SURVEY OF CYANOGENIC GLYCOSIDES IN PLANT-BASED FOODS IN AUSTRALIA AND NEW ZEALAND 2010-13

SUMMARY

Plants contain many substances that can pose potential risks to consumers, and one of these types of substances is cyanogenic glycosides. There are approximately 25 known cyanogenic glycosides and these are generally found in the edible parts of plants, including almonds, stone fruit, pome fruit, cassava, bamboo shoots, linseed/flaxseed and lima beans.

In 2010, a bi-national coordinated food survey of cyanogenic glycosides in plant-based foods commenced under the Food Regulation Standing Committee's (FRSC) Implementation Sub-Committee for Food Regulation's (ISFR) Coordinated Food Survey Plan. Food Standards Australia New Zealand (FSANZ) and the New Zealand Ministry for Primary Industries (NZ MPI) were the lead agencies, coordinating a total of three survey activities during 2010-2013 in consultation with Australian states and territories.

The survey identified that cyanogenic glycosides (measured as hydrocyanic acid or HCN) are present in a wide range of Australian and New Zealand plant-based foods at levels consistent with or lower than those reported in the scientific literature. Raw apricot kernels with skin contained HCN concentrations that were substantially higher than any other food analysed.

The survey also identified that the HCN levels detected in the analysed foods were within the regulatory limits for HCN (where such regulatory limits exist) in all but two cases. In the case of cassava roots, one sample did not comply with the regulatory requirements in Standard 1.4.4 of the Australia New Zealand Food Standards Code (the Code) covering prohibited and restricted plants, namely, it did not meet the criteria for 'sweet cassava' which may be sold in Australia and New Zealand, as it contained greater than the 50 mg HCN/kg limit that defines 'sweet cassava'. One sample of apricot nectar did not comply with the current Maximum Level (ML) of 5 mg HCN/kg permitted in stone fruit juices, as required in Standard 1.4.1.

Dietary exposure to elevated levels of some cyanogenic glycosides in food has the potential to cause acute cyanide poisoning or a debilitating irreversible neurological condition in the long term. A risk assessment was undertaken to determine whether there are any public health and safety issues associated with consuming foods containing cyanogenic glycosides and to assist in determining if any risk management options need to be considered.

Hazard assessments established two health-based guidance values (HBGVs). For chronic or long term effects, the provisional maximum tolerable daily intake (PMTDI)[1] of 20 µg cyanide/kg body weight, established by the Joint FAO/WHO Expert Committee on Food Additives (JECFA) was used. For acute effects, the acute reference dose (ARfD)[2] of 80 µg HCN/kg body weight previously established by FSANZ was used (FSANZ, 2008b).

The dietary exposure assessment included chronic and acute estimates. For the estimated chronic dietary exposure to HCN, there were no exceedances of the PMTDI for any population group assessed. On this basis, it is unlikely that there will be any public health

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[1] A health based guidance value used to estimate the tolerable level of exposure to a contaminant with no cumulative properties and is the maximum amount of a substance (expressed in milligrams per kilogram of body weight) that can be ingested on a daily basis over a period of time without appreciable health risk to the consumer.
[2] The estimated amount of a substance (expressed in milligrams per kilogram of body weight) that can be ingested in a period of 24 hours or less without appreciable health risk to the consumer.

and safety issues in relation to chronic dietary exposure to HCN for the Australian and New Zealand populations.

The acute dietary exposure assessment is conservative as it is an estimate of 'worst case' exposure, to be protective of the health and safety of consumers. The results of the acute dietary exposure assessment indicated that all population groups had estimated acute dietary exposures under the ARfD for the majority of foods.

The risk assessment identified that consumption of raw apricot kernels both with and without skin[3] can pose an acute public health and safety risk for all Australian and New Zealand population groups even at consumption levels for adults of 4 kernels/day (with skin), previously considered by FSANZ to be safe. For children, there are health risks even at consumption levels of one apricot kernel/day (with skin).

For cassava roots, there is potential for high consumers to exceed the ARfD. However, given the conservative assumptions made in the acute dietary exposure assessment, and the absence of any reports on poisonings in Australia or New Zealand following consumption of properly processed cassava, the estimated potential exposures to HCN are not considered to represent an appreciable health and safety risk.

For bread containing linseed, although the estimated acute dietary exposures resulted in potential exceedances of the ARfD for all population groups assessed, current exposures are not considered to represent a health and safety risk due to the absence of any clinical reports of poisonings or detectable levels of cyanide in the blood of human volunteers following consumption of ground linseed.

FSANZ has prepared Proposal P1016 – Hydrocyanic acid in apricot kernels and other foods, to consider options for the management of potential risks identified as a result of this ISFR survey and risk assessment.

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[3] Throughout this report the term 'raw apricot kernel' refers to the edible nut-like object found within the shell or stone of Prunus aremeniaca either unhulled (with skin) or hulled (without skin). Hulled, bitter apricot kernels are usually pale white in colour.

Key findings

- Survey findings confirmed that cyanogenic glycosides are present in a wide range of Australian and New Zealand plant-based foods. HCN levels are consistent with, or lower than, those reported in the scientific literature. Analysed levels met current regulatory requirements for HCN (where such regulatory limits exist), in all but two cases, namely, one sample of cassava root and one sample of apricot nectar did not comply with the regulatory requirements of the Code.
- A risk assessment based on these survey results indicated that there were no public health and safety issues in relation to the estimates of chronic dietary exposure to HCN for the Australian and New Zealand populations.
- There were a small number of foods (raw apricot kernels, cassava roots and bread containing linseed) where it was estimated that acute dietary exposure had the potential to exceed the ARfD. Of these foods, it is the consumption of raw apricot kernels both with and without skin that poses the greatest acute public health and safety risk for Australians and New Zealanders.

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Abbreviations

ANCNPAS	Australian National Children's Nutrition and Physical Activity Survey (2007)
ARfD	Acute Reference Dose
BMD	Benchmark Dose
bw	Body weight
CCCF	Codex Committee on Contaminants in Food
Codex	Codex Alimentarius Commission
DIAMOND	DIetAry Modelling Of Nutritional Data
EFSA	European Food Safety Authority
ESR	Institute of Environmental Science and Research (New Zealand)
EU	European Union
FAO	Food and Agriculture Organization of the United Nations
FRSC	Food Regulation Standing Committee
FSANZ	Food Standards Australia New Zealand
HBGV	Health-based guidance value
HCN	Hydrocyanic acid
HPLC	High-performance liquid chromatography
ISFR	Implementation Sub-Committee for Food Regulation
JECFA	Joint FAO/WHO Expert Committee on Food Additives
LB	Lower bound
LoD	Limit of Detection
LSA	Linseed, sunflower seeds and almonds
mg/kg	Milligrams per kilogram
µg/kg	Micrograms per kilogram
ML	Maximum Level
NNS	National Nutrition Survey
NOAEL	No-observed-adverse-effect level

NSW FA   NSW Food Authority

NZCNS   New Zealand National Children's Nutrition Survey (2002)

NZ MPI   New Zealand Ministry for Primary Industries

PMTDI   Provisional Maximum Tolerable Daily Intake

RTE   Ready-to-eat

the Code   Australia New Zealand Food Standards Code

UB   Upper bound

WHO   World Health Organization

Note: A glossary of terms can be found in Appendix 1.

1. OBJECTIVES

The objectives of the ISFR survey on cyanogenic glycosides were to:

- collect data on levels of HCN in plant-based foods, including cassava containing foods other than ready-to-eat (RTE) cassava chips, in the Australian and New Zealand food supply

- estimate dietary exposure and assess if there are any health and safety concerns from potential exposure to HCN from a range of foods, as well as inform any future standards development in Australia and New Zealand.

2.   BACKGROUND

Plants contain many substances that can pose potential risks to consumers, and one of these types of substances is cyanogenic glycosides. There are approximately 25 known cyanogenic glycosides and these are generally found in the edible parts of plants, including almonds, stone fruit, pome fruit, cassava, bamboo shoots, linseed/flaxseed and lima beans (Codex Committee on Contaminants in Foods, 2008; Haque and Bradbury, 2002).

2.1   Structure, breakdown and mechanism of toxicity

Cyanogenic glycosides are naturally occurring sugars that have the cyanide moiety in their structure. The basic structure and two examples of common cyanogenic glycosides are shown in Figure 1.

cyanogenic glycoside basic structure

linamarin

amygdalin
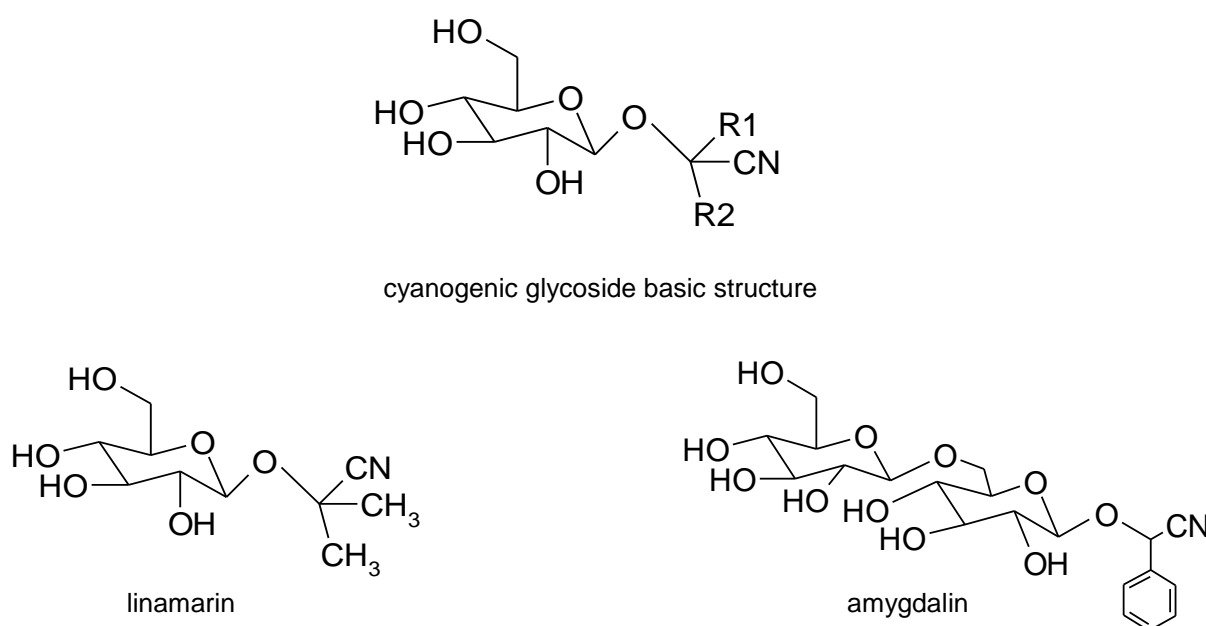
R1 may be a methyl group, a phenyl or *p*-hydroxyphenyl group. R2 is most commonly hydrogen, but may also be a methyl or ethyl group. The sugar moiety may be either glucose (monosaccharide) or gentiobiose (disaccharide).

Source: ESR, 2013a.

Figure 1: Cyanogenic glycoside general structure and examples

Cyanogenic glycosides are converted to the intermediate breakdown product cyanohydrins and hydrogen cyanide as a result of enzymatic action on plant tissue (through damage to the plant) or the action of gut microflora in animals or humans (at variable rates) after enzyme activity in the gut.

The cyanogenic glycoside content of a food is determined by measuring the free HCN and HCN evolved following enzyme or acid hydrolysis of the cyanogenic glycosides themselves (e.g. linamarin in cassava) and the intermediate breakdown product cyanohydrins. Together these two sources of HCN are expressed as 'total' HCN in this report (Codex Committee on Contaminants in Foods, 2013). The breakdown of cyanogenic glycosides to hydrogen cyanide is shown in Figure 2.
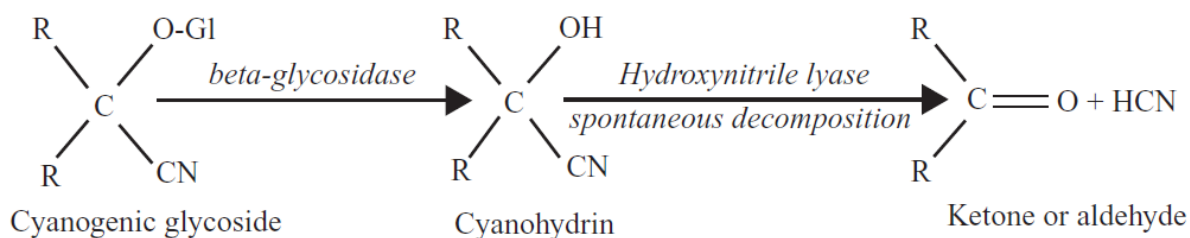
Gl = glucose
Source: FAO/WHO, 2012.

Figure 2: Breakdown of cyanogenic glycosides to hydrogen cyanide [or hydrocyanic acid (HCN)]

The toxicity of cyanogenic glycosides and their derivatives is dependent on the release of hydrogen cyanide. Toxicity may result in acute cyanide poisoning and has also been implicated in the aetiology of several chronic diseases (ESR, 2010 – unpublished; FAO/WHO, 2012).

2.2   Regulation of cyanogenic glycosides in Australia and New Zealand

Proposal P257 – Advice on the preparation of cassava and bamboo shoots (FSANZ, 2004a) examined the potential public health and safety risks associated with consumption of inadequately prepared cassava and/or bamboo shoots and determined whether risk management measures were necessary. This proposal indicated that there are public health and safety risks if these foods are not prepared properly, and that there may be insufficient knowledge in the general community of safe preparation techniques. As a result of this proposal, a number of changes were made to the Australia New Zealand Food Standards Code (the Code) as follows:

- Standard 1.4.4 – Prohibited and restricted plants and fungi: the sale of cassava, other than 'sweet cassava' or its intentional addition to foods is prohibited

- Standard 1.1.2 – Supplementary definitions of food: 'sweet cassava' is defined as those varieties that contain less than 50 mg/kg of HCN (fresh weight basis)

- Standard 1.2.6 – Directions for use and storage: the food (sweet cassava, bamboo shoots) must be labelled with, or accompanied by a statement indicating that they must be peeled and fully cooked before consumption (FSANZ, 2012a).

There are no provisions for bitter cassava in the Code. A report on the human health risk assessment undertaken as part of Proposal P257 was published under FSANZ's technical report series (FSANZ, 2004b).

Standard 1.4.1 – Contaminants and natural toxicants of the Code (FSANZ, 2012a) specifies Maximum Levels (MLs) for hydrocyanic acid from the addition of flavouring substances to food for the following foods:

- Confectionery   25 mg/kg
- Stone fruit juices   5 mg/kg
- Marzipan   50 mg/kg
- Alcoholic beverages   1 mg/kg per 1% alcohol content

In 2008, FSANZ was advised that HCN levels in some RTE cassava chips were much higher than what would occur if sweet cassava had been used. In response, the National Food Incident Response Protocol was activated.

FSANZ prepared a risk assessment in relation to linamarin in RTE cassava chips. FSANZ then prepared Proposal P1002 – Hydrocyanic acid in cassava chips to assess the public health risks associated with HCN in RTE cassava chips (FSANZ, 2008a). As a result of this assessment, FSANZ considered that regulatory measures in the Code were required to reduce levels of HCN in RTE cassava chips to protect public health and safety. In June 2009, an ML of 10 mg/kg for total HCN[4] in RTE cassava chips was established in Standard 1.4.1.

2.3   International regulations

The Codex Alimentarius Commission (Codex) has developed and published standards for Sweet Cassava, Edible Cassava Flour and Gari (a product obtained from processing cassava tubers) (also spelt as 'Garri').

The key aspects of these standards are:

- Sweet cassava is defined as a raw product containing less than 50 mg/kg of 'hydrogen cyanide, fresh weight' and that cassava must be peeled and fully cooked before being consumed (Codex, 2005)

- Edible cassava flour is defined as a product suitable for direct human consumption and the level of 'total hydrocyanic acid' in the flour must not exceed 10 mg/kg (Codex, 1995)

- For gari the 'total hydrocyanic acid' must not exceed 2 mg/kg determined as 'free' hydrocyanic acid (Codex, 1989).

For bitter varieties of cassava (greater than 50 mg/kg HCN), the Codex standard (Codex, 2010) states that the following information must be made available to consumers at the point of sale:

- Cassava must not be eaten raw

- Cassava shall be peeled, de-pithed, cut into pieces, rinsed and fully cooked before consumption

- Cooking or rinsing water must not be consumed or used for other food preparation purposes.

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[4] All hydrocyanic acid including hydrocyanic acid evolved from linamarin, lotaustralin, acetone cyanohydrin or butanone cyanohydrin during or following enzyme hydrolysis or acid hydrolysis.

2.4   ISFR coordinated food survey of cyanogenic glycosides in plant-based foods

As a result of Proposal P1002 on RTE cassava chips, FSANZ recognised that a more thorough review of cyanogenic glycosides in food should be undertaken. To support such a review, a strong scientific evidence base would be required. The collection of analytical data on levels of HCN in a range of plant-based foods other than RTE cassava chips contributes to this evidence base.

In 2009, a survey of cyanogenic glycosides in plant-based foods was included on the ISFR Coordinated Food Survey Plan (CFSP), to be led by FSANZ and the New Zealand Food Safety Authority [now New Zealand Ministry for Primary Industries (NZ MPI)]. This approach would ensure input from Australian states and territories on sampling and methodology. This survey is termed the ISFR survey and consists of three separate surveys. Each of these surveys were planned and implemented separately and therefore in this report they are referred to as Surveys 1, 2 and 3.

2.4.1 Survey 1

Survey 1 was conducted in March 2010 after NZ MPI, in conjunction with FSANZ, reviewed the occurrence of cyanogenic glycosides in plant-based foods and developed a sampling plan. The Institute of Environmental Science and Research (ESR) was engaged to undertake the analytical testing of the samples by acid hydrolysis of extracted cyanogens, followed by colourimetric determination (Haque and Bradbury, 2002).

