenium deficiency and infection with an enterovirus, possibly Coxsackie virus B3 (Beck *et al.*, 2003).
- Kashin-Beck disease is a chronic degenerative osteochondropathy occurring in pre-adolescents and adolescents in selenium-deficient areas of China, Mongolia, Siberia and North Korea. The aetiology of the disease is largely unknown, but possible risk

factors include mycotoxins in food, humic and fulvic acids in water, and iodine and selenium deficiency (Lei *et al.*, 2015; Yao *et al.*, 2011).

9.1.2 Selenium toxicity

Chronic excess selenium intake can give rise to selenosis, characterised by headache, loss of hair (alopecia), deformation and loss of nails, skin rash, unpleasant (garlic) odour on breath and skin, excessive tooth decay and discolouration, numbness, and paralysis or hemiplegia (Yang *et al.*, 1983). The mechanism of toxicity is not clear.

A case series of selenosis cases was examined after a misformulation in a dietary supplement resulted in users receiving approximately 200 times the specified amount of selenium (Aldosary *et al.*, 2012). Selenium intake was estimated to average 1.3 g over 37.5 days (approximately 35,000 μg/day). Symptoms of toxicity exhibited within 1 week of beginning ingestion of the supplement, with symptoms including alopecia, dystrophic fingernail changes, gastrointestinal symptoms and memory difficulties. Whole blood selenium concentrations were in the range 150-732 μg/L at an average of 27 days after ceasing ingestion of the supplement. Symptoms resolved with supportive care and no case required hospitalisation.

9.2 TOXICOLOGICAL ASSESSMENT

As selenium is an essential trace element, assessments are usually concerned with intakes of selenium that are required to achieve adequate nutrition. However, some of these assessments also include information relevant to the safety of selenium intake. Four reasonably recent assessments of selenium have been carried out (ATSDR, 2003; EFSA, 2014a; IARC, 1975; National Health and Medical Research Council/Ministry of Health, 2006).

9.2.1 National Health and Medical Research Council (NHMRC)

The NHMRC assessment derived Nutrient Reference Values (NRVs) for Australia and New Zealand (National Health and Medical Research Council/Ministry of Health, 2006). For selenium this included derivation of Estimated Actual Requirements (EAR), Recommended Daily Intakes (RDI) and Upper Levels of Intake (UL). Values for these NRVs were derived from:

- EARs for adults were based on GPx activity at various supplemental selenium intakes (Duffield *et al.*, 1999; Xia *et al.*, 2005). The RDI for adults was derived assuming a 10% coefficient of variation for the EAR
- EARs for children were extrapolated adult data on a metabolic body weight basis. The RDI for children was derived assuming a 10% coefficient of variation for the EAR
- For infants (0-6 months) an adequate intake (AI) was derived from the average intake of selenium from consumption of breast milk. For infants 7-12 months, the AI was derived by extrapolation from the AI for 0-6 month infants
- Additional selenium requirements were factored into EARs and RDIs for pregnant and lactating women
- ULs for adults were based on a rounded NOAEL of 800 μg/day for studies in a seleniferous region of China (Yang and Zhou, 1994) and application of an uncertainty factor of 2.
- UL for young infants was based on a study showing no adverse effects from consumption of human breast milk containing 60 μg/L (Shearer and Hadjimarkos,

1975). ULs for children and adolescents were extrapolated from the young infant UL on a body weight basis.

NRVs derived for selenium for Australia and New Zealand are summarised in Table 6.

Table 6. NRVs for selenium for Australia and New Zealand

Age group[a]	NRV (mg/day)			
	AI	**EAR**	**RDI**	**UL**
0-6 months	12			45
7-12 months	15			60
1-3 years		20	25	90
4-8 years		25	30	150
9-13 years		40	50	280
14-18 years				
- Boys		60	70	400
- Girls		50	60	400
- Pregnant		55	65	400
- Lactation		65	75	400
19-50 years				
- Men		60	70	400
- Women		50	60	400
- Pregnant		55	65	400
- Lactation		65	75	400
50+ years				
- Men		60	70	400
- Women		50	60	400

NRV: Nutrient reference value
AI: Adequate Intake
EAR: Estimated average requirement
RDI: Recommended daily intake
UL: Upper level of intake
[a] Unless specifically stated, NRVs apply to both males and females

9.2.2 International Agency for Research on Cancer (IARC)

The IARC monograph on selenium and selenium compounds was published over 40 years ago (IARC, 1975). It was concluded that:

- The available data were insufficient to allow an evaluation of the carcinogenicity of selenium compounds in animals
- The available data provide no suggestion that selenium is carcinogenic in humans
- Evidence for a negative correlation between regional cancer death rates and selenium intake was reviewed and it was concluded that the evidence was not convincing.

No update of this monograph has been carried out since.

9.2.3 European Food Safety Authority (EFSA)

EFSA were asked to derive dietary reference values (DRVs) for selenium (EFSA, 2014a). As such, the focus of this assessment was mainly on the nutritional aspects of selenium. EFSA concluded that:

- The levelling off of plasma selenoprotein P concentrations was considered to be indicative of an adequate selenium supply to all tissues
- There was insufficient data to derive an average daily requirement for selenium, but an adequate intake (AI) of 70 μg/day for adults was derived
- AIs for infants were extrapolated from estimated selenium intakes for fully breast-fed infants, while AIs for children and adolescents were extrapolated from AIs for adults.

A higher AI was derived for lactating women (85 μg/day) than for other adults, due to adaptive changes in the metabolism of selenium that occur during pregnancy
- A review of observational studies and randomised controlled trials that investigated the relationship between selenium and health outcomes (mortality, cancer, cardiovascular outcomes, diabetes and fertility) did not provide evidence for additional benefits associated with selenium intake beyond that required for the levelling off of selenoprotein P plasma concentrations.

9.2.4 Agency for Toxic Substances and Disease Registry (ATSDR)

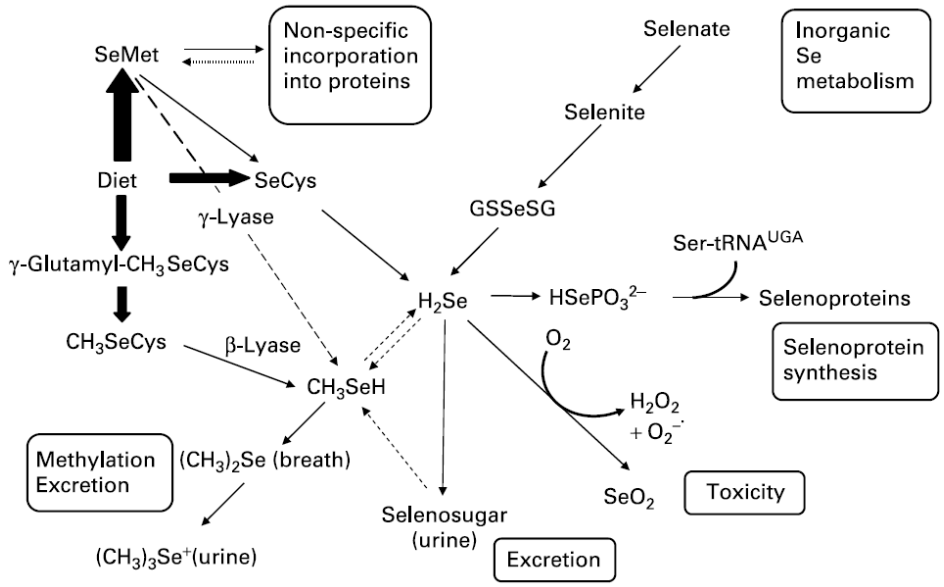
ATSDR noted that selenium could have nutritional or toxic effects and noted the Recommended Dietary Allowance (RDA; 55 μg/day for adult males and females) and the Tolerable Upper Intake Level (UL; 400 μg/day) derived by the National Academy of Sciences (NAS) (ATSDR, 2003). The ASTDR assessment focussed on toxicological aspects of selenium exposure. It was concluded that:

- Acute oral exposure to extremely high levels of selenium (several thousand times normal daily intake) could result in nausea, diarrhoea and vomiting, and occasionally cardiovascular symptoms, such as tachycardia. In laboratory animals, myocardial degeneration has been observed following very high acute or intermediate duration to selenium
- Chronic oral intake of very high levels of selenium (10-20 times normal daily intake) can produce selenosis, characterised by mainly dermal or neurological effects (see section 9.1.2 on this report). It was noted that selenosis appears to be more prevalent in the presence of other nutritional inadequacies
- Effects on the endocrine system have been noted in humans and animals, following long term exposure to elevated dietary levels of selenium. These effects were characterised by decreased thyroid T_3 hormone levels. However, hormone levels remained within the normal range and it is uncertain whether these effects can be classed as adverse
- No acute or intermediate minimal risk levels (MRLs) for oral exposure to selenium were derived due to insufficient information on adverse health effects. An MRL of 5 μg/kg bw/day was derived for chronic oral exposure. This was based on a NOAEL of 819 μg/day from a Chinese study (Yang and Zhou, 1994) of selenosis (15 μg/kg bw/day) and an uncertainty factor of 3, to account for human variability.

9.3 METABOLISM OF SELENIUM

The mechanisms of metabolism of various ingested forms of selenium in humans is summarised in Figure 1.

Figure 1. Metabolism of selenium in humans



Reproduced from Rayman et al. (2008)

These metabolic pathways indicate that all forms of ingested selenium may contribute to the synthesis of the functional selenoproteins. Synthesis of these proteins appears to be a saturable process, with body levels of the proteins plateauing once nutritional sufficiency is achieved. However, SeMet can continue to accumulate in the body through non-specific incorporation into general body proteins. Nutritionally, this creates a store of selenium, with SeMet released by catabolism of body proteins then available for the synthesis of selenoproteins. However, in selenium-replete individuals there also appears to be potential for released SeMet to contribute to the production of deleterious reactive oxygen species.

The metabolic pathways in Figure 1 suggest that excess inorganic selenium will either contribute to toxicity or be excreted, but will not be stored in the body.

10. EXPOSURE ASSESSMENT - SELENIUM

10.1 SELENIUM IN THE NEW ZEALAND FOOD SUPPLY

Selenium has been included as an analyte in five of the seven New Zealand Total Diet Studies (NZTDSs) carried out to date (ESR/MoH, 1994; Pickston *et al.*, 1985; Vannoort *et al.*, 2000; Vannoort and Thomson, 2005; Vannoort and Thomson, 2011). Table 7 summarises information on the selenium content of New Zealand foods from these studies. It should be noted that details of concentration data for selenium in foods are only available for three of these studies (Vannoort *et al.*, 2000; Vannoort and Thomson, 2005; Vannoort and Thomson, 2011).

Table 7. Selenium in foods available in New Zealand

Food/food group	Mean or Range of means (mg/kg)		
	1997/98 NZTDS	2003/04 NZTDS	2009 NZTDS
Grain products	0.01-0.20	0.01-0.10	<0.01-0.20
Fruits and vegetables	-	<0.01	<0.01
Nuts and nut products	0.09-0.11	0.06-0.13	0.06-0.08
Cheese	0.14	0.06	0.08
Other dairy products	0.01-0.02	<0.01	<0.01
Bacon	0.14	0.14	0.14
Ham	-	0.14	0.18
Pork	0.16	0.14	0.17
Chicken	0.21	0.20	0.34
Eggs	0.39	0.27	0.24
Fish and fish products	0.20-0.71	0.25-0.51	0.28-0.46
Shellfish	0.71-1.42	0.41-0.57	0.44-0.45
Lambs' liver	0.29	0.20	0.17
Other meat and meat products	0.05-0.11	0.04-0.08	0.06-0.10
Mushrooms	-	0.22	0.17
Yeast extract	-	0.02	0.12

NZTDS: New Zealand Total Diet Survey/Study

Birds preferentially accumulate selenium in egg yolk to support embryonic development (Pappas *et al.*, 2006). A study of selenium in egg yolks of wild bird species from New Zealand (n = 3), Canada (n = 8) and the United Kingdom (n = 3) found mean selenium concentration of 1147, 1194 and 522 μg/kg, respectively. The authors concluded that the selenium content of egg yolk was more strongly affected by the phylogenetic relatedness of the bird species than the geographical location. No impact of New Zealand's low selenium status was observed in egg selenium. The species sampled in New Zealand were blackbird (*Turdus merula*), thrush (*Turdus philomelos*) and starling (*Sturnus vulgaris*). There were no species in common across the three countries included in the study.

No reports were found of analyses of species forms of selenium in New Zealand foods.

10.2 SELENIUM IN THE AUSTRALIAN FOOD SUPPLY

Selenium was included as an analyte in the 19th, 20th, 22nd and 23rd Australian Total Diet Studies (ANZFA, 2003; FSANZ, 2003; 2008; 2011). Selenium data from these studies are summarised in Table 8. Data have been arranged to align as much as possible with those in Table 7.

