ELSEVIER

PII: S0308-8146(97)00084-8

Food Chemistry, Vol. 61, No. 1/2, pp. 145–151, 1998

0308-8146/98 $19.00 + 0.00

Carotenoid retention and vitamin A value in carrot (*Daucus carota* L.) prepared by food service

Helena Maria Pinheiro Sant'Ana,[a] Paulo César Stringheta,[b] Sebastião César Cardoso Brandão[b] & Raquel Monteiro Cordeiro de Azeredo[a]

[a]*Department of Nutrition and Health, Universidade Federal de Viçosa, MG-Brazil 36571-000*
[b]*Department of Food Technology, Universidade Federal de Viçosa, MG-Brazil 36571-000*

(Received 7 January 1997; accepted 2 April 1997)

The objective of this work was to study the influence of different methods of food preparation routinely used by catering and food services (in large quantities) on the stability of α-carotene, β-carotene and total carotenoids in carrots. Also, the values of vitamin A were evaluated. The methods of preparation studied were: raw shredded, steam cooking, water cooking with pressure, water cooking without pressure and moist/dry cooking. The quantification of α and β-carotenes was carried out by high-performance liquid chromatography (HPLC), using a reverse phase column (RP-18) with methanol: acetonitrile: ethyl acetate (80:10:10) as the mobile phase and a UV-Visible detector. Total carotenoids were quantified spectrophotometrically at 449 nm. The results showed a retention ranging from 56.0 to 89.1% for the carotenoids, with the moist/dry cooking causing the greatest losses in α and β-carotene. Considering the type of utensils used, the time and temperature, it was concluded that water cooking without pressure was best for reducing losses of carotenoids in carrots prepared by food service. Despite considerable losses in vitamin A value, carrots subjected to routine preparation methods by catering and food services remain a rich source of provitamin A. The methodology developed in this study is now being applied to carotenoid analysis in other vegetables prepared in large quantities. © 1998 Elsevier Science Ltd. All rights reserved

INTRODUCTION

Carotenoids are among the most important nutrients in food, owing to their diverse functions and actions. The attractive red or yellow colour conferred to many foods is their most apparent contribution to food quality. The provitamin A activity of some carotenoids and other potentially beneficial health effects, such as anticarcinogenic, antiulcer, anti-aging, antioxidant properties, as well as an increased immune response, add to their importance in the diet (Rodriguez-Amaya, 1993). It is well known that carotenoids are extremely susceptible to degradation. Their highly unsaturated structure makes them sensitive to heat, oxygen and light (Clydesdale and Francis, 1976; Britton, 1992). Their instability is of the utmost importance when the objective is to minimize these losses.

Catering and food services rank as the fourth largest industry in the US in volume sales and the first in terms of numbers of customers; they comprise 600,000 facilities (Lachance, 1975). The growing Brazilian catering food market prepares approximately 2 billion meals per year, with a potential to reach 14 billion meals per year. However, this increase will demand investments in specific facilities, require specialized professionals, and will need the development of new products (TODA, 1994). In spite of the evolution in many sectors, studies on retention of nutrients as a result of food preparation practices at food services have received very little attention.

It is necessary to establish nutritional standards for the meals served at the increasing number of food service facilities. This emphasizes the need for a deeper study of food preparation practices on a large scale and their implication for nutrient retention.

The objective of this study was to analyze the α-carotene, β-carotene and total carotenoids and their stability in carrots prepared in large quantities and in different manners at food service facilities.

MATERIALS AND METHODS

Raw material

Carrots (*Daucus carota* L.) of the variety Nantes, grown under standardized conditions (same type of soil, same planting techniques and same fertilizer treatment), were

used in this study at the Universidade Federal de Viçosa (UFV), MG, Brazil. The entire sample was obtained from the same planting date and harvested approximately 4 months later. The samples were taken from field to laboratory inside plastic containers at 25–26°C inside a closed vehicle. Samples were prepared the day after harvest.