2.4.2 Survey 2

Survey 2, conducted in 2012, examined the cyanogenic glycoside content of a large number of Australian and New Zealand apple juice samples, and was undertaken to confirm one positive result obtained in Survey 1. The analytical testing was again undertaken by ESR using the acid hydrolysis method.

An alternative method of analysis for total HCN agreed by the EU in 2012 as a standardised method and involving HPLC determination of the derivatised cyanide was subsequently used by ESR to confirm results obtained for several apple products analysed in Survey 2 and Survey 3 (see also below) (Animal feeding stuffs – Determination of hydrocyanic acid by HPLC. EN 16160:2012) (European Committee for Standardization, 2012). In this report it is termed the EU HPLC methodology.

2.4.3 Survey 3

Survey 3 was conducted in 2013 to obtain further data relating to the cyanogenic glycoside content of raw apricot kernels, apricot products and/or products containing apricot kernels and several other foods for which limited data were available in Australia and New Zealand. The results of this survey were used to augment results obtained in Survey 1 and to inform Proposal P1016, which was prepared in response to the hospitalisation of a consumer in Queensland in October 2011, after consuming raw apricot kernels with high levels of HCN.

The analytical testing was again undertaken by ESR using the acid hydrolysis method. The EU HPLC methodology was used to confirm results obtained for one apple product analysed in this survey.

3.   METHODOLOGY

3.1   Sampling

3.1.1 Survey of cyanogenic glycosides in plant-based foods (Survey 1)

For Survey 1, NZ MPI, in consultation with FSANZ, reviewed available information on the occurrence of HCN in foods and sampled a range of foods available in New Zealand that had the potential to contain naturally occurring cyanogenic glycosides. The plant-based foods analysed were: cassava and cassava products; bamboo shoots; almonds and almond products; apple products; linseed/flaxseed and products; stone fruit products; lima beans and various seeds; and several other miscellaneous products. A description of foods sampled and sample numbers are shown in Table 1.

All samples were collected from supermarkets or specialist retail outlets in Christchurch, New Zealand in March 2010. This was based on the assumption that the foods would be nationally distributed. It was also assumed that cyanogenic compounds would be present at reasonably consistent levels within each of the food groups sampled, and that levels in products available in New Zealand would be representative of those available in Australia.

At total of 100 individual samples were purchased. The number of samples for each food type varied from 1 to 11 primary purchase samples. Retail units or loose product were purchased to give a sample weight of at least 500 g. Exceptions were highly homogeneous, high-value foods, such as oils, for which a single retail unit (of at least 200 g) was purchased.

Table 1: Sample type and sample numbers for Survey 1 of cyanogenic glycosides in plant-based foods

Sample type	No. of samples	Description
Cassava	15	Raw (frozen) cassava roots Cassava flour/tapioca products
Bamboo shoots	10	Canned bamboo shoots Other forms (i.e. pickled bamboo shoots)
Almonds and almond products	15	Almonds (whole, flaked, ground or butter) Almond essence Almond confectionery (i.e. marzipan) Almond jelly Almond oil
Apple products	12	Apple juice Apple sauce Apple cider vinegar
Linseed/flaxseed and products	13	Linseed (whole and meal) LSA (i.e. a mixture of linseed, sunflower seeds and almonds) Bread containing linseed Flaxseed and LSA oil
Stone fruit products	10	Canned apricots Prune juice Cherry juice Cherry brandy
Lima beans and various seeds	15	Raw lima beans Canned butter/lima beans Canned mixed beans Pumpkin seeds Sunflower seeds
Miscellaneous products	10	Passionfruit Passionfruit drink Passionfruit sauce Taro leaves Spinach Canned stuffed vine leaves
Total number of samples	**100**	

3.1.2 Follow-up survey of apple juice (Survey 2)

Survey 2, which focussed on apple juice, collected a total of 108 individual samples in Christchurch, New Zealand and in the capital cities of Australian states and territories and country NSW (ACT – Australian Capital Territory, QLD – Queensland, NSW – New South Wales, NT – Northern Territory, SA – South Australia, TAS – Tasmania, VIC – Victoria and WA – Western Australia) during April/May 2012.

Three sets of apple juice samples were collected. These comprised two retail sets of juice samples—one from Australia (n=48); one from New Zealand (n=50); and one juice manufacturer set (n=10). Retail samples were obtained as normal retail units of at least 100 mL. Information on the samples is summarised in Table 2

Table 2: Sample type and sample numbers for Survey 2 of cyanogenic glycosides in apple juice

Apple juice sample type	No. of samples	Description
New Zealand retail samples	50	Samples purchased principally in Christchurch. Retail outlets included supermarkets, greengrocers and specialist food retailers (i.e. organics).
Australian retail samples	48	Samples purchased in nine geographical locations covering the eight Australian states and territories, making up a representative sample of the range of apple juice available on the marketplace. Retail outlets included supermarkets, greengrocers, markets and cafes.
New Zealand juice manufacturer samples	10	Five samples of juice were taken at the juicing press of a juice manufacturer. Five samples of juice were made in the juice manufacturer's laboratory, after ensuring apple pips were ruptured (cyanogenic glycosides are understood to be concentrated in apple pips).
Total number of samples	**108**	

Further information on some of the characteristics of the two sets of retail samples included in Survey 2 is shown in Table 3. Samples comprised juices made with local ingredients and a combination of both local and imported ingredients, reconstituted and non-reconstituted juices, and shelf stable juices as well as juices requiring refrigeration. For each country, approximately the same proportion of products were made locally from local ingredients. A higher proportion of products manufactured from reconstituted apple juice concentrate were sampled in Australia. A higher proportion of shelf stable products were sampled in New Zealand.

Table 3: Characteristics of retail apple juice samples included in Survey 2 of cyanogenic glycosides in apple juice[5]

Characteristic	No. of samples (%)	
	Sampled in Australia (*n*=48)	Sampled in New Zealand (*n*=50)
Origin of product		
Australia (local ingredients)	20 (42)	2 (4)
Australia (imported ingredients)	8 (17)	-
Australia (local and imported ingredients)	16 (33)	3 (6)
New Zealand (local ingredients)	-	23 (46)
New Zealand (imported ingredients)	-	2 (4)
New Zealand (local and imported ingredients)	1 (2)	18 (36)
South Africa	1 (2)	1 (2)
Not Stated	2 (4)	1 (2)
Juice material		
Apple juice	18 (38)	29 (58)
Reconstituted apple juice	27 (56)	19 (38)
Apple juice and reconstituted apple juice	3 (6)	-
Apple juice and puree	-	2 (4)
Storage		
Shelf stable	22 (46)	48 (96)
Require refrigeration	26 (54)	2 (4)

3.1.3 Follow-up survey of apricot kernels and other plant-based foods (Survey 3)

Survey 3 foods were sampled in Christchurch, New Zealand and in the capital cities of Australian states and territories. All samples were collected from supermarkets or specialist retail outlets (including Asian, Fijian Indian and continental supermarkets and health food shops) in February/March 2013. A total of 88 individual samples were purchased, making up 69 analytical samples. The number of samples for each food type varied from 3 to 18 primary purchase samples. Retail units or loose product were purchased to give a sample weight of at least 500 g. Sample types and sample numbers are shown in Table 4 and examples of apricot kernels with and without skin are shown in Figure 3.

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[5] Juice manufacturer samples are not included in this table.

Table 4: Sample type and sample numbers for Survey 3 of cyanogenic glycosides in apricot kernels and plant-based foods

Sample type	No. of samples	Description
Apricot kernels	28	Apricot kernels with skin Apricot kernels without skin
Apricot products/ Products containing apricot kernels	37	Amaretti biscuits Almond fingers/apple fingers Apricot jam Apricot nectar
Apple products	8	Infant apple puree
Cassava	9	Frozen cassava roots
Linseed/flaxseed products	3	Bread containing linseed
Bamboo shoots	3	Fresh and frozen bamboo shoots
Total number of samples	**88**	

Figure 3: Examples of apricot kernels with and without skin

3.2   Sample preparation

Samples of fresh foods (including apple juice) were frozen or assayed immediately to avoid break-down of cyanogenic glycosides by endogenous enzymes. Solid samples were homogenised in a blender.

In Survey 3, cassava roots and bamboo shoots were analysed in both their raw and cooked states. All samples of cassava roots were purchased ready peeled. The frozen roots were cut to a standard size of approximately 10 cm in length and 4cm in width. Groups of three samples of frozen cassava pieces were cooked by either steaming (washed and soaked for

two hours and then steamed for 20-25 minutes or until tender), boiling (20-25 minutes or until tender) or frying (sliced into 1-1.5 cm strips and then fried for 15 minutes in a deep fryer at 180ºC or until golden brown and tender). Fresh or frozen samples of bamboo shoots were cut lengthwise into two equal portions and one portion was cooked by boiling for approximately one hour or until tender. See Appendix 2 for further details on the food preparation instructions given to ESR for cassava roots and bamboo shoots.

3.3 Analysis

3.3.1 Analytical methods

ESR was engaged to analyse total HCN in the samples by acid hydrolysis of extracted cyanogens, followed by colourimetric determination (Haque and Bradbury, 2002). The analysis of cyanogens using this method involved three steps: (i) extraction from the foodstuff; (ii) hydrolysis of cyanogenic compounds to cyanide; and (iii) colourimetric analysis of cyanide. This method has also been used in another New Zealand survey of RTE cassava chips, also conducted by ESR in 2011 (ESR, 2011 - unpublished).

ESR was subsequently engaged to reanalyse a small number of samples (i.e. apple juice and infant apple puree) using an alternative method of analysis agreed by the EU in 2012 as a standardised method for animal feeding materials (Animal feeding stuffs – Determination of hydrocyanic acid by HPLC. EN 16160:2012) (European Committee for Standardization, 2012). This method involved enzymatic hydrolysis of cyanogenic glycosides, distillation of cyanide, derivatisation and HPLC determination of the derivatised cyanide.

3.3.2 Application of analytical methods to various foods

Analyses were carried out on the normal edible portion of the food. Specifically, analysis of canned foods was carried out on the drained contents, with the exception of the canned palusami (taro leaves in coconut cream). The edible flesh of the passionfruit was analysed with the skin being discarded. Cooked cassava and bamboo shoots were analysed after draining off the cooking water/frying oil. All other foods were analysed as 100% of the sample as received.

For Survey 1 and 2, each food and beverage was analysed individually for HCN. For Survey 3, the majority of foods were analysed as individual samples, however, multiple retail samples were analysed as one composite sample for amaretti biscuits, almond fingers/apple fingers and apricot jam.

3.3.3 Method comments

The EU HPLC method for HCN analysis was found to be more sensitive, accurate and reliable than other methods being used in Australasia and, in particular, to have superior performance characteristics in the following areas:

- Sensitivity – the method is highly sensitive, leading to substantially lower limit of detections (LoDs) and, as such, it is recommended for testing of samples containing total HCN concentrations up to about 10 mg HCN/kg

- Specificity – the method is highly specific to HCN, in contrast to the acid hydrolysis method which might give a positive response with certain non-HCN chemical species

- Validation – it is the only cyanide method where an appropriate inter-laboratory assessment has been carried out and reported.

There was poor agreement between results for apple juice samples analysed by the two different methods, suggesting that the acid hydrolysis method was not appropriate for the measurement of very low HCN concentrations, as detected in apple juice and infant apple puree.

However, the acid hydrolysis method is still considered robust for the concentration range of the majority of samples in the surveys, with results for samples such as apricot kernels (which were found to have very high concentrations of HCN —see Section 4.3.1), cassava roots and bamboo shoots being consistent within the survey and with results reported from other studies. Further, for each of the three surveys, a proportion of samples were analysed in duplicate and matrix spike recovery tests were carried out on various food matrices, in line with ESR analytical quality control. For the EU HPLC method to be used for samples with high concentrations of HCN such as apricot kernels, extracts would need to be diluted significantly to bring them within the working concentration range of this method.

3.3.4 Limits of detection (LoDs)

For Surveys 1, 2 and 3, the LoDs for single and duplicate analyses using the acid hydrolysis method were different. The LoDs for duplicate analyses were lower than those for single analyses.

LoDs using the acid hydrolysis method also differed across the three surveys. This is also the case with the LoDs achieved using the EU HPLC method in the reanalysis of apple juice (from Survey 2) and infant apple puree (from Survey 3). A summary of the LoDs achieved in the three surveys and for the two analytical methods is provided in Table 5.

As the LoDs for single and duplicate analyses differed within each survey, for some individual foods within some food types, the LoD varied. For example, in Survey 1, 'apple juice' was made up of 8 primary purchase samples. Within these 8 samples there were two different LoDs: 3 mg HCN/kg (for duplicate analyses) and 4 mg HCN/kg (for single analyses).

Table 5: Summary information on limits of detection (LoDs) achieved in Surveys 1, 2 and 3 using the acid hydrolysis and EU HPLC methods of analysis for total HCN

Survey	Analytical method	Limit of detection (LoD) (mg HCN/kg)	
		Single analysis	Duplicate analysis
Survey 1	Acid hydrolysis	4	3
	EU HPLC	NA	NA
Survey 2	Acid hydrolysis	2	1.5
	EU HPLC*	0.06	NA
Survey 3	Acid hydrolysis	5	4
	EU HPLC[#]	0.1	NA

NA = not applicable.
* reanalysis of apple juice.
[#] reanalysis of infant apple puree.

4. ANALYTICAL SURVEY RESULTS AND DISCUSSION

4.1 Survey of cyanogenic glycosides in plant-based foods (Survey 1)

Results of Survey 1 were reported in *Survey of cyanogenic glycosides in plant-based foods* (ESR, 2010 – unpublished) and are summarised in Appendix 3. In general, the cyanogenic glycoside contents (measured as total HCN) were consistent with or lower than those reported in the scientific literature. Linseed/flaxseed and the associated product LSA were found to contain consistently high concentrations (>50 mg HCN/kg) of total HCN. Other foods with higher levels of HCN included cassava roots (mean of 21 mg HCN/kg), pickled bamboo shoots (mean of 21 mg HCN/kg) and one sample of small green lima beans (32 mg HCN/kg).

Findings indicate that the foods surveyed were within regulatory limits for HCN, where such regulatory limits exist. In the case of cassava, the samples met the regulatory requirements covering prohibited and restricted plants; namely they met the criteria for 'sweet cassava', which may be sold in Australia and New Zealand, as they contained less than 50 mg/kg of HCN.

Eight samples of apple juice were analysed in Survey 1. Of these, only one was found to have measurable amounts of HCN. This sample had a total HCN content of 5.4 mg/kg, which was close to the LoD (4 mg HCN/kg). As there was some uncertainty associated with this one positive result, Survey 2 was undertaken to examine the cyanogenic glycoside content of a much larger sample of Australian and New Zealand apple juices.

4.2 Follow-up survey of apple juice (Survey 2)

Results of Survey 2 were reported in *Determination of presence of cyanogenic residues in apple juices in Australia and New Zealand* (ESR, 2012 – unpublished) and are summarised in Appendix 3. Of the 108 individual samples collected, measurable amounts of HCN were found in four of the retail samples but none in the set of juice manufacturer's samples.

4.2.1 Retail samples

For Survey 2, total HCN concentrations were above the LoD in four of 98 retail samples of juice, ranging from 1.5-4.2 mg/kg. The characteristics of these samples and total HCN levels detected are summarised in Table 6.

Table 6: Summary information for four apple juice samples available in Australia and New Zealand analysed which had detectable levels of total HCN

Apple juice type	Country of purchase	Origin of product	Total HCN (mg HCN/kg)*	Comments
Shelf-stable organic juice	New Zealand	New Zealand	2.6	No further batches or products from the manufacturer of this product were collected as part of this survey.
Shelf-stable, reconstituted juice	New Zealand	Made from imported and local ingredients	4.2	A second batch of the same product and a further five apple juices from the same manufacturer did not contain detectable levels of total HCN.

Subsequent to this survey, two retention samples for batches of apple juice concentrate that were used in producing this retail sample and two retention samples for batches made just prior to this analysed retail sample were tested. All retention samples contained no detectable levels of total HCN. |
| **Shelf-stable, reconstituted juice** | Australia | Made from imported and local ingredients | 1.5[#] | Analysed in duplicate.

No further products from the same manufacturer were collected as part of this survey. |
| **Reconstituted organic apple juice requiring refrigeration** | Australia | Australia | 2.1 | Two other batches of the same product and a further two apple juices from the same manufacturer did not contain detectable levels of total HCN. |

* LoD of 2 mg/kg for single analyses and 1.5 mg/kg for duplicate analyses.
[#] Total HCN at the LoD for duplicate analyses of 1.5 mg/kg.

4.2.2 Manufacturer samples

Cyanogenic glycosides are understood to be concentrated in apple pips (Haque and Bradbury, 2002). Therefore, ten samples of apple juice were obtained directly from a major New Zealand juice manufacturer to explore the suggestion that the presence of HCN in apple juice may be due to rupture of the pips during juice extraction, even though the pips are hardy and difficult to crush (ESR, 2010 – unpublished). Five samples of juice were taken from the juicing press, which achieves approximately a 70% juice extraction rate; and five samples were obtained in the juice manufacturer's laboratory, after ensuring pips were ruptured. Of the ten juice manufacturer samples obtained, none contained detectable concentrations of HCN.

Further details regarding the analytical results obtained for apple juice and subsequent investigations are at Section 4.4.