Table 8. Selenium in foods available in Australia

Food/food group	Mean or Range of means (mg/kg)			
	19th ATDS	20th ATDS	22nd ATDS	23rd ATDS
Grain products	0.02-0.15	<0.01-0.16	0.02-0.16	0.02-0.19
Fruits and vegetables	<0.01-0.04	<0.01-0.04	<0.01-0.03	<0.01-0.07
Nuts and nut products	<0.01-0.02	0.07-0.11	<0.01-0.18	0.03-0.23
Cheese	0.07	0.10	0.06-0.11	0.18
Other dairy products	<0.01-0.07	<0.01-0.04	<0.01-0.07	0.02-0.05
Bacon	-	0.20	0.25	0.34
Ham	0.15	0.22	0.15	0.43
Pork	0.30	-	0.34	-
Chicken	0.31	0.25	0.24	0.30-0.32
Eggs	0.27	0.28	0.24	0.31
Fish and fish products	0.36-0.53	0.28-0.89	0.29-0.63	0.39-0.87
Shellfish and crustaceans	0.33-0.67	0.60	0.34	0.56
Offals	0.41-1.44	0.40	0.47	0.37
Other meat and meat products	0.06-0.12	0.14-0.20	0.09-0.17	0.12-0.23
Mushrooms	0.16	0.18	0.12	0.18
Yeast extract	-	-	-	-

ATDS: Australian Total Diet Study

The patterns of selenium concentrations in foods are very similar from survey to survey and between the Australian and New Zealand surveys.

No information was found on selenium species in Australian retail foods.

10.3 OVERSEAS CONTEXT

Given the satisfactory data on the total selenium content of the New Zealand and Australian food supplies, the following section will focus on studies that have determined the species composition in foods. International data on selenium species in foods and beverages are summarised in Table 9.

Table 9. Selenium species in foods and beverages

Country	Food	Selenium species	Concentration, mean (range) (μg/kg or g/L)	Percent of total selenium	Reference
Argentina	Tap water	Se (IV)	0.37	24	Escudero *et al.* (2015)
		Se (VI)	1.17	76	
	Mineral water	Se (IV)	0.29	26	
		Se (VI)	0.84	74	
	Energy drink	Se (IV)	0.36	20	
		Se (VI)	1.43	80	
	Soft drink	Se (IV)	0.49	41	
		Se (VI)	0.70	59	
	Wine – Chablis	Se (IV)	1.04	65	
		Se (VI)	0.56	35	
	Wine – Sauvignon	Se (IV)	0.64	24	
		Se (VI)	2.05	76	
	Wine – Malbec	Se (IV)	1.70	89	
		Se (VI)	0.21	11	
	Yerba-Mate infusion	Se (IV)	0.47	31	
		Se (VI)	1.05	69	
	Tea, black, infusion	Se (IV)	0.77	31	
		Se (VI)	1.70	69	
	Tea, green, infusion	Se (IV)	0.79	46	
		Se (VI)	0.94	54	
	Tea, red, infusion	Se (IV)	0.76	22	
		Se (VI)	2.66	78	
Australia	Mushroom (*Agaricus bisporus*)		Caps Stalks		Maseko *et al.* (2013)
	- Control	SeMet	0.08 0.081		
		SeCys	4.16 5.35		

Country	Food	Selenium species	Concentration, mean (range) (μg/kg or g/L)	Percent of total selenium	Reference
	- 40 mg Se/L in irrigation water	SeMeSeCys SeMet SeCys SeMeSeCys	0.031 0.061 0.31 0.11 8.35 6.60 0.08 0.051		
China	Tea	Total Organic Se(IV) Se(VI)	14.3 11.5 2.08 0.10		Chen *et al.* (2015)
China	Rice	Total SeMet	2700-4500	>50-57%	Beilstein *et al.* (1991)
France			Control Enriched		Bierla *et al.* (2008)
	Chicken breast	Total	540 14,100		
		SeMet	360 14,000		
		SeCys	110 1,110		
	Chicken leg	Total	570 12,000		
		SeMet	320 12,000		
		SeCys	180 130		
	Lamb heart	Total	1260 13,000		
		SeMet	250 4,200		
		SeCys	750 3,400		
	Lamb liver	Total	1410 24,000		
		SeMet	250 2,600		
		SeCys	850 22,000		
	Lamb kidney	Total	4530 19,000		
		SeMet	400 3,300		
		SeCys	4000 17,000		
	Lamb muscle (*Psoas major)*	Total	420 9,900		
		SeMet	240 5,300		
		SeCys	150 930		
	Lamb muscle (*Longissimus dorsi*)	Total	310 8,900		
		SeMet	140 5,800		
		SeCys	130 740		
Spain	Cabbage				Funes-Collado *et al.* (2015)
	- Low Se	Total	11,000		
		$SeCys_2$	ND		
		SeMet	ND		
		Se(IV)	ND-500		
		Se(VI)	2800-5000		
	- Medium Se	Total	98,000		
		$SeCys_2$	ND		
		SeMet	700-2500		
		Se(IV)	500-1100		
		Se(VI)	29,000-40,000		
	- High Se	Total	952,000		
		$SeCys_2$	2800-3300		
		SeMet	2500-13,000		
		Se(IV)	2000-27,000		
		Se(VI)	307,000-347,000		
Spain	Wheaten flour	Se(IV)	ND		Moreno *et al.* (2004)
		Se(VI)	ND		
		$SeCys_2$	130		
		SeMet	360		
		TNSe+	ND		
	Oyster	Se(IV)	ND		
		Se(VI)	ND		
		$SeCys_2$	ND		
		SeMet	370-510		
		TNSe+	77-250		
	Mussel	Se(IV)	ND		
		Se(VI)	ND		
		$SeCys_2$	ND		
		SeMet	170-310		
		TNSe+	60-71		
	Tuna	Se(IV)	ND		
		Se(VI)	ND		
		$SeCys_2$	ND		
		SeMet	510-1100		
		TNSe+	ND		
	Trout	Se(IV)	ND		
		Se(VI)	ND		
		$SeCys_2$	ND		

Country	Food	Selenium species	Concentration, mean (range) (μg/kg or g/L)	Percent of total selenium	Reference
	Yeast	SeMet TNSe+ Se(IV) Se(VI) $SeCys_2$ SeMet TNSe+	180-580 22-61 280-3350 8010-8300 ND 102000-570000 ND		
Spain[c]	Chicken muscle Chicken liver Chicken kidney	Total SeMet Total SeMet Total SeMet	880 450 2340 300 3310 260	 51 13 8	Cabanero *et al.* (2005b)
Spain/ Portugal[c]	Swordfish Sardine Tuna	Total SeMet Total SeMet Total SeMet	2090 1950 1810 510 2320 1070	 93 28 46	Cabanero *et al.* (2005a)
Taiwan	Rice Wheat flour	Se(IV) Se(VI) $SeCys_2$ SeMet Se(IV) Se(VI) $SeCys_2$ SeMet	0.52 0.47 1.8 17.8 1.0 4.6 9.7 63.1	2 2 8 82 1 6 12 78	Tsai and Jiang (2011)
United Kingdom[a]	Bovine milk - Control - Feed supplemented with selenite - Feed supplemented with Se yeast Cheese - Control - Feed supplemented with selenite - Feed supplemented with Se yeast	 Total SeMet SeCys Other Se species Total SeMet SeCys Other Se species Total SeMet SeCys Other Se species Total SeMet SeCys Other Se species Total SeMet SeCys Other Se species Total SeMet SeCys Other Se species	 150 46 52 39 110 36 20 40 250 111 58 43 190 52 46 59 180 57 52 50 340 153 92 63		Phipps *et al.* (2008)
United Kingdom	White flour Wholemeal flour White bread 	SeMet SeCys SeMeSeCys Se (IV) Se (VI) Unknown SeMet SeCys SeMeSeCys Se (IV) Se (VI) Unknown SeMet SeCys SeMeSeCys Se (IV) Se (VI) Unknown		77-83 1.1-7.6 ND-8.6 ND-6.7 2.1-8.9 ND-8.8 72-87 4.6-10.8 ND-4.2 ND-6.9 1.6-8.2 ND-9.9 65-84 1.5-7.2 ND-4.4 2.2-7.7 2.0-7.1 ND-16.5	Hart *et al.* (2011)

Country	Food	Selenium species	Concentration, mean (range) (μg/kg or g/L)	Percent of total selenium	Reference
	Wholemeal bread	SeMet SeCys SeMeSeCys Se (IV) Se (VI) Unknown		71-87 2.6-11.4 ND-3.6 ND-7.7 1.6-4.8 ND-13.7	
United Kingdom	Se-rich wheat flour Se-enriched wheat flour Se-enriched bread	Total Se(VI) SeMet Total Se(VI) SeMet Total Se(VI) SeMet	452 5.3 256 1301 13.4 975 892 8.0 370	 1.2 59.6 1.0 74.5 0.9 42	Warburton and Goenaga-Infante (2007)
United Kingdom	Potato flesh Potato skin	Total Total soluble SeMet Total Total soluble SeMet	670 400 320 1990 815 180		Infante *et al.* (2009)
USA	Brazil nuts			Only SeMet detected	Vonderheide *et al.* (2002)
USA	Brazil nuts, with shells Brazil nuts, shelled Walnuts (black) Walnuts (white) Cashews Pecans	Total SeMet Total SeMet Total SeMet Total SeMet Total SeMet Total SeMet	35,100 8300 380 200 270 100 	 25[b] 21 19 23 22 25	Kannamkumarath *et al.* (2002)
USA	Tuna, canned Shark Marlin	Total extractable SeMet Total extractable SeMet Total extractable SeMet	5500 1600 900 500 3500 1600		Reyes *et al.* (2009)
USA	Onion - Control - Se-enriched Garlic - Control - Se-enriched Yeast - Control - Se-enriched	 Se(VI) Se(VI) γ-GSeMeSeCys SeMet γ-GSeMeSeCys γ-GSeMeSeCys SeMeSeCys SeMet SeMet Se(VI) SeMet Se(IV)		 100 10-33 35-63 53 31 8-73 3-60 13-28 90 10 60-85 1-15	Kotrebai *et al.* (2000)
Spain	Wheat Yeast	SeMet		59 68	Huerta *et al.* (2003)
Spain	Seed sprouts Alfalfa - Control - Se-enriched	 Total Se(IV) Se(VI) $SeCys_2$ SeMet Total Se(IV) Se(VI) $SeCys_2$ SeMet	 1500 ND ND ND ND 132,000-284,000 19,000-21,1000 21,400-30,000 4000-4250 13,600-20,100		Funes-Collado *et al.* (2013)

Country	Food	Selenium species	Concentration, mean (range) (μg/kg or g/L)	Percent of total selenium	Reference
	Lentil				
	- Control	Total	2400		
		Se(IV)	150		
		Se(VI)	300		
		$SeCys_2$	100		
		SeMet	1150		
	- Se-enriched	Total	98,000-111,000		
		Se(IV)	5,610-8,500		
		Se(VI)	31,000-53,300		
		$SeCys_2$	4300-10,100		
		SeMet	16,100-24,000		
	Soy				
	- Control	Total	800		
		Se(IV)	70		
		Se(VI)	520		
		$SeCys_2$	ND		
		SeMet	107		
	- Se-enriched	Total	158,000-188,000		
		Se(IV)	16,100-17,500		
		Se(VI)	70,000-85,000		
		$SeCys_2$	1900-2600		
		SeMet	14,900-29,100		

Se(IV): selenite Se(VI): selenate SeMet: selenomethionine
$SeCys_2$: selenocystine SeCys: selenocysteine SeMeSeCys: selenomethylselenocysteine
TNSe+: trimethylselenonium cation γ-GSeMeSeCys: γ-glutamyl-selenomethylselenocysteine

[a] Results are expressed on a dry weight basis. Results given here are after 112 days of supplemental feeding. Supplementation rates were 0.3 mg Se/kg dry matter
[b] Selenium was not completely recovered from the separation column and these percentages should be viewed as minimum values
[c] Results are expressed on a dry weight basis

The impact of soil selenium status on the selenium content of foods has been demonstrated in Finland (Alfthan *et al.*, 2015). During the 1980s, Finland responded to the low selenium status of the country by addition of selenium to all fertilisers at a rates in the range 6 to 15 mg/kg. Due to this intervention, the selenium content of spring cereals increased about 15-fold, while mean increases in beef, pork and milk selenium were 6-, 2- and 3-fold, respectively.

The development of selenium species was followed in broccoli grown in media enriched with sodium selenite (Pedrero *et al.*, 2007). After 40 days, SeMet was the dominant species in plant roots, followed by SeMeSeCys and Se(IV). However, SeMeSeCys dominated in the broccoli fruit, followed by SeMet and Se(IV). SeMeSeCys is a selenium detoxification product in plants and the dominance of this compound and the lower proportion of SeMet in broccoli fruit suggests that the ability of the fruit to metabolise selenium to SeMet may have become saturated. Approximately 70% of the SeMet present in the fruit was estimated to be incorporated into peptides or proteins.

