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Short communication

Total cyanide content of cassava food products in Australia

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ABSTRACT

Cassava products obtained in two major Australian cities, Melbourne and Canberra, were analysed for total cyanide content using the picrate method. In Melbourne in 2010, ready to eat cassava chips were found to contain large amounts of cyanide with a mean value of 91 mg HCN equivalents/kg fresh weight = ppm. In Canberra, similar values were found over a six-year study with cassava chip samples, except for one sample that gave 7 ppm, which was obtained in 2011 after the introduction by Food Standards Australia and New Zealand of a 10 ppm maximum limit. In Melbourne, the highest value obtained was 262 ppm. A calculation based on this very high cyanide sample and using the lethal dose of cyanide for humans, shows that a child of 20 kg body weight would only need to eat 40–270 g of these chips to reach the lethal dose. Frozen cassava roots gave a mean value of 52 ppm total cyanide, which is also a cause for concern. In contrast, more highly processed foods contained < 1 ppm total cyanide.

1. Introduction

Cassava (*Manihot esculenta* Crantz, Euphorbiaceae) is the sixth most important food crop globally, in terms of annual production (FAOSTAT, 2010), and is a staple food for approximately 800 million people (FAO/IFAD, 2000; Lebot, 2009). This perennial root crop is grown in the tropics, including sub-Saharan Africa, Asia, the Pacific Islands, and Central and South America (Hillocks et al., 2002; Lebot, 2009; McKey et al., 2010; Burns et al., 2010). The starchy tuberous roots are the main food source, but the young leaves, which are high in protein, are also consumed, particularly in Africa (Achidi et al., 2005; Lebot, 2009; Montagnac et al., 2009a).

Cassava contains two cyanogenic glucosides, linamarin and a small amount of lotaustralin, which are catalytically hydrolysed to release toxic hydrogen cyanide (HCN) when the plant tissue is crushed (Conn, 1981; Balagopalan et al., 1988; McMahon et al., 1995). Cyanide inhibits cellular respiration of all aerobic organisms by blocking mitochondrial electron transport and preventing oxygen uptake (Solomonson, 1981). High exposure to cyanide in humans causes nausea, vomiting, diarrhoea, dizziness, weakness and sometimes death (Akintonwa et al., 1994; Cliff and Coutinho, 1995; Jones, 1998; Merck, 2008). Medical conditions caused by degeneration of the nervous system, konzo and tropical ataxic neuropathy (TAN), affect people who consume a monotonous diet of bitter (high cyanide) cassava (Cliff et al., 1985; Tylleskar et al., 1992; Osuntokun, 1994; Cliff and Coutinho, 1995; McKey et al., 2010; Burns et al., 2010). The low content of protein in cassava roots and the deficiency of sulphur amino acids in this protein can affect the health of regular consumers and may also be a risk factor for konzo (Nhassico et al., 2008; McKey et al., 2010).

The range of total cyanide contents of different varieties of cassava is 1–1550 mg HCN equivalents/kg fresh material = ppm, in the root parenchyma and 900–2000 ppm in the root cortex (peel) (Mlingi and Bainbridge, 1994; Nambisan, 1994; Nambisan and Sundaresan, 1994; Cardoso et al., 2005). Cassava leaves contain 20–1860 ppm of total cyanide (Lancaster and Brooks, 1983; Bradbury and Denton, 2011). The World Health Organisation (WHO) set a safe limit of 10 ppm total cyanide for cassava flour (FAO/WHO, 1995). This maximum limit of 10 ppm has been adopted in Australia for cassava chips by Food Standards

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Australia and New Zealand (FSANZ, 2009), while 40 ppm is the limit in Indonesia (Djazuli and Bradbury, 1999). Internationally, the Codex Standard for 'sweet cassava' (those varieties with low levels of cyanogens) is 50 ppm (fresh weight basis, FAO/WHO, 2005), but many countries have yet to formally adopt recommended limits (Kolind-Hansen and Brimer, 2009).

