J. Braz. Chem. Soc., Vol. 20, No. 10, 1856-1861, 2009.
Printed in Brazil - ©2009 Sociedade Brasileira de Química
0103 - 5053 $6.00+0.00

Article

Bioactive Compounds from Several Tropical Fruits and Correlation by Multivariate Analysis to Free Radical Scavenger Activity

Gisela P.M. Barreto,[a] Marta T. Benassi[b] and Adriana Z. Mercadante*,[a]

[a]Department of Food Science, Faculty of Food Engineering, University of Campinas (UNICAMP), P.O. Box 6121, 13083-862 Campinas-SP, Brazil

[b]Department of Food Science and Technology, State University of Londrina (UEL), P.O. Box 6001, 86051-970 Londrina-PR, Brazil

O alto consumo de frutas tem sido associado à baixa incidência de doenças crônico-degenerativas, provavelmente devido à presença de compostos bioativos, como os antioxidantes, nestes alimentos. Os teores de compostos bioativos -ácido ascórbico, fenólicos totais, flavonóides e carotenóides totais - foram determinados em polpas obtidas a partir de 18 frutas tropicais adquiridas no Brasil. A atividade anti-radical livre foi avaliada pelo método ABTS. As frutas que apresentaram maior teor de compostos bioativos foram buriti, cajá-manga, canistel, murici, physalis, piquia e tucumã. Considerando a composição dos compostos bioativos analisados e as atividades anti-radical livre, as frutas foram divididas em 5 grupos, segundo Análise Hierárquica de Agrupamentos. Aplicando a Análise de Componentes Principais, a atividade anti-radical livre mostrou alta correlação com compostos fenólicos totais (r = 0,99) e flavonóides (r = 0,86); porém, a correlação encontrada foi muito pequena para ácido ascórbico (r = 0,02) e carotenóides totais (r = 0,08).

High ingestion of fruits has been associated with low incidence of chronic-degenerative diseases, probably due to the presence of bioactive compounds in these foods, such as antioxidants. The levels of bioactive compounds - ascorbic acid, total phenolics, flavonoids and carotenoids - were determined in 18 pulps obtained from tropical fruits acquired in Brazil. The free radical scavenger activity was evaluated by the ABTS assay. The fruits that showed higher levels of bioactive compounds were buriti, otaheite apple, egg-fruit, golden spoon, physalis, piquia and star nut palm. Considering the composition of bioactive compounds and free radical scavenger capacities the fruits were divided into five groups, according to Hierarchical Cluster Analysis. Applying Principal Component Analysis, free radical scavenger showed high correlation with total phenolic compounds (r = 0.99) and flavonoids (r = 0.86); however, the correlation was found to be very poor with ascorbic acid (r = 0.02) and with total carotenoids levels (r = 0.08).

Keywords: ascorbic acid, carotenoids, phenolic compounds, radical scavenger activity, Principal Component Analysis

Introduction

Brazil stands out as the third largest world producer of fresh fruits due to a favorable climate and vast territorial land. Fruit growing agribusiness is a rapidly growing activity, present in all Brazilian states, accounting for 25% of the value of domestic agricultural production. In Brazil fruit growing has experienced a remarkable expansion of the harvested area in the latest years, including the native and exotic fruits. The commercialization directed almost exclusively to the internal market, has led to gains in foreign markets, which has been crucial to the improvement of production systems and of the fruit quality.[1] Despite the enormous fruit diversity, Brazil still needs to improve in this area, since the Brazilian participation in world exports of fresh fruit and fruit products amounted only to 5.7% in 2007.[2]

The high ingestion of fruits has been associated with low incidence of degenerative diseases.[3] This effect is associated not only with the presence of antioxidants such as vitamins A, C and E, but also with other natural substances, such as carotenoids, flavonoids and other phenolic compounds, that show free radical scavenger and singlet oxygen quencher activities, or that are able to chelate metals.