Šindelářová *et al.* (2015) also enriched the selenium content of broccoli by foliar application of aqueous sodium selenate (Se(VI)). Total selenium in broccoli heads increased 15-18 fold at the highest application rate of sodium selenite. SeMet was the dominant species in heads from control plants (24-34%), followed by $SeCys_2$, Se(VI), SeMeSeCys and an unidentified selenium compound. The relative proportions of the selenium species were similar in the heads from plants receiving foliar selenium, with SeMet remaining the main selenium species.

Selenium species were quantified in pak choi, kale and broccoli grown in selenate (Se(VI)) enriched media (Thosaikham *et al.*, 2014). The authors reported virtually quantitative extraction of selenium species. Inorganic selenium (not specified, but presumably Se(VI)) was the main species in enriched pak choi (45% of total selenium), followed by SeMeSeCys

(22%) and SeMet (14%). In kale and broccoli, SeMeSeCys was the dominant species. It should be noted that there is a lack of agreement amongst the three studies that have considered selenium speciation in enriched broccoli (Pedrero *et al.*, 2007; Šindelářová *et al.*, 2015; Thosaikham *et al.*, 2014). While two of these studies identify SeMeSeCys as the main species, the third reports SeMet to be the main species.

Mushrooms (*Agaricus bisporus*) were grown on a media enriched with sodium selenite (Se(IV)) (Stefánka *et al.*, 2001). The only selenium species detected in an enzymatic extract (75% of selenium extracted) were Se(IV) and $SeCys_2$, with approximately two-thirds of the identified selenium present as Se(IV).

Selenium speciation was carried out on three samples of selenium-enriched rice by enzymatic extraction followed by CE-ICP-MS (Zhao *et al.*, 2011). Only SeMet was detected in rice extracts, with concentrations found in the range 0.136 to 0.143 mg/kg selenium on a dry weight basis. Detected SeMet accounted for approximately 96% of total selenium for all three samples analysed.

Wheat varieties (bread and durum) were enriched with selenium (sodium selenate and selenite) by either foliar or soil application (Galinha *et al.*, 2015). Species were quantified by enzymatic extraction HPLC-ICP-MS, with SeMet and Se(VI) detected in enriched wheat. SeMet was the dominant species, accounting for 70-100% of total selenium, while Se(VI) accounted for no more than 5% of total selenium.

Wheat samples from a region of India (Punjab) were found to have particularly high selenium content (up to 185,000 μg/kg dry weight) (Cubadda *et al.*, 2010). Selenium species were determined in samples, covering a range of total selenium concentrations, by HPLC-ICP-MS following ultrasonication-assisted enzymatic extraction. Overall, 70-89% of total selenium was able to be speciated. SeMet accounted for 72-85% of the speciated selenium, while Se(VI) accounted for 2-6%. SeMeSeCys was present at low levels (<0.5%), but was more apparent in high selenium wheat samples. SeMeSeCys is a selenium detoxification product in plants and the presence of this compound and the higher proportion of Se(VI) and lower proportion of SeMet in high selenium wheat suggests that the ability of the wheat to metabolise selenium to SeMet may have become saturated.

Pumpkins were enriched with selenium by foliar application of sodium selenate (Smrkolj *et al.*, 2005b). Enriched pumpkins contained 5-8-fold higher selenium than untreated pumpkins. Following enzymatic treatment, 90% of selenium was found in the soluble fraction. SeMet accounted for 77-85% of total selenium.

Eggs were separated into white and yolk and extracted and analysed for SeMet, SeCys and Se(IV) (Lipiec *et al.*, 2010). Extraction allowed speciation of 62-100% of total egg selenium. Different speciation profiles were observed between egg white and yolk. In egg white the dominant species was SeMet (52-80%), with similar proportions of SeCys (9-30%) and Se(IV) (11-20%). In egg yolk, the dominant species was SeCys (73-81%), followed by SeMet (12-20%) and Se(IV) (6-8%). Selenium supplementation of layer hens affected mainly the egg white selenium, with SeCys and SeMet concentrations in the egg white approximately doubling, while Se(IV) concentration were unchanged.

10.4 ESTIMATES OF DIETARY EXPOSURE

10.4.1 New Zealand

A number of regional and national estimates of dietary selenium exposure/intake have been derived in New Zealand. Estimates are summarised in Table 10.

Table 10. Estimate dietary selenium intakes for New Zealand

Region	Population group	Selenium intake, mean (μg/day)	Reference
Otago	Males (*n* = 15) - Duplicate diet - Dietary record - Food frequency Females (*n* = 28) - Duplicate diet - Dietary record - Food frequency	 35 30 60 26 27 46	(Duffield and Thomson, 1999)
South Island	Infants, 6 to <12 months Toddlers, 12 to 24 months Women	7.9 13.7 38	(McLachlan *et al.*, 2004)
National (1982 NZTDS)	Adult males (11.3 MJ/day) Adult females (8.4 MJ/day) Young adult males (16.7 MJ/day)	61 45 90	(Pickston *et al.*, 1985)
National (1987/88 NZTDS)	Adult males, 25+ years Adult females, 25+ years Young males, 19-24 years Children, 4-6 years Young children, 1-3 years	63 45 80 33 27	(ESR/MoH, 1994)
National (1997/98 NZTDS)	Adult males, 25+ years Adult females, 25+ years Young males, 19-24 years Children, 4-6 years Young children, 1-3 years	78 55 82 42 31	(Vannoort *et al.*, 2000)
National (2003/04 NZTDS)	Adult males, 25+ years Adult females, 25+ years Young males, 19-24 years Boys, 11-14 years Girls, 11-14 years Children, 4-6 years Toddlers, 1-3 years Infants, 6-12 months	67 49 71 55 41 32 21 16	(Vannoort and Thomson, 2005)
National (2009 NZTDS)	Adult males, 25+ years Adult females, 25+ years Young males, 19-24 years Boys, 11-14 years Girls, 11-14 years Children, 4-6 years Toddlers, 1-3 years Infants, 6-12 months	78 56 82 70 51 41 26 21	(Vannoort and Thomson, 2011)

NZTDS: New Zealand Total Diet Study

Information from the periodic NZTDSs suggests that New Zealanders' dietary exposure to selenium has increased since the 1980s. This is plausible given the increasing diversification of the New Zealand food supply, including a wide range of imported foods from regions with higher soil selenium levels than New Zealand.

The major contributors to selenium intake in New Zealand (2009 NZTDS) are grain products, chicken, takeaway foods, seafood, beef, pork and eggs. SeMet is likely to be the predominant form of selenium in these foods.

10.4.2 Australia

Dietary exposure to selenium has been estimated for the Australian population as part of the 19th, 20th, 22nd and 23rd ATDSs (ANZFA, 2003; FSANZ, 2003; 2008; 2011). Results are summarised in Table 11. It should be noted that for the 19th and 20th ATDSs exposure/intake estimates were presented in terms of μg/kg bw/day. RDIs for each age-gender group were

used to calculate mean body weights for each age-gender group, which were subsequently used to convert intake estimates to μg/day.

Table 11. Estimate dietary selenium intakes for Australia

ATDS	Age-gender groups	Intake estimate, mean (95th percentile), μg/day	Main contributing foods	References
19th (1998)	Adult males, 25-34 years Adult females, 25-34 years Boys, 12 years Girls, 12 years Toddlers, 2 years Infants, 9 months	90-128 58-84 60-80 46-63 31-44 9-26		ANZFA (2003)
20th (2000/01)	Adult males, 25-34 years Adult females, 25-34 years Boys, 12 years Girls, 12 years Toddlers, 2 years Infants, 9 months	96-116 63-78 73-81 59-68 37-42 20-22		(FSANZ, 2003)
22nd (2004)	Males - 70+ years - 50-69 years - 30-49 years - 19-29 years - 14-18 years - 9-13 years - 4-8 years - 2-3 years Females - 70+ years - 50-69 years - 30-49 years - 19-29 years - 14-18 years - 9-13 years - 4-8 years - 2-3 years Infants	 67 (99) 75 (117) 81 (130) 90 (143) 84 (124) 63 (120) 48 (84) 37 (70) 52 (69) 54 (74) 55 (80) 57 (88) 56 (88) 48 (70) 44 (57) 41 (52) 14 (36)	Bread (infant formula for infant age group)	(FSANZ, 2008)
23rd (2008)	Males - 70+ years - 50-69 years - 30-49 years - 19-29 years - 17-18 years - 14-16 years - 9-13 years - 4-8 years - 2-3 years Females - 70+ years - 50-69 years - 30-49 years - 19-29 years - 17-18 years - 14-16 years - 9-13 years - 4-8 years - 2-3 years Infants	 130 (184) 143 (215) 155 (232) 171 (266) 167 (249) 150 (204) 113 (178) 98 (146) 87 (124) 97 (131) 103 (147) 106 (157) 111 (183) 108 (197) 101 (142) 94 (136) 87 (120) 82 (114) 38 (76)	Milk and cream, bread, meat (infant formula for infant group)	(FSANZ, 2011)

Estimated selenium intakes for the Australian population are generally higher than equivalent estimates for the New Zealand population. However, estimates more than doubled for most age-gender groups from the 22nd to the 23rd ATDS. These differences were considered to be, at least in part, due to the inclusion of coffee in the 23rd ATDS, in place of tea. In the ATDSs, coffee was found to contain more selenium than tea.

10.4.3 Overseas estimates of dietary exposure

Total selenium

Table 12 summarises results of overseas studies that have derived estimates of dietary exposure to selenium.

Table 12. International estimates of dietary exposure to selenium

Country	Population group	Mean (high percentile) exposure, µg/day[a]	Main foods contributing	Reference
Denmark	General population - Males (*n* = 566) - Females (*n* = 691) Vegans - Males (*n* = 33) - Females (*n* = 37)	 52 (Range 44-60) 39 (33-46) 33 (25-40) 25 (19-30)		Kristensen *et al.* (2015)
Finland	Males 1 to <3 years 3 to <10 years 10 to <18 years 18 to <65 years 65 to <75 years Females 1 to <3 years 3 to <10 years 10 to <18 years 18 to <65 years 65 to <75 years	 36.3 (61.6) 45.9 (68.5) 60.3 (95.5) 65.6 (113.0) 54.1 (95.5) 35.8 (56.9) 41.1 (62.3) 46.9 (76.4) 49.6 (81.1) 42.7 (68.7)		EFSA (2014a)
France	Males 3 to <10 years 10 to <18 years 18 to <65 years 65 to <75 years ≥75 years Females 3 to <10 years 10 to <18 years 18 to <65 years 65 to <75 years ≥75 years	 33.2 (51.4) 41.6 (66.5) 48.8 (78.8) 49.8 (80.0) 44.1 29.7 (46.5) 33.9 (55.9) 37.9 (62.7) 38.8 (62.8) 35.2		EFSA (2014a)
Germany	Males 1 to <3 years 3 to <10 years 10 to <18 years Females 1 to <3 years 3 to <10 years 10 to <18 years	 19.8 (32.2) 22.5/41.4 (33.9/71.5) 42.2 (67.4) 17.2 (28.3) 20.6/34.9 (31.3/57.1) 39.0 (63.8)		EFSA (2014a)
Greece	Total population	39.3	Bread, meat	Pappa *et al.* (2006)
Guatemala	Institutionalised children (duplicate diet)	58.7-69.6		Cabrera-Vique *et al.* (2015)

Country	Population group	Mean (high percentile) exposure, μg/day[a]	Main foods contributing	Reference
Ireland	Males			EFSA (2014a)
	18 to <65 years	59.0 (92.2)		
	65 to <75 years	58.2 (104.4)		
	≥75 years	47.6		
	Females			
	18 to <65 years	44.0 (70.4)		
	65 to <75 years	47.2 (83.5)		
	≥75 years	40.2		
Italy	Males			EFSA (2014a)
	1 to <3 years	25.3		
	3 to <10 years	37.6 (66.3)		
	10 to <18 years	45.9 (88.4)		
	18 to <65 years	42.7 (82.3)		
	65 to <75 years	41.6 (93.4)		
	≥75 years	38.8 (64.6)		
	Females			
	1 to <3 years	24.9		
	3 to <10 years	34.5 (65.2)		
	10 to <18 years	39.6 (87.8)		
	18 to <65 years	35.8 (70.9)		
	65 to <75 years	35.0 (67.8)		
	≥75 years	31.0 (49.6)		
Latvia	10 to <18 years	50.9		EFSA (2014a)
	18 to <65 years	50.3 (82.9)		
Netherlands	Males			EFSA (2014a)
	3 to <10 years	35.5 (59.4)		
	10 to <18 years	45.5 (77.5)		
	18 to <65 years	56.9 (97.1)		
	65 to <75 years	53.3 (94.0)		
	Females			
	3 to <10 years	32.8 (51.3)		
	10 to <18 years	36.8 (59.6)		
	18 to <65 years	43.9 (72.9)		
	65 to <75 years	40.7 (70.9)		
Slovenia	Military personnel	87 (range 34-163)		Smrkolj *et al.* (2005a)
Sweden	Males			EFSA (2014a)
	18 to <65 years	63.3 (111.5)		
	65 to <75 years	59.6 (93.9)		
	≥75 years	61.7		
	Females			
	18 to <65 years	50.5 (85.4)		
	65 to <75 years	50.7 (88.6)		
	≥75 years	49.5		
United Kingdom	Males			EFSA (2014a)
	1 to <3 years	25.3 (41.1)		
	3 to <10 years	33.8 (53.4)		
	10 to <18 years	44.6 (74.3)		
	18 to <65 years	51.3 (86.8)		
	65 to <75 years	52.2 (84.9)		
	≥75 years	44.3		
	Females			
	1 to <3 years	24.2 (38.0)		
	3 to <10 years	30.5 (49.2)		
	10 to <18 years	35.2 (56.6)		
	18 to <65 years	41.6 (71.2)		
	65 to <75 years	42.6 (68.2)		
	≥75 years	39.6 (63.5)		

[a] High percentile dietary exposure estimates are usually expressed as the 95th percentile, but may occasionally be expressed as the 90th or 97.5th percentile. Where a range is presented for the exposure estimate, this usually refers to different treatments of left-censored analytical data

[b] Published results expressed as 'μg/day'. A body weight of 60 kg was used to convert exposures to 'μg/kg bw/day'

Selenium Species

No dietary exposure estimates were found for selenium species. However, information on the species forms of selenium in foods suggest that the majority of the dietary exposure summarised in Table 9 will be exposure to SeMet.