4.3   Follow-up survey of apricot kernels and other plant-based foods (Survey 3)

Results of Survey 3 were reported in *Determination of presence of cyanogenic glycosides in apricot kernels and other plant-based foods in Australia and New Zealand* (ESR, 2013a – unpublished) and are summarised in Appendix 3. In general, the cyanogenic glycoside contents were consistent with or lower than those reported in the scientific literature, and levels detected were within the regulatory limits for HCN (where such regulatory limits exist) in all but two cases, namely, one sample of cassava root and one sample of apricot nectar did not comply with the regulatory requirements of the Code. Further information regarding

the results for several of the surveyed products where HCN was detected is provided in Sections 4.3.1 – 4.3.5.

4.3.1 Apricot kernels

All apricot kernel samples analysed contained detectable levels of HCN. However, there was a significant difference in HCN concentrations between samples with skin and those without. Samples with skin contained HCN concentrations that were substantially higher than those without skin and indeed substantially higher than any other food analysed. Levels ranged from 1,240-2,820 mg HCN/kg. On average, apricot kernels without skin contained less than 10% of the HCN of those with skin.

Higher concentrations of HCN detected in samples with skin may indicate that the cyanide-producing compounds are concentrated within the skin. The processing methods used to remove the skin may also contribute to a reduction in HCN concentrations.

Concentration of cyanide-producing compounds in the skin may support the prevailing theory that these compounds have a role in plant defence (Vetter, 2000), in that a concentration of these compounds at the point at which pests are first likely to attack would be of most benefit to the plant (ESR, 2013a – unpublished).

4.3.2 Apricot nectar

HCN was detected in one of four samples of apricot nectar analysed. The total HCN concentration in this sample was 6.5 mg/kg. This sample did not comply with the current ML of 5 mg HCN/kg permitted in stone fruit juices in Standard 1.4.1 of the Code.

4.3.3 Cassava roots

HCN was detected in all nine samples of raw frozen cassava roots. One raw sample had a maximum total HCN concentration of 81 mg HCN/kg and therefore did not comply with the regulatory requirements set out in the Code of less than 50 mg HCN/kg[6].

HCN was detected in all nine samples of cooked cassava roots. However, all cooking methods resulted in a marked reduction in the HCN content of cassava roots, with a mean reduction of 56% (range of 28-83%). The HCN concentration of the non-compliant sample decreased from 81 mg HCN/kg to 37 mg HCN/kg after cooking.

Unfortunately, the sample numbers were too small to determine if there is any statistically significant difference between the HCN reductions that occurred by different cooking methods.

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[6] Thirteen samples of frozen cassava root were purchased but the agreed sampling/analytical plan required that only nine of these be analysed. However, label information was recorded for all 13 samples. Only four of the 13 samples included directions for use, as required under Standard 1.2.6 of the Code.

4.3.4 Bamboo shoots

Of the three samples of raw bamboo shoots collected, two were purchased fresh and one was purchased frozen. Fresh varieties proved difficult to locate and do not appear to be regularly available in major retail outlets.

All three samples of raw bamboo shoots contained detectable concentrations of HCN. The two fresh samples contained significantly higher concentrations than the frozen sample. It is likely that the lower concentration of HCN was due to further processing (i.e. blanching before freezing) [7].

HCN was detected in all three samples of cooked bamboo shoots. Cooking the raw samples resulted in an approximate 90% reduction in the cyanide generating potential of the two high-cyanide samples. For two of three samples, the reduction in HCN upon cooking was within the range of concentrations reported for canned/pickled varieties in Survey 1.

4.3.5 Bread containing linseed

The source of HCN in bread containing linseed is assumed to be solely from the linseeds. For Survey 3, all three samples of bread containing linseed were collected in Australia. HCN was detected in all of these.

HCN concentrations were found to be higher than those reported in Survey 1 for bread containing linseed from New Zealand.

The higher HCN content of Australian bread containing linseed may be due to higher HCN concentrations in Australian-sourced linseed, as an Australian study of cyanogenic plant materials reported higher concentrations of total HCN in linseeds of up to 390 mg HCN/kg (Haque and Bradbury, 2002), compared with concentrations of 180 mg HCN/kg for equivalent analyses in New Zealand (ESR, 2010 – unpublished).

Australian breads analysed in Survey 3 contained 5-7% linseed according to the label, which was similar to the New Zealand breads analysed in Survey 1 (7-8%). In both cases, HCN concentrations in bread containing linseed were relatively consistent with linseed being the only source of HCN, the stated linseed content of the breads according to the label, and the reported concentrations of HCN in linseed, for each country.

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[7] None of the three samples of fresh/frozen bamboo shoots included directions for use, as required under Standard 1.2.6 of the Code.

4.4   Apple juice and apple products

4.4.1 Survey results for apple juice

Results of Surveys 1 and 2 suggest that there is a potential for apple juice to occasionally contain measurable amounts of HCN ranging from 1.5-5.4 mg/kg. However, this occurred in isolated cases and did not appear to be linked to any particular manufacturer or brand, and/or storage or processing practice. Adding to the uncertainty associated with these results, analyses of additional batches of the same product and retention samples (where available) for samples collected in both Surveys 1 and 2 resulted in no detections of HCN.

Analyses of apple juice samples taken directly from the juice press or made in the laboratory (after ensuring the cyanogen-containing pips were ruptured) (Survey 2) also resulted in no detections. This indicates that the occasional detection of HCN in apple juice may not be due to rupture of the pips at high extraction rates, as was initially proposed (ESR, 2010 – unpublished).

FSANZ carried out a number of investigations to validate the results of Surveys 1 and 2 and identify the source of the cyanogenic glycosides detected in the juice, as described in the sections below.

4.4.2 Analysis of apple aroma extract

It was suggested that the source of the cyanogenic glycosides present in apple juice was the apple aroma (flavour) extract that is collected from the vapour during the production of the apple juice concentrate. This product may be sold separately by juice manufacturers for adding back into a juice concentrate when it is being diluted, to achieve the original apple flavour. It is added back into the juice for restorative purposes and the addition rate is typically less than 1%. Therefore, the HCN concentration of the apple aroma extract would need to be high to be detected in the juice.

To explore this suggestion, three samples of composite batches of apple aroma extract were tested. All samples showed no detections of HCN, casting doubt on the extract being the source of HCN. As this activity was undertaken separate to the ISFR survey, detailed results are not provided in this report.

4.4.3 Reanalysis of apple product samples

In late 2012, ESR had conducted an extensive assessment of the currently available methods of analysis of total HCN in foods. As a result of this assessment, ESR determined that a more sensitive method of analysis agreed by the EU as a standardised method and involving HPLC, be used to reanalyse the positive samples (see Section 3.3).

Only three of the four samples of apple juice analysed in Survey 2 were available for reanalysis using the EU HPLC method, with a lower LoD of 0.06 mg HCN/kg. Results of the reanalysis indicated a very low concentration of HCN in one of the three apple juices; HCN was not detected in the other two.

Given its much greater sensitivity, the EU HPLC method was also used to confirm results obtained for all eight samples of infant apple puree analysed in Survey 3. Although HCN was not detected above the LoD of 5 mg HCN/kg in any sample of infant apple puree in Survey

3, reanalysis using the EU HPLC method (LoD of 0.1 mg HCN/kg) demonstrated that a very low concentration of HCN was present in seven of the eight infant apple purees.

Given the increased sensitivity, specificity and reliability of the EU HPLC method, a good degree of confidence can be placed in the results obtained for apple juice and infant apple puree using this method. FSANZ considers that the results are suitable for generating estimates of exposure with an acceptable level of uncertainty.

A comparison of levels of total HCN obtained for juice and apple puree in Surveys 2 and 3 using the acid hydrolysis method against those obtained using the EU HPLC method is provided in Table 7.

Table 7: Comparison of levels of total HCN (mg HCN/kg) obtained for apple products using acid hydrolysis and EU HPLC analytical methods

Apple product type	Country of purchase	Total HCN (mg HCN/kg)	
		Acid hydrolysis method*	EU HPLC method[#]
Shelf-stable organic juice	New Zealand	2.6	N/A
Shelf-stable, reconstituted juice	New Zealand	4.2	<0.06
Shelf-stable, reconstituted juice	Australia	1.5	0.17
Reconstituted organic apple juice requiring refrigeration	Australia	2.1	<0.06
Infant apple puree	Australia	<5	0.9
Infant apple puree	Australia	<5	1.1
Infant apple puree	Australia	<5	1.2
Infant apple puree	New Zealand	<5	1.3
Infant apple puree	Australia	<4	<0.1
Infant apple puree	Australia	<5	0.1
Infant apple puree	Australia	<5	0.8
Infant apple puree	Australia	<4	0.7

* LoD of 2 mg HCN/kg for single analyses and 1.5 mg HCN/kg for duplicate analyses for apple juice and
   LoD of 5 mg HCN/kg for single analyses and 4 mg HCN/kg for duplicate analyses for infant apple puree.

[#] LoD of 0.06 mg HCN/kg for apple juice and
   LoD of 0.1 mg HCN/kg for infant apple puree.

4.5 Summary of results from Surveys 1-3

The results of the survey indicate that cyanogenic glycosides are present in a wide range of Australian and New Zealand plant-based foods, at levels that are consistent with, or lower than, those reported in the scientific literature. Raw apricot kernels with skin contained HCN concentrations that were substantially higher than those without skin and indeed substantially higher than any other food analysed. Other foods with relatively high levels of HCN included cassava roots, bamboo shoots, linseeds (including LSA) and bread containing linseed. Many of the remaining foods analysed were found to occasionally contain measurable amounts of HCN, but at lower levels.

The survey also identified that the HCN levels detected in the analysed foods were within the regulatory limits for HCN (where such regulatory limits exist) in all but two cases. In the case of cassava roots, one sample contained greater than the 50 mg HCN/kg limit that defines 'sweet cassava' permitted for sale in Australia and New Zealand. One sample of apricot

nectar did not comply with the current Maximum Level (ML) of 5 mg HCN/kg permitted in stone fruit juices.

5.   HUMAN HEALTH SIGNIFICANCE OF SURVEY RESULTS

A risk assessment was undertaken based on the results of the ISFR survey to determine if there were any public health and safety issues from consuming foods with the concentrations of total HCN determined in Surveys 1-3.

5.1   Hazard Assessment

In 2008 FSANZ completed a comprehensive hazard assessment on the cyanogenic glycoside linamarin (the predominant cyanogenic glycoside in cassava) as part of Proposal P1002. FSANZ established an ARfD based on the no-observed-adverse-effect level (NOAEL) of 70 mg/kg body weight (bw) for clinical signs in hamster dams in a developmental toxicity study following a single dose of linamarin. Application of a 100-fold uncertainty factor to this NOAEL to enable extrapolation to humans gave an ARfD for linamarin of 0.7 mg/kg bw. The linamarin units of this ARfD were converted to total HCN— the actual compound measured in analytical assays. The ARfD was calculated to be equivalent to 80 µg HCN/kg bw (FSANZ, 2008b). FSANZ did not establish a tolerable daily intake for humans (for chronic risk assessment purposes) due to a paucity of suitable data. However, it was noted that there was no available evidence in adequately nourished humans to show that chronic intake of cyanogenic glycosides causes a cumulative hazard above that of repeated acute toxicity.

In 2011, JECFA re-evaluated the public health implications of cyanogenic glycosides and their derivatives in food. Benchmark dose (BMD) modelling was performed on dose-response data from the same study used by FSANZ to establish its ARfD. However, in contrast to FSANZ, JECFA used a secondary toxicological endpoint, namely skeletal defects in the fetuses of dams who exhibited clinical signs of toxicity. The lower limit of the BMD for a 10% response (BMDL10) for linamarin was 85 mg/kg bw for an increased incidence of skeletal defects in developing hamster fetuses following acute exposure of maternal animals. Application of a 100-fold uncertainty factor established an ARfD for linamarin of 0.09 mg/kg bw (equivalent to 90 µg cyanide/kg bw).

At this time, JECFA also established a PMTDI of 20 µg/kg bw per day as cyanide for use in chronic dietary risk assessment. This value was based on a BMDL10 of 1.9 mg/kg bw per day for reduced absolute cauda epididymis (in testes) weights from a 13-week drinking water study in rats with sodium cyanide and application of a 100-fold uncertainty factor. JECFA decided that it was not necessary to apply an additional uncertainty factor to account for the absence of a long-term study, considering the generally acute nature of cyanide toxicity and the sensitivity of the effect (i.e. the reduction of absolute cauda epididymis weight).

Given that both chronic (long term) and acute (short term) HBGVs have been established, both a chronic and acute dietary exposure assessment was conducted for this risk assessment. To determine the risk associated with the estimated dietary exposures based on the current survey data, the ARfD of 80 µg HCN/kg bw established by FSANZ for P1002 and the JECFA PMTDI of 20 µg/kg bw per day as cyanide were used.

5.2   Concentration data used in the dietary exposure assessment

Concentration data were derived primarily from the ISFR survey as well as several other surveys of RTE cassava chips conducted in recent years in Australia and New Zealand. Further details regarding how these data were used in the dietary exposure assessment are provided in Section 5.2.1 and 5.2.2.

5.2.1 Concentration data from Surveys 1-3

Concentration data from Surveys 1-3 were used, as appropriate. For apple juice and infant apple puree, only the concentration data obtained using the EU HPLC method were used as inputs into the dietary exposure estimates. For cassava root, the one sample (Survey 3) that did not comply with the regulatory requirements for sweet cassava was excluded. The next highest concentration of 26 mg HCN/kg for cooked cassava determined in a steamed sample (the total HCN concentration for this same sample in its raw state was 40 mg/kg) was used. Similarly, for apricot nectar, the one sample (Survey 3) that did not comply with the current ML permitted in stone fruit juices was excluded from the acute dietary exposure assessment.

For bamboo shoots, a preliminary dietary exposure assessment including Survey 3 samples that were purchased raw (and subsequently cooked) indicated that one population group, Australians aged 17 years and over, would exceed the ARfD for consuming bamboo shoots (110%). However, fresh/frozen varieties of bamboo shoots, as sampled in Survey 3, proved difficult to locate as part of the sampling plan. From this, it was assumed that it is unlikely that they are commonly available in the Australian and New Zealand marketplace. Therefore, it was considered reasonable to estimate dietary exposures using values from Survey 1 only (i.e. canned/pickled bamboo shoots).

For the purposes of the chronic dietary exposure assessment, lower bound mean and upper bound mean concentrations were derived from the raw survey data. When deriving the lower bound mean concentrations, results lower than the LoD ('not detected' or <LoD) were assigned a concentration of zero. For the upper bound mean not detected results were assigned a concentration equal to the LoD. See Table 5 for the LoDs achieved for Surveys 1-3.

For the acute dietary exposure assessment, maximum concentrations for each food were determined. If the food was analysed as a composite sample, the concentration determined was used. If all of the samples for the food were analysed as containing levels less than the LoD, the value of the highest LoD was used; similarly, the highest value was used in the upper bound scenario for the chronic dietary exposure estimates.

A summary of the concentration data from Survey 1, 2 and 3, as prepared for the dietary exposure assessment is shown in Appendix 4. Detailed information about the concentration data for some specific foods for the purpose of the dietary exposure assessment can be found in Appendix 5.

5.2.2 Concentration data from surveys of RTE cassava chips

Since the ML for RTE cassava chips was introduced in 2009, the NSW Food Authority (NSW FA) and NZ MPI, have continued to monitor HCN concentrations in RTE cassava chips through analytical surveys. The data from these surveys were reviewed to determine a representative (mean) concentration for use in the chronic dietary exposure calculations for this report. These data were summarised and used separately for each country. This was because the surveys were done by independent agencies with potential differences in the sampling and analysis. A summary of the concentration data used in the chronic dietary exposure calculations is shown in Appendix 6. A comprehensive acute dietary exposure assessment was conducted for RTE cassava chips in P1002 (FSANZ, 2008a) and therefore was not undertaken in this study.

5.3 Dietary exposure assessment

The dietary exposure assessment was conducted to estimate the level of chronic and acute dietary exposure to HCN.

5.3.1 Dietary exposure assessment methodology

A dietary exposure assessment is the process of estimating how much of a food chemical a population, or population sub-group, consumes. FSANZ's approach to dietary modelling is based on internationally accepted procedures for estimating dietary exposure to food chemicals. Different dietary modelling approaches may be used depending on the assessment, the type of food chemical, the data available and the risk assessment questions to be answered. Further information on how FSANZ conducts dietary exposure assessments is not provided in this report but can be found in the document *FSANZ principles and practices of dietary exposure assessment for food regulatory purposes* (FSANZ, 2009). Specific details related to this assessment are outlined below.

5.3.1.1 Chronic dietary exposure assessment

Chronic dietary exposure estimates are used to represent the long term, usually life-long, dietary exposure for the population from the range of foods containing the chemical of interest.

The chronic dietary exposure assessment for HCN includes the range of foods analysed in the ISFR survey as well as RTE cassava chips. It was not possible to include raw apricot kernels in the chronic dietary exposure assessment, as this product was not recorded as having been consumed in any of the national nutrition surveys.

The chronic dietary exposure assessments were conducted using a semi-probabilistic method, where a distribution of consumption values for each food derived from individual records was combined with a single concentration value of HCN for each food. Summary population statistics were then calculated from the distribution of individual exposures to HCN at the mean and 90th percentile of consumption. For risk characterisation purposes, the estimated dietary exposures to HCN were converted to exposures as cyanide to allow direct comparison with the PMTDI which is expressed as cyanide. The conversion of HCN to CN⁻ was done using molecular weights (HCN = 27.03; cyanide = 26).

5.3.1.2 Acute dietary exposure assessment

Acute dietary exposure estimates are used to represent the high consumer and a high exposure, from a single food or food group, from one meal or over one day.

A deterministic acute dietary exposure assessment was undertaken where a single food consumption value at the 97.5th percentile for each food (consumers of the food only) was combined with the maximum concentration of HCN for the food from the ISFR survey. A deterministic acute dietary exposure assessment is intended to be conservative as it is an estimate of 'worst case' exposure, to be protective of the health and safety of consumers. For risk characterisation purposes, the estimated acute dietary exposures were then compared with the ARfD.