*e-mail: azm@fea.unicamp.br

The ascorbic acid (vitamin C) has many biological functions and plays an important role as an antioxidant, preventing cell damages caused by oxidation.[4] Some carotenoids, such as β-carotene, show provitamin A activity and are capable to act as an antioxidant either as free radical scavenger or singlet oxygen quencher.[5] The phenolic compounds also act as antioxidants, since they are known as scavenger of free radicals.[5]

Foods generally contain a variety of compounds with free radical scavenger activity some of which are hydrophilic, for example, ascorbic acid, and others that are lipophilic, such as carotenoids, so it is interesting to use an assay capable of evaluating the antioxidant capacity of compounds with different polarities. A large number of assays have been used to evaluate the free radical scavenger activity, including the indirect $ABTS^{\bullet+}$ assay, which can be applied in the study of both hydrophilic and lipophilic antioxidant compounds, either pure or as extracts.[6,7]

The essential role of biodiversity for its sustainable use in food security and nutrition is world-wide recognized,[8] and Brazil has a wide variety of native, wild and non-commercially cultivated fruits; although few information about their constituents are available. Thus, this paper has three-fold objectives: conduct an exploratory evaluation on some bioactive chemical constituents, as ascorbic acid, phenolic compounds, total flavonoids and total carotenoids, in 18 tropical fruit pulps, verify the free radical scavenger activity in these fruits, and apply multivariate statistical analysis, specifically Principal Component Analysis (PCA) and Hierarchical Cluster Analysis (HCA), to classify the fruits according to their functional characteristics and to verify the correlation between the chemical constituents and free radical scavenger activity. Sporadic information on bioactive compounds can be found in different studies. For bacuri, cubiu red and yellow, egg fruit, golden spoon and pequia, no data related to the levels of total phenolic compounds, flavonoids, total carotenoids and free radical scavenger activity have been found in the literature so far.

Experiments

Samples

The fruits obtained in the Northern (Manaus) and in the Northeastern (Fortaleza) regions of Brazil were: bacuri, buriti, cubiu red, cubiu yellow, golden spoon, marimari, otaheite apple, piquia, star nut palm. The other fruits were acquired in the Southeastern region (Campinas): banana, egg fruit, jackfruit, loquat, nectarines, plum varieties D´Agem and Larri Ann, physalis and starfruit. The common name, the scientific one, the family and other designations used for these fruits are shown in Table 1S in the Supplementary Information.

The batches of each fruit weighted from 300 to 600 g. Different fruits were chopped, the peels and seeds removed and the pulps were homogenized in a domestic processor and then frozen. These pulps were stored at −20 °C until the analysis.

Reagents and equipments

The reagents as methanol, ethanol, acetone, petroleum ether, diethyl ether, sodium carbonate, aluminum chloride, potassium hydroxide, sodium hydroxide, all analytical grade were acquired from LabSynth (Diadema, Brazil); oxalic acid, 2,6-dichlorophenol indophenol, sodium nitrate and ascorbic acid were from Merck (Darmstadt, Germany); Folin-Ciocalteu reagent from Dinâmica Química (Diadema, Brazil) and gallic acid from Extrasynthèse (Genay, France); 2,2,9-azinobis (3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt (ABTS), potassium persulfate, catechin and 6-hydroxy-2,5,7,8-tetramethychroman-2-carboxylic acid (Trolox) were from Sigma (St. Louis, USA). The analyses were carried out on a diode array spectrophotometer from Agilent, model 8453 (Palo Alto, USA).

Extraction

The extraction was carried out with 50 mL solution of methanol/water (8:2) added to every 10 g of each sample, in a ultrasound for 20 min. This mixture was filtered through the Buchner funnel and washed with a small amount of methanol. These steps were repeated two more times by adding 20 mL of methanol/water (8:2). The three filtrates were mixed and transferred to a 100 mL volumetric flask. After 1 h in the freezer, the solution was centrifuged at 2000 rpm for 20 min, the supernatant was removed and kept in the freezer (**extract A**)**.** For each fruit, the extraction was performed in duplicate.

Determination of ascorbic acid

Ascorbic acid (AA) was determined in both the pulp and in the **extract A** by titration with 0.02% 2,6 dichlorophenol indophenol.[9] The analysis in **extract A** was done only to check possible interference in the analysis of total phenols. The analysis was performed in duplicate.