10.4.4 Biomarkers of exposure

There are currently a number of biomarkers of selenium exposure in use. However, the choice of biomarker will depend on the specific requirements of the biomonitoring study.

Selenium can be detected in blood and blood components, faeces, urine, hair and nails of exposed individuals (ATSDR, 2003). However, hair selenium can be affected by use of selenium sulphide as an anti-dandruff treatment. Toenail selenium has been used in epidemiological studies, as an indicator of long-term selenium intake (Cai *et al.*, 2016). However, toenail selenium does not represent any functional aspect of selenium and should be viewed predominantly as a form of selenium excretion (Combs, 2015).

Plasma selenium has been reported to be a good indicator of recent selenium exposure when selenium is in organic forms (SeMet or high-selenium yeast), but not when inorganic selenium is the intake form (Combs, 2015; Fairweather-Tait *et al.*, 2010). This is likely to be due to the non-specific incorporation of SeMet into plasma proteins, in place of methionine. However, in selenium-replete individuals, inorganic selenium will be processed by the liver and largely excreted (Combs, 2015).

There is evidence that plasma selenium concentrations correlate with GPx activity at low blood selenium levels, but not at high levels (ATSDR, 2003). GPx activities reach a plateau, presumably due to homeostatic control of the synthesis of this enzyme. Similar observations have been made for selenoprotein P (Duffield *et al.*, 1999). This suggests that plasma selenium may be a suitable biomarker of selenium exposure, while GPx or selenoprotein P concentrations in blood or blood components may be useful biomarkers for selenium nutritional status. EFSA noted that plasma selenoprotein P status is saturable, but this occurs at higher intake levels to the plateauing of GPx activity (EFSA, 2014a). EFSA have proposed plasma selenoprotein P as a biomarker of selenium functional status.

Urinary selenium has also been used as a useful indicator of selenium status, particularly in the toxic range (Kobayashi *et al.*, 2002). Selenium is detoxified by methylation. In the range of sufficient to marginally toxic intake, selenium is excreted in a monomethylated form, while at higher, toxic intakes selenium is excreted in di- or trimethylated forms. However, urinary excretion of selenium also appears to be impacted by gender, with women excreting more selenium than men at similar intakes, and genetics, with different genotypes of tissue GPx genes resulting in different selenium excretion patterns (Combs, 2015).

Biomonitoring equivalents values have been derived to relate biomarker levels of selenium to health-based exposure limits or nutrient reference values (Hays *et al.*, 2014). An estimated average requirement (EAR) of 45 μg/day was determined to be equivalent to whole blood, plasma and urinary concentrations of selenium of 100, 80 and 10 μg/L, respectively. An upper level of intake (UL) of 800 μg/day was determined to be equivalent to whole blood, plasma and urinary selenium concentrations of 480, 230 and 110 μg/L, respectively.

11. RISK CHARACTERISATION - SELENIUM

11.1 ADVERSE HEALTH EFFECTS IN NEW ZEALAND

While a number of studies have noted the low selenium intake and low selenium status of some New Zealanders, clinical signs of deficiency have not been reported (Duffield and Thomson, 1999; Thomson and Robinson, 1996; Thomson, 2004), except in one case associated with total parenteral nutrition (van Rij *et al.*, 1979). No reports of selenium toxicity in New Zealand were found.

11.1.1 Risk assessment

The periodic NZTDSs provide an assessment of nutritional risks associated with selenium intake in New Zealand, through comparison of estimate dietary intakes with dietary reference values (DRVs) (ESR/MoH, 1994; Pickston *et al.*, 1985; Vannoort *et al.*, 1995; Vannoort and Thomson, 2005; Vannoort and Thomson, 2011). Table 13 summarises selenium dietary intake estimates from the most recent NZTDS and the associated DRVs.

Table 13. Dietary selenium intakes

Selenium intake (µg/day)	25+ years Males	25+ years Females	19-24 years Young Males	11-14 years Boys[d]	11-14 years Girls[d]	5-6 years Children[e]	1-3 years Toddlers	6-12 month Infants[f]
2009 NZTDS[a]	**78**	**56**	**82**	**70**	**51**	**41**	**26**	**21**
EAR[b] (µg/day)	60	50	60	40	40	25	20	15 (AI)
UL[c] (µg/day)	400	400	400	280	280	150	90	60

Reproduced from Vannoort and Thomson (2011)

[a] The intake for each age-gender cohort is based on assigning not detected results to half LOD

[b] Estimated Average Requirement

[c] Upper Level of Intake

[d] Nutrient reference values (NRV) for 11-14 year boys/girls extrapolated from values for 9-13 year children

[e] NRVs for 5-6 year children extrapolated from 4-8 year children

[f] NRVs for 6-12 month infants from 7-12 month infants

Based on the information in Table 13, current New Zealand mean dietary intakes of selenium exceed the EAR, but are well below the UL.

11.2 HEALTH EFFECTS OVERSEAS

11.2.1 Epidemiological studies

A huge number of epidemiological studies have been carried out relating selenium status and/or selenium supplementation to a wide range of health states. It is not possible to summarise all of these studies here and the following summaries should be considered to be representative, rather than exhaustive. However, an attempt has been made to summarise meta-analyses, systematic reviews and large intervention studies.

Nutritional status

A meta-analysis of animal and human studies was carried out to determine the impact of different forms of selenium on the activity of the selenoenzyme GPx (Bermingham *et al.*, 2014). For animals, selenium-enriched foods were more effective in increasing GPx activity than SeMet. In humans, GPx response appears to be dependent on both the form of

selenium ingested and the baseline selenium status of the subject. SeMet is able to increase plasma selenium concentrations in subjects with low or optimal selenium status, while selenite is less effective in subjects that do not have low selenium status. Selenium-enriched protein sources appear to enhance the rate of GPx increase compared to selenium-yeast.

Three groups of men (*n* = 10 per group) with low selenium status received supplementary selenium (200 μg/day) as sodium selenate, Se-rich yeast or Se-rich wheat (Levander *et al.*, 1983). Plasma selenium increased steadily across the study period for subjects receiving yeast or wheat, but plateaued after 4 weeks in the selenate-supplemented group. Platelet GPx activity increased rapidly in the wheat and selenite-supplemented groups, then plateaued. Platelet GPx activity increased more slowly in the yeast group, but 10 weeks after the end of the supplementation period platelet GPx activities remained higher in the wheat and yeast-supplemented groups. It is likely that some of the SeMet from wheat and yeast would have been incorporated non-specifically into proteins and subsequently released through catabolism and was utilised for production of platelet GPx.

A cohort (*n* = 120) from a selenium-deficient region of China were randomised into groups receiving either no supplemental selenium or up to 66 μg/day for 20 weeks, as SeMet or selenite (Xia *et al.*, 2005). Plasma selenium increased with increasing supplement concentration, with a greater increase seen with SeMet than selenite. Plasma GPx sufficiency, as assessed by plateauing of GPx activity, was achieved with SeMet supplementation of 37 μg/day or greater, while selenite supplementation at 66 μg/day was required to achieve GPx sufficiency. Selenoprotein P sufficiency was not achieved under any of the supplementation protocols employed.

Cancer protection

Selenium exposure, usually at levels above nutritional sufficiency have been reported to have a protective effect with respect to cancer.

A meta-analysis of 69 studies indicated that high selenium exposure had a protective effect on cancer risk, with a pooled odds ratio of 0.78 (95th percentile confidence interval 0.73-0.83) compared to the lowest selenium exposure category (Cai *et al.*, 2016). It should be noted that this study did not define ‘high selenium exposure’ Dose-response analysis indicated a significant impact of plasma/blood selenium or toenail selenium on overall cancer risk, but no significant efficacy trend for supplemental selenium. At the cancer site level, there were protective effects for breast, lung, oesophageal, gastric, and prostate cancers, but not for colorectal, bladder, or skin cancer.

The SELECT study was an intervention study, to examine the potential impact of selenium (200 μg/day as SeMet) and vitamin E supplementation on prostate cancer risk (Lippman *et al.*, 2009). The study was randomised and placebo controlled and included approximately 35,000 males. At the planned 7-year interim analysis (median follow-up period 5.46 years), the study was discontinued as there was no significant benefit from either study agent for prostate cancer risk. Further analysis also showed no significant benefit with respect to lung, colorectal or other primary cancers.

These results are largely consistent with a recent systematic review (Vinceti *et al.*, 2014). The review considered 55 observational studies (total of more than 1.1 million participants) and 8 randomised controlled trials (RCTs; total of approximately 45,000 participants, including the SELECT study). The observational studies found a lower incidence (summary odds ratio 0.69, 95% CI 0.53-0.91) and lower cancer mortality (summary odds ratio 0.60, 95% CI 0.39-0.93) associated with higher selenium exposure, with the most pronounced effect seen for stomach, bladder and prostate cancers. However, these findings have

limitations due to study design, quality and heterogeneity. The RCTs found no clear evidence of a reduced risk of cancer or reduction in cancer-related mortality due to selenium supplementation.

A meta-analysis of studies relating selenium exposure to gastric cancer risk found in overall protective effect (odds ratio significantly less than one) for selenium in case-control studies (OR 0.62, 95% CI 0.44-0.89) and cohort studies (OR 0.87, 95% CI 0.78-0.97) (Gong *et al.*, 2016). High selenium status was also associated with a decreased risk of gastric cancer mortality.

A review and meta-analysis of studies investigating associations between selenium supplementation and lung cancer concluded that for those with serum selenium below 106 μg/L may reduce the risk of lung cancer, but may increase the risk of lung cancer for those with baseline serum selenium greater than 122 μg/L (Fritz *et al.*, 2011). Pooled data from two trials found no impact of selenium supplementation on lung cancer risk.

Cancer causation

Several studies have suggested an association between selenium status and risk of melanoma. A small case-control study (*n* = 54 cases, 56 controls) was carried out in northern Italy (Vinceti *et al.*, 2012). Selenium status was assessed by plasma selenium, toenail selenium and dietary selenium intakes, as assessed by a food frequency questionnaire. Plasma selenium, but not toenail or dietary selenium, was significantly associated with increased risk of melanoma. The authors noted that no clear mechanism for causation of melanoma by selenium has been established and that dietary intakes and plasma selenium levels were within the range usually associated with nutritional adequacy.

An earlier and larger case-control study (*n* = 278 cases, 278 controls) investigating risk factors for melanoma amongst Hawaiian Caucasians found no significant associations between plasma, erythrocyte or toenail selenium and melanoma risk (Le Marchand *et al.*, 2006). A meta-analysis of 9 prospective studies also found no association between selenium and other antioxidants, individually or in combination, and risk of melanoma (Miura and Green, 2015).

A small Argentine case-control study (*n* = 27 cases, 86 control) found a significant positive association between occurrence of oral squamous cell carcinoma and selenium intake, as assessment by semi-quantitative food frequency questionnaire (Secchi *et al.*, 2015). It should be noted that all selenium intakes were quite high; the controls had a mean intake of 107 μg/day, while cases had a mean intake of 143 μg/day. While selenium intake was also significant in a linear regression model, the odds ratio was only marginally above unity (1.02; 95th percentile confidence interval 1.00-1.03).