If there were insufficient consumers (i.e. <39 consumers) to derive a robust 97.5th percentile consumption figure, a median consumption value was used (consumers of the food only). Median consumption values may be used for occasionally consumed foods where it is considered unrealistic to assume anyone in the population consumes these foods at the

97.5th percentile level every day over a lifetime (ANZFA, 1999). Occasionally consumed foods are defined as those that are consumed less than once a week by 75% of the population or more. Data were available on the frequency of consumption from the 1995 Australian National Nutrition Survey for broad food groups or similar foods (e.g. 'legumes' and 'sweet potato') but not for foods specifically analysed. However, the data for the broader food group justified the assumption that the relevant surveyed foods would be occasionally consumed and a median consumption value was appropriate to use for these foods. These foods include linseed oil, taro and lima beans, among others, depending on the population group, and are all identified in the results tables in Appendix 10 as P50, with a summary of consumption levels used for each food provided in Table A10.8 of Appendix 10.

If there were not enough consumers for a 97.5th percentile for a commonly consumed food for a given population group (e.g. bread containing linseed for 2002 NZCNS), the 97.5th percentile for a broader group of that food (e.g. all grain and wholemeal breads) was used as a proxy. The data for the two days of consumption for the 2007 ANCNPAS were pooled to derive a larger dataset (i.e. not averaged across the two days) from which to derive consumption values with less uncertainty.

For several foods where there were no consumers in the nutrition survey or an appropriate broader food category, a different approach was taken. For example, for biscuits containing apricot kernels, the serve size on the label was used as the consumption amount; for apricot kernels recommended maximum consumption levels were used in the calculation. In other cases, where concentration data only were available, a back calculation was used to determine how much of each of these foods could be consumed before the ARfD would be exceeded for each population group. The exception was cherry brandy for children, which was not included as they are not expected to consume this food (see Section 5.4.2.2).

5.3.2 Food consumption data

FSANZ uses food consumption data from the most recent NNSs to estimate dietary exposure to food chemicals for the Australian and New Zealand populations.
In the assessment, for Australia, the following nutrition surveys were used:

- 2007 Australian National Children's Nutrition and Physical Activity Survey (ANCNPAS), which surveyed 4,487 children 2-16 years, collecting two days of food consumption data for all respondents (CSIRO, 2008)

- 1995 National Nutrition Survey (NNS), which surveyed 13,858 respondents aged 2 years and above, collecting one 24-hour recall on all respondents (McLennan & Podger, 1997); only the respondents aged 17 years and above (*n*=11,129) from this survey were used in this assessment

- 2009 NOURISH study for infants under 12 months of age sampled at three time periods (Daniels et al, 2009, for details see Appendix 7). As this study had an education intervention included, only the food consumption data from controls was obtained for use by FSANZ for risk assessment purposes. For this dietary exposure assessment, only the data from Time 1 were used (respondents aged 2-11 months).

For New Zealand, two nutrition surveys were used:

- 2002 National Children's Nutrition Survey (NZCNS), which surveyed 3,275 children aged 5-14 years, with one 24-hour recall conducted on all respondents (Ministry of

Health, 2002)

- 1997 NNS, conducted on 4,636 respondents aged 15 years and over, with a single 24-hour recall conducted for all respondents (Ministry of Health, 1999).

Survey weights are used in NNSs to ensure that the results derived from the survey sample are adjusted to be reflective of the population. Survey weights were used in the dietary exposure assessment for the 2007 ANCNPAS and the 2002 NZCNS. The 1995 and 1997 surveys were used unweighted as per FSANZs standard exposure assessment conventions (FSANZ, 2009).

5.3.2.1   Chronic dietary exposure assessment

For the chronic dietary exposure estimates, the distribution of consumption values from individuals in the nutrition surveys for each food was used in the calculations. As two 24-hour recalls were available for the 2007 ANCNPAS, both days were used for the chronic dietary exposure assessment. Estimated dietary exposures were then averaged across the two days for each respondent before population summary statistics were derived.

Specific consumption of RTE cassava chips (crisps) was not captured, or not captured to a large extent, from respondents in the nutrition surveys used in this risk assessment. As RTE cassava chips can contain high HCN concentrations, two options were investigated for the chronic dietary exposure assessment:

- Option 1 assumed all savoury snacks, such as potato chips, extruded snacks, soy chips and tapioca chips, were RTE cassava chips thus avoiding underestimation of chronic exposure to cyanogenic glycosides in the diet as all these products were assigned the HCN concentration derived for RTE chips. This assumes these are similar foods and are likely to be consumed in similar ways and in similar amounts

- Option 2 took account of the market share of RTE cassava chips in the savoury snacks category to give a more realistic estimation of population exposure to RTE cassava chips.

In the second option, approximate market share of salty snacks held by RTE cassava chips was determined and applied to the concentration of HCN in RTE cassava chips. Specifically, the grocery volume of salty snacks (including potato, corn, cereal and 'other' salty snacks) available in Australia is approximately 49,377 tonnes per year, with the volume of 'other' salty snacks (presumably including RTE cassava chips) being 2,534 tonnes per year. Therefore, the volume share of 'other' salty snacks is approximately 5% (Retail Media, 2010). It should be noted that this is an approximation only, used in the absence of actual market share data. The HCN concentrations derived incorporating the approximate market shares of 5% are shown in Appendix 6.

This method of using a broader food group and applying a market share value was also used for other foods for which there were no consumers, including bread containing linseed (no reported consumers in the 1995 and 1997 surveys), whereby all grain and wholemeal breads were used as a proxy, and amaretti biscuits and almond fingers/apple fingers, whereby all nut containing biscuits were used as a proxy (see Appendix 7 for further information).

5.3.2.2   Acute dietary exposure assessment

For the acute dietary exposure assessments, 97.5th percentile or median consumption values were derived for each relevant food using the individual dietary records from each consumer of that food for each nutrition survey, as described in Section 5.3.1.2.

Consumption amounts for each food represented the total amount of that food eaten, irrespective of whether it was consumed alone and/or as an ingredient in mixed foods. For example, the total consumption of passionfruit included passionfruit eaten fresh, in fruit salad, in cakes, slices and other desserts, as a topping on pavlova or in mixed juices.

The only exception to the method of deriving consumption amounts for an acute dietary exposure estimate (as described above) was apple juice. Only apple juice and blended juices containing apple juice were included in the total consumption amounts for juice and it was assumed that the concentration of HCN in unblended apple juice was equivalent to that in the apple juice within the juice blends. This exception was made because the consumption amount of the juice itself was deemed to present the greatest acute risk. This approach is considered valid, because this food is not commonly used as an ingredient in mixed foods or further cooked/prepared before consumption.

The recommended maximum consumption levels of raw apricot kernels, as provided on websites associated with the product (32 kernels/day) and consumption levels previously considered by FSANZ to be safe (4 kernels/day) were used in the acute dietary exposure estimates as consumption data were not available from the NNSs (see Appendix 7 for further information).

To assess the risk to infants for specific foods (apple juice, infant apple puree), NOURISH data for the first survey time period were used, with between 1 and 3 days of consumption data per respondent treated as separate days of data. A summary of the food consumption data used for infants in the assessment are shown in Table A10.1 and further details on the NOURISH data are provided in Appendix 7.

Food consumption data from all four national nutrition surveys were on a 24-hour basis. The data from the NOURISH study were on a 'per eating occasion' basis, which is also appropriate for use in an acute dietary exposure assessment.

5.3.3 Food mapping

Mapping is the process of matching the foods analysed for HCN to the foods consumed in the NNSs. Direct mapping was used in this exposure estimate. Direct mapping requires the analysed foods to be matched to the same foods or to appropriately similar foods consumed in the surveys where possible. For example, the analysed food 'Apple sauce' was only mapped to apple sauce in the surveys. Details of the analysed foods and the nutrition survey foods to which they were mapped are provided in Appendix 8. As previously mentioned, if a food was not consumed in a nutrition survey (e.g. amaretti biscuits), then consumption data were derived from other sources.

5.3.4 Population groups assessed

Chronic dietary exposure assessments are usually conducted for the whole population. Therefore, the four nutrition surveys were used in total. This was with the exception of the 1995 NNS where only data from respondents aged 17 years and over were used. This is because the Australian population younger than 17 years was included in the 2007 ANCNPAS.

Acute dietary exposure estimates were derived for adults and children. Children up to and including 6 years of age as well as older children were assessed using the available age groupings in the 2007 ANCNPAS and 2002 NZCNS surveys. The age range for the NZCNS was from 5-14 years only so acute estimates for the 5-6 year old age group only could be reported for younger children.

Acute dietary exposure estimates for infants 2-11 months old, as surveyed through the NOURISH study, were included for selected foods (apple juice, infant apple puree).

A summary of the population groups examined in this risk assessment are shown in Table 8. Mean body weights are used in the calculations of acute dietary exposure. The mean body weights for each of the population groups assessed are shown in Table 9.

Table 8: Population groups assessed for acute and chronic dietary exposure assessments

Exposure assessment type	Infants 2-11 months	Australia			New Zealand		
		2-6 years	2-16 years	17 years and above	5-6 years	5-14 years	15 years and above
Acute	✓*	✓	✓	✓	✓	✓	✓
Chronic			✓	✓		✓	✓

* Selected foods only

Table 9: Mean body weights for population groups used in the acute dietary exposure assessment

Country	Age group	Nutrition survey	Mean body weight (kg)
Australia	2-6 years	2007	19
	2-16yrs	2007	38
	17 years and above	1995	74
New Zealand	5-6 years	2002	23
	5-14yrs	2002	40
	15 years and above	1997	71
All	2-11 months	NOURISH	7.2

5.3.5 Assumptions made and limitations of the dietary exposure assessment

The aim of dietary exposure assessments is to make an estimate of dietary exposure to the food chemical of interest that is as realistic as possible. Where uncertainties in the data existed, a conservative approach was taken to be protective of the health and safety of consumers.

Many of the limitations associated with dietary exposure assessments conducted by FSANZ are common across most assessments. Limitations include: the age of the consumption data; that one/two days of consumption data are used to estimate chronic dietary exposure; and that concentration data for Australian foods reflects that for New Zealand foods and vice versa etc. Such limitations are outlined in detail in the document *FSANZ principles and*

practices of dietary exposure assessment for food regulatory purposes (FSANZ, 2009) and will not be elaborated on here.

Limitations specific to this assessment are outlined in more detail below:

- Concentration data – only a limited number of samples were analysed for each food. For some foods, only one sample was analysed. As such, there is less certainty around whether the analysed values are representative of that food and it is impossible to reliably determine the degree of variation of HCN concentrations in that food. This is not as much of an issue in cases where the concentrations of HCN obtained reflect those in the scientific literature e.g. cassava starch products, which had no detectable HCN.

- Consumption data – there were some foods that were consumed by only a small number of consumers. These foods include marzipan, lima beans and linseed products. These foods have detectable concentrations of HCN, however, the resulting estimates of dietary exposure may not accurately reflect current levels of exposure to this food.

- Food consumption data for infants – the food consumption data for infants was obtained from the NOURISH study. Although this study was conducted in Australia, the results were assumed representative of infants in New Zealand.

Despite the assumptions made and the limitations described above, the results of the dietary exposure assessment provide a basis from which to identify the potential for any risks and possible areas for more detailed work to be conducted in the future.

5.3.6 Chronic dietary exposure assessment

The results of the chronic dietary exposure assessment are shown in Table 10. For respondents, estimated mean dietary exposures ranged from 1 to 2 µg/kg bw/day for the lower bound concentrations and from 1 to 3 µg/kg bw/day for the upper bound. For consumers of foods containing HCN, estimated mean dietary exposures ranged from 1 to 2 µg/kg bw/day for the lower bound and from 1 to 3 µg/kg bw/day for the upper bound. For consumers, estimated 90th percentile dietary exposures were 3 to 4 µg/kg bw/day for the lower bound and from 3 to 5 µg/kg bw/day for the upper bound.

As illustrated in Table 10, there are different numbers of consumers of foods containing HCN for lower bound and upper bound results. This is because for lower bound concentrations, a mean of zero was applied for foods where all concentrations were below the LoD. Even if there were respondents in the nutrition surveys who had consumed these foods, they were not counted as a consumer of foods containing HCN as the concentration was zero and therefore their exposure was zero from that food. One impact of this is that the exposures for the lower and upper bound concentrations cannot be presented as a range.

The major contributors to estimated chronic dietary exposure to HCN (≥5% contribution for at least one population group) are shown in Table 11. Bread containing linseed was a major contributor in all four population groups assessed (24-75%), as was RTE cassava chips (5-22%). For Australian and New Zealand children another high contributor was linseeds (26-32%). Cassava roots were a major contributor for New Zealanders (7-13%), but not Australians. A full list of contributions for all foods can be found in Appendix 9.

Table 10: Estimated chronic dietary exposures for consumers of foods containing cyanogenic glycosides (measured as total HCN) for Australian and New Zealand population groups

Population group	Number of respondents	Number of consumers		Exposure units	Estimated dietary exposure					
					All respondents		Consumers only			
		(% consumers)			Mean		Mean		90th percentile	
		LB	UB		LB	UB	LB	UB	LB	UB
Australia 2-16 years	4,487	3,859	4,456	µg/day	47	81	55	82	81	146
		(86.0)	(99.3)	µg/kg bw/day	2	3	2	3	3	5
Australia 17 years and above	11,129	6,146	10,186	µg/day	50	87	90	95	188	213
		(55.2)	(91.5)	µg/kg bw/day	1	1	1	1	3	3
New Zealand 5-14 years	3275	2,105	3,146	µg/day	48	74	74	77	154	166
		(64.3)	(96.1)	µg/kg bw/day	1	2	2	2	4	5
New Zealand 15 years and above	4,636	2,296	4,237	µg/day	58	85	118	93	206	205
		(49.5)	(91.4)	µg/kg bw/day	1	1	2	1	3	3

Note: Lower bound (LB) results are calculated by assigning concentrations below the LoD as zero; upper bound (UB) results are are calculated by assigning concentrations below the LoD as a concentration equal to the LoD.

Table 11: Major contributors (≥5%) to chronic dietary exposure to total HCN for the population groups assessed

Food	Contribution* (%)			
	Australia		New Zealand	
	2-16 years	17 years and above	5-14 years	15 years and above
Almonds	<5	8	<5	<5
Apple juice[a]	6	<5	<5	<5
Bamboo shoots[b]	<5	<5	5	<5
Bread containing linseed	37	75	24	75
Cassava, roots, cooked[c]	<5	<5	7	13
Cassava chips, ready-to-eat	15	5	22	5
Linseeds	26	NC	32	NC
Passionfruit	6	<5	<5	<5

Shaded cells are where contribution is ≥5%.
* Based on lower bound (LB) scenario where not detected results were assigned a zero concentration of HCN.
NC = not consumed (i.e. not recorded as being consumed in the NNS).
[a] Survey 2, EU method.
[b] Survey 1 only.
[c] Survey 3, sweet cassava only.

5.3.7 Acute dietary exposure assessment

Estimated acute dietary exposures to HCN from individual foods for different age groups of Australian and New Zealand consumers are presented in Appendix 10.

There was a broad range of estimates of acute dietary exposure given the different foods with very different concentrations of HCN, the consumption level used, the consumption amount and the numerous population groups assessed. Exposures ranged from <1 to 755 µg/kg bw/day across all of the foods and population groups assessed.

For Australian and New Zealand adults, the highest estimated acute exposures to HCN were associated with the consumption of raw apricot kernels with skin. Based on a consumption level of 32 kernels/day, estimated acute exposures ranged from 724-755 µg/kg bw/day. For raw apricot kernels without skin, acute dietary exposures at consumption levels of 32 kernels/day were lower, ranging from 113-118 µg/kg bw/day. For all or most population groups, there were high estimated acute exposures for bread containing linseed (163-511 µg/kg bw/day) and cassava roots (up to 80 µg/kg bw/day).

5.4   Risk characterisation

5.4.1 Characterisation of chronic risk

To characterise the chronic exposure risk, the estimated chronic dietary exposures to HCN in cyanogenic glycoside containing foods were converted to a cyanide ion concentration for each age group and compared with the PMTDI of 20 µg CN⁻/kg bw.

The chronic dietary exposures of cyanogenic glycoside containing food expressed as a per cent of the PMTDI are shown in Table 12. There were no exceedances of the PMTDI for any population group assessed, even based on the upper bound concentrations and estimated 90th percentile dietary exposures for consumers of foods containing HCN. The highest level of estimated chronic

dietary exposure as a per cent of the PMTDI was 25% PMTDI for Australians aged 2-16 years, based on the upper bound concentrations and 90th percentile dietary exposures. On this basis, it is unlikely that there will be any public health and safety issues in relation to chronic dietary exposure to HCN for the Australian and New Zealand populations.

Table 12: Estimated chronic dietary exposures to HCN for Australian and New Zealand population groups as a per cent of the provisional maximum tolerable daily intake (PMTDI) (%)

Population group	% PMTDI*					
	All respondents		Consumers only			
	Mean		Mean		90th percentile	
	LB	UB	LB	UB	LB	UB
Australia 2-16 years	8	15	9	15	15	25
Australia 17 years and above	3	6	6	6	15	15
New Zealand 5-14 years	7	10	10	10	20	20
New Zealand 15 years and above	4	6	8	6	15	15

* PMTDI: 20 µg CN⁻/kg bw (FAO/WHO, 2012).
Note: Lower bound (LB) results are based on where concentrations below the LoD were assigned a zero; upper bound (UB) results are based on where concentrations below the LoD were assigned a concentration equal to the LoD.

5.4.2 Characterisation of acute risk

5.4.2.1 Estimated acute dietary exposures to HCN for Australian and New Zealand populations (from NNS data[8])

The estimated acute dietary exposures for individual foods were compared to the ARfD of 80 µg HCN/kg bw. Results are shown in Table 13 for all population groups assessed (with the exception of infants) and Table 16 for infants, for selected apple-based foods.

