Assessing the ascorbic acid contents in beverages and powdered juices: comparison between the experimental data and the values displayed on the product label

Avaliação do conteúdo de ácido ascórbico em bebidas e refrescos em pó: comparação entre valores experimentais e os declarados no rótulo

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ABSTRACT

The determination of ascorbic acid (AA) contents in beverages is a challenging and necessary task because consumers have to know the exact amount of this component in the product as displayed on the container label. Moreover, the food industries have to add the exact (± 20%) quantity of ascorbic acid as it is stated on the label. This study aimed at evaluating 60 ready-to-drink beverages and powdered juices marketed in Curitiba (PR, Brazil). The results were compared with the values declared on the label, and in 76.7% of the samples no statistically similar values ($p < 0.05$) were found by comparing with those stated on the packaging label. Moreover, the AA values found in this analysis were from 12 to 90% lower than those reported by manufacturers. For that reason, the ascorbic acid contents in these products should be monitored by the National Health Surveillance Agency (ANVISA).

Keywords. vitamin C, quality control, beverages.

RESUMO

A determinação do conteúdo de ácido ascórbico (AA) em bebidas é uma tarefa desafiadora e indispensável, uma vez que os consumidores necessitam ter conhecimento da quantidade exata de AA declarada no rótulo da embalagem do produto. As indústrias de alimentos devem adicionar a quantidade exata (± 20%) de AA declarada no rótulo. Neste trabalho, foram avaliadas 60 amostras de bebidas prontas para beber e sucos em pó comercializados em Curitiba (PR, Brasil). Os resultados obtidos foram comparados estatisticamente com o valor declarado no rótulo. Em 76,7% das amostras, não foram encontrados valores similares ($p < 0,05$) aos declarados nas embalagens. Além disso, os teores de AA encontrados no estudo foram de 12% a 90% menores do que os valores declarados pelos fabricantes. Estes achados indicam que os teores de vitamina C em produtos dessa natureza necessitam ser monitorados pela Agência Nacional de Vigilância Sanitária (ANVISA).

Palavras-chave. vitamina C, controle de qualidade, bebidas.

INTRODUCTION

Ascorbic acid (AA), also known as vitamin C, is a water-soluble vitamin that can be found in many biological systems and foodstuffs (fresh vegetables and fruits). Ascorbic acid plays an important role in collagen biosynthesis, iron absorption, and immune response activation[1]. It also acts as a powerful antioxidant in numerous *in vivo* protocols by buffering free-radicals that are closely associated to aging, neurodegenerative and other non-transmissible chronic diseases[2]; moreover, ascorbic acid has been proved to suppress sarcoma cell death in rats[3]. In this regard, it is noteworthy that AA has been added as an antioxidant (fruit juices, for example) in many foods, being used in medicine in pill form as a component of multivitamin tablets[4].

In order to analyze the ascorbic acid content in beverages, different analytical methods have been used to quantify the ascorbic acid present in foods and beverages such as UV-VIS spectrophotometry, high performance liquid chromatography, electrophoresis, enzymatic methods, amperometry, and titametric methods[5], in which one of the most employed oxidant agents is the iodine[6]. Iodine is a mild oxidizing agent that is capable of oxidizing only quantitatively strongly reducing substances such as ascorbic acid.

According to the Brazilian Ministry of Health[7], Ordinance nº 31 January 1998, a food is considered "source of vitamins" since 100 mL or 100 g of the product ready for consumption provide at least 7.5% of the daily recommended value of intake (DRV) for liquids, and 15% of the DRV in case of solids. To use high content or rich in vitamins 'claim' it is necessary that 100 ml or 100 g of the product ready for consumption provide at least 15% of the DRV in case of liquids, and 30% of DRV in the case of solids.

Consumers demand for safe, functional, and fresh products has been increasing continuously, contributing to the increasing consumption of fruit juices and fruit juice-based drinks. Nutritional information related to these products is necessary to guide consumers towards the best options of beverages available in the marketplace. In this context, this study aimed at evaluating 60 ready-to-drink beverages, including fruit juices, powdered juices, teas, and soy/juice blends, marketed in Curitiba (PR, Brazil).

MATERIAL AND METHODS

Sampling

A total number of 60 samples of soft drinks, powder juices, green teas, juice concentrates, ready-to-drink natural juices, and soy beverages (blended with fruit juices) were purchased from five different local markets in Curitiba, PR. A total number of 19 beverages (soy blends, teas, sodas, fruit juices) and 41 powder juices from different trademarks were acquired. Attention to the expiration date and condition of the packaging was paid for each product. A quantity of about 200 g of powder juice from the same lot and 1 liter of beverage were used for each product.

Assessment of the ascorbic acid content displayed on the label

In order to assess the adequacy of the information on the label, firstly a worksheet was assembled using the software Microsoft Excel version 2003, having the ascorbic acid content (AA) (mg/100 g) of each sample. The Resolution number 269 from 22 September 2005 from the National Agency of Sanitary Vigilance[8] was taken into consideration to compare the content of AA displayed on the label with the one determined in our study.