Non-cancer endpoints

The involvement of selenium in thyroid enzymes suggests that selenium deficiency may contribute to thyroid pathology. A study compared people in a selenium-adequate region of China (*n* = 3038) to people from a low-selenium region (*n* = 3114) (Wu *et al.*, 2015). The prevalence of pathological thyroid conditions was significantly lower in the selenium-adequate area (18.0% compared to 30.5%, p<0.001). Higher serum selenium was associated with lower odds ratio (95th percentile confidence interval) of autoimmune thyroiditis (0.47; 0.35-0.65), subclinical hypothyroidism (0.68; 0.58-0.93), hypothyroidism (0.75; 0.63-0.90), and enlarged thyroid (0.75; 0.59-0.97).

A cohort of elderly healthy Swedes (*n* = 668) was followed for an average of 6.8 years (Alehagen *et al.*, 2016). The cohort were found to have serum selenium concentrations corresponding to low selenium intake. Participants in the lowest quartile of serum selenium were found to have a 43% and 56% greater risk of all-cause and cardiovascular mortality, respectively. The authors recognised that the differences of serum selenium could have been driven by pathological conditions. However, inflammatory markers were not significantly different across the four quartiles of serum selenium.

Other studies have reported both negative and positive associations between selenium exposure and diabetes. A cross-sectional study (*n* = 5423) of older Chinese (40+ years) was carried out (Wei *et al.*, 2015). The prevalence of diabetes in the study population was 9.7%. The multivariate adjusted odds ratio for diabetes for the highest quartile of selenium intake compared to the lowest quartile was 1.52 (95th percentile confidence interval 1.01-2.28). The overall trend in diabetes risk across quartiles was also significant. However, it should be noted that other potential risk factors, such as body mass index (BMI), waist circumference, and drinking and smoking prevalence also increased across quartiles of selenium intake.

In contrast, a meta-analysis of 6 observational studies found that the serum selenium concentrations of women with gestational diabetes was significantly lower than normal pregnant women (Askari *et al.*, 2015). A meta-analysis of 4 RCTs (20,294 participants) found no significant impact of selenium supplementation on the risk of developing type 2 diabetes (Mao *et al.*, 2014).

The impact of selenium status and selenium supplementation on the risk of developing hypertensive conditions (pre-eclampsia and pregnancy-induced hypertension) was examined as part of the SPRINT (Selenium in PRegnancy INTervention) study (Rayman *et al.*, 2015). A randomised cohort of 230 primiparous UK women received either 60 μg/day of supplemental selenium or placebo from 12 weeks of gestation. In a stepwise logistic regression model, toenail selenium was associated with a significant reduced odds ratio for development of hypertensive conditions (OR 0.38, 95th percentile confidence interval 0.17-0.87). The cohort was found to have generally low selenium status at baseline and selenium supplementation was also associated with reduced risk of hypertensive conditions (OR 0.30, 95th percentile confidence interval 0.09-1.00).

A meta-analysis of 13 observational studies and 3 RCTs found that observational studies indicated an inverse association of blood selenium level and the risk of pre-eclampsia, while supplementation with selenium was significantly associated with a reduced incidence of pre-eclampsia (Xu *et al.*, 2015).

A meta-analysis of studies (16 prospective observational studies and 16 RCTs) investigating the relationship between selenium and cardiovascular disease (CVD) risk found a significant negative association between blood selenium and CVD risk, particularly across the concentration range 55-145 μg/L (Zhang *et al.*, 2016). However, while selenium supplementation significantly increased blood selenium, no significant decrease in CVD risk was found (relative risk 0.91, 95% CI 0.74-1.10). This is in agreement with the findings of a systematic review, that selenium supplementation did not significantly decrease the relative risk of all-cause mortality, CVD mortality, non-fatal CVD events or all CVD events (Rees *et al.*, 2013).

11.2.2 Risk assessments

Due to the large amount of epidemiological evidence related to selenium exposure, quantitative risk assessments are generally unnecessary. However, nutritional risks due to selenium intake are usually assessed as part of dietary intake studies, by comparison to

nutrient reference values. As dietary intake study comparisons of this type are available for New Zealand (Section 11.1.1), no international examples were summarised.

12. RISK MANAGEMENT INFORMATION - SELENIUM

12.1 RELEVANT FOOD CONTROLS: NEW ZEALAND

12.1.1 Establishment of regulatory limits

Standard 1.4.1 of the Australia New Zealand Food Standards Code does not specify maximum limits (ML) for selenium in any food.[26] However, the Animal Products (Contaminant Specifications) Notice 2008 defines a Maximum Permissible Level (MPL) for selenium (inorganic and organic selenium, as selenium) in 'meat, fat and offal of any animal' and 'eggs' of 2 mg/kg (NZFSA, 2008).

12.1.2 Imported Food Requirements (IFR)

There are currently no IFRs that require products imported into New Zealand to be monitored for selenium.

12.2 RELEVANT FOOD CONTROLS: OVERSEAS

12.2.1 Establishment of regulatory limits

While a number of countries have defined nutrient reference values for selenium as a nutrient, no references were found to regulatory limits for selenium as a contaminant.

12.2.2 Agricultural controls

In response to a naturally low selenium environment and increasing evidence of the essentiality of selenium, Finland established a Selenium Working Group in 1983 (Alfthan *et al.*, 2015). Initially, sodium selenate was added to multinutrient fertilisers used in agriculture and horticulture at rates of 16 mg Se/kg of fertiliser for cereals and 6 mg/kg for grasses. In 1990, the quantity of Se allowed in all solid multinutrient fertilisers was reduced to 6 mg/kg of fertiliser. In 1998, the quantity of selenium to be added to inorganic compound fertilisers for agriculture was increased to 10 mg Se/kg and since 2007 has been added as selenate at 15 mg Se/kg fertiliser. This has resulted in a substantial increase in the selenium content of many foods, an approximate doubling of the estimated dietary intake of selenium and an increase in mean plasma selenium concentrations from 70 μg/L to 111 μg/L.

12.3 INFLUENCE OF FOOD PROCESSING ON SELENIUM SPECIES LEVELS

12.3.1 Cooking

Boiling of cabbage (*Brassica oleracea*) resulted in decreases in the concentration of $SeCys_2$, Se(IV) and Se(VI), but increases in the concentration of SeMet (Funes-Collado *et al.*, 2015). Most of the selenium (65-100%) leached into the cooking water. The authors of this study did not clearly identify why the concentration of SeMet increased in cabbage extracts after boiling and SeMet was also detected in the water used to boil the cabbage. Se(VI) was the

[26] https://www.comlaw.gov.au/Details/F2015C00052 Accessed 2 February 2016

dominant selenium species in the raw cabbage, but concentrations decreased by approximately 75% in boiled cabbage.

Boiling of broccoli results in the formation of an unknown seleno-compound, probably formed from SeMeSeCys (Pedrero *et al.*, 2007). Se(VI) is also apparent in the cooked fruit, while it was not observed in the raw product. The water used to cook the broccoli contained a similar selenium profile to the raw broccoli fruit.

12.3.2 Brewing

Two-row barley was grown under five fertiliser regimes (control, 10 or 20 g/ha selenium as selenate or selenite) (Rodrigo *et al.*, 2014). Barley was then malted (germination and drying), before production of wort (aqueous extract of mashed malt) and beer. Selenate increased the selenium content of the barley to a greater extent than selenite (approximately 3-times at the same level of fertilisation). Selenate-fertilised barley also contained a greater proportion of selenium as SeMet (90%) than selenite-fertilised barley (~70%) or control barley (~60%). The balance of selenium was present as selenite in all cases.

Malting produced little change in the selenium content or species distribution of all barley samples. Wort production resulted in a substantial decrease in selenium content, by greater than 90% in most cases, with the majority of the SeMet apparently remaining in the spent mash. Only selenite was detected in wort from control and selenite-fertilised barley, while wort from selenate-fertilised barley still contained approximately two-thirds of selenium as SeMet. Finished beer samples contained total selenium in the range 12-73 μg/kg, with all selenium as selenite, except for one sample that contained about one-third of selenium in an unknown form. It should be noted that for some samples only about 20% of selenium was able to be extracted for speciation analysis.

13. CONCLUSIONS - SELENIUM

13.1 DESCRIPTION OF RISKS TO NEW ZEALAND CONSUMERS

Selenium is an essential trace element, with a relatively narrow gap between intake levels providing nutritional sufficiency and levels causing toxic effects. There is also some evidence to suggest a beneficial impact of selenium intakes between nutritional and toxic levels on a number of disease states, including cancer. However, the evidence for such beneficial effects are equivocal. There are reasonably consistent associations between higher selenium status and reduced disease risk. However, the ability of supplementation, in the form of selenite, selenate or SeMet or in the form of selenium enriched foods, to reduce disease risk is not supported by the weight of evidence, as measured by meta-analyses.

Despite New Zealand being recognised as a low-selenium environment, supported by studies showing low dietary intake of selenium and low levels of biomarkers, adverse effects due to selenium deficiency have not been identified. There is evidence that the selenium intake and status of New Zealanders has increased in the last 30 years.

SeMet is the main form of selenium present in most plant foods and is almost the sole form in animal products, except for the small amounts of SeCys present in functional selenoproteins. SeMet has some nutritional advantages, as its nonspecific incorporation into body proteins represents a storage form of selenium, in the event of periods of insufficient selenium dietary intake. It is unclear what the toxicological implications are of intake of different selenium species.

13.2 COMMENTARY ON RISK MANAGEMENT OPTIONS

There is currently a maximum permissible level for selenium in animal meat, fat or offal and eggs in New Zealand. No other risk management measures are currently in place to manage potential risks of selenium deficiency or toxicity. Current information on the selenium status of New Zealanders does not suggest that further risk management measures would be justified.

13.3 DATA GAPS

While there is reasonable data on the selenium content of the New Zealand food supply, no information is available on the species forms of selenium in New Zealand foods. Species in New Zealand foods are unlikely to be different to those seen internationally. However, international information on the species forms of selenium in foods is still fragmentary.

Information on adverse effects associated with marginal selenium deficiency or toxicity are currently lacking. Similarly, biomarkers or other clinical indicators of marginal deficiency/toxicity states are not currently available.

14. HAZARD IDENTIFICATION - VANADIUM

Vanadium is widely distributed in the earth's crust, with an average concentration of about 100 mg/kg (0.01%) (ATSDR, 2012a; EFSA, 2004). Vanadium occurs in over 70 minerals, carnatite and vanadinite being the most important from the point of view of mining (IPCS, 1988). Metallic vanadium does not occur in nature. Production of vanadium is linked with that of other metals such as iron, uranium, titanium, and aluminium. Extraction of vanadium from fossil fuels, including vanadium-rich oil and coal, tars, bitumens and asphaltites, is important in several countries.

Vanadium is mainly used (75-85% of production) as an additive to iron in the production of steel (IPCS, 1988).

Vanadium may be present in air, due to combustion of vanadium-rich fossil fuels (IPCS, 1988; USEPA, 1987). Levels in water are usually less than 10 μg/L. The main source of vanadium exposure for the general population is from ingestion of food.

14.1 STRUCTURE AND NOMENCLATURE

Vanadium can exist in six oxidation states; -1, 0, +2, +3, +4 and +5 (IPCS, 1988). The +3, +4 and +5 oxidation states are the most common, with the +4 oxidation state being the most stable. Organic forms of vanadium are generally unstable and vanadium in foods and the human body will generally be in free ionic form. Common vanadium compounds for the two major oxidation states are shown in Table 14.

Table 14. Common vanadium compounds

Chemical Name	Formula	Oxidation state	CAS No.
Vanadium pentoxide	V_2O_5	+5	1314-62-1
Sodium metavanadate	$NaVO_3$	+5	13718-26-8
Ammonium metavanadate	NH_4VO_3	+5	7803-55-6
Sodium orthovanadate	Na_3VO_4	+5	13721-39-6
Vanadyl sulphate	$VOSO_4$	+4	27774-13-6

CAS No.: Chemical Abstract Service Registry Number

14.2 OCCURRENCE

Vanadium occurs in most, if not all, foods, although concentrations are usually less than 30 μg/kg fresh weight (EFSA, 2004). The lowest concentrations (1-5 μg/kg) are found in fats and oils, fruits and vegetables. Higher concentrations (5-30 μg/kg) are found in grains, seafood, meat and dairy products. The highest reported levels (up to 1 mg/kg) are in seed products, such as dill seed and black pepper.

14.3 ANALYTICAL METHODS

As for other elements, analysis of vanadium and its species forms can be considered in terms of three processes; extraction, separation and detection. For total vanadium determination the processes of extraction and separation are replaced by a process of matrix destruction and solubilisation.

There has been considerably more research on speciation of vanadium in water and environmental samples than in food samples (Chen and Owens, 2008; Pyrzyńska and Wierzbicki, 2004). However, the following sections will focus on methods applied to food.

14.3.1 Sample extraction for vanadium species

Extraction procedures for vanadium speciation may involve extraction of total vanadium for subsequent separation or selective extraction of either V(IV) or V(V).