There is a higher degree of uncertainty in acute dietary exposure estimates if based on a small number of consumers of the food of interest. As described in Section 5.3.1.2, where there were fewer than 39 consumers of a food, the 50th percentile (median) rather than the 97.5th percentile consumption was derived for use in the acute dietary exposure estimate. For all these foods, except cassava roots, where the 50th percentile was derived, the acute estimated dietary exposures were well below the ARfD. Median consumption of cassava roots by New Zealand adults may result in acute dietary exposures at 100% of the ARfD.

The consumption of a serving of raw apricot kernels and 97.5th percentile consumption of bread containing linseed may lead to exceedances of the ARfD in some or all Australian and New Zealand population groups. All other foods assessed at the 97.5th percentile level of consumption, or by using recommended serving sizes, had estimated exposures below the ARfD.

Further information regarding the acute dietary exposure estimates is provided below for cassava roots and bread containing linseed, as well as bamboo shoots (traditionally considered a high risk food in regards to HCN). For raw apricot kernels, further information regarding the acute dietary exposure estimates is provided in Section 5.4.2.2.

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[8]This is with the exception of apricot kernels, amaretti biscuits and almond fingers/apple fingers, as no NNS data were available. For these foods, recommended serving sizes (and, in the case of apricot kernels, consumption amounts equating to what FSANZ had previously considered to be safe) were used.

Table 13: Estimated acute dietary exposures to total HCN for Australian and New Zealand population groups as a per cent of the acute reference dose (ARfD) (%)

Food	% ARfD*					
	Australia			New Zealand		
	2-6 years	2-16 years	17 years and above	5-6 years	5-14 years	15 years and above
Almonds	10	7	8	1[#]	8	7
Almond oil	NC	NC	NC	NC	NC	NC
Apple juice[a]	6	4	2	8	4	3
Apple sauce	NC	NC	1[#]	4[#]	2[#]	2[#]
Apple cider vinegar	4	2	1	2	2	1
Apricots, canned	30	15	15	3[#]	15	15
Apricot jam	2	3	2	NC	<1[#]	<1[#]
Apricot kernels, with skin (32/d)	NA	NA	910	NA	NA	940
Apricot kernels, with skin (4/d)	NA	NA	110	NA	NA	120
Apricot kernels, without skin (32/d)	NA	NA	140	NA	NA	150
Apricot kernels, without skin (4/d)	NA	NA	20	NA	NA	20
Apricot nectar	35[#]	7[#]	20[#]	3[#]	35	NC
Bamboo shoots[b]	20	15	65	25[#]	45	3[#]
Biscuits, amaretti	65	35	15	55	30	20
Biscuits, almond fingers/apple fingers	40	20	10	35	20	10
Bread containing linseed	520	350	200	639	360	250
Butter beans	8[#]	5[#]	4[#]	2[#]	<1[#]	NC
Cassava roots, cooked[c]	9[#]	80	85[#]	60[#]	35[#]	100[#]
Cassava starch	9	5	1[#]	2	1	<1[#]
Cherry brandy	<1[#]	6	15	NC	<1[#]	10
Lima beans	20[#]	10[#]	75	NC	NC	4[#]
Linseeds	55	30	NC	35	20	NC
Linseed oil	3[#]	2[#]	NC	NC	NC	NC
LSA mix	20[#]	10[#]	NC	NC	NC	NC
Marzipan	NC	2[#]	4[#]	NC	NC	NC
Passionfruit	20	10	6	8	9	4
Prune juice	40[#]	20[#]	35[#]	NC	NC	15[#]
Pumpkin seeds	<1[#]	<1[#]	NC	2[#]	<1[#]	NC
Spinach	50	25	25	2[#]	20	15
Sunflower seeds	3	2	1	2	<1	1
Taro in mixed foods	NC	NC	NC	15[#]	8[#]	15[#]
Taro, leaves only	NC	NC	NC	15[#]	9[#]	5[#]
Vine leaves, stuffed, canned	65[#]	25[#]	8[#]	NC	NC	NC

NC = not consumed.
NA = not applicable. Dietary exposure assessments not conducted for children for this food.
* ARfD = 80 µg HCN/kg bw (FSANZ, 2008).
[#] P50 Median consumption level used for consumers only, rather than 97.5th percentile (number of respondents <39).
[a] Survey 2, EU method.
[b] Survey 1.
[c] Survey 3, sweet cassava only.

Cassava roots

Acute dietary exposure estimates fell below the ARfD except for the New Zealand adult population, which was at 100% of the ARfD (Table 13), based on median levels of consumption for consumers of cassava.

Based on the maximum consumption amount of cassava reported by New Zealand adults in the 1997 NNS of approximately 220 g per day, back calculations were also undertaken to determine the %ARfD reached at different concentrations of HCN. Table 14 indicates that cooked cassava with concentrations of total HCN approaching 30 mg HCN/kg leads to acute dietary exposure estimates that exceed the ARfD. The mean reduction of HCN upon cooking was 56%, with a range of 28-83%. If the per cent HCN reduction is at the lower end of this range, it is possible that a concentration of 30 mg HCN/kg in cooked cassava (and thus an exceedance of the ARfD) could occur in cases where the raw cassava contained just below the maximum level of 50 mg total HCN/kg permitted in regulations.

Table 14: %ARfD reached at different concentrations of total HCN in cooked cassava roots*

Concentration in cooked cassava (mg HCN/kg)	%ARfD[#] New Zealand 15 years and above
15	55
20	75
25	95
30	110
35	130

* Based on a consumption amount of 220 g of cooked cassava root.
[#] ARfD = 80 µg HCN/kg bw (FSANZ, 2008).
Shaded cells are ≥100% ARfD.

The acute dietary exposure assessment is intended to be conservative as it is an estimate of 'worst case' exposure, but is still protective of the health and safety of consumers. In this instance, it was assumed: (1) HCN levels in the raw product approach the maximum levels for sweet cassava, as defined in the regulations; (2) minimum losses of HCN after cooking (28%); together with (3) high consumption amounts (median amount for consumers of cassava). It is considered unlikely that these three events will occur simultaneously.

The following issues also impact on characterisation of the risk from consumption of cassava:

- FSANZ is not aware of any data in the scientific literature that suggests cases of acute cyanide poisoning have occurred in Australia or New Zealand as a result of consuming properly processed cassava

- The availability of non-compliant raw bitter cassava (>50 mg HCN/kg) in the market place (which may cause exceedances of the ARfD if consumed, despite conventional processing) is not known

- The available data on cassava consumption patterns are limited, making accurate estimates of the likely exposure for high consumers resulting in an exceedance of the ARfD more difficult to determine.

Although there are certain gaps in the data that impact on characterisation of the risk, given the conservative nature of the acute dietary exposure assessment, together with the absence of any reports on poisonings in Australia and New Zealand, the estimated acute dietary exposures to HCN are not considered to represent an appreciable health and safety risk.

FSANZ will continue to consider the risk characterisation of this food as further information becomes available.

Bamboo shoots

The acute dietary exposure estimates were calculated using the maximum concentration (44 mg HCN/kg, Appendix 3) for bamboo shoots from Survey 1 (i.e. canned/pickled varieties). This resulted in exposure estimates below the ARfD for all population groups (Table 13).

Bread containing linseed

For bread containing linseed, and using the maximum concentration of HCN detected, the ARfD was exceeded for all population groups assessed (200-639% ARfD) (Table 13).

The consumption data for this food are robust, with high numbers of consumers in most cases. (Note that for some population groups, where specific consumption data were unavailable, grain and wholemeal breads were used as a proxy for bread containing linseed as consumption amounts recorded for these are likely to be similar). In terms of the analytical data, HCN concentrations are considered to be realistic as they are consistent with concentrations of HCN in linseeds themselves (as sourced/analysed in each respective country) and the stated linseed content of the bread according to label information. Therefore, despite the small number of bread samples (*n*=6), it is reasonable to assume that the dietary exposure estimates are realistic.

There are no reports in the scientific literature for human poisonings following linseed consumption. Studies of the toxicokinetic profile of cyanide following consumption of 100 g of ground linseed show an undetectable amount of cyanide in the blood of human volunteers (Schilcher et al, 1986). These data suggest that there is unlikely to be an acute public health and safety risk associated with the consumption of bread containing linseed.

5.4.2.2 Maximum possible consumption before exceeding the acute reference dose for specific foods (no NNS data)

For some foods and population groups, there were no consumers to enable a deterministic acute dietary exposure to be calculated. These were indicated by the use of 'NC' (not consumed) in Table A10.2 to A10.7 in Appendix 10 in the column 'Number of consumers'. Therefore, a calculation was done using the maximum concentration of HCN in the food from the available ISFR survey data and the mean body weight of the population group, to determine the maximum amount of the food that could be consumed before the ARfD is exceeded. This calculation was also done for raw apricot kernels and biscuits containing apricot kernels, in addition to the deterministic acute dietary exposure estimates discussed above. The results are shown in Table 15.

The resulting consumption values were evaluated to determine whether these levels of consumption were likely. With the exception of raw apricot kernels and linseeds, for the majority of the foods assessed, the amount that would need to be consumed to exceed the ARfD seems unrealistic for the types of foods in question and the manner in which they would normally be consumed. Further comments regarding raw apricot kernels and other foods of interest are provided below.

Table 15: Maximum amount of food that can be consumed (g/day) based on the ISFR survey concentrations, before the acute reference dose (ARfD) is exceeded

Food	Concentration (mg HCN/kg)	Maximum consumption (g/day)					
		Australia			New Zealand		
		2-6 years	2-16 years	17 years and above	5-6 years	5-14 years	15 years and above
Almond oil	4	380	760	1480	460	840	1420
Apple sauce	4	371	741				
Apricot jam	4				460		
Apricot kernels (with skin)	2820	0.5 (<1 kernel*)	1.1 (1 kernel*)	2.1 (3 kernels*)	0.7 (1 kernel*)	1.2 (1 kernel*)	2.0 (3 kernels*)
Apricot kernels (without skin)	440	6.9 (11 kernels*)	13.5 (22 kernels*)	13.5 (22 kernels*)	4.2 (6 kernels*)	7.6 (12 kernels*)	12.9 (21 kernels*)
Apricot nectar	-5						1136
Biscuits, amaretti	34	44	89	174	54	94	167
Biscuits, almond fingers/apple fingers	13	116	233	455	141	246	436
Butter beans	7						835
Lima beans	32				57	104	
Linseeds	178			33			32
Linseed oil	4			1480	460	840	1420
LSA mix	60			99	31	56	95
Marzipan	5	287		1117	347	634	1072
Prune juice	4				460	840	
Pumpkin seeds	4			1480			1420
Taro, in mixed foods	4	380	760	1480			
Taro, leaves only	4	380	760	1480			
Vine leaves, stuffed, canned	4				460	840	1420

Acute reference dose = 80 µg HCN/kg bw.
Blank cells: A deterministic acute dietary exposure was able to be done for this population group therefore this calculation was not required.
* Weight of 1 kernel = 0.6 grams, rounded down to nearest whole kernel.

Apricot kernels

A deterministic calculation of acute dietary exposure was conducted for raw apricot kernels for adults only (see Table 13). However, as no nutrition survey data were available for this product, the assessment was based on estimated consumption of recommended serve sizes as provided by websites associated with the product and from what FSANZ had previously considered to be safe (FSANZ, 2011).

Apricot kernels with skin had a much higher HCN concentration than those without. Based on recommended serve sizes as provided on various websites (32 kernels/day), estimated dietary exposures ranged between 910-940% ARfD for kernels with skin and 140-150% ARfD for kernels without skin. Based on the available data at the time, suggesting that the maximum total HCN content of raw apricot kernels was around 2,300 mg HCN/kg, FSANZ had advised that no more than 4 kernels per day should be consumed (FSANZ, 2011). With the higher total HCN levels in

raw apricot kernels being reported in Survey 3 (2,820 mg HCN/kg), dietary exposure from 4 kernels slightly exceeded the ARfD for kernels with skin (110-120%) but not for kernels without skin (20% ARfD) (see Table 13).

The exceedance of the ARfD by up to nine-fold is of considerable concern for public health and safety because, there are multiple published cases of adult and child cyanide poisoning resulting from eating cooked and/or ground apricot kernels (or pits). These reported cases ranged from clinical signs of mild cyanide poisoning (headache, nausea) right through to severe intoxication resulting in hypotension, coma, convulsions and death (Sayre & Kaymakcalan 1964; Lasch et al 1981; Suchard et al 1998; Akyildiz et al 2010; Cigolini et al 2011; Akıl et al 2013). Almost invariably these published cases only describe presentations at hospitals necessitating clinical intervention.

An estimation of the maximum number of raw apricot kernels that could be consumed before exceeding the ARfD was also undertaken for this product using maximum mean levels derived from the analytical data from Survey 3, for both adults and children (Table 15). For apricot kernels with skin, it was estimated that adults could consume 3 kernels and children could not even consume 1 kernel/day before the ARfD is exceeded. For apricot kernels without skin, adults could consume 21 kernels and children could consume 6 kernels/day before the ARfD is exceeded.

It is recognised this survey analysed a relatively small sample of raw apricot kernels and that the levels of HCN/kg may be more variable. However it is noted that literature reports suggest the mean range of HCN/kg found in kernels from a variety of different apricot cultivars can be up to 4,090 mg/kg (e.g. Holzbecher et al, 1984). A random chance selection of kernels with a higher HCN content would increase the potential health risk.

Apricot nectar

There were no consumption data for New Zealand adults and so a back calculation was done for this group to determine the maximum amount of apricot nectar that could be consumed before the ARfD is exceeded. The maximum amount of this food that could be consumed before exceeding the ARfD is 1,136 g/day, assuming HCN is present at the ML of 5 mg/kg (Table 15). Based on 50th percentile consumption levels recorded for Australian adults (223 g) (Table A10.4), this level of consumption is unlikely to occur.

Amaretti biscuits

A deterministic calculation of acute dietary exposure was conducted, however, it was based on estimated consumption amounts from serve sizes on the product label as no food consumption data were available. Therefore, an estimation of the maximum amount that could be consumed before exceeding the ARfD was also undertaken (Table 15). For children aged 2-6 years, it was estimated that the maximum amount of amaretti biscuits that could be consumed before exceeding the ARfD was only 44 g. However, this value is higher than the serve size of 30 g, as given on labels. In addition, these types of biscuits tend to have a slightly bitter taste, which are less likely to be palatable to children and, as such, they would be less likely to be consumed in large quantities.

Almond fingers/apple fingers

Like amaretti, deterministic acute dietary exposures were calculated using serve sizes provided on labels as an indication of likely consumption amounts. In this case the serve size given was 47 g. The maximum concentration of HCN detected in almond fingers/apple fingers was less than half that of amaretti biscuits. Therefore, the estimated maximum consumption amounts before exceeding the ARfD were calculated as being much higher than amaretti biscuits—116 g for children aged 2-6 years, much higher than the 47 g serve size (Table 15).

Linseed

There were no consumption data for New Zealand and Australian adults for linseeds consumed directly as seeds and therefore a back calculation was done for these groups. The maximum amount of linseeds that could be consumed before exceeding the ARfD is 32-33 g/day (Table 15). Consumption at levels near this maximum calculated amount is feasible, however, like bread containing linseed, the toxicokinetic profile suggests that there is no acute public health and safety risk associated with the consumption of linseeds.

Prune juice

There were no consumption data for New Zealand children and so the back calculation method was used to determine that the maximum amount of prune juice that could be consumed by New Zealand 5-6 year olds before exceeding the ARfD. The maximum amount of prune juice that could be consumed by New Zealand 5-6 year olds before exceeding the ARfD is 460 g/day. This amount might seem realistic for a juice. However, prune juice is not usually consumed in as large quantities as other juices such as orange and apple juice, probably in large part due to its laxative properties. The 50th percentile consumption for prune juice for Australian children 2-16 years was 157 g/day, suggesting that consumption at levels near the maximum calculated amount is unlikely (Table A10.3).

5.4.3 Characterisation of acute risk – infant foods

For selected infant foods, the estimated acute dietary exposures as a per cent of the ARfD are shown in Table 16.

Table 16: Estimated acute dietary exposures to total HCN for Australian and New Zealand infants (2-11 months old) for selected apple based foods as a per cent of the acute reference dose (ARfD) (%)

Food	Estimated exposure to total HCN (%ARfD)*
Apple juice (Survey 2, EU method)	4
Infant apple puree (Survey 3, EU method)	30

* ARfD = 80 µg HCN/kg bw (FSANZ, 2008).

Using the maximum concentration of 0.17 mg HCN/kg (EU HPLC method), estimated exposure to HCN as a per cent of the ARfD was 4% for infants.

For infant apple puree, using the maximum concentration of 1.3 mg HCN/kg (EU HPLC method), estimated exposure to HCN as a per cent of the ARfD was 30%.

5.6   Summary of risk assessment

For the chronic dietary exposure assessment, the highest estimated dietary exposure to HCN across all population groups was 5 µg/kg bw/day for consumers at the 90th percentile (upper bound concentrations). The major contributors to estimated chronic dietary exposure to HCN included bread containing linseed and RTE cassava chips. For Australian and New Zealand children another high contributor was linseeds. Cassava roots were a major contributor for New Zealanders, but not Australians.

For the estimated chronic dietary exposure to HCN, there were no exceedances of the PMTDI for any population group assessed. On this basis, it is unlikely that there will be any public health and safety issues in relation to chronic dietary exposure to HCN for the Australian and New Zealand populations.

For the acute dietary exposure assessment, there was a broad range of estimates of exposure given the variety of foods and levels of consumption. For Australian and New Zealand adults, the highest acute exposures to HCN were associated with the consumption of raw apricot kernels with skin, followed by the consumption of raw apricot kernels without skin. For all or most population groups, there were high acute exposure estimates for consumers of bread containing linseed and cassava roots.