The aim of this study was to determine the amounts of cyanogens present in cassava products in Melbourne and Canberra, Australia, and the possibility of deleterious effects as a result of consumption of high cyanide products.

2. Materials and methods

2.1. Materials

Products containing cassava as the primary or secondary ingredient were obtained from supermarkets and specialist grocery stores in Melbourne in February 2010. These products included flour, starch (in the form of powder and pearls), chips (i.e. crisps), and frozen roots (peeled and chopped into large pieces or grated, see Table 1). Three samples of each product were tested ($n = 3$), except for the chips from India where only 2 samples were available. Cassava chips were obtained over a period of six years in Canberra and analysed for total cyanide in each year. The survey was limited in scope, reflecting the relatively recent appearance of cassava products other than tapioca flour in Australia. Chips and biscuits were crushed in a mortar and pestle and the powder was used as is. Frozen roots were sampled by cutting a transverse section (approx. 1–2 mm thick) from the middle of the root piece and then cutting a triangular wedge (following Bradbury et al., 1999), blotted dry with paper towel, weighed and used in the assays. Samples of grated frozen roots were also blotted dry with paper towel before testing. Data were analysed using MSExcel2007 and Minitab 16® using one-way ANOVA, and pair-wise comparison was made *post hoc* using Tukey *t*-tests.

2.2. Methods

Duplicate or triplicate 100 mg samples of cassava products were added to a small plastic bottle, a buffer/enzyme paper was added, followed by 1 mL of 1 M pH 6 phosphate buffer, a picrate paper and a screw cap lid. The bottles were allowed to stand overnight at 30 °C, the picrate papers were removed from the plastic support and 5.0 mL of water added to elute the colour. The absorbance was measured in a spectrophotometer at 510 nm and the total cyanide content in mg HCN equivalents/ kg fresh weight = ppm calculated by multiplying the absorbance by 396 (Egan et al., 1998; Bradbury et al., 1999). This gives an accurate total cyanide analysis down to a minimum of 1 ppm total cyanide (Haque and Bradbury, 2002; Bradbury, 2009). The cyanide present is primarily linamarin (Jørgensen et al., 2005). The assay used here measures both linamarin and acetone cyanohydrin, but the concentration of the latter is extremely small compared to linamarin. Any HCN released from acetone cyanohydrin is of relevance to human health, as it breaks down completely in the alkaline condition in the gut to give CN (Bradbury, 2009). The amount of lotaustralin and free cyanide (HCN and CN) is negligible (Jørgensen et al., 2005). Food was tested in the form that it is normally consumed, with cyanide expressed on a fresh weight (or 'as consumed') basis.

Table 1
Total cyanide content (mg HCN equivalents/kg fresh wt = ppm) of cassava products in Melbourne (2010).

Product	Origin	Total cyanide in ppm[a]
Vegetable chips	Australian and imported ingredients	26 (6)
Tapioca crisps (BBQ)	Singapore	42 (8)
Cassava chips	India	262 (34)
Frozen cassava roots, peeled, large pieces	Vietnam	52 (7.5)
Frozen cassava, peeled, grated	Fiji and Vietnam	12 (5.6)
Tapioca flour, starch and pearls	Thailand	<1
Soup powder and dipping biscuits (gluten free)	Australian and imported ingredients	<1
Shrimp meat chip	Korea	<1

[a] Standard deviation in brackets.

3. Results and discussion

The results of the analyses of cassava products obtained in Melbourne are shown in Table 1. Cassava chips contained by far the largest amount of cyanide, with the highest value of 262 ppm from one product and an overall mean of all cassava chip products of 91 (±106 SD) ppm. Whole and grated frozen cassava roots contained, on average, 52 ppm and 12 ppm cyanide on a fresh-weight basis (equivalent to approximately 69 and 15 on a dry-weight basis). Products that contained tapioca flour and starch and other specialised products (Table 1) were essentially cyanide free, which supports other work that most cyanogens are removed during starch production from cassava (Montagnac et al., 2009b)