Selective extraction

Extraction of plant material with 1M $(NH_4)_2HPO_4$ (boiling for 15 minutes) has been reported to selectively extract V(V) (Mandiwana and Panichev, 2006; Tian *et al.*, 2014). The authors of these studies were unable to validate this procedure due to the lack of any suitable certified reference materials. An alternative extractant to solubilise V(V) from plant material was 0.1-0.5M Na_2CO_3 (shaking and ultrasonication at 80ºC for 10-60 minutes) (Khan *et al.*, 2011). This procedure was also not able to be validated due to the lack of suitable certified reference materials.

General extraction

While not strictly a food species, a study analysing vanadium species in the crustacean species, *Hyallela azteca*, analysed sample extracts for both V(IV) and V(V) (Jensen-Fontaine *et al.*, 2014). Dried samples (60ºC for 72 hours) of whole ground crustacea were extracted with 2.5 mM Na_2EDTA and the extracts were clarified by centrifugation and membrane filtration. Comparing the sum of the concentrations of the species forms to total vanadium, determined by atomic absorption spectroscopy, extract of vanadium by this method was in the range 11 to 71%.

14.3.2 Separation of vanadium species

One report of chromatographic separation of extracted vanadium species was identified (Jensen-Fontaine *et al.*, 2014). It should be noted that the matrix was not strictly a food, but a crustacean species, *Hyallela azteca.* Samples extracts were separated by HPLC on a strong anion exchange column under isocratic conditions. The mobile phase was 3% acetonitrile, 2mM Na_2EDTA, 80 mM ammonium bicarbonate. Detection of vanadium species was achieved by interfacing the HPLC with an ICP-MS (hyphenated technique).

14.3.3 Detection of vanadium species or total vanadium

Vanadium can be detected and quantified by:

- flame atomic absorption spectroscopy (Iwegbue *et al.*, 2010);
- electrothermal (graphite furnace) atomic absorption spectrophotometry (Bu-Olayan and Al-Yakoob, 1998; Fard *et al.*, 2015; Filik and Aksu, 2012; Khan *et al.*, 2011; Mandiwana and Panichev, 2006; Naeemullah *et al.*, 2015; Pekiner *et al.*, 2014; Sepe *et al.*, 2003; Wadhwa *et al.*, 2013; Zeeb *et al.*, 2014);
- inductively-coupled plasma mass spectrometry (ICP-MS) (Agusa *et al.*, 2005; Arnich *et al.*, 2012; Bocio *et al.*, 2005; Chen *et al.*, 2014; Copat *et al.*, 2013; Gimou *et al.*, 2014; Hassan *et al.*, 2012; Horner and Beauchemin, 2013; Iamiceli *et al.*, 2015; Llobet *et al.*, 1998; Marti-Cid *et al.*, 2009; Millour *et al.*, 2012; Nisianakis *et al.*, 2009; Tu *et al.*, 2008; Turconi *et al.*, 2009); and
- ICP-atomic emission spectroscopy (ICP-OES) (Dolan and Capar, 2002; Gummow *et al.*, 2005; Iyengar *et al.*, 2000; Oliveira *et al.*, 2009), in samples following wet or dry matrix destruction.

Less commonly used techniques for total vanadium include adsorptive stripping voltammetry (Abbasi *et al.*, 2012).

The performance of these methods can be improved in some cases by concentration of vanadium from the sample digest, usually by complexation and extraction. Cloud point extraction has been used, based on the formation of a ternary complex between vanadium, 2-(2´-thiazolylazo)-p-cresol, and ascorbic acid, with subsequent extraction–pre-concentration of the formed complexes using Triton X-100 (Filik and Aksu, 2012). In this technique, ascorbic acid reduces V(V) to V(IV) before complexation.

Alternative pre-concentration techniques include:

- Complexation of vanadium with 4-(2-pyridylazo) resorcinol, followed by separation into the ionic liquid, 1-butyl-3-methylimidazolium hexafluorophosphate [C4MIM][PF6] (Naeemullah *et al.*, 2015). A variation on this method involved complexation with N-benzoyl-N-phenylhydroxylamine, followed by *in situ* formation of an ionic liquid (Zeeb *et al.*, 2014).
- Complexation with 4-(5-bromo-2-pyridylazo)-5-(diethylamino)-phenol, followed by addition of mixture of dispersant solvent (ethanol) and extractant solvent (trichloroethylene). Phases were separated by centrifugation (Santos and Lemos, 2015).
- Complexation with 8-hydroxyquinoline, followed by adsorption onto multi-walled carbon nanotubes and elution with 1M HCl (Wadhwa *et al.*, 2013).

Neutron activation analysis (NAA) is a sensitive method for vanadium determination. The method involves activation of vanadium in a neutron flux, followed by monitoring of the 1.44 MeV γ-rays of ^{52}V (Byrne and Kosta, 1979). NAA can be carried out on intact samples. However, sensitivity of the technique can be improved further by either pre- or post-irradiation separation of vanadium from the matrix. It should be noted that NAA requires access to a nuclear reactor. Byrne and Kosta (1979) employed post-irradiation separation, which involved matrix destruction with 5M HCl-1.8M H_2SO_4-30% H_2O_2, followed by pH adjustment, oxidation with potassium permanganate to ensure all vanadium is in the V(V) form and complexation with N-benzoyl-N-phenylhydroxylamine combined with extraction into toluene.

14.3.4 Certified reference materials

While no certified reference materials are available for the speciated forms of vanadium, materials are available with certified values for total vanadium. Appropriate materials are summarised in Table 15.

Table 15. Certified reference materials for vanadium

Designation	Matrix	Certified value (mg/kg)
NIST SRM 1515	Apple leaves	0.26
NIST SRM 1547	Peach leaves	0.37
NIST SRM 1570a	Spinach leaves	0.57
GBW 07605	Tea	0.86

NIST: National Institute of Standards and Technology
SRM: Standard reference material
GBW: Reference materials from Institute of Environmental Health Monitoring (China)

14.3.5 Vanadium and vanadium species testing in New Zealand

Several New Zealand laboratories are accredited for the analysis of total vanadium in foods.[27] However, no New Zealand laboratories are accredited for the analysis of species

[27] http://www.ianz.govt.nz/directory/ Accessed 17 March 2016

forms of vanadium. No evidence was found in the scientific literature of any New Zealand testing for vanadium species.

It should be noted that achieving a suitably low limit of detection is essential for analysis of vanadium in foods. The 23rd Australian Total Diet Study analysed all foods for vanadium, but did not detect the element in any foods. While the study did not report the analytical limit of detection for vanadium, it was clearly insufficiently sensitive for the levels present in foods.

15. HAZARD CHARACTERISATION: ADVERSE HEALTH EFFECTS - VANADIUM

15.1 CONDITIONS

Vanadium is considered to be of low toxicity. Vanadium compounds have exhibited insulin mimetic properties and have been investigated for the treatment of diabetes mellitus and for use as body building supplements (Barceloux, 1999; Fortoul *et al.*, 2014). For example, vanadyl sulphate is used as a supplement to improve performance in weight training athletes at doses up to 60 mg/day (Barceloux, 1999).

Patients (*n* = 8) with non-insulin-dependent diabetes mellitus were given vanadyl sulphate (50 mg twice daily, orally) for 4 weeks (Boden *et al.*, 1996). Gastrointestinal side-effects (diarrhoea, abdominal cramps, flatulence and nausea) were experienced by six of eight patients during the first week, but the vanadyl sulphate was well tolerated subsequently.

There is some evidence of vanadium causing haemolysis in animal studies (FNB/IOM, 2001). A double-blind placebo-controlled challenge was conducted in a group of weight training athletes (Fawcett *et al.*, 1997). The challenge groups received oral vanadyl sulphate (dose approximately 0.5 mg/kg bw/day) for 12 weeks. No treatment related effects on haematology or biochemistry were observed.

A human fatality was reported following intentional ingestion of vanadium compounds (Boulassel *et al.*, 2011). The exposure dose was unknown. Autopsy revealed widespread asphyxia syndrome and erosive gastritis.

Vanadate (+5) and vanadyl (+4) have been shown to be developmental and reproductive toxins in laboratory animals (mice, rats, hamsters), with effects including decreased fertility, embryotoxicity, foetotoxicity and teratogenicity (Domingo, 1996). However, there is no evidence of similar effects in humans.

15.2 TOXICOLOGICAL ASSESSMENT

Five reasonably recent toxicological assessments of vanadium have been carried out (ATSDR, 2012a; EFSA, 2004; FNB/IOM, 2001; IARC, 2006; USEPA, 1987).

15.2.1 Agency for Toxic Substances and Disease Registry (ATSDR)

ATSDR have derived estimates of exposure posing minimal risks to human (MRLs). An MRL is defined as an estimate of daily human exposure to a substance that is likely to be without an appreciable risk of adverse effects (non-carcinogenic) over a specified duration of exposure.

While a NOAEL for gastrointestinal disturbance in human volunteers, receiving vanadium in the form of vanadyl sulphate (+2 oxidation state), can be determined, it was considered that the effects seen were due to local irritation of gastrointestinal tract, rather than a systemic effect. ATSDR concluded that the available human data was not suitable for derivation of an acute MRL.

ATSDR derived an intermediate duration (15 to 364 days) MRL of 0.01 mg/kg bw/day. The MRL was based on a NOAEL of 0.12 mg/kg bw/day as vanadyl sulphate trihydrate, in a cohort of people involved in a weight training programme, with dosing continued for 12 weeks. It should be noted that this dose level was the limit dose for the trial and no adverse health effects due to vanadium were observed in the study. An uncertainty factor of 10 was applied to the NOAEL to account for inter-individual variability.

ATSDR did not derive a chronic MRL for vanadium, due to the lack of long-term studies in humans and the lack of observed adverse effects in long-term studies in animals.

15.2.2 International Agency for Research on Cancer (IARC)

The IARC assessment considered vanadium pentoxide, in which vanadium is present in the +5 oxidation state. IARC concluded that there was:

- Inadequate evidence that vanadium pentoxide is carcinogenic to human.
- Sufficient evidence in experimental animals for the carcinogenicity of vanadium pentoxide.

Based on these conclusions, vanadium pentoxide was classified as Group 2B (possibly carcinogenic to humans).

15.2.3 European Food Safety Authority (EFSA)

EFSA were asked to derive an upper level for the intake of vanadium from food that would be unlikely to pose a risk of adverse health effects (EFSA, 2004). EFSA concluded that:

- Vanadium has not been shown to be essential for humans
- Orally administered vanadium compounds produce adverse effects on kidneys, spleen, lungs and blood pressure in rats and show reproductive and developmental toxicity in rats and mice
- While gastrointestinal disturbance has been reported in humans following ingestion of vanadium compounds, the currently available data are inadequate to derive a tolerable upper level of intake
- While normal dietary intake of vanadium is about three orders of magnitude below daily doses causing adverse health effects, use of dietary supplements containing vanadium may result in daily doses similar to levels causing adverse health effects in humans and rodents.

15.2.4 Food and Nutrition Board, Institute of Medicine (FNB/IOM, USA)

FNB/IOM considered information of vanadium in order to set a tolerable upper intake level (UL).[28] It was concluded that there was no evidence of adverse effects associated with vanadium intake from food, but some data on adverse effects associated with vanadium intake from supplements and drinking water.

A UL was derived based on the Lowest Observed Adverse Effect Level (LOAEL) for renal effects in rats of 7.7 mg/kg bw/day. An uncertainty factor of 300 was applied, including a factor of three for extrapolation from the LOAEL to NOAEL and a factor of 100 to account for inter- and intra-species uncertainty. The resulting UL of 26 μg/kg bw/day was multiplied the average reference body weight for men and women (68.5 kg) and rounded to give a UL of

[28] The highest level of daily nutrient intake that is likely to pose no risk of adverse health effects for almost all individuals

1.8 mg/day. No separate UL was set for specific life stages, as there was no evidence of particular susceptibilities to vanadium toxicity in any particular life stage.

15.2.5 United States Environmental Protection Agency (USEPA)

USEPA considered sodium metavanadate, vanadium pentoxide and vanadyl sulphate separately, as evidence from rodent studies suggested that sodium metavanadate was more toxic than the other vanadium compounds.

USEPA derived a subchronic oral reference dose (RfD_{SO}) for vanadium as sodium metavanadate of 0.006 mg/kg bw/day, based on a NOAEL of 0.55 mg/kg bw/day for mild lesions of lung, spleen and kidney in a 90-day rat study and an uncertainty factor of 100. No suitable subchronic studies were available for derivation of RfD_{SO} for vanadium pentoxide or vanadyl sulphate and it was proposed that the chronic oral reference dose (RfD_O) for these compounds be used as the RfD_{SO}.

RfD_O values of 0.009 and 0.007 mg/kg bw/day were derived for vanadium as vanadium pentoxide and vanadyl sulphate, respectively, by applying an uncertainty factor of 100 to NOAELs from lifetime studies. A RfD_O for sodium metavanadate was derived from the RfD_{SO} by applying a further uncertainty factor of 10 to account for extrapolation from the subchronic to chronic time frame. The resultant RfD_O was 0.001 mg/kg bw/day.

