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Analytical Methods

Flow injection analysis of nitrate-N determination in root vegetables: Study of the effects of cooking

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ABSTRACT

Vegetables are the major vehicles for the entry of nitrate into the human system. Ever-increasing concern over nitrate toxicity has directed a number of countries to lay down maximum allowable threshold concentrations with regards to nitrate-N in vegetables. Fiji is an independent island nation, located in the southern Pacific Ocean, has a tropical oceanic climate and hence expected to have high nitrate-N levels in vegetables. Thus, the present study was devoted to establish a flow injection analysis (FIA) technique for nitrate-N determination in Fiji's commonly consumed fresh and cooked root vegetables such as potato (Solanum tuberosum), dalo (Colocasia esculenta), sweet potato (Ipomoea batatas) and carrot (Daucus carota L.). Activated carbon extraction technique was applied to extract nitrate-N. FIA with colorimetric detection technique having linear dynamic range of determination $1.0-20.0 \text{ mgL}^{-1}$ and detection limit of 0.042 mgL⁻¹ (0.34 mg kg⁻¹), using sulphanilamide and N-(1-naphthyl)ethylenediamine dihydrochloride as colour reagents, was used to determine nitrate-N contents in selected fresh and cooked root vegetables. The samples throughput was $38 h^{-1}$. The effects of various cooking (boiling, baking and frying) methods on nitrate-N contents in root vegetables have also been studied. The study shows that the nitrate content of fresh root vegetables ranges from 53.76–258.00 mg kg⁻¹ whereas boiling reduces nitrate content by 23.30-42.62%. The frying in soya bean oil elevates nitrate contents from 204.53-299.12% but after baking nitrate contents remains almost constant with slight increasing trend from 2.80-8.43%. A comparison of the nitrate obtained by standardised method and the nitrate contents in vegetables of other countries are also presented.

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1. Introduction

Nitrogen is an important nutrient for the existence of plants and animals as it is required in many crucial constituents, such as proteins, DNA, RNA, vitamins, hormones and enzymes. Plants absorb nitrogen from the environment in the form of simple nitrogenous compounds such as nitrates and ammonia whereas humans get the bulk of their nitrogen (as nitrates) from foods (meat and vegetables) and drinking water. The nitrates present in plants and drinking water can be termed as "naturally occurring" although this is not fully true as the nitrate content is dependent on the use of fertilizers and the conditions under which they are grown. harvested and stored (Walker, 1975). Nitrates form part of the essential chemistry of soils and plants. Thus vegetables plant root are able to absorb nitrate directly from the soil. Vegetables play an important role in human nutrition since they are an outstanding resource for vitamins, minerals and biologically active compounds (Kmiecik et al., 2004). Consequently vegetables are high-value crops and provide a consistent income for vegetable farmers in Fiji. Since nitrogen plays a key role in plant growth, the most readily available agricultural fertilizers contain nitrate. Nitrate content of vegetables may range from 1 to 10000 mg kg⁻¹ (Hill, 1996; Ximenes et al., 2000). The various reasons for this wide range are excessive use of fertilizer, crop variety, types of N-fertilizers, light and temperature conditions, lack of water, etc. (Corré & Breimer, 1979; Santamaria et al., 1999, 2001; Santamaria, 2006). A combination of these factors account for different nitrate values reported for vegetables in different countries. The complexity with regards to nutritional exploitation of vegetables is the presence of nitrate (nitrite) as antinutritional and toxic in nature.

Concern about the high content of nitrates in certain foodstuffs such as baby foods and drinking water has long been recognised as it causes methemoglobinemia due to its rapid conversion to nitrite (Ezeagu, 1996; Gangolli et al., 1994; Tannenbaum et al., 1978; Usher & Telling, 1975). Thus nitrate is seen as an undesirable component of meats, vegetables and drinking water because of its association with infantile methaemoglobinaemia and cancer. Approximately 5% of all dietary nitrates are reduced to nitrites in saliva and the gastrointestinal tract (Hill, 1996; Santamaria, 2006). Nitrites being highly unstable can be metabolised within the digestive tract to N-nitroso compounds (MAFF UK, 2001).





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N-nitroso compounds comprise of nitrosamines and nitrosamides (Ahn et al., 2002). Nitrosamines produced through acid catalysis of nitrite with certain nitrogen compounds are carcinogenic and volatile (Ezeagu, 1996; Perez-Olmos et al., 1997). Greater nitrite content thus could increase the likelihood of endogenous nitrosation reactions, which in turn may lead to a greater risk of cancer causation.