Sample preparation

After being washed and mechanically peeled with an industrial tuber peeler (model 5B 22D, Hobart), carrots were subjected to the preparation practices described in Table 1, according to a preparation routine commonly followed at the UFV cafeteria, a model facility for other catering and food services. Three replicates were performed for each treatment and the analyses were done in duplicate.

For the raw carrot method, an electric slicer model 4B, 22D, Hobart was utilized. Carrots (45 kg) were shredded and maintained at room temperature (27–28°C) for approximately 3 h and placed on workbenches under transparent plastic sheets. Waiting time between time of preparation and time of food distribution was thus simulated. Carrots were served raw, not being submitted to any cooking.

Cooking tests were previously conducted to establish the times and temperatures adequate for each cooking method. The start of cooking for each method (zero time) was established when carrots were added. Adequate cooking (tested during the cooking trials and sensorially evaluated, according to Brazilian eating habits) was verified by tasting and/or by texture assessment using a fork. For treatments which involved water-cooking without pressure, 45 kg of carrots and an American industrial pan of 300 litres capacity were used. For the water cooking method with pressure, 45 kg of carrots and a 250 litres autoclave pan were used. The moist/dry cooking (45 kg of carrots) was initiated with cooking without pressure using a 300 litres American industrial pan and concluded in an electric industrial oven. Steam cooking (45 kg of carrots) was performed in an industrial autoclave, with a thermometer and manometer. Temperature was controlled by a temperature register, Speedomax model, with thermocouples placed in pairs in each pan. After being cooked and cooled, the samples were cut into $2\times2\times3$ cm pieces using an industrial vegetable cutter.

Approximately 500 g of carrots were randomly collected from each treatment and the samples were codified and frozen by the quick freezing method (a method which uses liquid nitrogen aspersion over the samples, freezing them instantaneously) at −54°C in a cryogenic chamber, placed in transparent plastic bags, wrapped in aluminum foil and stored in a freezer at −18°C for further analyses.

Analysis of solids

The moisture content of the samples was initially determined and the total solids content was determined by subtraction (Silva, 1981). The insoluble solids were determined according to the analytical procedures of the Instituto Adolfo Lutz (1985). The soluble solids were also determined by a subtraction between the total and insoluble solids.

Preparation of standards

Stock solutions of α and β-carotenes (obtained from Basf do Brasil) were prepared by weighing 50 mg of each in stock solutions of 100 ml of the mobile phase used for HPLC analysis. Standard purity was checked in petroleum ether using the Lambert-Beer law. Increasing concentrations of stock solutions of carotenoids were prepared to build standard curves, which were used for determination of carotenoids in the samples. Sudan {1-(phenylazo) 2-naphthalenol} was used as internal standard according to Quackembush and Smallidge (1986). For its preparation, 0.1 g of this standard was weighed and diluted in a solution of 500 ml of methanol: chloroform (9:1) and further diluted for sample analysis.

Carotenoid extraction

The carotenoid extraction method was based on the procedure described by Rodriguez *et al.* (1976). Samples (5 g) were ground with the help of chilled acetone and a microgrinder model TE 102, Tecnal and vacuum-filtered using a Büchner funnel. This procedure was repeated until the residue became colourless and pigments were transferred to petroleum ether, each fraction being washed with distilled water for complete acetone removal.

Carotenoid analysis

After extraction, the total carotenoid content of the pigments extracted was determined by spectrophotometer at 449 nm, as proposed by Ramos (1991).