The available data were insufficient to establish whether any of the vanadium compounds considered were carcinogenic and USEPA did not derive a cancer potency for vanadium or its compounds.

15.3 METABOLITES OF VANADIUM AND THEIR RELATIVE TOXICITY

Vanadium is an element, and as such, is not metabolised (ATSDR, 2012a; EFSA, 2004). In oxygenated blood, vanadium circulates as a polyvanadate (isopolyanions containing pentavalent vanadium), but in tissues it is retained mainly as the vanadyl cation (tetravalent vanadium). When it enters cells pentavalent vanadium is reduced to tetravalent vanadium by glutathione (Barceloux, 1999).

Toxicity of vanadium is generally considered to be low, but to increase with increasing oxidation state (Barceloux, 1999; Ghosh *et al.*, 2015).

It has been speculated that toxicity due to vanadium could occur due its ability to act as a phosphate analogue and to inhibit the action of a number of enzyme systems, including ATPases, phosphatases and phosphate transfer enzymes (ATSDR, 2012a).

16. EXPOSURE ASSESSMENT - VANADIUM

16.1 VANADIUM IN THE NEW ZEALAND FOOD SUPPLY

No information was found on vanadium in the New Zealand food supply.

16.2 VANADIUM IN THE AUSTRALIAN FOOD SUPPLY

Vanadium was included as an analyte in the 23rd Australian Total Diet Study (FSANZ, 2011). However, vanadium was not detected in any of the food samples analysed. It should be noted that the limit of detection for vanadium was not given in the study report. It is not possible to say whether the fact that vanadium was not detected was due to very low vanadium concentrations or limitations with the analytical method.

16.3 OVERSEAS CONTEXT

Most studies in the current literature have analysed foods for total vanadium, rather than analysing for the species forms of vanadium, specifically V(IV) and V(V). The following section will summarise available information on total vanadium in foods, then discuss the implications of the few speciation studies.

16.3.1 Plant foods

Grain products

The second French Total Diet Study reported mean vanadium concentrations (ICP-MS) in grain products that ranged from 0.008 to 0.085 mg/kg fresh weight (Arnich *et al.*, 2012). The first Hong Kong Total Diet Study reported a mean vanadium concentration for cereal products at the bottom of the range seen in France (0.010 mg/kg fresh weight) (Chen *et al.*, 2014). However, concentrations (ICP-MS) up to 0.26 mg/kg were seen in cereal product samples.

A US study did not detect (ICP-AES) vanadium in corn (LOD 0.008 mg/kg), white bread (LOD 0.02 mg/kg) or spaghetti and meatballs (LOD 0.008 mg/kg) (Dolan and Capar, 2002). Trace amounts (0.021 mg/kg) were detected in pancakes, while a fruit-flavoured cereal contained 0.093 mg/kg.

Neutron activation analysis was used to determine vanadium concentrations in the range 0.001 to 0.040 mg/kg fresh weight in a variety of cereals (Byrne and Kosta, 1978). A higher concentration (0.27 mg/kg fresh weight) was detected in buckwheat, but this was considered to be due to soil contamination.

Higher concentrations were reported in an Iranian study, using adsorptive stripping voltammetry (Abbasi *et al.*, 2012). A rice sample was reported to contain 0.11 mg/kg vanadium, while a wheat flour sample was reported to contain 0.10 mg/kg vanadium.

Fruits, vegetables, pulses and nuts

Fruits and vegetables are generally considered to contain low concentrations of vanadium.

The second French Total Diet Study reported mean vanadium concentrations in vegetables, fruits, pulses, nuts and seeds in the range 0.015-0.035 mg/kg fresh weight (Arnich *et al.*, 2012). Even lower concentrations were reported for the first Hong Kong Total Diet Study with

mean vanadium concentrations for the same food groups in the range 0.0015 to 0.011 mg/kg fresh weight (Chen *et al.*, 2014).

A US study did not detect vanadium in peanut butter (LOD 0.04 mg/kg), broccoli (LOD 0.004 mg/kg), sweet potato (LOD 0.01 mg/kg), canned pears (LOD 0.005 mg/kg) or prune juice (LOD 0.008 mg/kg) (Dolan and Capar, 2002).

Very low vanadium concentrations (<0.001 mg/kg wet weight) were found in most vegetables, fruits and nuts (Byrne and Kosta, 1978). Higher concentrations were detected in carrot (0.0023-0.0024 mg/kg), spinach (0.035 mg/kg), potato (0.0013-0.0019 mg/kg) and hazelnuts (0.0037 mg/kg). Parsley (1.8 mg/kg dry weight) contained particularly high levels of vanadium.

A method using ionic liquid microextraction followed by GFAAS was used to determine vanadium in a range of vegetables (country of origin not stated) (Naeemullah *et al.*, 2015). Concentrations were generally higher than in other studies for tomatoes (0.10 mg/kg), spinach (0.25 mg/kg), lettuce (0.12 mg/kg), parsley (0.32 mg/kg) and cucumber (0.14 mg/kg). The same group also developed an analytical method for determination of vanadium by solid phase extraction on multi-walled carbon nanotubes followed by GFAAS (Wadhwa *et al.*, 2013). Concentrations of vanadium were determined in tomatoes (0.17 mg/kg), cabbage (<0.012 mg/kg), zucchini (0.10 mg/kg) and apple (0.25 mg/kg).

A further method for analysis of vanadium used cloud-point extraction followed by GFAAS (Filik and Aksu, 2012). Vanadium concentrations of 0.45 and 0.37 mg/kg were reported for two samples of tomatoes.

A single sample of potato was analysed by adsorptive stripping voltammetry in an Iranian study, with vanadium found a concentration of 0.44 mg/kg (Abbasi *et al.*, 2012).

Mushrooms

Concentrations of 0.05-2.0 mg/kg dry weight were reported in 26 species of mushrooms analysed by neutron activation analysis (Byrne and Kosta, 1978).

A mean concentration of vanadium in cultivated mushrooms of 0.17 mg/kg was reported from analysis by ionic liquid microextraction followed by GFAAS (Naeemullah *et al.*, 2015).

Other foods

The second French Total Diet Study reported vanadium concentrations greater than 0.05 mg/kg in chocolate (0.079 mg/kg), alcoholic beverages (0.066 mg/kg), pizzas, quiches and savoury pastries (0.054 mg/kg), sandwiches and snacks (0.054 mg/kg) and sauces and seasonings (0.096 mg/kg) (Arnich *et al.*, 2012).

The first Hong Kong Total Diet Study reported highest mean vanadium concentration in alcoholic beverages (0.043 mg/kg) and condiments, sauces and herbs (0.022 mg/kg) (Chen *et al.*, 2014).

A US study did not detect vanadium in lemonade (LOD 0.003 mg/kg) or mayonnaise (LOD 0.06 mg/kg), but did detect vanadium in beer (0.03 mg/kg) (Dolan and Capar, 2002).

Beverages contained vanadium concentrations in the range 0.0003-0.032 mg/kg, with the higher levels in wine and beer (Byrne and Kosta, 1978). Cocoa powder contained high

concentrations of vanadium (0.61 mg/kg). Fats and oils did not contain detectable vanadium (LODs 0.0002 to 0.003 mg/kg).

A Polish study reported the vanadium content of different types of beer (n = 6) to be in the range 0.018 to 0.031 mg/L (Krosniak *et al.*, 1998). A process analysis found that most of the vanadium entered the beer during the filtration step, when the brew was passed through diatomaceous earth.

GFAAS with cloud point extraction was used to determine vanadium in a sample of white wine (0.028 mg/L) and red wine (0.075 mg/L) (Filik and Aksu, 2012).

GFAAS with ionic liquid microextraction was used to determine vanadium in white (0.0163 mg/L) and red (0.0113 mg/L) wine (Naeemullah *et al.*, 2015). The same group also developed an analytical method for determination of vanadium by solid phase extraction on multi-walled carbon nanotubes followed by GFAAS (Wadhwa *et al.*, 2013). Concentrations of vanadium were determined in black tea (1.58 mg/kg) and coffee (0.40 mg/kg).

A single study was found that determined the vanadium content of a pooled sample of seaweed collected from coastal areas of Ceará, Brazil over a one year period (Oliveira *et al.*, 2009). The concentration of vanadium found in the pooled seaweed sample was 3.56 mg/100 g (35.6 mg/kg). While it is unclear from the publication, it appears that these results are expressed on a dry weight basis.

16.3.2 Foods of animal origin

Seafood

A Malaysian study determined vanadium (ICP-MS) in liver and fresh samples of fish species from the coastal waters of peninsular Malaysia (Agusa *et al.*, 2005). Geometric mean vanadium concentrations by species/sampling site were in the range 0.6 to 2.3 mg/kg dry weight (liver) and 0.033 to 0.18 mg/kg dry weight (flesh). If it is assumed that fish flesh and liver have an average moisture content of about 70% these ranges equate to 0.18 to 0.7 mg/kg wet weight (liver) and 0.01 to 0.054 mg/kg wet weight (flesh). The highest vanadium concentration reported in fish flesh was 0.21 mg/kg dry weight (0.063 mg/kg wet weight).

The same authors extended this study to include fish from the coastal waters of Cambodia, Thailand and Indonesia (Agusa *et al.*, 2007). Vanadium concentrations (geometric mean by species and sampling site) were in a similar range to those from Malaysian waters, with liver vanadium concentrations in the range 0.025 to 4.0 mg/kg dry weight and flesh vanadium concentrations in the range 0.008 to 0.20 mg/kg dry weight.

The second French Total Diet Study reported a mean vanadium concentration for fish of 0.029 mg/kg fresh weight and for crustaceans and molluscs of 0.224 mg/kg fresh weight (Arnich *et al.*, 2012).

The first Hong Kong Total Diet Study reported a mean vanadium concentration for fish and seafood and their products of 0.02 mg/kg fresh weight, although concentrations in individual samples were as high as 0.27 mg/kg (Chen *et al.*, 2014).

A US study did not detect vanadium in canned tuna (LOD 0.03 mg/kg), but detected trace amounts (0.034 mg/kg) in haddock (Dolan and Capar, 2002).

Concentrations of vanadium in fish were found in the range 0.0035 to 0.028 wet weight (Byrne and Kosta, 1978). A sample of dried oyster contained substantially higher concentrations (0.46 mg/kg).

Analysis of giant river prawns (*Macrobrachium rosenbergii*) from southern Vietnam found concentrations of vanadium (ICP-MS) in the range 0.006-0.036 mg/kg (abdominal muscle), 0.039-0.47 mg/kg (abdominal exoskeleton) and 0.012-0.27 (hepatopancreas) (Tu *et al.*, 2008).

Four fish species were collected from three sites on the coast of Saudi Arabia and analysed for contaminant elements, including vanadium, by GFAAS (Al-Saleh and Shinwari, 2002). Unfortunately, the high limit of detection for vanadium (0.11 mg/kg wet weight) meant that vanadium was only detectable for one fish species (rabbitfish; *Siganus canaliculatus*) at one location. The mean vanadium concentration was 0.114 mg/kg.

Six fish species and shrimp were sampled from markets in Kuwait and analysed for elemental contaminants, including vanadium, by GFAAS (Bu-Olayan and Al-Yakoob, 1998). Mean vanadium concentrations by species ranged from 0.48 mg/kg (sheim; *Acanthopagrus latus*) to 1.48 mg/kg (nakroor; *Pomadasys argenteus*). These vanadium concentrations appear very high and this may be related to the massive oil contamination that occurred during the 1991 Gulf War.

A survey of contaminant elements in six fish and shellfish species from the eastern Mediterranean (*n* = 30 samples per species) found considerably lower vanadium concentrations (ICP-MS), with mean concentrations ranging from 0.074 mg/kg wet weight (European anchovy; *Engraulis encrasicolus*) to 0.497 mg/kg (Wedge clam; *Donax trunculus*) (Copat *et al.*, 2013).

Another study of fish and crustacean species in the Mediterranean found vanadium concentrations (ICP-MS) in fish species ranging from <0.002 to 0.48 mg/kg fresh weight (Iamiceli *et al.*, 2015). The highest vanadium concentration was found in European anchovy, but there was considerable variation in vanadium concentrations within the same species, suggesting that vanadium content was not species specific. Vanadium concentrations in the crustacean species examined were within the range of concentrations seen in fish (0.06 to 0.12 mg/kg fresh weight). Parameters related to the size and/or age of the samples was not examined.

A study of elemental contaminants in six fish species in the Adriatic Sea (part of the Mediterranean) found highest vanadium concentrations in anchovies, with mean concentrations in the range 0.045 to 0.074 mg/kg fresh weight, depending on the location and time of sampling (Sepe *et al.*, 2003). Very low concentrations were found in angler, hake and sole (mostly less than 0.004 mg/kg fresh weight). Although no data were presented, the authors noted good correlations between fish length and weight and elemental concentrations, suggestion accumulation over time.