Nitrate occurrence in vegetables, foodstuffs and water is documented in literature from olden days till date where enormous variation in nitrate concentration exists between different food items and vegetables (Prasad & Chetty, 2008; Usher & Telling, 1975). The great variation in nitrate levels also exists between different samples of the same vegetables (Prasad & Chetty, 2008). Many analytical techniques like spectrophotometry, potentiometry, ion chromatography, polarography, capillary electrophoresis as well as high-performance liquid chromatography have been used for quantification of nitrate in vegetable samples (Prasad & Chetty, 2008). However, FIA with colorimetric analysis of nitrate/nitrite in different matrices has proved to be an inexpensive technique (Andrade et al., 2003; Burakham et al., 2004; Melchert & Rocha, 2005; Monser et al., 2002; Prasad & Chetty, 2008). Very recently we have reported the optimisation of a FIA method with colorimetric detection and applied it for the determination of the nitrate contents in commonly consumed fresh, cooked and frozen leafy vegetables marketed in Fiji (Prasad & Chetty, 2008). In continuation of our study the present report is devoted to assess the extent of nitrate contamination in commonly consumed root vegetables in Fiji such as potato (Solanum tuberosum), dalo (Colocasia esculenta), sweet potato (Ipomoea batatas) and carrot (Daucus carota L.).

2. Experimental

The details about the samples and reagents preparations, instrumentation, extraction and quantification procedures are reported in our previous communication (Prasad & Chetty, 2008). Hence, a brief about the samples and reagents preparations, instrumentation, extraction, etc. are presented in the following sections.

2.1. Study site, samples for testing and samples preparation

Suva is the capital city of Fiji. It has the highest urban population in the country. The Suva municipal market is located next to the bus stand, which makes it an ideal and convenient place for consumers to purchase fresh vegetable produce. Thus, Suva municipal market was chosen as the sampling site in Fiji. The four more popular and commonly consumed root vegetables available in the Suva market were identified and used in this study.

Vegetables sampling were done during the months of 2006. The six different lots of each variety of root vegetables (potato, dalo, sweet potato and carrot) were collected from Suva municipal market from the six different vendors at random on different days. In the laboratory, each vegetable sample was rinsed with water to remove any soil or wind borne particles on the vegetables. The fresh root vegetable samples were meticulously scrubbed and dried. They were peeled and sliced into little pieces with the ends and blemished pieces discarded before being freeze-dried.

For the study of the effects of cooking on nitrate-N concentration sub-samples were taken from the same lot of fresh root vegetables. For *boiling* all peeled root vegetable samples were evenly sliced before being placed (25.0 g) into a small aluminium pot of cold water. This was then brought to the boil and simmered for 15 to 20 min until cooked. The boiled samples were then cooled, drained on absorbent paper and then freeze-dried. For *baking* after being scrubbed and dried, all samples were pricked over their entire surface. The samples (25.0 g) were weighed into baking cups and placed in a preheated Omega mini kitchen oven immediately. The samples were baked at approximately 180 °C (20–25 min) since this temperature is common in domestic cooking. All samples were cooled before being freeze-dried. For *frying* all samples were sliced into chips and placed into a non-stick fry pan. The samples were fried in soya bean oil for 12 min (or until cooked). After frying all samples were cooled and drained on absorbent paper before being sliced into tiny cubes (<0.5 × 0.5 cm) prior to freeze-drying. After cooking all freezedried sub-samples were labelled and frozen (-20 °C) in acid pre-washed (snap-lock) plastic bags.

2.2. Reagents and standard solution preparation

All the reagents used in this study were of analytical-reagent grade and the distilled de-ionised water (DDW) was used for nitrate extraction, standard nitrate and other solutions preparation. The 0.004 gmL⁻¹ stock solution of nitrate was prepared by dissolving 0.25 g of KNO₃ (Biolab, Australia) in 250 mL water. The calibration standard solutions of 1.0, 4.0, 8.0, 12.0, 16.0 and 20.0 mgL⁻¹ nitrate-N were prepared from the stock solution. The colour reagents mixed solution of sulphanilamide (SA) (Ajax Finechem, Australia) and N-(1-naphthyl)ethylenediamine dihydrochloride (NED) (BDH, England) was prepared as reported (Prasad & Chetty, 2008) and stored in a dark amber colour bottle and discarded when pink. Ammonium chloride buffer was prepared from ammonium chloride (Asia Pacific Specialty Chemicals Ltd., Australia), and disodium ethylenediaminetetraacetic acid dihydrate (Sigma-Aldrich) whilst the pH adjusted to 8.5 with 10% NaOH (w/v) (Prasad & Chetty, 2008). The standard buffer solutions (Aldrich) were used to standardise the pH metre.