Table 1. Preparation methods of carrots in large quantities (45 kg for preparation)

Preparation	Water quantity (l)	Cooking time (min)	Cooking temperature (°C)
Shredded raw	—	—	—
Steam-cooking	—	15	115–120
Water-cooking with pressure	35	17	100
Water-cooking without pressure	35	21	99
Moist/dry-cooking	35/—	21/15	99/200

The α and β-carotene analysis was carried by HPLC. Pigments were clarified in a MgO:hyflosupercel (1:1) mini column, according to Carvalho (1993). This procedure was adopted to avoid overlapping of the internal standard's retention time with that of other pigments in the carrot. Clarification allows for the elimination of carotenoids other than the provitamin A ones. Samples thus obtained were evaporated under nitrogen, re-dissolved in a known acetone volume, a constant volume of internal standard added, filtered using 0.45 μm membrane and injected into the liquid chromatographic column. The following apparatus were used: liquid chromatograph, CG-480 (Instrumentos CG, São Paulo, Brazil) equipped with Rheodyne 7125 injector, 100 μl loop, UV-VIS detector (UV-50, Varian) at 449 nm and an integrator (CG 200). The column used was a 25 cm $\times$ 4 mm RP-18 (Lichrospher, 5 μm, Merck). The mobile phase was methanol:acetonitrile:ethyl acetate (80:10:10) with a flow rate of 2 ml min^{-1}. Solvents (Lichrosolv-Merck) were filtered immediately before use utilizing the Millipore vacuum filtration system, and degassed under an ultrasound system in order to remove air bubbles. Samples and standards were filtered through an FH 1300 Millipore membrane (0.45 μm).

Recovery experiments

Since carotenoids are rather unstable compounds, their stability during sample preparation was determined through addition and recovery of known quantities of α and β-carotene standards to the carrot samples, followed by HPLC analysis, as previously described.

Vitamin A value calculation

Calculation was performed based on the vitamin A activity of each carotenoid precursor (α and β-carotenes), according to Bauernfeind (1972), and the conversion factors provided by the National Academy of Sciences—National Council Research (NAS—NCR, 1980). Vitamin A value was expressed in RE (retinol equivalents) per 100 g of sample. It is known that 0.6 μg of β-carotene and 1.2 μg of α-carotene and other carotenoids are equivalent to 1 IU (International Unit), with 1 RE being equivalent to 10 IU.

Experimental design

The experiment was arranged in a completely randomized design. Statistical analyses were performed based on SAEG program, 5.0 version (System for Statistical and Genetic Analyses developed by the Data Processing Center of the UFV). Variance analysis was carried out in order to detect significant differences among the treatments. The Duncan test was applied to analyze the differences between treatment averages presenting significant differences (Pimentel-Gomes, 1984).

RESULTS AND DISCUSSION

Qualitative separation of α and β-carotenes by HPLC

The preparation methods and cooking conditions are shown in Table 1. Figure 1 shows chromatograms of carrot samples prepared in large quantities by five methods. As can be verified from Fig. 1(a), the preparation using raw ground carrots did not present any extra peaks after the β-carotene peak. The other methods (Fig. 1(b)–(e)) using higher temperatures and longer cooking times resulted in peak after the β-carotene peak. According to Table 1, the longer the cooking time and temperature, the higher is the resulting peak. Although a corroborative analysis has not been done, literature data suggest that these peaks are the result of thermal isomerization of β-carotene, with the production of the less potent *cis* isomer (O'Neill and Schwartz, 1992). It has been well documented that food processing and some preservation methods, especially those involving thermal treatments, induce formation of *cis* isomers (Almeida and Penteado, 1987; Chandler and Schwartz, 1987; O'Neill and Schwartz, 1992).

Recovery of added α and β-carotenes

The results on recovery of added α and β-carotenes to carrot samples are shown in Tables 2 and 3. The percentage recovery was very good. Hence, it can be concluded that the results obtained on the concentration of carotenoids are due to the treatments conducted.

Quantitative analysis of carotenoids

The contents of α-carotene, β-carotene and total carotenoids are presented in Tables 4–6. The highest α-carotene and β-carotene contents were obtained by using the method of water cooking without pressure which suggests that, under the conditions of this experiment, this method gave less reduction of these carotenoids. The steam-cooking method preserved the total carotenoids better, being the second best option for avoiding losses of α and β-carotene. In this method, when samples were not placed in water, losses due to leaching were prevented during cooking. As can be observed, the samples submitted to moist/dry cooking presented lower concentrations of α and β-carotene. In this method, the conditions of preparation (see Table 1) were more drastic (especially cooking time and temperature) thus explaining the higher reduction of carotenoids in these samples. However, no significant difference was found in the concentrations of α-carotene, β-carotene and total carotenoids for the methods studied.