Determination of total phenols

The quantification of total phenolic compounds was performed with 1 mL of **extract A** and 1 mL of Folin-Ciocalteu

reagent.[10] Concentrations between 50 and 200 mg L^{-1} of gallic acid were used for the standard curve. The analysis was carried out in triplicate and the result was expressed as gallic acid equivalent (GAE) *per* 100 g. The presence of ascorbic acid in the extracts (0.3 to 3.8 mg AA/100 g) did not have an influence on the results of reaction.

Determination of total flavonoids

Total flavonoids were quantified according to Zhishen *et al.*[11] using 1 mL of **extract A.** Solutions with concentration of catechin from 50 to 200 mg L^{-1} were used for the construction of the standard curve. The absorbance was read at 510 nm, and the result was expressed in catechin equivalent (CE) *per* 100 g. The analysis was carried out in triplicate.

Determination of free radical scavenger activity

The determination of free radical scavenger activity was carried out in **extract A**, according to the procedure described by Re *et al.*[7] An aliquot of 1 mL of **extract A** was completely dried under nitrogen flux and immediately dissolved in 1 mL of ethanol. The sample extract (20 μL) or Trolox standard (0-15 μmol L^{-1}, final concentration) were added to 2 mL of the radical $ABTS^{\bullet+}$ solution and kept in ultrasound for 30 s. The absorbance was read in a spectrophotometer after 6 min reaction for Trolox and 60 min for the samples. The analysis was done in triplicate.

The free radical scavenger activity was determined based on the curve of standard reference (Trolox). The inhibition percentage of the sample was determined and the Trolox concentration equivalent was calculated, through the standard curve of this standard. The TEAC value was expressed in mmol L^{-1} *per* 100 g of sample.

Determination of total carotenoids

The extraction according to De Rosso & Mercadante[12] was performed in duplicate. For quantification of total carotenoids, petroleum ether was added to the dried extract, followed by measurement of the maximum absorption wavelength (λ_{max}) in spectrophotometer in the range from 220 to 750 nm. The total carotenoid concentration was calculated considering the absorption coefficient of β-carotene in petroleum ether (2592).[13]

Statistical Analysis

The chemical constituents' data and free radical scavenger activity in fruits were evaluated by Principal Component and Cluster Analyses using the software Statistics 6.0 (STATSOFT, 2001). The dendrograms were obtained by unweighted pair-group average as the linkage rule, and the Euclidean distance was considered as coefficient of similarity.

Results and Discussion

The levels of ascorbic acid (AA), total phenols, flavonoids, carotenoids and free radical scavenger activity, expressed as TEAC, of the 18 pulps of tropical fruits are presented in Table 1. Among the fruit pulps analyzed, the pequia showed the highest values of TEAC, total phenols and flavonoids; the egg fruit presented the highest level of ascorbic acid, whereas the highest concentration of total carotenoids was detected in the pulp of buriti.

The levels of total carotenoids found in buriti, physalis, star nut palm and marimari were lower than those previously reported,[12] however, among several fruits from the Amazonia (Brazil), buriti was the fruit that had the highest concentration of total carotenoids and was considered the richest source of provitamin A.[12,14]

The levels of total phenols of plums (Table 1) were within the range found by Gil *et al.*,[15] whereas the total carotenoids contents were higher than those found by these authors for plum from California (United States). Furthermore, Kim *et al.*[16] and Chun *et al.*[17] found higher levels of total phenols in plum from New York, while the levels of flavonoids were within the range presented in this study. Gil *et al.*[15] observed that the total carotenoid contents of nectarines from United States were *ca.* 3 times lower and that the levels of total phenols were similar to those presented in Table 1.

The levels of ascorbic acid, total phenols and flavonoids detected in otaheite apple (Table 1) were in the same range as those previously reported for this fruit collected in Mauritius.[18] The contents of total phenols in starfruit and of ascorbic acid in bananas reported by these authors were close to those presented in Table 1.