2.3. Instrumentation and calibration curves

The glassware used in this study were scrupulously cleaned by soaking in 10% HCl for 24 h and rinsed several times with DDW prior to use. Hanna Instruments Microprocessor 8521 pH metre was used for all pH measurements. For extraction of nitrate shaking of all samples was carried out on a Stuart Scientific shaker model SF1. A Lachat QuickChem 8000 flow injection analyser, the schematic diagram of which is shown in Fig. 1, was used for the determination of nitrate. The FIA manifold is equipped with peristaltic pumps (P), an injection valve (V), a Cu–Cd reduction column (C) and a two state switching valve (SV). The teflon tubes R₁, R₂ and R₃ (1.07 mm ID) were used for the carrier water, buffer and reagent flow respectively whilst teflon tubes used for the manifold connections, the delay coil (DC, 70 cm) and the reaction coil (RC, 70 cm)

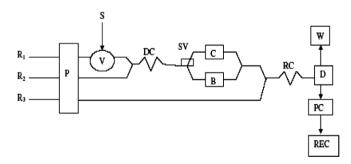


Fig. 1. Schematic diagram of the flow injection manifold used for determination of nitrate: R_1 , carrier stream (distilled deionised water); R_2 , buffer stream (NH₄Cl/EDTA); R_3 , colour reagent stream (SA/NED); P, peristaltic pump; V, injection valve; S, sample; DC, delay coil; SV, two state switching valve; C, copperised cadmium column; RC, reaction coil; D, detector; W, waste tank; PC, personal computer; REC, recorder.

were of 0.8 mm ID. The FIA is equipped with an auto-sampler (sample volume 200 μ L) with a 10 mm path length flow cell, a 60-position sample rack and a colorimetric detector (D) with UV filter of 520 nm.

The FIA instrument was operated at 25 °C at a flow rate of 2.6 mLmin⁻¹ in all three channels R₁, R₂ and R₃. The proposed FIA technique was applied for determination of nitrate-N in root vegetables. The typical FIA peaks profile for the calibration standards of 1.0–20.0 mgL⁻¹ nitrate were obtained under optimum conditions as reported by us (Prasad & Chetty, 2008). The calibration graphs were obtained for each run by injecting six different concentrations of nitrate (in duplicate) in the range 1.0-20.0 mgL⁻¹ and plotting peak area (volts.sec) against concentration (mgL^{-1}) . The calibration equations of the measured peak areas versus nitrate concentrations (mgL⁻¹) were used to calculate nitrate in root vegetable samples in mgL^{-1} and finally reported as mgkg⁻¹. Linearity of calibration curve in terms of correlation coefficient and precision in terms of relative standard deviation (RSD) for peak areas were 0.9974 and 2.22% in the range of $1-20 \text{ mgL}^{-1}$ of nitrate.

2.4. Sample extraction

The activated carbon and alkaline extraction techniques were assessed using four fresh and spiked vegetable samples (Chinese cabbage, celery, lettuce and tomato) where average recoveries of nitrate-N were 100.52 \pm 5.19% and 103.02 \pm 2.51%, respectively (Prasad & Chetty, 2008). The nitrate-N obtained from the two extraction techniques in fresh vegetables in mg kg⁻¹ is compared in Fig. 2 which clearly shows that both the extraction techniques gave almost similar results in terms of nitrate recovery as well as nitrate extracted from the fresh vegetables. Activated carbon extraction being cost effective was adopted for the nitrate determinations in selected root vegetables.

2.5. Quality control, method precision/validation and method detection limit

The quality control methods employed in this study, to guarantee the output of quality data, were method of standard addition (analysis of nitrate recovery data), analysis of blank samples, inter-

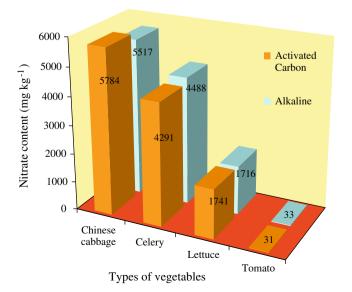


Fig. 2. Comparison of nitrate-N contents (mg kg⁻¹) obtained from the activated carbon and alkaline extraction techniques using fresh vegetable samples.

day repeatability study, analysis in duplicate and an interlaboratory comparison. A standard reference material (Fluka Nitrate Ion Standard Solution No. 72544) was used in the study of nitrate recovery as well as the interday repeatability study.