As can be observed from the table, the sum of α and β-carotene values does not correspond to that of the total carotenoid values. This can be explained on the basis of the methodologies used, different analysis techniques,

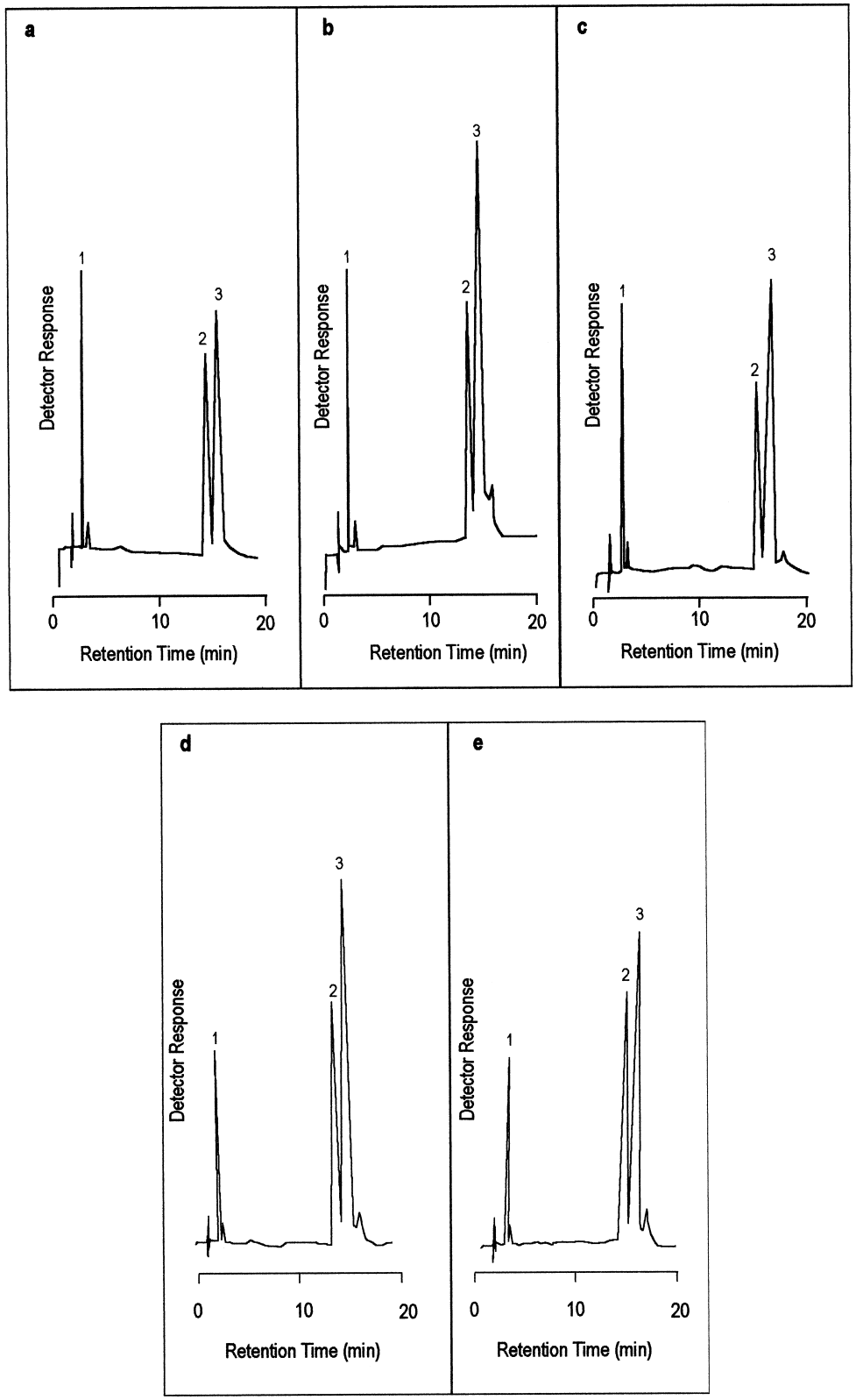

Fig. 1. HPLC analysis of the carotenes in carrot prepared in large quantities. Mobile phase: methanol:acetronitrile:ethyl acetate (80:10:10); flow rate: 2 ml min^{-1}; RP18 column; detection at 470 nm; absorbance range: 0.5.

Table 2. Percentage recovery of α-carotene added to carrots (results expressed on fresh weight basis)

Test	Sample without addition ($\mu g\,g^{-1}$)[b] ± SD	Range	Sample with addition ($\mu g\,g^{-1}$)[b] ± SD	Range	Percentage recovery[a]
1	40.2 ± 0.2	39.9–40.6	51.9 ± 0.4	51.5–52.4	97.8
2	32.6 ± 0.5	31.9–33.2	44.2 ± 0.6	43.4–45.0	96.6
3	36.4 ± 0.4	35.9–37.0	48.2 ± 0.4	47.8–48.8	98.1

[a]12 μg of α-carotene added per gram of sample.
[b]Average of three determinations in duplicate.

Table 3. Percentage recovery β-carotene added to carrots (results expressed on fresh weight basis)

Test	Sample without addition ($\mu g\,g^{-1}$)[b] ± SD	Range	Sample with addition ($\mu g\,g^{-1}$)[b] ± SD	Range	Percentage recovery[a]
1	79.5 ± 1.4	78.1–81.7	99.0 ± 1.8	97.0–102.0	97.3
2	62.6 ± 0.8	61.8–64.1	81.9 ± 2.1	79.2–84.9	96.7
3	71.6 ± 0.6	71.1–72.5	91.1 ± 1.9	89.1–94.3	97.3

[a]20 g of β-carotene added per gram of sample.
[b]Average of three determinations in duplicate.

Table 4. Concentration of α-carotene in carrot samples prepared in large quantities (expressed on insoluble solids basis), as analysed by HPLC