The content of total carotenoids of loquat (Table 1) was within the range found by Faria *et al.*[19] in four cultivars of loquat cultivated in São Paulo State (Brazil). On the other hand, the total carotenoids in jackfruit (Table 1) was lower than those found in fruits harvested in India,[20] but higher as compared to those collected in Brazil.[21]

The comparison with literature data makes clear that the edaphic-climatic conditions of the raw material (place of harvest, soil and climate) are a source of considerable variability for micronutrient composition. However, according to the wide range of values reported in some studies, variables such as stage of fruit ripeness, cultivar/variety, harvest season, temperature, among others, may have also contributed to these differences.

Table 1. Bioactive compound levels and free radical scavenger activity in tropical fruit pulps

Fruits	ascorbic acid[a] (mg AA/100 g)	total phenols[b] (mg GAE/100 g)	total flavonoids[b] (mg CE/100 g)	total carotenoids[c] (mg/100 g)	TEAC[b] ((mmol L^{-1}) /100 g)
bacuri (A)	0.5 ± 0.0	266.8 ± 3.3	103.8 ± 0.3	0.1 ± 0.0	1640 ± 30
banana (B)	0.6 ± 0.0	117.8 ± 2.4	62.8 ± 2.7	0.2 ± 0.0	180 ± 30
buriti (C)	0.7 ± 0.0	108.1 ± 6.8	71.3 ± 3.6	14.2 ± 0.5	540 ± 10
cubiu - red (D)	3.0 ± 0.4	71.9 ± 1.8	56.6 ± 4.4	0.6 ± 0.0	140 ± 10
cubiu - yellow (E)	0.4 ± 0.0	76.0 ± 1.5	54.4 ± 1.9	0.1 ± 0.0	70 ± 10
egg fruit (F)	36.5 ± 0.7	187.5 ± 1.7	90.6 ± 0.6	2.1 ± 0.0	140 ± 10
otaheite apple (G)	28.2 ± 0.3	99.4 ± 2.8	18.2 ± 0.3	0.3 ± 0.0	210 ± 10
golden spoon (H)	0.4 ± 0.0	384.5 ± 7.7	319.4 ± 3.6	1.2 ± 0.1	1840 ± 10
jackfruit (I)	1.0 ± 0.0	34.1 ± 1.0	18.3 ± 2.9	0.3 ± 0.0	50 ± 3
loquat (J)	0.6 ± 0.0	33.6 ± 0.5	24.3 ± 0.2	1.4 ± 0.0	100 ± 10
marimari (K)	0.5 ± 0.0	109.3 ± 3.9	57.8 ± 6.7	1.8 ± 0.0	230 ± 10
nectarine (L)	0.5 ± 0.0	32.0 ± 1.7	23.7 ± 1.2	0.6 ± 0.0	80 ± 10
pequia (M)	5.9 ± 0.5	4623.4±102.4	741.2 ± 36.6	0.4 ± 0.0	25280±1570
physalis (N)	20.6 ± 0.5	39.4 ± 0.7	14.4 ± 1.0	1.7 ± 0.0	130 ± 10
Plum variety D'agem (O)	1.8 ± 0.0	77.1 ± 0.5	67.4 ± 3.7	0.5 ± 0.0	100 ± 10
Plum variety Larri Ann (P)	0.8 ± 0.1	105.5 ± 5.6	78.6 ± 3.2	0.2 ± 0.0	210 ± 30
star nut palm (Q)	4.6 ± 0.1	456.8 ± 5.2	433.2 ± 10.4	4.3 ± 0.1	2150 ± 40
starfruit (R)	0.7 ± 0.0	51.9 ± 2.1	42.6 ± 2.3	0.8 ± 0.0	770 ± 70

The data correspond to the average and standard deviation of: a- four values obtained from pulps; b- six values obtained from extract A; c- two values obtained from pulps.
AA: ascorbic acid, GAE: gallic acid equivalent; CE: catechin equivalent.
The letters between parentheses refer to **Figure 2**.