The precision of the method was evaluated based on ten replicate measurements from an interday repeatability study and analysing the results in terms of the standard deviation (SD) and RSD. The RSD of ten repeated determinations of 2.70 mgL⁻¹ standard solution of nitrate (Fluka Nitrate Ion Standard Solution No. 72544) was evaluated consequently for three days (results not shown here). The average SD and RSD values were 0.06 mgL⁻¹ and 2.22%, respectively and thus the method precision is well acceptable.

For interlaboratory precision/comparison of the reproducibility of the proposed method four freeze-dried vegetable samples (Chinese cabbage. English cabbage. lettuce and celery) in duplicate (n = 2) were sent to the Commonwealth Scientific and Industrial Research Organisation (CSIRO) laboratory, Canberra, Australia for nitrate-N analysis. The vegetables samples sent to the CSIRO were subjected to the heat treatment by the CSIRO laboratory for quarantine purposes. At the CSIRO laboratory, the nitrate was extracted using the activated carbon method, as employed in this study at the University of the South Pacific (USP) laboratory, and the analysis was performed using similar instrument, Lachat QuickChem 8500 FIA system. The comparison of nitrate-N obtained in the vegetables analysed at the USP and the CSIRO laboratories show that the nitrate-N values for Chinese cabbage, English cabbage, lettuce and celery determined at the USP laboratory were slightly higher than those analysed at the CSIRO laboratory (Chetty, 2008). The slight difference in values may be attributed to the delay in analysis as a result of the time taken for samples to arrive at the CSIRO and subjected to a heat treatment. The delay in analysis might have resulted in decreased nitrate content. Despite these facts, interlaboratory comparison may be viewed as being quite satisfactory as the nitrate-N for each sample compared falls in a close range.

Method detection limit (MDL) was calculated using the expression: MDL = $t \times$ SD, where SD is the standard deviation of the replicate analyses and t (= 3.143 for six degrees of freedom) is Student's t value at the 99% confidence limit. The MDL for nitrate determination in selected root vegetables was found to be 0.042 mgL⁻¹ (0.34 mgkg⁻¹). To have a check on the quality control, the MDL was determined regularly or when there is a noteworthy change in instrumental (FIA) response observed. A comparison of the detection limits with those reported by earlier workers showed that 10-fold lower detection limit was obtained by the standard-ised method (Chetty, 2008).

3. Results and discussion

3.1. General

Root vegetables are a great source of carbohydrates and are consistently consumed in Fiji. The most commonly consumed four root vegetables have been assayed in the present study. The nitrite levels in fresh root vegetables are much less than 2 mgkg⁻¹ (Prasad & Chetty, 2008; Ximenes et al., 2000). Therefore, the contribution of nitrite towards the total nitrate load was not considered. The methodology employed to quantify nitrate is based on the FIA with colorimetric detection. The calibration graphs obtained using peak areas *versus* concentrations of nitrate were linear up to 20.0 mgL⁻¹ of nitrate-N. The determination of nitrate-N in root vegetables was carried out after extraction by activated carbon technique and employing the automated calibration during each run of samples. Regression equations relating peak area to the concentration of nitrate were automated with very high correlation coefficients.

Table 1

Nitrate contents in selected fresh root vegetables using activated carbon extraction technique.

Vegetable	Samples analysed $(n)^{a}$	Nitrate cont	Nitrate content (mgkg ⁻¹)				R^2
		Min	Max	Mean	SD		
Dalo (Colocasia esculenta)	6	118.32	817.52	258.00	275.74	106.88	0.9973
Potato (Solanum tuberosum)	6	102.91	119.85	110.79	6.23	5.63	0.9932
Carrot (Daucus carota L.)	6	81.03	99.97	91.31	7.40	8.10	0.9998
Sweet potato (Ipomoea batatas)	6	41.06	62.96	53.76	8.28	15.40	0.9967

SD, standard deviation.

RSD, relative standard deviation.

^a Each sample was analysed in duplicate.