Preparation	α-carotene ($\mu g\,g^{-1}$) ± SD	Ranges
Shredded raw	1057.1[a] ± 223.7	766.4–1319.6
Steam-cooking	1139.7[a] ± 400.2	663.6–1650.8
Water-cooking with pressure	1037.5[a] ± 213.6	630.2–1254.1
Water-cooking without pressure	1326.3[a] ± 583.7	857.1–2372.8
Moist/dry-cooking	951.9[a] ± 494.9	574.6–1796.2

Means followed by same letter in column do not differ at 5% probability by the variance analysis.

Table 5. Concentration of β-carotene in carrot samples prepared in large quantities (expressed on insoluble solids basis), as analysed by HPLC

Preparation	β-carotene ($\mu g\,g^{-1}$) ± SD	Ranges
Shredded raw	1960[a] ± 438	1451–2493
Steam-cooking	2220[a] ± 634	1313–3125
Water-cooking with pressure	2110[a] ± 519	1342–2975
Water-cooking without pressure	2345[a] ± 744	1534–3296
Moist/dry-cooking	1810[a] ± 684	1233–2969

Means followed by same letter in column do not differ at 5% probability by the variance analysis

Table 6. Concentration of total carotenoids in carrot samples prepared in large quantities (expressed on insoluble solids basis), as analysed by spectrophotometry

Preparation	Total carotenoids ($\mu g\,g^{-1}$) ± SD	Ranges
Shredded raw	2921[a] ± 979	2237–4714
Steam-cooking	3767[a] ± 1186	2357–5192
Water-cooking with pressure	3173[a] ± 757	2137–4323
Water-cooking without pressure	3586[a] ± 494	3097–4437
Moist/dry-cooking	3431[a] ± 663	2682–4507

Means followed by same letter in column do not differ at 5% probability by the variance analysis.

higher sensitivity of the HPLC method and the fact that, besides α and β-carotenes, other carotenoids are present in carrots (not quantified by HPLC).