The results obtained from Cluster Analysis joined the pulps into 5 distinct groups, as can be seen in Figure 1. The first group was formed by a sample of pequia characterized by a very high free radical scavenger activity probably due to the high level of phenolic and flavonoids compounds. The second group represented by samples of golden spoon and of star nut palm showed intermediate free radical scavenger activity (1840 and 2150 mmol L^{-1} *per* 100 g, respectively) with values 12.5 times lower than that of pequia. Fruits belonging to the third group, such as plum, cubiu red and yellow, banana, starfruit, jackfruit, nectarine, marimari, loquat and bacuri, were characterized by free radical scavenger activity values (50 to 1600 mmol L^{-1} *per* 100 g) smaller than those found for the previous groups. The other groups also showed low free radical scavenger activity (130 to 540 mmol L^{-1} *per* 100 g), but the fourth group, which included otaheite apple, egg fruit and physalis, had the highest level of ascorbic acid (20 to 40 mg *per* 100 g) and the fifth group, composed only by buriti, showed the highest level of total carotenoids (14 mg *per* 100 g).

The Principal Component Analysis (PCA) separated the samples according to the composition of bioactive compounds and free radical scavenger activity, supporting the formation of the same five groups described above (Figure 2). The first principal component (PC1) (57% of the variance) was correlated mainly with the contents of total phenols, flavonoids and total free radical scavenger activity, while the second component (PC2, 21% of variability) was correlated with AA and total carotenoids

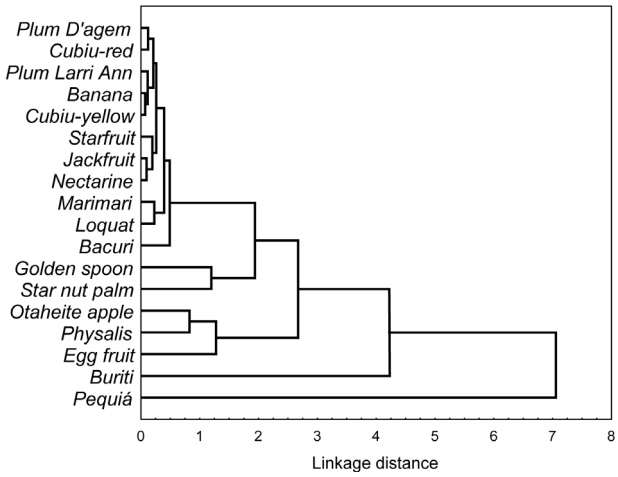

Figure 1. Dendrogram, taking into consideration the composition of bioactive compounds (AA, phenols, flavonoids and carotenoids) and free radical scavenger capacity of the 18 fruit pulps analyzed.

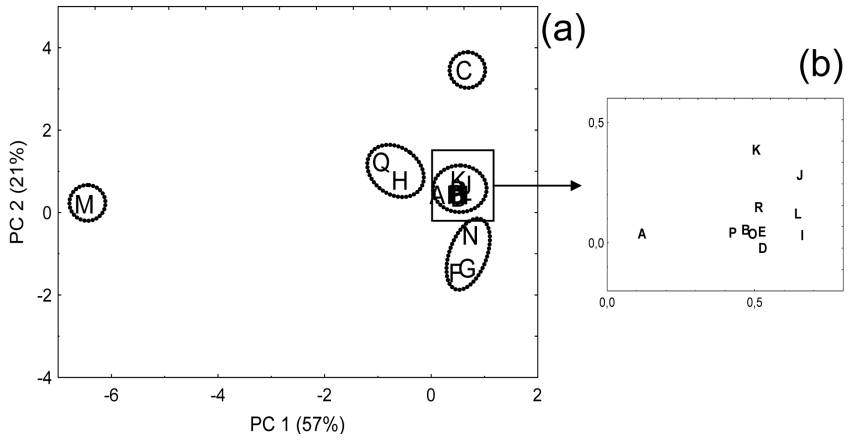

Figure 2. Configuration of the samples, obtained by Principal Component Analysis based on bioactive compounds composition and free radical scavenger capacity of 18 different fruit pulps. General projection (a) and expanded detail of the region surrounded by the square (b).