3.2. Procedure

For the determination of nitrate-N in the root vegetables the extracted sample solutions were placed in sample tubes on the 60-position rack along with the six calibration standards. The auto-sampler filled the solutions into injection valve (V) and then injected into the carrier flow stream (R_1) that merges with the buffer (pH 8.5) stream (R_2) at a flow rate of 2.6 mLmin⁻¹. The buffered sample passes through the delay coil (DC) and flows to the two state column switching valve (SV). The SV is turned on to pass the stream of buffered sample through the Cu-Cd reduction column (C) where the nitrate is reduced to nitrite. Under non-injection of standard and samples the carrier and buffer passes through a parallel channel (B) and not through C. The nitrite stream then combines with the colour-developing reagent solution passing through R₃. The diazo-coupling reaction takes in the reaction coil (RC) forming a purplish-pink dye (λ_{max} 520 nm) giving peaks on passing through the detector (D). The peaks were recorded in a PC connected to the FIA and the solution goes to waste tank (W) (Fig. 1). The nitrate-N concentration was determined by measuring the peak area of the dye formed at 520 nm.

3.3. Determination of nitrate in fresh root vegetables

The nitrate levels in fresh and cooked root vegetables were determined using FIA as discussed in Section 3.2. The results of the investigation of the nitrate contents (mean) of the selected fresh root vegetables, their range, variability (SD) and corresponding correlation coefficient are reported in Table 1. The mean nitrate content of the four root vegetables ranges from 54 to 258 mg kg⁻¹. Dalo has the highest nitrate content within the studied vegetables followed by potato, carrot and sweet potato. It is evident from the results shown in Table 1 that only dalo, in the present studied root vegetables, shows considerable variation of nitrate-N contents between different samples of the same vegetable. RSD values of potato, carrot and sweet potato can be considered to be minimal.

3.4. Nitrate in cooked root vegetables

The nitrate-N contents in cooked (boiled, backed and fried) root vegetables including their range, mean values and variability are presented in Table 2. The percentage gain (+) or loss (-) of nitrate on different cooking methods employed are also presented in Table 2. The comparison of the nitrate contents in Table 1 and 2 clearly shows that the nitrate contents are significantly decreased on boiling in all studied root vegetables and similar observation has also been reported by MAFF UK (1999) and Prasad and Chetty (2008). The highest nitrate loss in case of root vegetables after boiling was found for sweet potato (42.62%), followed by dalo (41.88%), carrot (32.92%) and potato (23.30%). The loss of nitrate on boiling is justified as nitrate being water soluble can easily be leached into cooking water.

The nitrate-N contents in the root vegetables on baking and frying in sova bean oil including their range, mean values and variability are also presented in Table 2. In case of baking and frying of the root vegetables the similar trends as observed in case of leafy vegetables were found (Prasad & Chetty, 2008). Table 2 indicates that the nitrate values remain relatively constant after baking. The highest nitrate gain was found in carrot (8.43%) followed by sweet potato (7.23%), dalo (6.87%) and potato (2.10%). The results of the investigation of nitrate contents in studied root vegetables after frying in soya bean oil presented in Table 2 shows 2-3 fold increase in nitrate content. The amazing increase may be attributed to the loss of moisture from the vegetable upon frying and the small sample size and high amount of oil used in frying since soya possesses nitrogenous species, ammonia (Hill, 1996). Critical frying times, oil temperature, initial moisture content and oil absorption are some of the factors that need to be analysed in order to fully justify the findings of the present study with regards to the effects of frying on nitrate content of vegetables. To compare the mean nitrate-N values and the variability for the fresh root vegetables and after they have been subjected to three widespread cooking methods (boiling, baking and frying), a bar diagram was constructed as shown in Fig. 3. Fig. 3 clearly shows an increasing trend of nitrate-N levels as sweet potato < carrot < potato < dalo and well below 300 mg kg^{-1} (fresh sample).

Table 2

Nitrate contents in selected fresh root vegetables after boiling, baking and frying using activated carbon extraction technique.