Evaluation of carotenoid stability

The results of the effect of large-scale preparation methods on the stability of α and β-carotenes and total carotenoids can be observed in Table 7.

Lower retention levels show a lower stability of α-carotene as a function of preparation (56.0–78.0%). β-carotene was more stable in the treatments presenting a retention percentage of 68.8–89.1%. Water-cooking without pressure resulted in higher retention of α and β-carotenes (78.0 and 89.0%, respectively). This effect can be explained on the basis of the lower temperature used in the cooking (Table 1). After water-cooking without pressure, steam-cooking produced the highest retention

Table 7. Influence of cooking method (in large quantities) on stability of α-carotene, β-carotene and total carotenoids in carrot samples in relation to control sample (100%) (expressed on insoluble solid basis)

Cooking method	Percentage retention of carotenoids after treatments		
	α-carotene[a]	β-carotene[b]	Total carotenoids[b]
Shredded raw	62.2[c]	74.5[d]	59.0[e]
Steam-cooking	67.1[b]	84.4[b]	76.1[a]
Water-cooking with pressure	61.0[c]	80.2[c]	64.1[d]
Water-cooking without pressure	78.0[a]	89.1[a]	72.4[b]
Moist/dry-cooking	56.0[d]	68.8[e]	69.3[c]

[a]Analyzed by HPLC.
[b]Analyzed by spectrophotometry.
Means followed by same letter in column do not differ at 5% probability by the Duncan Test.

for α and β-carotenes (67.0 and 84.4%, respectively) and also presented the highest stability for total carotenoids (76.0%). It was expected that the absence of water in this method would reduce losses. However, the higher temperature used (Table 1) contributed more than the absence of water. Significant differences among the retention levels were found. Tannenbaum (1976) has reported that high temperatures provoke higher degradation.

Krehl and Winters (1950) studied the effect of cooking on losses of total carotenoids in vegetables, using different volumes of water. The results clearly showed that the highest retention was obtained when the vegetables were cooked without addition of water and that the lowest retention was associated with the use of larger amounts of water during cooking. Intermediary losses were verified in cooking with pressure or when a small amount of water was used. The studies showed that steam-cooking resulted in retention values similar to those obtained from cooking with pressure or using a small quantity of water. Intermediary losses verified in this study were also a result of water-cooking with pressure (61.0; 80.21 and 64.1% of retention to α-carotene, β-carotene and total carotenoids, respectively).

Harris (1960) reviewed the effects of water-cooking in large quantities on the nutrient content. The results showed that 81–99% of the carotenoids in carrots were retained, while steam-cooking resulted in 91–93% retention. However, cooking temperature and time were not reported.

As can be verified from Table 7, samples submitted to moist/dry cooking showed the lowest α and β-carotene retention. This result can be explained on the basis of the higher cooking time and temperature used in oven cooking (Table 1). Spiers (1945) reported retention values of 76.0–90.0% of total carotenoids in baked and water-cooked potatoes, respectively. In our study, a retention value of 69.3% for total carotenoids was observed in carrots cooked in water followed by baking.

Low retentions of α-carotene, β-carotene and total carotenoids (62.2, 74.5 and 60.0%, respectively) were observed in samples obtained by the raw peeling method. Since the samples were exposed to light and oxygen for approximately 3 h, conditions favorable to carotenoid degradation were verified. It is worth mentioning that the samples were not submitted to a blanching process before storage. Thus, it is evident that the enzymatic process continued during storage, leading to carotenoid degradation. Booth (1960), cited by Edwards and Lee (1986) reported that some enzymatic systems in plants containing chlorophyll are 'carotenoid destroyers'. Edwards and Lee (1986) reported carotenoid losses due to enzymatic activity during pigment extraction procedures in fresh peas. It is, thus, recommended that the carrots, if shredded, be served raw immediately after preparation, since excessive cutting greatly exposes their surfaces to oxygen and light, leading to carotenoid losses. An alternative to serving the carrots raw would be cutting them vertically into large pieces, thus avoiding excessive exposure to oxygen and light, ensuring a larger carotenoid consumption.