The total cyanide content of cassava chips obtained at food outlets in Canberra over a period of six years are given in Table 2. The high values obtained from 2005 to 2008 are similar to those obtained in Melbourne in February 2010 (Table 1), after the introduction by FSANZ of a 10 ppm safe limit in 2009. However, we note a very significant reduction in total cyanide content of a recent sample of cassava chips to 7 ppm, which is below the safe limit, and shows the importance of the development of enforceable standards (Kolind-Hansen and Brimer, 2009). Apart from this sample, the toxicity of all samples of cassava chips and frozen cassava roots was much greater than the FSANZ limit of 10 ppm. Frozen roots are usually boiled, baked or fried before consumption, which can reduce the concentration of total cyanide in these products by 10–75% (Nambisan and Sundaresan, 1985; Montagnac et al., 2009b), but may not reduce the concentration below the safe limit. Consumption of such cyanide-containing products, therefore, poses a health risk, especially to consumers who are unaware of the need for detoxification of cassava.

The toxic effect of cyanide on humans depends on body size, health status, the dose of cyanide ingested and the time duration over which it is ingested. The acute lethal dose of hydrogen cyanide for humans is 0.5–3.5 mg kg^{-1} body weight (Halstrøm and Møller, 1945). The average total cyanide concentration of

Table 2
Total cyanide content in ppm of cassava chips (i.e. crisps) and crackers purchased in Canberra over six years.

Product	Date of purchase (month/year)	Total cyanide in ppm[a]
Vegetable chips	10/05	68 (20)
Vegetable chips	10/06	83 (5)
Vegetable chips	10/07	84 (1)
Vegetable crackers	10/08	51 (2)
Vegetable chips	2/11	7 (4)

[a] Standard deviation in brackets.

Table 3
Minimum lethal doses of cyanide[a] and amount of cassava product containing[b] 50 ppm and 262 ppm cyanide required to reach the lethal doses.

Body weight (kg)	Lethal range of cyanide (mg)	Lethal amount of cassava (kg) containing	
		50 ppm	262 ppm
10	5–35	0.1–0.7	0.02–0.13
20	10–70	0.2–1.4	0.04–0.27
40	20–140	0.4–2.8	0.08–0.54
60	30–210	0.6–4.2	0.12–0.81
80	40–280	0.8–5.6	0.15–1.08
100	50––350	1.0–7.0	0.19–1.35

[a] From Halstrøm and Møller (1945) and Jones (1998).
[b] Cassava with 50 ppm cyanide is classified as 'sweet' by the International Codex Standard. 262 ppm is the maximum concentration detected in any product in this study.

262 ppm found in one brand of cassava chips is alarming. A child of 20 kg body weight would only need to eat 40–270 g of these very high cyanide chips to reach the lethal dose (Table 3). Since each bag of these chips was 200 g net weight, it would be possible for a child to ingest a lethal dose of cyanide from one bag of these chips. An adult of 80 kg could eat 150–1000 g of the same chips before reaching the lethal limit. These calculations do not take into account the general health and nutritional status of the consumer, which may alter the effective lethal limit.

Cassava products are increasingly being imported into countries that do not have experience in growing and processing cassava, and thus lack knowledge about the health risks associated with consumption of these food products. Based on the results presented here it is strongly advisable that the maximum 10 ppm safe level for total cyanide in cassava products introduced by FSANZ (FSANZ, 2009) be monitored to ensure that cassava chips will be safe, as shown by the most recent result given in Table 2, and that frozen cassava root parenchyma will also be safe in Australia.

4. Conclusion

This survey revealed a wide range of cyanide concentrations in commonly available cassava-based products in Australia. As the negative impacts of excess cyanide consumption are well known, it is clear that careful regulation of the importation of cassava products may be necessary to monitor and control the amount of total cyanogens in ready-to-eat food products.