Vegetable	Samples analysed (n) ^a	Nitrate content (mgkg ⁻¹)				RSD	Nitrate loss (-)	
		Min	Max	Mean	SD	(%)	or gain (+) (%)	
Boiling								
Dalo	6	66.06	478.85	149.96	130.34	107.95	-41.88	
Potato	6	72.31	95.10	84.98	9.39	11.05	-23.30	
Carrot	6	48.34	71.43	61.26	8.66	14.14	-32.92	
Sweet potato	6	15.07	42.32	30.85	9.98	32.35	-42.62	
Baking								
Dalo	6	129.17	903.68	275.72	250.45	112.10	+6.87	
Potato	6	103.96	121.33	113.12	7.16	6.33	+2.10	
Carrot	6	86.11	109.09	99.01	8.08	8.16	+8.43	
Sweet potato	6	50.43	68.41	57.65	8.35	14.49	+7.23	
Frying								
Dalo	6	526.18	1467.59	785.67	240.92	43.39	+204.53	
Potato	6	320.61	481.04	360.37	60.03	16.66	+225.27	
Carrot	6	303.14	384.21	347.60	33.42	9.61	+280.67	
Sweet potato	6	175.16	245.63	214.58	26.23	12.22	+299.12	

SD, standard deviation. RSD, relative standard deviation.

^a Each sample was analysed in duplicate.

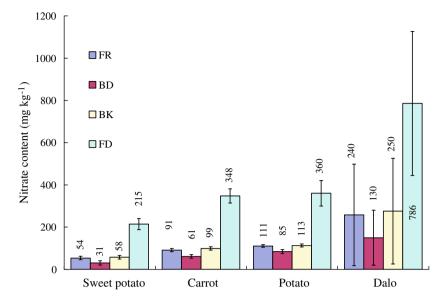


Fig. 3. Comparison of mean (n = 6) nitrate-N contents in fresh and cooked root vegetables (FR, fresh; BD, boiled; BK, baked and FD, fried).

3.5. Vegetables grouping, comparison of nitrate levels in fresh vegetables and analytical figures of merit

Corré and Breimer (1979) classified different vegetables into five groups with respect to their increasing nitrate contents.

Table 3

Comparison of nitrate values of the root vegetables studied with those of other countries.

Country	Root vegetables nitrate-N content (mgkg ⁻¹)					
	Carrot	Potato	Sweet potato	Dalo		
Fiji (present study)	91.3	110.8	53.8	258.0		
New Zealand ^b	48.0	107 ^a	-	-		
Estonia ^c	148.0	-	-	-		
Slovenia ^d	264.0	158.0	-	-		
Korea ^e	316.0	452.0	-	-		
UK ^f	97.0	155.0	-	-		
Greece ^g	87.0	32.0	-	-		
The Netherlands ^h	340.0	280.0	-	-		
Belgium ⁱ	278.0	154.0	-	-		
India ^j	170.0	80.0	110.0	-		
Italy ^k	17.0	-	-	-		
Australia ¹	8.4	-	-	-		
USA ^{m,n}	72.0 ^m	120.0 ^m	65.0 ⁿ	-		
China ^o	-	164.0	-	-		
Croatia ^p	-	196.0	-	-		
Denmark ^q	-	229.0	-	-		
Poland ^r	-	174.0	-	-		

Determined after boiling.

- Thomson et al. (2007).
- Tamme et al. (2006).
- d Sušin et al. (2006).

Chung et al. (2003).

- Ysart et al. (1999).
- Anastasios and Constantinos (1999).
- van der Schee (1998).
- Dejonckherre et al. (1993).
- Gundimeda et al. (1993).
- Consalter et al. (1992).
- Lyons et al. (1991).
- ^m Siciliano et al. (1975). n National Academy of Sciences (1981).
- Zhong et al. (2002).
- Sebecic and Vedrina-Dragojevic (1999). q Peterson and Stoltze (1999).
- Cieslik and Sikora (1998).

The vegetables studied in present case along with the range of the nitrate contents have been grouped according to Corré and Breimer (1979) classification. Only dalo falls in group 2 whilst sweet potato, potato and carrot fall in group 1. The studied root vegetables very well follow Corré and Breimer's grouping criteria.

The comparison of nitrate-N contents in selected Fiji's fresh root vegetables with the literature values available on similar root vegetables in sixteen other countries is presented in Table 3. The results from the present study are comparable or lower than the nitrate-N obtained in the root vegetables of foreign countries. Out of the root vegetables studied carrot and potato are the most investigated root vegetables in terms of nitrate-N determination. There was no data available on dalo as it is only common in the Pacific Islands. The proposed and applied FIA-colorimetric method for the determination of nitrate in vegetable was also compared with other methods in literature along with their MDL, dynamic range of detection (DRD) and other characteristics reported elsewhere (Chetty, 2008). It was found that the proposed and applied FIA-colorimetric technique for the determination of nitrate in root vegetables has much lower detection limit, higher DRD and sample throughput.