Moreover, it has become evident that the shorter the cooking time, temperature and the contact of the samples with water, the higher is the content of α-carotene, β-carotene and total carotenoids preserved during cooking.

Provitamin A loss evaluation

The results obtained show that losses follow the same profile of those verified for β-carotene (Table 8) since this is the main carotenoid giving provitamin A activity in carrots. Thus, the smallest losses in vitamin A values were obtained by water-cooking without pressure (13.6%), followed by steam-cooking (19.8%), water-cooking with pressure (25.0%), shredded raw (28.5%) and moist/dry-cooking (34.3%). Hence, according to the conditions used in this experiment, water-cooking without pressure proved to be the most adequate method to prevent provitamin A losses in carrots. The moist/dry-cooking was the least beneficial for carrot preparation. It is important to emphasize that the carrot samples submitted to the preparation methods routinely used by catering and food services remain good sources of provitamin A, despite losses.

Table 8. Vitamin A values of carrots prepared by food service (expressed on insoluble solid basis)

Cooking method	Vitamin A values[a] (RE 100 g^{-1})			Losses in relation to the total (%)[b]
	α-carotene[b]	β-carotene[c]	Total[c] ($\alpha + \beta$)	
Control sample	14165[a]	43852[a]	58017[a]	0.0[f]
Shredded raw	8809[cd]	32668[a]	41477[a]	28.5[b]
Steam-cooking	9498[c]	37007[a]	46505[a]	19.8[d]
Water-cooking with pressure	8646[cd]	35173[a]	43520[a]	25.0[c]
Water-cooking without pressure	11053[b]	39088[a]	50141[a]	13.6[e]
Moist/dry-cooking	7932[d]	30167[a]	38099[a]	34.3[a]

[a]10 Retinol Equivalents (RE) = 1 International Unit (IU).
[b]Means followed by same letter in column do not differ at 5% probability by Duncan Test.
[c]Means followed by same letter in column do not differ at 5% of probability by variance analysis.

CONCLUSIONS

The results obtained in this study showed that the preparation methods used by catering and food services allowed for a retention ranging from 56.0 to 89.1% of the carotenoids. The moist/dry-cooking method caused the greatest losses in α and β-carotenes due to the more drastic preparation conditions. Water-cooking without pressure (a method routinely used by catering and food services in Brazil) was the method which allowed the greatest stability of α-carotene, β-carotene and total carotenoids. This shows that water-cooking without pressure is the most appropriate method for cooking whole carrots in large quantities, provided cooking time and temperature are adequately controlled. Provitamin A losses were significant, following the profile of β-carotene losses. In spite of this, carrots submitted to routine preparation methods by catering and food services remain a very good source of provitamin A. The methodology developed for this study is being presently used for carotenoid analysis in other vegetables prepared by catering and food services.

ACKNOWLEDGEMENTS

We gratefully thank Prof. Marcelo José Vilela for review of this manuscript and Prof. Alice Jham and Valéria Vitarelli de Queiróz for its translation into English.

REFERENCES

Almeida, L. B. and Penteado, M. V. C. (1987). Carotenóides com atividade pró-vitamínica A de cenouras (*Daucus carota* L.) comercializadas em São Paulo, Brasil. *Revista de Farmácia e Bioquímica de Universidade de São Paulo*, **23**(2), 133–141.

Bauernfeind, J. C. (1972). Carotenoid vitamin A precursors and analogs in foods and feeds. *Journal of Agricultural and Food Chemistry*, **20**, 456–473.