The acute dietary exposure assessment identified that all population groups had estimated acute dietary exposures under the ARfD of 80 µg/kg bw/day for the majority of foods. However, the consumption of raw apricot kernels and bread containing linseed may lead to exceedances of the ARfD in some or all Australian and New Zealand population groups. Median consumption of cassava roots by New Zealand adults resulted in estimated acute dietary exposures at 100% of the ARfD.

The consumption of raw apricot kernels both with and without skin poses the greatest acute public health and safety risk for all Australian and New Zealand population groups. For cassava roots estimated acute dietary exposures to HCN are not considered to represent an appreciable health and safety risk although these estimates may reach 100% ARfD for high consumers (median level of consumption). It is considered unlikely that the 'worst case' exposure scenario used in this calculation would occur (maximum level of HCN in raw products followed by minimum losses of HCN after cooking and a high consumption amount) and FSANZ is not aware of any reports on poisonings in Australia or New Zealand following consumption of properly processed cassava. For bread containing linseed, although estimated acute dietary exposures do exceed the ARfD, there were no detectable levels of cyanide in the blood of human volunteers consuming ground linseeds, so consumption of linseeds or products containing linseeds is not considered to represent an appreciable health and safety risk.

6. RISK MANAGEMENT

The Code currently includes a range of regulatory measures that are intended to limit and manage HCN in cassava, cassava based foods (such as RTE cassava chips) and bamboo shoots. The Code also includes the MLs for a range of foods that may contain HCN as a result of use or presence of apricot kernels in their production. However, there are currently no risk management measures in the Code to manage HCN levels in raw apricot kernels.

FSANZ has prepared Proposal P1016 to consider options for the management of potential risks identified from the consumption of foods containing HCN that have been identified in the analysis of the ISFR survey results.

For cassava roots, it was estimated that for high consumers there was a potential to exceed the ARfD, based on an assumption of a high level of HCN in raw cassava and minimal losses on cooking. However, in the absence of any reports on poisonings in Australia and New Zealand following consumption of properly processed cassava, FSANZ concludes that there is no appreciable health and safety risk for consumers and that no additional regulatory risk management measures for cassava roots are needed at this time.

FSANZ has advised the relevant enforcement agencies that one sample of cassava root analysed did not meet the definitional requirements for sweet cassava, and that a number of samples of frozen cassava root and fresh/frozen bamboo shoots did not have directions for use on the label or accompanying the product, as required under Standard 1.2.6 – Directions for use and storage. Therefore, while FSANZ may not need to consider additional risk management measures for cassava (and bamboo shoots) at this time, food enforcement agencies may need to consider whether any actions need to be taken in relation to compliance with Standard 1.2.6 of the Code. Upon release of this survey and risk assessment report, a link to a FSANZ fact sheet giving advice to consumers on the safe preparation of these foods and the requirements for these foods to include directions for use will be provided.

Taking into account the most up-to-date risk assessment conclusions, the focus of FSANZ's risk management measures should be on managing the risk arising from high concentrations of HCN in raw apricot kernels. In the interim, whilst this measure is being considered, updated advice on the maximum amount of raw apricot kernels that can safely be consumed has been provided by a new fact sheet on the FSANZ website.

7. CONCLUSIONS

The findings from the ISFR survey identified that cyanogenic glycosides are present in a wide range of Australian and New Zealand plant-based foods, at levels that were within regulatory limits for HCN (where such regulatory limits exist) in all but two cases and at levels consistent with or lower than those reported in the scientific literature.

In considering the characterisation of the risk, it is important to note the limitations associated with the dietary exposure estimates, including the limited concentration data available for most foods and the small number of consumers that reported consuming some of the surveyed foods. Where uncertainties in the data existed, a conservative approach was taken to estimate dietary exposure and to be protective of the health and safety of consumers.

A risk assessment based on these survey results indicated that there were no public health and safety issues in relation to the estimates of chronic dietary exposure to HCN for the Australian and New Zealand populations.

There were a small number of foods (raw apricot kernels, cassava roots and bread containing linseed) where it was estimated that acute dietary exposure had the potential to exceed the ARfD.

Consumption of raw apricot kernels both with and without skin can pose an acute public health and safety risk for all Australian and New Zealand population groups even at levels well below the serve sizes that accompany the sale of the product. Risk management options for managing this risk will be considered as part of the FSANZ Proposal P1016 in addition to providing advice for consumers on the FSANZ website.

For cassava roots, there is potential for high consumers to exceed the ARfD. However, due to the conservative approach taken in calculating estimated dietary exposures and the absence of any reports on poisonings in Australia and New Zealand, FSANZ concludes no additional risk management measures for cassava are needed at this time and risk management actions should focus on enforcement of the current standards. Food enforcement agencies may also wish to consider whether any other enforcement and compliance actions are needed in relation to compliance with labelling requirements. This may also be relevant for bamboo shoots.

For bread containing linseed, although the estimated acute dietary exposures resulted in exceedances of the ARfD for all population groups assessed, current exposures are not considered to represent a health and safety risk based on the absence of any clinical reports of poisonings or detectable levels of cyanide in the blood of human volunteers following consumption of ground linseed.

8. ACKNOWLEDGEMENTS

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Appendix 1. Glossary of terms

Acute reference dose (ARfD)

An estimate of the amount of a substance in food and/or drinking-water, normally expressed on a body-weight basis, that can be ingested in a period of 24 hours or less, without appreciable health risk to the consumer.

Benchmark dose (BMD)

The term Benchmark Dose (BMD) is used generically to refer to the BMD assessment method. The BMD assessment method involves fitting a mathematical model with appropriate statistical measures to all the dose-response data within a study and therefore potentially incorporates more relevant information into the resulting estimates of health-based guidance values (e.g., ADI or ARfD).

Dietary exposure

The amount of a chemical that is ingested by a consumer as part of their diet (e.g. via food, beverages and drinking water).

Dose-response

The relationship in which a change in the magnitude of exposure to a chemical agent is associated with a change in the manifestation and magnitude of physiological effects.

Exposure assessment

The evaluation of the magnitude, frequency and duration of exposure to a chemical agent via food (as well as exposures from other sources, if relevant).

Hazard

A chemical agent in food with the potential to cause an adverse health effect.

Health-based guidance value (HBGV)

A numerical value that reflects the level of a chemical that can be ingested over a defined time period (e.g. daily for a lifetime or a single meal) without appreciable health risk.

Limit of detection (LoD)

The lowest concentration of a specific chemical that can be qualitatively detected using a specified laboratory method and/or item of laboratory equipment (i.e. its presence can be detected but not quantified).

Lower bound mean

An estimate of the mean concentration of a chemical in a food or dietary intake assuming analytical results reported as being below the LoD equal zero.

Maximum level (ML)

The maximum level of a contaminant or natural toxicant that is permitted to be present in a nominated food.

Natural toxicant

A chemical hazard naturally present in a particular food.

No-observed-adverse-effect-level (NOAEL)

The highest dose level of a substance that produces no adverse effects in the most sensitive test species.

Provisional maximum tolerable daily intake (PMTDI)

The maximum permissible human daily exposures to food contaminants unavoidable associated with the consumption of otherwise wholesome and nutritious food. The tolerable intake is referred to as 'provisional' as there is often a lack of data on the consequences of human exposure at low levels and new data may result in changes to the tolerable intake.

Upper bound mean

An estimate of the mean concentration of a chemical in a food or dietary intake assuming analytical results reported as below the LoD are equal to the value of the LoD.

Appendix 2. Food preparation instructions for foods requiring cooking

Frozen cassava root

Dividing retail samples into groups

1. The nine retail samples were divided into three groups ensuring that no two of the same brand were in one group.
2. Group 1 (retails samples 1, 2 & 3) were prepared using method 1 (boiling); Group 2 (retail samples 4, 5 & 6) were prepared using method 2 (frying); Group 3 (retail samples 7, 8 & 9) were prepared using method 3 (steaming).

Sizing

Cassava roots were cut to a 'standard' size prior to being cooked.

For whole tubers:

1. Each tuber was cut lengthways into four pieces. The thickness of the pieces for each tuber could vary depending on the diameter of the cassava tubers in the pack. Therefore, if necessary, large tubers were cut again (still lengthways), so that the largest section did not exceed about 4 cm.
2. Each piece was shortened to approximately 10 cm long.

For ready cut pieces:

1. Where necessary, pieces were cut again, lengthways, so that the largest section did not exceed about 4 cm.
2. Where necessary, each piece was shortened to approximately 10 cm long.

The inner thread was removed in cases where it appeared stringy and inedible. Every effort was made to proceed with cooking as soon as possible after cutting.

Cooking

For each of the three retail samples in each group, half of the packet's contents was set aside to analyse raw and half of the contents to analyse cooked. Each portion was clearly labelled to allow for later comparison of raw and cooked values (and calculation of adjustment factors).

Group 1/Method 1 - Boiling

1. Cassava pieces were not washed before boiling.
2. Pieces were placed in a saucepan and boiling water was poured over them, enough to cover.
3. The pieces were boiled for 20-25 minutes OR until tender (like boiling potatoes).
4. The cooked cassava pieces were drained immediately and completely, using a colander. Cooking water was discarded.

Group 2/Method 2 - Frying

1. Cassava pieces were not washed before frying.
2. Cassava pieces had to be sufficiently thawed to enable further cutting into strips.
3. Oil was heated in a deep fryer to approximately 350° F (180° C). The oil had to be hot enough to prevent the pieces from soaking up the oil and enable them to cook through until they were golden brown in colour.

4. In the meanwhile, cassava pieces were further cut into thin strips between 1-1.5 cm thick.
5. Strips were placed between sheets of paper towel and pat dried to remove any excess moisture.
6. Strips were gradually added to the hot oil and fried for about 15 minutes, or until deep golden and tender.
7. Strips were removed from the oil and drained.

Group 3/Method 3 - Steaming

1. Cassava pieces were washed thoroughly.
2. Pieces were soaked to allow to thaw for approximately 2 hours before steaming.
3. The soaking water was changed once during that time.
4. A saucepan was filled with enough water so that it just reached the bottom of the steamer basket and water was brought to the boil.
5. Cassava pieces were placed into the steamer basket and a loose fitting lid was placed on top to cover.
6. Pieces were steamed for approximately 20-25 minutes OR until tender (like boiling potatoes).
7. Cooked cassava pieces were removed from the steamer using a slotted spoon. Cooking water was discarded.

Bamboo shoots

For each of the retail samples:

1. The tough outer skin was peeled off (if present).
2. The tough root end was cut off (if present).
3. Each bamboo shoot was cut lengthwise into two pieces. Half was set aside to analyse raw and the other half to analyse cooked. Each half was clearly labelled to allow for later comparison of raw and cooked values (and calculation of adjustment factors).

Boiling

1. The bamboo shoot was placed in a saucepan with enough water to cover.
2. Water was brought to the boil, and simmered for at least an hour, until the bamboo shoot was tender enough to poke a skewer through.
3. Bamboo shoots were allowed to cool in the water; because peeling it while still hot could cause the shoot to split.
4. The bamboo shoots were drained.
5. Any remaining outer skin was peeled off.

Appendix 3. Analytical results

Table A3.1. Total HCN concentration of selected foods available in Australia and New Zealand analysed using the acid hydrolysis method

Food type	Number of samples analysed	Number positive for total HCN (%)	Total HCN in positive samples, mean (range) (mg HCN/kg)	Comparison of levels to the available scientific literature	Comparison to standards in the Code

Apricot kernels and apricot kernel products
Survey 3
    Apricot kernels, with skin
    Apricot kernels, without skin
    Amaretti biscuits˜
    Almond fingers/apple fingers˜
	
	18
	10
	1
	1
		
		18 (100)
		10 (100)
		1 (100)
		1 (100)
			
			2,120 (1,240-2,820)
			190 (49-440)
			34
			13
				Apricot kernels – range of 122-4,090 mg/kg (Holzbecher et al, 1984).
				
				Amaretti biscuits – 44 mg/kg (EFSA, 2004).
					There are no set maximum levels for total HCN for apricot kernels.

Cassava and cassava products
Survey 1
    Cassava, frozen root, raw (whole or grated)
    Cassava flour/tapioca products
 	
Survey 3
    Cassava, frozen root, raw
    Cooked – method 1 (steaming)
    Cassava, frozen root, raw
    Cooked – method 2 (boiling)
    Cassava, frozen root, raw
    Cooked – method 3 (frying)
	
	11
	
	4
	
	3
	3
	3
	3
	3
	3
		
		11 (100)
		
		0 (0)
		
		3 (100)
		3 (100)
		3 (100)
		3 (100)
		3 (100)
		3 (100)
			
			21.0 (8.6-43.6)
			
			-
			
			37.3 (34-40)
			16.0 (9.0-26)
			23.6 (16-32)
			15.0 (7.3-23)
			50.9 (31-81)
			18.8 (5.4-37)
				Raw – concentrations range from 10 mg/kg for sweet cassava up to 1,120 mg/kg for bitter cassava (EFSA, 2004).
				
				Reductions in HCN differ depending on the cooking method.
				
				Montagnac (2009) reports reductions of 10% for frying, 13% for baking or steaming and 45% for boiling.
				
				Other literature indicates reductions of 20% for baking, frying and steaming and 80-99% for boiling (CCCF, 2013).
					All but one sample in Survey 3 met the requirements of Standard 1.1.2 for 'sweet cassava'.
					
					Levels for all but one sample in Survey 3 complied with Standard 1.4.4 (<50 mg/kg HCN for 'sweet cassava').

Bamboo shoots
Survey 1
    Canned
    Other (pickled)
 	
Survey 3
    Fresh/frozen shoots, raw
    Cooked (boiling)
	
	7
	3
	
	3
	3
		
		7 (100)
		3 (100)
		
		3 (100)
		3 (100)
			
			10.0 (3.7-24.5)
			21.1 (9.6-44.0)
			
			320 (24-550)
			46 (28-73)
				Canned – 27 mg/kg (Ferreira et al, 1995).
				
				Raw – concentrations are within the range previously reported for bamboo shoots of 551 mg/kg (Chandra et al, 2004); 139-1,164 mg/kg (Haorongbam et al, 2009); 114-1,460 mg/kg (Haque and Bradbury, 2002).
					There are no set MLs for HCN for this product.

Food type	Number of samples analysed	Number positive for total HCN (%)	Total HCN in positive samples, mean (range) (mg HCN/kg)	Comparison of levels to the available scientific literature	Comparison to standards in the Code

Almonds and almond products
Survey 1
    Almonds (whole, flaked, ground or butter)
    Almond essence
    Marzipan
    Other (almond oil, almond jelly)
	6
	
	3
	4
	2
		3 (50)
		
		0 (0)
		1 (25)
		0 (0)
			8.4 (4.8-12.4)
			
			-
			5.3
			-
				Almonds – usually less than 10 mg/kg for 'sweet almonds' however higher concentrations may occur in some varieties (Karkocha, 1973; EFSA, 2004).
				
				Marzipan and other similar products made from apricot kernels – range of 15-50 mg/kg (EFSA, 2004).
					Levels comply with Standard 1.4.1.

Apple products
Survey 1
    Apple juice
    Apple sauce
    Apple cider vinegar
 	
Survey 2
    Apple juice
 	
Survey 3
    Infant apple puree
	8
	3
	1
	
	108
	
	8
		1 (13)
		2 (66)
		0 (0)
		
		4 (4)
		
		0 (0)
			5.4
			3.9 (3.6-4.1)
			-
			
			2.6 (1.5-4.2)
			
			-
				Apple seeds – range of 566-690 mg/kg (Haque and Bradbury, 2002; Holzbecher, 1984; WHO, 2012).
				
				There does not appear to be any published data on the total HCN content of apple juice or apple sauce.
					No standards established.

Linseed/flaxseed and products
Survey 1
    Linseed (whole or meal)
    LSA^
    Bread containing linseed
    Flaxseed and LSA oil
 	
Survey 3
    Bread containing linseed
	5
	2
	3
	3
	
	3
		5 (100)
		2 (100)
		3 (100)
		0 (0)
		
		3 (100)
			127 (91-178)
			58 (55-60)
			9.0 (5.4-12.6)
			-
			
			37 (30-49)
				Linseed and LSA – results consistent (but generally at the low end) with previous published material for linseed/flaxseed (Haque and Bradbury, 2002; Kobaisy et al, 1996; WHO, 2012).
				
				Bread containing linseed – 9 mg/kg (WHO, 2012). Results consistent with the stated linseed content of the breads and the HCN concentrations in linseed for each country.
					No standards established.

Food type	Number of samples analysed	Number positive for total HCN (%)	Total HCN in positive samples, mean (range) (mg HCN/kg)	Comparison of levels to the available scientific literature	Comparison to standards in the Code

Stone fruit products
Survey 1
    Canned apricots
    Prune or cherry juice
    Cherry brandy/liqueur
 	
Survey 3
    Apricot jam˜
    Apricot nectar
	
	4
	3
	3
	
	1
	4
		
		0 (0)
		0 (0)
		0 (0)
		
		0 (0)
		1 (25)
			
			-
			-
			-
			
			-
			6.5
				Canned stone fruit – up to 4 mg/kg (EFSA, 2004).
				
				Stone fruit juices (including cherry, plum, apricot and peach juices) and stone fruit liqueurs – 0.3-12 mg/L and <10 mg/L of total HCN, respectively (EFSA, 2004).
					No standards established other than levels for stone fruit juices and alcoholic beverages in Standard 1.4.1.
					
					Levels detected in one sample of apricot nectar do not comply with Standard 1.4.1, which includes an ML of 5 mg HCN/kg in stone fruit juices.