4. Conclusion

The present study has employed FIA and colorimetric detection technique to quantify the nitrate-N contents in four types of common root vegetables available to consumers in Fiji. The effects of various cooking methods were also assessed. On boiling the nitrate-N content is reduced by an appreciable amount. The fresh root vegetables analysed in this study did not present unpredictably high values. The average nitrate-N values are comparable or lower than overseas data. Thus there is no cause for concern regarding the nitrate-N content of fresh root vegetables marketed in Fiji.

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References

- Ahn, H. S., Yook, H. S., Rhee, M. S., Lee, C. H., Cho, Y. J., & Byun, M. W. (2002). Application of gamma irradiation on breakdown of hazardous volatile Nnitrosamines. *Journal of Food Science*, 67, 596–599.
- Anastasios, S. S., & Constantinos, C. D. (1999). Nitrates in vegetables produced in Greece. Journal of Vegetable Crop Production, 5, 3–13.
- Andrade, R., Viana, C. O., Guadagnin, S. G., Reyes, F. G. R., & Rath, S. (2003). A flowinjection spectrophotometric method for nitrate and nitrite determination through nitric oxide generation. *Food Chemistry*, 50, 597–602.
- Burakham, R., Oshima, M., Grudpan, K., & Motomizu, S. (2004). Simple flowinjection system for the simultaneous determination of nitrite and nitrate in water samples. *Talanta*, 64, 1259–1265.
- Chetty, A. (2008). Study of the effects of cooking and deep-freezing on the nitratenitrogen concentration in Fiji's vegetables using flow injection analysis (FIA). Suva, Fiji: M.Sc. Thesis, submitted to the Faculty of Science and Technology, the University of the South Pacific.
- Chung, S. Y., Kim, J. S., Kim, M., Hong, M. K., Lee, J. O., Kim, C. M., et al. (2003). Survey of nitrate and nitrite contents grown in Korea. *Food Additives and Contaminants*, 20, 621–628.
- Cieslik, E., & Sikora, E. (1998). Correlation between the levels of nitrates and nitrites and the contents of potassium, calcium and magnesium in potato tubers. *Food Chemistry*, 63, 525–528.
- Consalter, A., Rigato, A., Clamor, L., & Giandon, P. (1992). Determination of nitrate in vegetables using an ion-selective electrode. *Journal of Food Composition and Analysis*, 5, 252–256.
- Corré, W. J., & Breimer, T. (1979). Nitrate and nitrite in vegetables. Literature Survey No. 39 (p. 85). Wageningen, The Netherlands: Centre for Agricultural Publishing and Documentation.
- Dejonckherre, W., Stearbaut, W., Drieghe, S., Verstraeten, R., & Braeckman, H. (1993). Nitrate in food commodities of vegetable origin and the total diet in Belgium (1992–1993). *Microbiologie–Aliments–Nutrition*, *12*, 359–370.
- Ezeagu, I. E. (1996). Nitrate and nitrite contents in ogi and the changes occurring during storage. Food Chemistry, 56, 77–79.
- Gangolli, S. D., van de Brandt, P. A., Feron, V. J., Janzowski, C., Koeman, J. H., Spijers, G. J. A., et al. (1994). Nitrate, nitrite, and N-nitroso compounds. *European Journal* of Pharmacology–Environment Toxicology and Pharmacology Section, 292, 1–38.
- Gundimeda, U., Naidu, A. N., & Krishnaswamy, K. (1993). Dietary intake of nitrate in India. Journal of Food Composition and Analysis, 6, 242–249.
- Hill, M. J. (1996). Nitrates and nitrites in food and water. Cambridge, England: Woodhead Publishing Limited.
- Kmiecik, W., Lisiewska, Ž., & Slupski, J. (2004). Effects of freezing and storage of frozen products on the content of nitrate, nitrites, and oxalates in dill (*Anethum* graveolens L.). Food Chemistry, 86, 105–111.
- Lyons, D. J., McCallum, E. L., Osborne, W. J., & Nobbs, P. E. (1991). Assessment of procedures for the determination of nitrate and nitrite in vegetable extracts. *Analyst*, 116, 153–157.
- MAFF UK (Ministry of Agriculture, Fisheries and Food of United Kingdom). (1999). Nitrate in vegetables. Food Surveillance Information Sheet No. 158, September 1998, London, England.
- MAFF UK. (2001). UK monitoring programme for nitrate in Lettuce and Spinach. Food Survey Information Sheet No. 16/01, July 2001, London, England.
- Melchert, W. R., & Rocha, P. R. F. (2005). A green analytical procedure for flowinjection determination of nitrate in natural waters. *Talanta*, 65, 461–465.