Britton, G. (1992). Carotenoids. In *Natural Foods Colorants*, ed. G. F. Hendry, pp. 141–148. G. F. Blackie, New York.

Carvalho, P. R. N. (1993). *Análise de Vitaminas em Alimentos*. Ital, Campinas.

Chandler, L. A. and Schwartz, S. J. (1987). HPLC separation of *cis-trans* carotene isomers in fresh and processed fruits and vegetables. *Journal of Food Science*, **52**(3), 669–672.

Clydesdale, F. M. and Francis, F. J. (1976). Pigments. In *Principles of Food Science—Food Chemistry*, ed. O. R. Fennema. pp. 417–430. Marcel Dekker, New York.

Edwards, C. G. and Lee, C. Y. (1986). Measurement of provitamin A and carotenoids in fresh and canned carrots and green peas. *Journal of Food Science*, **51**(2), 534–535.

Harris, R. S. (1960). Effects of large-scale preparation on nutrients of foods of plant origin. In *Nutritional Evaluation of Food Processing*, eds R. S. Harris and H. von Loesecke, pp. 455–515. AVI, Westport, CN.

Instituto Adolfo Lutz (1985). *Normas Analíticas do Instituto Adolfo Lutz*. 3rd edn. São Paulo.

Krehl, W. A. and Winters, R. W. (1950). Effect of cooking methods on retention of vitamins and minerals in vegetables. *Journal of American Dietetic Association*, **26**, 966–1003.

Lachance, P. A. (1975). Effects of food preparation procedures on nutrient retention with emphasis upon food service practices. In *Nutritional Evaluation of Food Processing*, 2nd edn, eds R. S. Harris and E. Karmas 2nd edn, pp. 463–528. AVI, Westport.

National Academy of Science/National Council Research (NAS–NCR). (1980). *Recommended Dietary Allowances*. 9. edn. Washington, DC, pp. 51–71.

O'Neill, C. A. and Schwartz, S. J. (1992). Chromatographic analysis of cis/trans carotenoid isomers. *Journal of Chromatography*, **624**, 235–252.

Pimentel-Gomes, F. (1984). *A Estatística moderna na Pesquisa Agropecuária*. Piracicaba, São Paulo.

Quackenbush, F. W. and Smallidge, R. L. (1986). Nonaqueous reverse phase liquid chromatographic system for separation and quantification of provitamins A. *Journal of Association of Official Analytical Chemistry*, **69**(5), 767–773.

Ramos, D. M. R. (1991). Avaliação das perdas de carotenóides e valor de vitamina A durante a desidratação e a liofilização industrial de cenoura e espinafre. (Tese M.S.), Universidade Estadual de Campinas, Campinas.

Rodriguez, D. B., Raymundo, L. C., Lee, T., Simpson, K. L. and Chichester, C. O. (1976). Carotenoid pigment changes in ripening *Momordica charantia* fruits. *Annals of Botany*, **40**, 615–624.

Rodriguez-Amaya, D. B. (1993). Stability of carotenoids during the storage of foods. In *Shelf Life Studies of Foods and Beverages – Chemical, Biological, Physical and Nutritional Aspects*, ed. F. Charalambous, pp. 591–624. Elsevier Science, Amsterdam.

Silva, D. J. (1981). *Análise de Alimentos (Métodos Químicos e Biológicos)*. UFV, Impr. Univ., Viçosa, MG.

Spiers, M. (1945). The effects of fertilizer treatments, curing storage, and cooking on the carotene and ascorbic acid content of sweet potatoes. *Southern Cooperative Serial Bulletin*, **3**, 34–41.

Tannenbaum, S. R. (1976). Vitamins and minerals. In *Principles of Food Science — Food Chemistry*, ed. O. R. Fennema. pp. 417–450. Marcel Dekker, New York.

TODA Atenção ao Institucional (1994). *Revista Cozinha Industrial*, **39**, 56–64.