Lima bean and various seeds
Survey 1
    Lima beans, raw
    Butter/lima/mixed beans, canned
    Pumpkin seeds
    Sunflower seeds
	
	3
	6
	4
	2
		
		1 (33)*
		1 (17)#
		0 (0)
		0 (0)
			
			32
			6.8
			-
			-
				Beans – means range from 100 mg/kg for white American variety to 3000 mg/kg for black Puerto Rican variety (EFSA, 2004).
				
				Processing techniques such as soaking, autoclaving and toasting, and processing into bean paste can significantly reduce cyanide levels (Kawamura, 1993; Adeparusi, 2001).
				
				Sunflower seeds – cyanogenesis is not known to occur in the genera Helianthus L. (sunflowers) (Jones, 1997).
					No standards established.

Miscellaneous foods
Survey 1
    Passionfruit and passionfruit products
    Taro leaves and taro leaf products
    Spinach
    Vine leaves, canned
	
	5
	
	2
	2
	1
		
		2 (40)
		
		0 (0)
		0 (0)
		0 (0)
			
			5.7 (4.7-6.6)
			-
			-
			-
				Passionfruit (Passiflora edulis) juice – approximately 6-8 mg/kg total HCN (Chassagne et al, 1996).
				
				Giant taro leaves – 29 mg/kg (Haque and Bradbury, 2002).
					No standards established.

^ LSA - a mixture of linseed, sunflower seeds and almonds.
* Small green lima beans.
# Canned bean salad containing green lima beans.
˜ Multiple retail samples were analysed as one composite sample.

Appendix 4. Concentration data used in the dietary exposure assessment

Table A4.1. Concentration of total HCN in selected foods as used in the dietary exposure assessments derived from Australian and New Zealand analytical survey data

Food	Survey	Number of samples	Number of non-detects (% samples)	Limit of detection (LoD) (mg HCN/kg)	Mean concentration (mg HCN/kg)[#]		Maximum concentration (mg HCN/kg)[##]
					Lower bound (not detected = 0)	Upper bound (not detected = LOD)	
Almonds	1	7	4 (57)	4	3.6	5.9	12.4
Almond oil	1	1	1 (100)	4	0	4.0	**4.0
Apple juice	2 (EU method)	3	2 (67)	0.06	0.06	0.10	0.17
Apple sauce	1	3	1 (33)	4	2.6	3.9	4.1
Apple cider vinegar	1	1	1 (100)	4	0	4.0	**4.0
Apple puree, infant food	3 (EU method)	8	1 (13)	0.1	0.8	0.8	1.3
Apricots, canned	1	4	4 (100)	4	0	4.0	**4.0
Apricot jam˜	3	8	1 (100)	4	0	4.0	**4.0
Apricot kernels, with skin	3	18	0 (0)	NA	NA	NA	2820.0
Apricot kernels, without skin	3	10	0 (0)	NA	NA	NA	440.0
Apricot nectar^	3	4	3 (75)	5	1.6	5.4	**5.0
Bamboo shoots	1 (prepared only)	10	0 (0)	NA	13.4	13.4	44.0
Biscuits, amaretti˜	3	12	0 (0)	NA	34.0	34.0	34.0
Biscuits, almond fingers/apple fingers˜	3	13	0 (0)	NA	NA	NA	13.0
Bread containing linseed	1 & 3	6	0 (0)	NA	23.0	23.0	49.0
Butter beans*	1	6	5 (83)	3 (4)	1.1	4.1	6.8
Cassava roots, cooked	3 (sweet only)	8	0 (0)	NA	14.1	14.1	26.0
Cassava starch	1	4	4 (100)	4	0	4.0	**4.0

Food	Survey	Number of samples	Number of non-detects (% samples)	Limit of detection (LoD) (mg HCN/kg)	Mean concentration (mg HCN/kg)[#]		Maximum concentration (mg HCN/kg)[##]
					Lower bound (not detected = 0)	Upper bound (not detected = LOD)	
Cherry brandy	1	3	3 (100)	4	0	4.0	**4.0
Lima beans*	1	3	2 (67)	3 (4)	10.8	13.1	32.3
Linseeds	1	5	0 (0)	NA	127.0	127.0	178.0
Linseed oil	1	3	3 (100)	4	0	4.0	**4.0
LSA mix	1	2	0 (0)	NA	57.5	57.5	60.0
Marzipan	1	4	3 (75)	4	1.3	4.3	5.3
Passionfruit*	1	5	3 (60)	3 (4)	2.3	4.5	6.6
Prune juice	1	3	3 (100)	4	0	4.0	**4.0
Pumpkin seeds	1	4	4 (100)	4	0	4.0	**4.0
Spinach*	1	2	2 (100)	3 (4)	0	3.5	**4.0
Sunflower seeds	1	2	2 (100)	4	0	4.0	**4.0
Taro, in mixed foods	1	1	1 (100)	4	0	4.0	**4.0
Taro, leaves only	1	1	1 (100)	4	0	4.0	**4.0
Vine leaves, stuffed, canned	1	1	1 (100)	4	0	4.0	**4.0

* Indicates a food consisting of samples with different limits of detection. Number in brackets indicates the higher limit of detection.
** At the LoD.
[#] Mean concentrations used in the chronic dietary exposure assessment.
[##] Maximum concentrations used in the acute dietary exposure assessment.
˜ One composite analysis.
^ Sample that did not comply with the current ML permitted in stone fruit juices was excluded from the acute dietary exposure assessment.
NA = not used for the assessment.

Appendix 5. Total hydrocyanic acid concentrations for specific foods for the dietary exposure assessment

Apple juice and Infant apple puree

For apple juice and infant apple puree, results using the EU HPLC method were used as inputs into the dietary exposure estimates.

Three apple juice samples were analysed using this method (n=3) with two samples showing no detection (i.e. <0.06 mg HCN/kg) and one detection at 0.17 mg HCN/kg. Means were calculated for the LB and UB and used as inputs for the chronic dietary exposure estimates and the maximum of 0.17 mg HCN/kg was used as the input for the acute dietary exposure estimates.

For infant apple puree the following data was used: n=8; detections – 7; non-detections – 1 (<0.1 mg/kg). Means were calculated for the LB and UB and used as inputs for the chronic dietary exposure estimates and the maximum of 1.3 mg HCN/kg was used as the input for the acute dietary exposure estimates.

Apricot kernels

Raw apricot kernels were analysed in Survey 3. Only acute dietary exposure assessments were conducted for raw apricot kernels because there were no consumption data in any of the nutrition surveys to allow them to be included in the chronic dietary exposure assessment. Both kernels with and without skin were sampled. The two types had very different concentrations of total HCN (the mean for kernels with skin was 2,120 mg HCN/kg (n=18); the mean for kernels without skin was 190 mg HCN/kg (n=10)). Therefore, the two types were kept separate for the acute dietary exposure assessment.

Bamboo shoots

For bamboo shoots, samples in Survey 1 included canned or pickled varieties. In Survey 3, both frozen and fresh bamboo shoots were sampled, with the samples being split and half analysed raw and half boiled until tender and analysed. Only the results from Survey 1 (n=10) were used for the chronic and acute dietary exposure assessments. This is because fresh/frozen varieties of bamboo shoots, as sampled in Survey 3, proved difficult to locate as part of the sampling plan. From this, it was assumed that it is unlikely that they are commonly available in the Australian and New Zealand marketplace and that bamboo shoots are more likely to be consumed in their canned/pickled form.

Biscuits containing apricot kernels

Amaretti biscuits and almond fingers/apple fingers were each analysed as a composite sample. Amaretti biscuits were found to contain a higher HCN concentration than almond fingers/apple fingers. The foods mapped to both of these types of biscuits were the same i.e. *Sweet biscuits containing nuts or fruit and nuts, macaroons; commercial or homemade.* Therefore, for the chronic dietary exposure assessment, the HCN concentration for amaretti biscuits was used as a worst case scenario.

For the acute dietary exposure assessment, each biscuit type was assessed separately. The HCN concentration determined from the composite analysis of each type of biscuit was used in the calculations as there was no distribution of results to determine a maximum concentration.

Bread containing linseed

Bread containing linseed was analysed in both Survey 1 (n=3) and Survey 3 (n=3). All Survey 1 samples were collected in Zealand. All Survey 3 samples were collected in Australia. As the same analytical methodology was used for both surveys, it was valid for all six samples to be pooled for the purposes of the chronic and acute dietary exposure assessments.

There were differences in the range of HCN concentrations obtained for Australian and New Zealand samples, in that concentrations were found to be higher in Australian Survey 3 samples. Literature indicates that this may be due to higher HCN concentrations in Australian-sourced

linseed. However there was no information to indicate where bread manufacturers source their seeds from. Therefore, all data were pooled together.

Cassava roots
For cassava, only raw samples were analysed in Survey 1, however cassava roots are not consumed raw. In Survey 3, the nine frozen samples that were purchased were split into three groups and cooked using three different methods, based on typical cooking methods used in Australia and New Zealand (steaming, boiling and frying), then analysed individually in their cooked state. All of the data from Survey 3 for the cooked cassava (irrespective of cooking method) were pooled for use in the chronic and acute dietary exposure assessments. However, the one non-compliant bitter cassava was excluded from the data set.

Appendix 6. Concentration data for RTE cassava chips used in the dietary exposure assessment

Table A6.1. Summary of total HCN concentrations in RTE cassava chips collected since 2008 and used in the chronic dietary exposure assessment

Country	Number of samples	Number of non-detects (% samples)	Limit of detection (LoD) (mg/kg)	Concentration result	Mean concentration of total HCN		Maximum concentration of total HCN (mg/kg)*
					Lower bound (not detected = 0) (mg/kg)	Upper bound (not detected = LOD) (mg/kg)	
Australia	195	135 (69)	10	Actual survey data	20.7	27.6	268
				With 5% market share of RTE cassava chips applied	1.0	1.4	NA
New Zealand	165	93 (56)	9 and 10	Actual survey data	18.1	23.4	360
				With 5% market share of RTE cassava chips applied	0.9	1.2	NA

* Reported for information, not used in acute dietary exposure assessment as an acute assessment was not required for RTE cassava chips.

Appendix 7. Consumption data - additional information about specific foods

Apricot kernels
Raw apricot kernels were not consumed in any of the NNSs used by FSANZ for conducting dietary exposure assessments. Therefore, the chronic dietary exposure assessment did not include raw apricot kernels and the acute dietary exposure assessment was based on estimated consumption amounts from recommended serve sizes as provided by websites associated with the product and from what FSANZ had previously considered to be safe. An estimation of the maximum number of raw apricot kernels that could be consumed before exceeding the ARfD was also undertaken for this product, for both adults and children

Advice on the FSANZ website indicates that adults should not consume more than 4 kernels per day and this used as one consumption amount. Advice on the FSANZ website also indicates that children should not consume any apricot kernels (FSANZ, 2011) and so an equivalent consumption amount for children was not determined.

Advice on the internet from companies selling apricot kernels recommends higher consumption amounts. Recommended consumption amounts range from 1 to 2 kernels per 10 pounds (4.5 kg) of body weight (Nu-Gen Nutrition, 2013a; Apricot Seeds Australia, 2013); and 1 kernel per 5 kg body weight, 3 times per day (b17, 2013). For Australians 17 years and over, the mean body weight is 74 kg (see Table 11). A second consumption value was calculated for adults, based on 2 kernels per 4.5 kg body weight, for a body weight of 74 kg. On this basis, the consumption value was approximately 32 kernels per day.

The weight of one kernel was calculated to translate the per kernel consumption amount into a gram amount. Information on the internet indicated that a 1-pound (450 g) bag contains between 700-750 kernels (Nu-Gen Nutrition, 2013b; Rainrock Nutritionals, 2013; My Apricot Seeds, 2013). This equates to around 0.6 g per kernel. This would therefore equate to 19 grams per day for 32 kernels and 2.4 grams per day for 4 kernels.

Biscuits containing apricot kernels
Specific consumption data for amaretti biscuits or almond fingers/apple fingers was not recorded in any of the nutrition surveys. For the chronic dietary exposure assessment, consumption values for all nut-containing biscuits were included as the closest match to the food analysed. To obtain a realistic representation of population exposure, a 1% market share was used as a realistic percentage of all nut biscuits that amaretti and almond fingers/apple fingers represent. In deciding on this percentage, market share data from *Retail World* (Retail Media, 2012) was reviewed. Data indicated that out of the top eight sweet biscuits (excluding chocolate biscuits) the lowest had a volume share of 1.7%. It would seem realistic to assume that amaretti biscuits and almond fingers/apple fingers would be even lower than this figure. Therefore a rounded down market share value to the nearest whole percentage (i.e. 1%) was used in the chronic dietary exposure assessment. This is still likely to be an overestimate.

For the acute dietary exposure assessment, deriving a 97.5[th] percentile consumption value based on all nut containing biscuits was considered likely to overestimate consumption because, due to the bitter nature of biscuits containing apricot kernels, it could be assumed that they are not typically consumed in as large quantities as other sweet biscuits. Therefore, a consumption value equivalent to one serve, according to label information on each biscuit type, was used. One serve of amaretti biscuits was generally listed as being 20-30 g. Therefore, a 30 g amount was used as a worst case scenario. The typical serve size for almond fingers/apple fingers was 43 g. However, a slightly larger serve size of 47 g was used as the worst case scenario.

Bread containing linseed
In the 2002 and 2007 surveys, bread containing linseed was coded separately from other breads and, as such, the consumption data are specifically for bread containing linseed. There were no

consumers of linseed breads in the 1995 and 1997 nutrition surveys. Therefore, for adults, all grain and wholemeal breads were used as a proxy.

No market share information was available indicating what proportion of all grain and wholemeal breads is made up of linseed breads to apply to the chronic dietary exposure assessment for adults, to make it more representative of breads containing linseed. However, in the 2007 survey, the consumption of breads containing linseed as a proportion of all grain and wholemeal breads was estimated to be 3.4%. This estimate was based on the mean consumption amount for all respondents. It is likely that adult females consume more bread containing linseed than children. Therefore, a rounded value of 5% was used in all the chronic dietary exposure assessments for all surveys.

For the acute dietary exposure assessments, the 97.5[th] percentile consumption value for grain and wholemeal breads was used for each of the Australian and New Zealand adult populations. This was based on the assumption that the number of slices of bread (and therefore grams) that would be consumed would be the same whether the bread were wholemeal, grain or linseed containing.

NOURISH data for infants
Food consumption data for infants (under 12 months of age) for relevant foods was obtained from the NOURISH study conducted in Australia from 2009 (Daniels et al, 2009). This study intended to contain three specific survey periods. The aim of the survey was to sample infants at 4-7 months (Time 1, 'Baseline'), 13-16 months (Time 2) and 22-25 months (Time 3) to obtain one 24-hour recall and two 24-hour records for each respondent at each time period (*n*=9 days of data in total). However, the reported age of the respondents at each time period differed from the survey design. At Time 1, the age range based on the date of birth reported was 2-11 months.

As this study had an education intervention included, only the food consumption data from controls was obtained for use by FSANZ for risk assessment purposes. For this assessment, only the data from Time 1 were used (2-11 months). Despite the aims of the study, there were some respondents for which only one or two days of consumption data were collected. In addition, in some cases the three days of consumption data were not collected within the required 12-day period.

For any survey day, if the respondents' body weight was not recorded, then the record was not included in the dataset. Each of the (up to) three survey days for each respondent were treated in isolation and counted as a single consumption day. The consumption data for infant juices (or blended juices) or foods made primarily from juice (e.g. fruit gels) were subset, as were consumption data for infant apple puree, including home prepared cooked apples. All consumption data points used were on a 'per eating occasion' basis, not summed for a 24-hour period. This is because these data were still in a non-edited state at the time the dietary exposure assessment was undertaken and FSANZ had not yet compiled the data into per day consumption amounts.

Label information from commercial 'infant' juices indicates that they contain juice diluted with water (around 30% juice). Parents also dilute regular juices prior to consumption, as evidenced by recipes provided in the NOURISH dataset. Both of these situations were taken into account in the estimation of apple juice consumption.