- Monser, L., Sadok, S., Greenway, G. M., Shah, I., & Uglow, R. F. (2002). A simple simultaneous flow-injection spectrophotometric method based on phosphomolybdenum chemistry for nitrate and nitrite determinations in water and fish samples. *Talanta*, 57, 511–518.
- National Academy of Sciences (1981). The health effects of nitrate, nitrite and Nnitroso compounds. Washington, DC, USA: National Academy Press.
- Perez-Olmos, R., Herrero, R., Lima, J. L. F. C., & Montenegro, M. C. B. S. M. (1997). Sequential potentiometric determination of chloride and nitrate in meat products. *Food Chemistry*, 59, 305–311.
- Peterson, A., & Stoltze, S. (1999). Nitrate and nitrite in vegetables on the Danish market: Content and intake. Food Additives and Contaminants, 16, 291–299.
- Prasad, S., & Chetty, A. A. (2008). Nitrate-N determination in leafy vegetables: Study of the effects of cooking and freezing. *Food Chemistry*, 106, 772–780. and references cited therein.
- Santamaria, P. (2006). Review Nitrate in vegetables: Toxicity, content, intake and EC regulation. *Journal of the Science of Food and Agriculture*, 86, 10–17. and references cited therein.
- Santamaria, P., Elia, A., Gonnella, M., Parente, A., & Serio, F. (2001). Ways of reducing rocket salad nitrate content. Acta Horticulture, 548, 529–537.
- Santamaria, P., Elia, A., Serio, F., & Todaro, E. (1999). A survey of nitrate and oxalate content in retail fresh vegetables. *Journal of the Science of Food and Agriculture*, 79, 1882–1888.
- Sebecic, B., & Vedrina-Dragojevic, I. (1999). Nitrate and nitrite in vegetables from areas affected by war-time operation in Croatia. *Nahrung*, 43, 284–287.
- Siciliano, J., Krulick, S., Schwartz, J. H., & White, J. W. Jr., (1975). Nitrate and nitrite content of some fresh and processed market vegetables. *Journal of Agriculture* and Food Chemistry, 23, 461–463.
- Sušin, J., Kmecl, V., & Gregorčič, A. (2006). A survey of nitrate and nitrite content of fruits and vegetables grown in Slovenia during 1996–2002. Food Additives and Contaminants, 23, 385–390.
- Tamme, T., Reinik, M., Roasto, M., Juhkam, K., Tenno, T., & Kiis, A. (2006). Nitrates and nitrites in vegetables and vegetable based-products and their intake by the Estonian population. *Food Additives and Contaminants*, 23, 355–361.
- Tannenbaum, S. R., Fett, D., Young, V. R., Land, P. D., & Bruce, W. R. (1978). Nitrite and nitrate are formed by endogenous synthesis in the human intestine. *Science*, 200, 1487–1489.
- Thomson, B. M., Nokes, C. J., & Cressey, P. (2007). Intake and risk assessment of nitrate and nitrite from New Zealand foods and drinking water. *Food Additives* and Contaminants, 24, 113–121.
- Usher, D., & Telling, G. M. (1975). Analysis of nitrate and nitrite in foodstuffs: A critical review. Journal of the Science of Food and Agriculture, 2, 1793–1805.
- van der Schee, H. A. (1998). *The nitrate content of vegetables of the Dutch market in 1996*. Amsterdam, The Netherlands: Inspectorate for Health Protection.
- Walker, R. (1975). Naturally occurring nitrate/nitrite in foods. Journal of the Science of Food and Agriculture, 26, 1735–1742.
- Ximenes, M. I. N., Rath, S., & Reyes, F. G. R. (2000). Polarographic determination of nitrate in vegetables. *Talanta*, 51, 49–56.
- Ysart, G., Miller, P., Barette, G., Farrington, D., Lawrence, P., & Harrison, N. (1999). Dietary exposures to nitrate in the UK. *Food Additives and Contaminants*, 16, 521–532.
- Zhong, W., Hu, C., & Wang, M. (2002). Nitrate and nitrite in vegetables from North China: Content and intake. Food Additives and Contaminant, 19, 1125–1129.