Table 2. Correlations between the variables obtained in the multivariate analyses

Variables	AA	Phenols	Flavonoids	Carotenoids	ABTS
AA	1.000	0.000	–0.068	–0.042	–0.023
Phenols	0.000	1.000	0.871	–0.086	0.999
Flavonoids	–0.068	0.871	1.000	0.015	0.866
Carotenoids	–0.042	–0.086	0.015	1.000	–0.083
ABTS	–0.023	0.998	0.866	–0.083	1.000

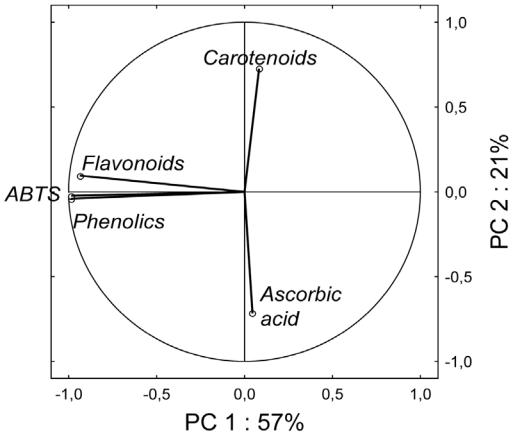

Figure 3. Projection of variables, obtained by Principal Component Analysis, from bioactive compounds composition and free radical scavenger capacity of 18 different fruit pulps.

levels (Figure 3). With regard to the correlations between variables, total phenolic compounds (r = 0.99) and flavonoids (r = 0.86) parameters were positively correlated with free radical scavenger activity measured with ABTS$^{\bullet+}$ (Table 2 and Figure 3).

According to the literature, several authors also reported high correlation between free radical scavenger activity and total phenols. Chun *et al.*[17] and Cai *et al.*[22] found a correlation of r = 0.99 and r = 0.96, respectively, both using ABTS$^{\bullet+}$ to determine the free radical scavenger activity. Silva *et al.*[23] found a better correlation by using ABTS$^{\bullet+}$ (r = 0.88) compared to the ORAC assay (r = 0.70). These authors also reported a good correlation between flavonoids and free radical scavenger activity, measured by both the ABTS$^{\bullet+}$ (r = 0.75) and the ORAC assays (r = 0.74).

The correlations obtained were very similar to those reported in the literature. Gil *et al.*[15] described good correlation between total phenols and free radical scavenger activity, using both DPPH$^{\bullet}$ as well as FRAP assays (r = 0.78 to 0.96 and r = 0.86 to 0.99, respectively), whereas a good correlation of free radical scavenger activity with the levels of ascorbic acid and carotenoids was not found. Luximon-Ramm, Bahorun & Crozier[18] observed the same behavior, with high correlation between total phenols and free radical scavenger activity (r = 0.98 with ABTS$^{\bullet+}$ and r = 0.95 through FRAP assays) and low correlation between ascorbic acid and free radical scavenger activity, either with ABTS$^{\bullet+}$ (r = 0.07) or FRAP (r = 0.04) methods.

Conclusions

Fruits such as pequia, star palm nut and golden spoon showed the highest levels of total phenolics and flavonoids, as well as free radical scavenger activity. The buriti showed the highest level of total carotenoids, but presented lower free radical scavenger activity among the fruits cited above. The study has confirmed the high positive correlation between free radical scavenger activity and phenolic compounds and flavonoids contents, besides indicating that, for the pulps studied, the carotenoids and ascorbic acid presented a smaller contribution than the phenolic compounds to free radical scavenger activity measured by the ABTS$^{\bullet+}$ assay.

Acknowledgments

The authors acknowledge FAPESP and CAPES for financial support.

Supplementary Information

The common name, the scientific one, the family and other designations used for the fruits analyzed in the present study are shown in Table 1S; and selected literature survey of chemical constituents is shown in Table 2S, both available free of charge at http://jbcs.sbq.org.br, as PDF file.