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References

Achidi, A.U., Ajayi, O.A., Bokanga, M., Maziya-Dixon, B., 2005. The use of cassava leaves as food in Africa. Ecology of Food and Nutrition 44, 423–435.
Akintonwa, A., Tunwashe, O., Onifade, A., 1994. Fatal and non-fatal acute poisoning attributed to cassava-based meal. Acta Horticulturae 375, 285–288.
Balagopalan, C., Padmaja, G., Nanda, S.K., Moorthy, S.N., 1988. Cassava nutrition and toxicity. In: Balagopalan, C. (Ed.), Cassava in Food, Feed and Industry. CRC Press, Florida, USA, pp. 13–36.
Bradbury, J.H., 2009. Development of a sensitive picrate method to determine total cyanide and acetone cyanohydrin contents of gari from cassava. Food Chemistry 113, 1329–1333.
Bradbury, J.H., Denton, I.C., 2011. Mild methods of processing cassava leaves to remove cyanogens and conserve key nutrients. Food Chemistry 127, 1755–1759.
Bradbury, M.G., Egan, S.V., Bradbury, J.H., 1999. Picrate paper kits for determination of total cyanogens in cassava roots and all forms of cyanogens in cassava products. Journal of the Science of Food and Agriculture 79, 593–601.
Burns, A., Gleadow, R.M., Cavagnaro, T.R., 2010. Cassava: the drought, war and famine crop in a changing environment. Sustainability 2, 3572–3607.
Cardoso, A.P., Mirione, E., Ernesto, M., Massaza, F., Cliff, J., Haque, M.R., Bradbury, J.H., 2005. Processing of cassava roots to remove cyanogens. Journal of Food Composition and Analysis 18, 451–460.
Cliff, J., Coutinho, J., 1995. Acute intoxication from newly-introduced cassava during drought in Mozambique. Tropical Doctor 25, 193–1193.
Cliff, J., Martensson, J., Lundqvist, P., Rosling, H., Sorbo, B., 1985. Association of high cyanide and low sulfur intake in cassava-induced spastic paraparesis. Lancet 326, 1211–1213.
Conn, E.E., 1981. Biosynthesis of cyanogenic glycosides. In: Vennesland, V., Conn, E.E., Knowles, C.J., Westly, J., Wissing, F. (Eds.), Cyanide in Biology. Academic Press, New York, NY, USA, pp. 1–10.
Djazuli, M., Bradbury, J.H., 1999. Cyanogen content of cassava roots and flour in Indonesia. Food Chemistry 65, 523–525.
Egan, S.V., Yeoh, H.H., Bradbury, J.H., 1998. Simple picrate paper kit for determination of the cyanogenic potential of cassava flour. Journal of the Science of Food and Agriculture 76, 39–48.
FAO/IFAD, 2000. The World Cassava Economy: Facts, Trends and Outlooks. Food and Agriculture Organization of the United Nations and International Fund for Agricultural Development, Rome.
FAO/WHO, 1995. Codex Standard for Edible Cassava Flour. Codex Standard 176-1989. Food and Agriculture Organisation and World Health Organisation of the United Nations, Rome, Italy.
FAO/WHO, 2005. Codex Standard for Sweet Cassava. Codex Standard 238-2003. Food and Agriculture Organisation and World Health Organisation of the United Nations, Rome.
FAOSTAT, 2010. Food and agricultural commodities production. Food and Agriculture Organization of The United Nations Statistics Database Retrieved 1 April, 2010 from: www.faostat.fao.org.
FSANZ, 2009. Proposal P1002 Hydrocyanic Acid in Ready-to-Eat Cassava Chips, First Review Report. Food Standards Australia New Zealand.
Halstrøm, F., Møller, K.D., 1945. The content of cyanide in human organs from cases of poisoning with cyanide taken by mouth. With a contribution to the toxicology of cyanides. Acta Pharmacalogica et Toxicologica 1, 18–28.
Haque, M.R., Bradbury, J.H., 2002. Total cyanide determination of plants and foods using the picrate and acid hydrolysis methods. Food Chemistry 77, 107–114.