Appendix 8. Food mapping

Table A8.1. Foods from the national nutrition surveys that are mapped to analysed foods for the purpose of the dietary exposure assessment

Food Analysed	Nutrition Survey Foods Included	Food Name for Reporting
Almonds, whole, flaked, ground and butter, almond jelly	Almonds whole, slivered, raw, roasted, almond meal, almond butter	Almonds
Almond oil	Almond oil	Almond oil
Apple cider vinegar	Vinegar	Apple cider vinegar
Apple juice	Apple juice (includes where apple juice used in a blended juice), fruit gels (for infant assessment only)	Apple juice
Apple sauce	Apple sauce	Apple sauce
Apple puree	Infant apples, canned; infant fruit not further specified	Apple puree, infant food
Apricot jam	Apricot jam, and/or stone fruit jam, or if specific jams not available, generic jams; sugar sweetened, reduced sugar and intense sweetened varieties	Apricot jam
Apricot nectar	Apricot nectar, juice containing apricot nectar, apricot juice, juice drinks containing apricot nectar or apricot juice	Apricot nectar
Canned apricots	Canned apricots in water, juice, and syrup	Apricots, canned
Bamboo shoots (canned, boiled in water, in brine, soured, pickled)	Bamboo shoots, cooked from fresh and canned	Bamboo shoots
Amaretti biscuits	Sweet biscuits containing nuts or fruit and nuts, macaroons; commercial or homemade	Biscuits (amaretti)
Almond fingers and apple fingers	Sweet biscuits containing nuts or fruit and nuts, macaroons; commercial or homemade	Biscuits (almond fingers/apple fingers)
Canned butter beans, canned four bean mix and mixed bean salad	Butter bean, fresh, cooked or canned	Butter beans
Lima beans, white and green (raw)	Lima bean, cooked, canned or dried	Lima beans
Ready-to-eat cassava chips (crisps)	Potato crisps, soy crisps, cheese and non-cheese flavoured extruded snacks, tapioca based snacks	Cassava chips, RTE
Cassava roots, cooked	Cassava (Manioc, Tapioca)	Cassava roots, cooked
Tapioca pearls, starch and flour; gluten free all purpose flour	Tapioca pearls, tapioca starch, sago	Cassava starch
Cherry brandy	Brandy and premixed drinks containing brandy	Cherry brandy
Infant apple puree	Infant apple puree, home stewed apples	Infant apple puree
Linseed, ground, powder and whole	Linseed (Flax-seed)	Linseed
Flaxseed oil, LSA oil	Linseed oil, linseed oil supplements	Linseed oil
Soy and linseed bread	Linseed containing bread (where no linseed containing breads were consumed, all grain and wholemeal breads were used as a proxy)	Bread containing linseed
LSA	LSA mix	LSA mix
Marzipan (including chocolate covered), almond flavoured icing	Marzipan, almond paste	Marzipan

Food Analysed	Nutrition Survey Foods Included	Food Name for Reporting
Passionfruit pulp, passionfruit drink, passionfruit in sweetened syrup, passionfruit sauce	Passionfruit pulp and juice	Passionfruit
Prune juice, red cherry juice	Prune juice	Prune juice
Pumpkin seeds	Pumpkin seed	Pumpkin seed
Spinach	Spinach, English spinach	Spinach
Sunflower seeds	Sunflower seed	Sunflower seed
Taro root and leaves cooked, baked, etc. in recipe foods e.g. with coconut, pawpaw and other ingredients	Taro in mixed dishes such as when cooked with coconut or paw paw	Taro in mixed foods
Taro leaves	Taro leaves	Taro leaves only
Canned stuffed vine leaves	Vine leaf stuffed with rice and/or lamb	Vine leaves, canned, stuffed

Appendix 9. Foods contributing to chronic dietary exposure

Table A9.1. Contributions of each food group to chronic dietary exposure to total HCN

Food Name	Contribution*(%)			
	Australia		New Zealand	
	2-16 years	17 years and above	5-14 years	15 years and above
Almonds	2	8	1	3
Almond oil	NC	NC	NC	NC
Apple juice	6	1	3	1
Apple puree, infant food	<1	<1	NC	<1
Apple sauce	NC	<1	<1	2
Apple cider vinegar	<1	<1	<1	<1
Apricots, canned	<1	<1	<1	<1
Apricot jam	<1	<1	<1	<1
Apricot nectar	1	2	NC	NC
Bamboo shoots	4	3	5	<1
Biscuits (amaretti and apple/apricot fingers)	<1	<1	<1	<1
Bread containing linseed	37	75	24	75
Butter beans	<1	<1	<1	NC
Cassava chips	15	5	22	5
Cassava roots, cooked	<1	<1	7	13
Cassava starch	<1	<1	<1	<1
Cherry brandy	<1	<1	<1	<1
Lima beans	<1	2	NC	<1
Linseeds	26	NC	32	NC
Linseed oil	<1	NC	NC	NC
LSA mix	<1	NC	NC	NC
Marzipan	<1	<1	NC	NC
Passionfruit	6	2	3	<1
Prune juice	<1	<1	NC	<1
Pumpkin seeds	<1	NC	<1	NC
Spinach	<1	<1	<1	<1
Sunflower seeds	<1	<1	<1	<1
Taro, in mixed foods	NC	NC	<1	<1
Taro, leaves only	NC	NC	<1	<1
Vine leaves, stuffed, canned	<1	<1	NC	NC

* Based on lower bound (LB) scenario where not detected results were assigned a zero concentration of HCN.
NC = not consumed.

Appendix 10. Estimated acute dietary exposures to HCN

Table A10.1. Estimated dietary exposures to total HCN for infants (2-11 months old) for selected apple based foods

Food	Number eating occasions	Consumption level used	Consumption (g/eating occasion)*	Estimated exposure to total HCN (µg/kg bw/eating occasion)
Apple juice [a]	78	P97.5	110	3
Apple puree, infant food[b]	687	P97.5	120	22

* For reference, a small can contains 120 grams, a juice popper 150 ml and a small juice bottle 250 mL.
[a] Survey 2, EU method.
[b] Survey 3, EU method.

Table A10.2. Estimated acute dietary exposures to total HCN for young Australian children aged 2-6 years

Food	Number of consumers	Consumption level used	Consumption amount (g/day)	Estimated acute exposure to total HCN (µg/kg bw/day)
Almonds	362	P97.5	12	8
Almond oil	NC	NC	NC	NC
Apple juice[a]	981	P97.5	524	5
Apple sauce	NC	NC	NC	NC
Apple cider vinegar	2406	P97.5	15	3
Apricots, canned	49	P97.5	108	23
Apricot jam	58	P97.5	8	2
Apricot nectar	8	P50	106	28
Bamboo shoots[b]	119	P97.5	7	15
Biscuits, amaretti[d]		1 serve	30	54
Biscuits, almond fingers/apple fingers[d]		1 serve	47	32
Bread containing linseed	1058	P97.5	160	413
Butter beans	13	P50	17	6
Cassava roots, cooked[c]	23	P50	5	7
Cassava starch	128	P97.5	36	8
Cherry brandy	33	P50	1	<1
Lima beans	1	P50	9	15
Linseeds	864	P97.5	5	45
Linseed oil	7	P50	13	3
LSA mix	1	P50	5	16
Marzipan	0		NC	
Passionfruit	336	P97.5	45	16
Prune juice	8	P50	157	33
Pumpkin seeds	21	P50	3	<1
Spinach	96	P97.5	183	39
Sunflower seeds	245	P97.5	12	2
Taro, in mixed foods	NC	NC	NC	NC
Taro, leaves only	NC	NC	NC	NC
Vine leaves, stuffed, canned	1	P50	240	51

NC = not consumed.
[a] Survey 2, EU method.
[b] Survey 1.
[c] Survey 3, sweet cassava only.
[d] Consumption level based on recommended serves/recommended maximum consumption levels.

Table A10.3. Estimated acute dietary exposures to total HCN for Australian children aged 2-16 years

Food	Number of consumers	Consumption level used	Consumption amount (g/day)	Estimated acute exposure to total HCN (µg/kg bw/day)
Almonds	1394	P97.5	18	6
Almond oil	NC	NC	NC	NC
Apple juice[a]	2514	P97.5	630	3
Apple sauce	NC	NC	NC	NC
Apple cider vinegar	7623	P97.5	19	2
Apricots, canned	167	P97.5	115	12
Apricot jam	171	P97.5	24	3
Apricot nectar	33	P50	44	6
Bamboo shoots[b]	270	P97.5	10	11
Biscuits, amaretti[d]		1 serve	30	27
Biscuits, almond fingers/apple fingers[d]		1 serve	47	16
Bread containing linseed	78	P97.5	216	279
Butter beans	30	P50	22	4
Cassava roots, cooked[c]	73	P97.5	92	63
Cassava starch	608	P97.5	39	4
Cherry brandy	100	P97.5	43	4
Lima beans	1	P50	9	8
Linseeds	2518	P97.5	5	23
Linseed oil	13	P50	12	1
LSA mix	1	P50	5	8
Marzipan	2	P50	10	1
Passionfruit	993	P97.5	55	10
Prune juice	10	P50	157	17
Pumpkin seeds	36	P50	3	<1
Spinach	379	P97.5	200	21
Sunflower seeds	817	P97.5	17	2
Taro, in mixed foods	NC	NC	NC	NC
Taro, leaves only	NC	NC	NC	NC
Vine leaves, stuffed, canned	9	P50	180	19

NC = not consumed.
[a] Survey 2, EU method.
[b] Survey 1.
[c] Survey 3, sweet cassava only.
[d] Consumption level based on recommended serves/recommended maximum consumption levels.

Table A10.4. Estimated acute dietary exposures to total HCN for Australians 17 years and over

Food	Number of consumers	Consumption level used	Consumption amount (g/day)	acute exposure to total HCN (μg/kg bw/day)
Almonds	2036	P97.5	36	6
Almond oil	NC	NC	NC	NC
Apple juice[a]	838	P97.5[a]	819	2
Apple sauce	35	P50	22	1
Apple cider vinegar	8468	P97.5	20	1
Apricots, canned	168	P97.5	231	12
Apricot jam	2100	P97.5	27	1
Apricot kernels, with skin[d]		32 kernels/d	19.0	724
		4 kernels/d	2.4	91
Apricot kernels, without skin[d]		32 kernels/d	19.0	113
		4 kernels/d	2.4	14
Apricot nectar	30	P50	223	15
Bamboo shoots[b]	58	P97.5	86	51
Biscuits, amaretti[d]		1 serve	30	14
Biscuits, almond fingers/apple fingers[d]		1 serve	47	8
Bread containing linseed	3887	P97.5	246	163
Butter beans	34	P50	35	3
Cassava roots, cooked[c]	1	P50	195	69
Cassava starch	21	P50	16	<1
Cherry brandy	117	P97.5	258	14
Lima beans	45	P97.5	135	59
Linseeds	NC	NC	NC	NC
Linseed oil	NC	NC	NC	NC
LSA mix	NC	NC	NC	NC
Marzipan	3	P50	48	3
Passionfruit	404	P97.5	54	5
Prune juice	6	P50	540	29
Pumpkin seeds	NC	NC	NC	NC
Spinach	183	P97.5	380	21
Sunflower seeds	744	P97.5	19	1
Taro, in mixed foods	NC	NC	NC	NC
Taro, leaves only	NC	NC	NC	NC
Vine leaves, stuffed, canned	9	P50	112	6

NC = not consumed.
[a] Survey 2, EU method.
[b] Survey 1.
[c] Survey 3, sweet cassava only.
[d] Consumption level based on recommended serves/recommended maximum consumption levels.

Table A10.5. Estimated acute dietary exposures to total HCN for young New Zealand children aged 5-6 years

Food	Number of consumers	Consumption level used	Consumption amount (g/day)	Estimated acute exposure to total HCN (µg/kg bw/day)
Almonds	21	P50	2	1
Almond oil	NC	NC	NC	NC
Apple juice[a]	70	P97.5	830	6
Apple sauce	3	P50	17	3
Apple cider vinegar	567	P97.5	11	2
Apricots, canned	21	P50	14	2
Apricot jam	NC	NC	NC	NC
Apricot nectar	18	P50	11	2
Bamboo shoots[b]	4	P50	10	19
Biscuits, amaretti[d]		1 serve	30	44
Biscuits, almond fingers/apple fingers[d]		1 serve	47	27
Bread containing linseed	157	P97.5	240	511
Butter beans	5	P50	4	1
Cassava roots, cooked[c]	1	P50	44	50
Cassava starch	113	P97.5	10	2
Cherry brandy	NC	NC	NC	NC
Lima beans	NC	NC	NC	NC
Linseeds	112	P97.5	4	28
Linseed oil	NC	NC	NC	NC
LSA mix	NC	NC	NC	NC
Marzipan	NC	NC	NC	NC
Passionfruit	49	P97.5	23	7
Prune juice	NC	NC	NC	NC
Pumpkin seeds	3	P50	8	1
Spinach	12	P50	9	2
Sunflower seeds	90	P97.5	8	1
Taro, in mixed foods	1	P50	68	12
Taro, leaves only	6	P50	72	13
Vine leaves, stuffed, canned	NC	NC	NC	NC

NC = not consumed.
[a] Survey 2, EU method.
[b] Survey 1.
[c] Survey 3, sweet cassava only.
[d] Consumption level based on recommended serves/recommended maximum consumption levels.

Table A10.6. Estimated acute dietary exposures to total HCN for New Zealand children aged 5-14 years

Food	Number of consumers	Consumption level used	Consumption amount (g/day)	Estimated acute exposure to total HCN (µg/kg bw/day)
Almonds	128	P97.5	21	6
Almond oil	NC	NC	NC	NC
Apple juice[a]	350	P97.5	741	3
Apple sauce	12	P50	17	2
Apple cider vinegar	2882	P97.5	13	1
Apricots, canned	75	P97.5	116	11
Apricot jam	2	P50	1	<1
Apricot nectar	39	P97.5	244	29
Bamboo shoots[b]	39	P97.5	33	35
Biscuits, amaretti[d]		1 serve	30	24
Biscuits, almond fingers/apple fingers[d]		1 serve	47	15
Bread containing linseed	784	P97.5	248	289
Butter beans	23	P50	4	<1
Cassava roots, cooked[c]	7	P50	47	29
Cassava starch	585	P97.5	10	<1
Cherry brandy	9	P50	5	<1
Lima beans	NC	NC	NC	NC
Linseeds	545	P97.5	3	14
Linseed oil	NC	NC	NC	NC
LSA mix	NC	NC	NC	NC
Marzipan	NC	NC	NC	NC
Passionfruit	250	P97.5	48	8
Prune juice	NC	NC	NC	NC
Pumpkin seeds	12	P50	3	<1
Spinach	67	P97.5	148	14
Sunflower seeds	479	P97.5	6	<1
Taro, in mixed foods	4	P50	68	7
Taro, leaves only	23	P50	72	7
Vine leaves, stuffed, canned	NC	NC	NC	NC

NC = not consumed.
[a] Survey 2, EU method.
[b] Survey 1.
[c] Survey 3, sweet cassava only.
[d] Consumption level based on recommended serves/recommended maximum consumption levels.

Table A10.7. Estimated acute dietary exposures to total HCN for New Zealanders 15 years and above

Food	Number of consumers	Consumption level used	Consumption amount (g/day)	Estimated acute exposure to total HCN (µg/kg bw/day)
Almonds	378	P97.5	30	5
Almond oil	NC	NC	NC	NC
Apple juice[a]	244	P97.5	855	2
Apple sauce	33	P50	33	2
Apple cider vinegar	3591	P97.5	18	1
Apricots, canned	141	P97.5	240	14
Apricot jam	31	P50	<1	<1
Apricot kernels, with skin[d]		32 kernels/d	19.0	755
		4 kernels/d	2.4	95
Apricot kernels, without skin[d]		32 kernels/d	19.0	118
		4 kernels/d	2.4	15
Apricot nectar	NC	NC	NC	NC
Bamboo shoots[b]	14	P50	4	2
Biscuits, amaretti[d]		1 serve	30	14
Biscuits, almond fingers/apple fingers[d]		1 serve	47	9
Bread containing linseed	1714	P97.5	288	199
Butter beans	NC	NC	NC	NC
Cassava roots, cooked[c]	9	P50	217	80
Cassava starch	13	P50	7	<1
Cherry brandy	39	P97.5	145	8
Lima beans	13	P50	6	3
Linseeds	NC	NC	NC	NC
Linseed oil	NC	NC	NC	NC
LSA mix	NC	NC	NC	NC
Marzipan	NC	NC	NC	NC
Passionfruit	153	P97.5	36.0	3
Prune juice	3	P50	216	12
Pumpkin seeds	NC	NC	NC	NC
Spinach	92	P97.5	218	12
Sunflower seeds	214	P97.5	16	<1
Taro, in mixed foods	5	P50	243	14
Taro, leaves only	7	P50	77	4
Vine leaves, stuffed, canned	NC	NC	NC	NC

NC = not consumed.
[a] Survey 2, EU method.
[b] Survey 1.
[c] Survey 3, sweet cassava only.
[d] Consumption level based on recommended serves/recommended maximum consumption levels.

Table A10.8. Summary of consumption levels used in acute dietary exposure assessments

Food	Consumption level used					
	Australia			New Zealand		
	2-6 years	2-16 years	17 years and above	5-6 years	5-14 years	15 years and above
Almonds	P97.5	P97.5	P97.5	P50	P97.5	P97.5
Almond oil	NC	NC	NC	NC	NC	NC
Apple juice[a]	P97.5	P97.5	P97.5	P97.5	P97.5	P97.5
Apple sauce	NC	NC	P50	P50	P50	P50
Apple cider vinegar	P97.5	P97.5	P97.5	P97.5	P97.5	P97.5
Apricots, canned	P97.5	P97.5	P97.5	P50	P97.5	P97.5
Apricot jam	P97.5	P97.5	P97.5	NC	P50	P50
Apricot nectar	P50	P50	P50	P50	P97.5	NC
Bamboo shoots[b]	P97.5	P97.5	P97.5	P50	P97.5	P50
Biscuits, amaretti[d]	1 serve	1 serve	1 serve	1 serve	1 serve	1 serve
Biscuits, almond fingers/apple fingers[d]	1 serve	1 serve	1 serve	1 serve	1 serve	1 serve
Bread containing linseed	P97.5	P97.5	P97.5	P97.5	P97.5	P97.5
Butter beans	P50	P50	P50	P50	P50	NC
Cassava roots, cooked[c]	P50	P97.5	P50	P50	P50	P50
Cassava starch	P97.5	P97.5	P50	P97.5	P97.5	P50
Cherry brandy	P50	P97.5	P97.5	NC	P50	P97.5
Lima beans	P50	P50	P97.5	NC	NC	P50
Linseeds	P97.5	P97.5	NC	P97.5	P97.5	NC
Linseed oil	P50	P50	NC	NC	NC	NC
LSA mix	P50	P50	NC	NC	NC	NC
Marzipan	NC	P50	P50	NC	NC	NC
Passionfruit	P97.5	P97.5	P97.5	P97.5	P97.5	P97.5
Prune juice	P50	P50	P50	NC	NC	P50
Pumpkin seeds	P50	P50	NC	P50	P50	NC
Spinach	P97.5	P97.5	P97.5	P50	P97.5	P97.5
Sunflower seeds	P97.5	P97.5	P97.5	P97.5	P97.5	P97.5
Taro, in mixed foods	NC	NC	NC	P50	P50	P50
Taro, leaves only	NC	NC	NC	P50	P50	P50
Vine leaves, stuffed, canned	P50	P50	P50	NC	NC	NC

NC = not consumed.
[a] Survey 2, EU method.
[b] Survey 1.
[c] Survey 3, sweet cassava only.
[d] Consumption level based on recommended serves/recommended maximum consumption levels.