References

1. Lacerda, M. A. D.; Lacerda, R. D.; Assis, P. C. O.; *Rev. Biol. Ciênc. Terra*, **2004**, *4*, 1.
2. http://www.agricultura.gov.br/, accessed in September 2008.
3. Feskanich, D.; Ziegler, R. G.; Michaud, D. S.; Giovannucci, E. L.; Speizer, F. E.; Willett, W. C.; Colditz, G. A.; *J. Natl. Cancer Inst.* **2000,** *92*, 1812; Michels, K. B.; Giovannucci, E.; Joshipura, K. J.; Rosner, B. A.; Stampfer, M. J.; Fuchs, C. S.; Colditz, G. A.; Speizer, F. E.; Willett, W. C.; *J. Natl. Cancer Inst.* **2000**, *92*, 1740; Núnez-Sélles, A. J.; *J. Braz. Chem. Soc.* **2005**, *16*, 699.
4. Block, G.; *Age* **1993**, *16***,** 55.
5. Rice-Evans, C. A.; Miller, N. J.; Paganga, G.; *Free Radical Biol. Med.* **1996**, *20*, 933; Montenegro, M. A; Nazareno, M. A.; Durantini, E. N.; Borsarelli, C. D.; *Photochem. Photobiol.* **2002**, *75*, 353; Rios, A. de O.; Mercadante, A. Z.; Borsarelli, C. D.; *Dyes Pigm.* **2007**, *74*, 561.
6. Roginsky, V.; Lissi, E. A.; *Food Chem.* **2005***, 92*, 235.
7. Re, R.; Pellegrini, N.; Proteggente, A.; Pannala, A.; Yang, M.; Rice-Evans, C.; *Free Radical Biol. Med.* **1999**, *26*, 1231.
8. CBD (Secretariat of the Convention on Biological Diversity); Handbook of the Convention on Biological Diversity Including its Cartagena Protocol on Biosafety, 3rd ed., Montreal: 2005.
9. Benassi, M. T.; Antunes, A. J.; *Arq. Biol. Tecn.* **1988**, *31*, 507.
10. Kim, D. O.; Jeong, S. W.; Lee, C. Y.; *Food Chem.* **2003**, *81*, 321.
11. Zhishen, J.; Mengcheng, T.; Jianming, W.; *Food Chem.* **1999**, *64*, 555.
12. De Rosso, V. V.; Mercadante, A. Z.; *J. Agric. Food Chem.* **2007**, *55*, 5062.
13. Britton, G.; In *Carotenoids* vol 1B; Britton, G.; Liaaen-Jensen, S.; Pfander, H., eds.; Birkhauser: Basel, 1995, ch. 2.
14. Godoy, H. T.; Rodriguez-Amaya, D. B.; *J. Agric. Food Chem.* **1994**, *42*, 1306.
15. Gil, M. I.; Tomás-Barberán, F. A.; Hess-Pierce, B.; Kader, A. A.; *J. Agric. Food Chem.* **2002**, *50*, 4976.
16. Kim, D. O.; Chun, O. K.; Kim, Y. J.; Moon, H. Y.; Lee, C. Y.; *J. Agric. Food Chem.* **2003**, *51*, 6509.
17. Chun, O. K.; Kim, D. O.; Moon, H. Y.; Kang, H. G.; Lee, C. Y.; *J. Agri. Food Chem.* **2003**, *51*, 7240.
18. Luximon-Ramma, A.; Bahorun, T.; Crozier, A.; *J. Sci. Food Agric.* **2003**, *83*, 496.
19. de Faria, A. F.; Hasegawa, P. N.; Chagas, E. A.; Pio, R.; Purgatto, E.; Mercadante, A. Z.; *J. Food Compos. Anal.*, **2009**, *22,* 196.
20. Jagadeesh, S. L.; Reddy, B. S.; Swamy, G. S. K.; Gorbal, K.; Hegde, L.; Raghavan, G. S. V.; *Food Chem.* **2007**, *102*, 361.
21. de Faria, A. F.; De Rosso, V. V.; & Mercadante A. Z.; *Plant Foods Hum. Nutr.* **2009**, *64*, 108.
22. Cai, Y.; Luo, Q.; Sun, M.; Corke, H.; *Life Sci.* **2004**, *74*, 2157.
23. Silva, E. M.; Souza, J. N. S.; Rogez, H.; Rees, J. F.; Larondelle, Y.; *Food Chem.*, **2007**, *101*, 1012.

Received: October 28, 2008
Web Release Date: November 12, 2009

FAPESP helped in meeting the publication costs of this article.