Neutron activation analysis was used to analyse for vanadium in brown mussels (*Perna perna*) from Brazilian coastal waters (Seo *et al.*, 2013). Concentrations of vanadium were reported in the range 0.77 to 3.56 mg/kg, on a dry weight basis.

Samples of the fish species *Johnius belangerii* (Belanger's croaker) (*n* = 67) were taken from the Musa Estuary, a shallow estuarine region of the Persian Gulf (Fard *et al.*, 2015). The mean vanadium concentration (GFAAS) was 2.92 mg/kg wet weight, with a maximum concentration of 5.95 mg/kg wet weight. The Persian Gulf is a major transport waterway for

the oil industry and the Musa Estuary has limited recharge capacity. These high vanadium concentrations are probably the result of petrochemical pollution.

Meat and meat products (including poultry and eggs)

The second French Total Diet Study reported mean vanadium concentrations in meat (including offals), poultry and egg samples in the range 0.013 to 0.079 mg/kg fresh weight (Arnich *et al.*, 2012). The higher concentrations were in processed meats.

The first Hong Kong Total Diet Study reported lower mean vanadium concentrations for meat, poultry and eggs than seen in France (range 0.004 to 0.013 mg/kg fresh weight) (Chen *et al.*, 2014).

A US study did not detect (ICP-AE) vanadium in bacon (LOD 0.04 mg/kg), boiled eggs (LOD 0.02 mg/kg) or a beef-based baby food (LOD 0.01 mg/kg) (Dolan and Capar, 2002).

In contrast to the results summarised above, muscle meat from cattle raised adjacent to a vanadium processing plant had a median vanadium content of 0.25-0.28 mg/kg (maximum 1.77-2.55 mg/kg) (Gummow *et al.*, 2005). Organ meats contained even higher concentrations (liver median 1.34 mg/kg, kidney median 1.09 mg/kg).

Poultry, red meat and egg samples contained vanadium concentrations in the range 0.0002 to 0.038 mg/kg wet weight by neutron activation analysis (Byrne and Kosta, 1978). The higher concentrations were in the lung and liver, while the lowest concentrations were in fat. Muscle meats contained 0.0004 to 0.0017 mg/kg wet weight of vanadium, while in eggs the highest concentration were encountered in the egg yolk (0.002 to 0.0036 mg/kg wet weight).

A Greek study compared the trace element concentrations (ICP-MS) of egg components from different poultry species (Nisianakis *et al.*, 2009). Little difference in the vanadium content of egg yolk was observed, with a range from 0.0105 mg/kg (goose) to 0.0154 mg/kg (pigeon). It should be noted that the feed given to all birds contained quite high concentrations of vanadium (2.09 mg/kg).

Vanadium was determined by ICP-MS in muscle of semi-domesticated reindeer (n = 100) from Norway (Hassan *et al.*, 2012). The maximum concentration reported was 0.0083 mg/kg wet weight, with the majority of results below 0.001 mg/kg.

An analytical method was developed for determination of vanadium by solid phase extraction on multi-walled carbon nanotubes followed by GFAAS (Wadhwa *et al.*, 2013). The concentration of vanadium in chicken was determined as 0.13 mg/kg. It should be noted that this is markedly higher than other vanadium concentrations determined for muscle meats.

Dairy products

The second French Total Diet Study reported mean vanadium concentrations in the range 0.014 to 0.065 mg/kg fresh weight (Arnich *et al.*, 2012). The higher concentrations were found in cheese and butter. The first Hong Kong Total Diet Study reported only an overall mean value for vanadium in dairy products of 5 mg/kg wet weight (Chen *et al.*, 2014).

A US study did not detect (ICP-AE) vanadium in evaporated milk (LOD 0.01 mg/kg) or cheddar cheese (LOD 0.03 mg/kg) (Dolan and Capar, 2002).

Neutron activation analysis detected generally low concentrations of vanadium in milk (<0.0002 mg/kg) (Byrne and Kosta, 1978).

GFAAS with ionic liquid microextraction was used to determine vanadium in cow's (0.0257 mg/L) and sheep (0.0408 mg/L) milk (Naeemullah *et al.*, 2015).

In contrast to the results summarised above, milk from cattle raised adjacent to a vanadium processing plant had a median vanadium content of 0.23 mg/kg (maximum 1.92 mg/kg) (Gummow *et al.*, 2005).

The vanadium content of infant formula available in Nigeria was determined by atomic absorption spectroscopy (Iwegbue *et al.*, 2010). While the authors do not specify, it appears probable that the formulas were powdered and not made up prior to analysis. The mean vanadium content of all milk-based formulas (from birth and follow-on) was 0.04 mg/kg, with slightly higher mean concentrations in soy-based formulas (0.06 mg/kg). Given that powdered infant formulas are usually made up at a rate of approximately 12 to 13 g of powdered formula to give 100 mL of prepared formula, these concentrations are not inconsistent with other results published for dairy products.

16.3.3 Speciation of vanadium in foods

Very little information is available on the form of vanadium present in foods. A study on Chinese cabbage determined that 60-80% of vanadium in leaves was in the tetravalent form, whereas the mobile vanadium in soils is believed to be predominantly in the pentavalent form (Tian *et al.*, 2014). The authors concluded that these results were indicative of bio-reduction of vanadium in the plant.

Speciation of vanadium was carried out on vegetable samples grown near a thermal power station in the Sindh province of Pakistan and from an agricultural site not near a power station (Khan *et al.*, 2011). Total vanadium was higher in samples from the vicinity of the power station (8.7 to 14.0 mg/kg dry weight) than from the agricultural land (3.0 to 4.9 mg/kg dry weight). V(IV) was the dominant species in all vegetable samples, with V(IV)/V(V) ratios consistently in the range 1.4 to 1.9 (58-66% as V(IV)). Approximately 70-90% of total vanadium was able to be accounted for by species analysis. Vegetables examined included bitter gourd, carrots, cluster beans, coriander, okra, onion, peppermint, potatoes, spinach and peas.

16.4 ESTIMATES OF DIETARY EXPOSURE

16.4.1 New Zealand

No estimates of dietary exposure to vanadium have been made for New Zealanders.

16.4.2 Australia

The 23rd Australian Total Diet Study included analyses of vanadium (FSANZ, 2011). However, vanadium was not detected in any sample analysed and no estimate of dietary exposure was possible.

16.4.3 Overseas estimates of dietary exposure

Total vanadium

Table 16 summarises results of overseas studies that have derived estimates of dietary exposure to vanadium.

Table 16. International estimates of dietary exposure to vanadium

Country	Population group	Mean (high percentile) exposure, μg/kg bw/day[a]	Main foods contributing	Reference
Cambodia	General population	0.008 (max 0.057)[b]	Only fish included	Agusa *et al.* (2007)
Cameroon	Adult	0.71-1.03 (1.45-1.82)	Fish, cereals and cereal products, beverages	Gimou *et al.* (2014)
France	Children (3-17 years) Adults (18-79 years)	1.06 (2.10) 0.86 (1.47)	Water Water, alcoholic beverages	Arnich *et al.* (2012)
Hong Kong	Adult	0.13 (0.27)	Cereals, vegetables	Chen *et al.* (2014)
Indonesia	General population	0.009 (max 0.098)[b]	Only fish included	Agusa *et al.* (2007)
Italy (North)	Adult	0.20[b]	Cereals and tubers, fruits and vegetables	Turconi *et al.* (2009)
Malaysia	General population	0.036-0.043 (max 0.16)[b]	Only fish included	Agusa *et al.* (2007)
Nigeria	Infants 0-6 months 7-12 months	 1.05 1.64	Only infant formula included	Iwegbue *et al.* (2010)
Spain (Tarragona)	Adult male (70 kg)	0.41	Vanadium only detected in fish and seafood	Bocio *et al.* (2005)
Spain (Tarragona)	Duplicate diet, restaurant meals	2.6[b]	Not determined, whole meals analysed	Domingo *et al.* (2012)
Thailand	General population	0.016-0.025 (max 0.11)[b]	Only fish included	Agusa *et al.* (2007)
USA	General population	0.17-1.0[b]	Not stated	Harland and Harden-Williams (1994)
USA	Male, 25-30 years	0.26[b]	Estimate based on composite total diet samples	Iyengar *et al.* (2000)

[a] High percentile dietary exposure estimates are usually expressed as the 95th percentile, but may occasionally be expressed as the 90th or 97.5th percentile. Where a range is presented for the exposure estimate, this usually refers to different treatments of left-censored analytical data

[b] Published results expressed as 'μg/day'. A body weight of 60 kg was used to convert exposures to 'μg/kg bw/day'

16.4.4 Biomarkers of exposure

While elevated concentrations of vanadium have been observed in urine and serum of occupationally exposed workers, relationships between exposure levels and blood/serum or urine vanadium levels have not been established (ATSDR, 2012a). Tang *et al.* (2012) used urine vanadium as a marker of exposure in a case-control study of the association between vanadium exposure and breast cancer risk.

17. RISK CHARACTERISATION - VANADIUM

17.1 ADVERSE HEALTH EFFECTS IN NEW ZEALAND

17.1.1 Risk assessment

No risk assessments for vanadium have been conducted in New Zealand.

17.2 ADVERSE HEALTH EFFECTS OVERSEAS

17.2.1 Epidemiological studies

A case-control study (*n* = 240 cases, 246 controls) carried out in Guangzhou, China measured urinary concentrations of vanadium and titanium by ICP-MS (Tang *et al.*, 2012). Women in the second and highest tertiles of urinary vanadium showed a decreased risk of breast cancer compared to women in the lowest tertile, after adjustment for known risk factors. However, it should be noted that there was no clear biological gradient, with women in the second tertile of urinary vanadium having lower risk of breast cancer than women in the highest tertile.

A cohort of workers involved in manufacture of vanadium-containing products (*n* = 463) was compared to a control group not in contact with vanadium-containing products (*n* = 251) (Li *et al.*, 2013). There was no attempt to match exposed and control cohorts, although the general characteristics of the groups (age, socioeconomic status, background environmental factors) were reported to be similar. The vanadium-exposed cohort performed significantly worse in a range of neurobehavioural tests, including mood states, reaction times, digit span, visual retention and pursuit aiming. When the cohorts were segregated by length of service (<10 years or ≥10 years), significant differences in test performance persisted for the longer service group, but not the shorter service group. The authors suggested that the effect seen could be due to oxidative stress on the central nervous system. It should be noted that, in this study, exposure to vanadium is likely to be largely due to inhalation exposure.

17.2.2 Risk assessments

Due to a lack of health-based exposure limits for vanadium, exposure assessments carried out have not been used to characterise risk.

18. RISK MANAGEMENT INFORMATION - VANADIUM

18.1 RELEVANT FOOD CONTROLS: NEW ZEALAND

18.1.1 Establishment of regulatory limits

Standard 1.4.1 of the Australia New Zealand Food Standards Code does not specify maximum limits (ML) for vanadium in any food.[29]

18.1.2 Imported Food Requirements (IFR)

There are currently no IFRs that require products imported into New Zealand to be monitored for vanadium.

18.2 RELEVANT FOOD CONTROLS: OVERSEAS

18.2.1 Establishment of regulatory limits

No maximum limits for vanadium in food was found for any country.

18.2.2 Codes of Practice

No Codes of Practice specific for the controls of vanadium in foods were identified.

18.3 INFLUENCE OF FOOD PROCESSING ON VANADIUM AND VANADIUM SPECIES LEVELS

18.3.1 Brewing

A Polish study determined vanadium at various stages through the beer production process (Krosniak *et al.*, 1998). The concentration of vanadium was fairly consistent, at about 1 μg/L up to the filtration step, where it increased to about 20 μg/L. This concentration was maintained into the final product. Filtration involved passing the brew through diatomaceous earth and it was concluded that this was the source of the increase in the vanadium content.

[29] https://www.comlaw.gov.au/Details/F2015C00052 Accessed 2 February 2016

19. CONCLUSIONS - VANADIUM

19.1 DESCRIPTION OF RISKS TO NEW ZEALAND CONSUMERS

While there is suggestive information in some animal species, it has still not been established that vanadium is an essential trace element in humans. Vanadium compounds have been used as supplements by body builders and in clinical trials for people with diabetes mellitus at daily doses three orders of magnitude above international estimates of dietary exposure. Adverse effects have generally been confined to gastrointestinal systems and it is uncertain whether these symptoms are due to a toxicological response or to local irritation of the gastrointestinal tract.

There is insufficient data to comment on the potential impact of vanadium speciation on risks associated with dietary exposure to vanadium.

19.2 COMMENTARY ON RISK MANAGEMENT OPTIONS

No specific risk management measures for control of vanadium concentrations in food have been identified.

19.3 DATA GAPS

The following data gaps were identified:

- Vanadium content of New Zealand foods
- Species forms of vanadium in foods
- Adverse health effects related to oral vanadium exposure
- Evidence or essentiality or otherwise of vanadium to humans
- Impact of food processing on vanadium content of foods

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