Hillocks, R.J., Thresh, J.M., Bellotti, A.C. (Eds.), 2002. Cassava: Biology, Production and Utilization. CABI Publishing, Oxon, UK.
Jørgensen, K., Bak, S., Busk, P.K., Sørensen, C., Olsen, C.E., Puonti-Kaerlas, J., Møller, B.L., 2005. Cassava (*Manihot esculenta* Cranz.) plants with a depleted content of cyanogenic glucosides in leaves and tubers. Distribution of cyanogenic glucosides, their site of synthesis and transport and blockage of the biosynthesis by RNAi technology. Plant Physiology 139, 363–374.
Jones, D.A., 1998. Why are so many food plants cyanogenic? Phytochemistry 47, 155–162.
Kolind-Hansen, L., Brimer, L., 2009. The retail market for fresh cassava root tubers in the European Union (EU): the case of Copenhagen, Denmark – a chemical food safety issue? Journal of the Science of Food and Agriculture 90, 252–256.
Lancaster, P.A., Brooks, J.E., 1983. Cassava leaves as human food. Economic Botany 37, 331–348.
Lebot, V., 2009. Tropical Root and Tuber Crops: Cassava, Sweet Potato, Yams and Aroids. CABI, Wallingford, UK.
McKey, D., Cavagnaro, T.R., Cliff, J., Gleadow, R.M., 2010. Chemical ecology in coupled human and natural systems: people, manioc, multitrophic interactions and global change. Chemoecology 20, 109–133.
McMahon, J.M., White, W.L.B., Sayre, R.T., 1995. Cyanogenesis in cassava (*Manihot esculenta* Crantz). Journal of Experimental Botany 46, 731–741.
Merck, 2008. Cyanide Poisoning: Introduction. The Merck Veterinary Manual. Retrieved 4 November, 2010 from: http://www.merckvetmanual.com/mvm/index.jsp?cfile=htm/bc/210800.htm&word=cyanide%2cpoisoning.
Mlingi, N.L.V., Bainbridge, Z., 1994. Reduction of cyanogen levels during sun-drying of cassava in Tanzania. Acta Horticulturae 375, 233–239.
Montagnac, J.A., Davis, C.R., Tanumihardjo, S.A., 2009a. Nutritional value of cassava for use as a staple food and recent advances for improvement. Comprehensive Reviews in Food Science and Food Safety 8, 181–194.
Montagnac, J.A., Davis, C.R., Tanumihardjo, S.A., 2009b. Processing techniques to reduce toxicity and antinutrients of cassava for use as a staple food. Comprehensive Reviews in Food Science and Food Safety 8, 17–27.
Nambisan, B., 1994. Evaluation of the effect of various processing methods on cyanogen content reduction in cassava. Acta Horticulturae 375, 193–201.
Nambisan, B., Sundaresan, S., 1985. Effect of processing on the cyanoglucoside content of cassava. Journal of the Science of Food and Agriculture 36, 1197–1203.

Nambisan, B., Sundaresan, S., 1994. Distribution of linamarin and its metabolizing enzymes in cassava tissues. Journal of the Science of Food and Agriculture 66, 503–507.

Nhassico, D., Muquingue, H., Cliff, J., Cumbana, A., Bradbury, J.H., 2008. Rising African cassava production, diseases due to high cyanide intake and control measures. Journal of the Science of Food and Agriculture 88, 2043–2049.

Osuntokun, B.O., 1994. Chronic cyanide intoxication of dietary origin and a degenerative neuropathy in Nigerians. Acta Horticulturae 375, 311–321.

Solomonson, L.P., 1981. Cyanide as a metabolic inhibitor. In: Vennesland, B., Conn, E.E., Knowles, C.J., Westley, J., Wissing, F. (Eds.), Cyanide in Biology. Academic Press, London, UK, pp. 11–28.

Tylleskar, T., Banea, M., Bikangi, N., Cooke, R.D., Poulter, N.H., Rosling, H., 1992. Cassava cyanogens and konzo, an upper motoneuron disease found in Africa. Lancet 339, 208–211.