Determination of the ascorbic acid content

The ascorbic acid content was determined by the redox titration method, in which a 0.01 M potassium iodate (KIO_3) solution was prepared according to the method 364/IV from the Instituto Adolfo Lutz[9]. In order to perform the ascorbic acid determination, the ready-to-drink beverages were not diluted, whereas the powdered juices were diluted 5 times using Mili-Q ultra-pure water. The ascorbic acid content in every sample was obtained from three parallel determinations and were expressed as mg/100 g or mg/100 mL.

The end-point of the reaction between AA and the potassium iodate is indicated by the production of a blue-black product. As long as the ascorbic acid is present in the test sample, the triiodine is quickly converted to iodide ion, and no blue-black iodine-starch product is formed in the medium. However, when all ascorbic acid has been oxidized, the excess of triiodide (in equilibrium with iodine) reacts with the starch to form the expected blue-black color. This method was validated and showed a good linearity, precision and accuracy when ready-to-drink beverages were assessed[10]. In comparison to other analytical procedures, this titration method is considered simple, inexpensive, and no time-consuming once it does not require preliminary extraction.

Statistical evaluation

Results were presented as mean ± standard deviation. The *t*-Student test for a single sample was applied to the data in order to compare the value displayed on the label with the one determined experimentally. The probability level (p-value) for each evaluation was also provided. For this analysis, the software Statistica v. 7 (StatSoft, Tulsa, OK, USA) was used. A p-value below 0.05 was considered significant, that is, a p-value higher than 0.05 means that no statistical difference was found between the experimental value and the one on the label.

RESULTS AND DISCUSSION

In our study, a great number of products (n = 60) available on the marketplace was selected in order to confer reliability and provide scientifically sound results. Table 1 summarizes the data obtained from the determination of the ascorbic acid content and the statistical comparison between experimental results and the values displayed on the label of each product. It is possible to observe that only 6 samples out of 60 presented a non-significant p-value ($p > 0.05$) in accordance with the Student *t*-test, while 8 products did not present nay information about the ascorbic acid content on the label. It means that 76.7% of the products did not contain a similar ($p > 0.05$) ascorbic acid content as compared to the value displayed on the label. Likewise, Quirós et al.[11] measured the ascorbic acid content in fruit juices and soft drinks using the same experimental procedures adopted in this study. By applying this method, they found that the amounts of vitamin C declared on the labels of many drinks are not what they obtained experimentally. Among a total of 17 fruit juices, soft drinks and isotonic drinks, only 2 samples presented a similar value to the one indicated on the label. The authors also evaluated how the vitamin C content present in orange juices and tea drinks varies during the shelf life. After a six-day storage period, the juice barely lost 8% of its ascorbic acid amount, while in teas the vitamin C content fell by 54% at 4 °C and practically disappeared at room temperature.

From the comparison between the mean AA contents and the value indicated on the label it is possible to observe that the ascorbic acid content remained between 12 to 90% lower than the values declared on the label of the products. Although 6 samples showed no significant difference in the assessment, 4 samples were liquid juice, and only two were powdered juices.

Other authors have already analyzed the ascorbic acid content in fruit juices, sodas, and other beverages marketed in Brazil and verified that a great part of the products does not contain the specified content displayed on the label. A study conducted by José et al.[12] showed that powdered juices marketed in Brazil did not contain the content of AA displayed on the label: the contents varied from 3.45 to 12.12 mg/100 mL. The study emphasized that powdered juices are not sources of AA and that a tougher control and monitoring should be performed by governmental agencies; Silva et al.[13] concluded that powdered juices cannot be considered sources of AA, once the samples presented low AA contents (0.67 – 32.00 mg/100 g of powder). They also verified that eight out of eleven samples presented wrong information about the AA content displayed on the label, and it would be necessary to intake a total number of 4 to 67 glasses (200 mL) of diluted powdered juice to provide the daily recommended value of intake for adults (≥ 19 years old); Abreu et al.[14] evaluated the AA content of soy extracts blended with fruit juices (mango, coconut, pineapple, passion fruit, and guava) and found AA values ranging from 0.09 to 3.30 mg/100 g. These values were also lower than the ones declared on the label. The authors also mentioned that for some samples the difference was 10 times lower than the value declared on the packaging.

Other authors such as Santos et al.[15] observed that among six brands of mango nectar only two had lower values than those provided on the label. In the same sense, other fruit nectars presented higher (up to 65%) AA levels than the values indicated on the label. Yamashita et al.[16] investigated the vitamin C stability in fresh acerola, pasteurized acerola pulp, and acerola juice, stored at -12 °C/18 °C for up 4 months. The ascorbic acid content of acerola juice at the beginning of the experiment was 1,511 mg/100 g, and after the storage period at -12 °C this content decreased to 869 mg/100 g, while the sample stored at -18 °C had 1,223 mg/100 g. The initial ascorbic acid content in the pasteurized pulp was 1360 mg/100 g and at the end of 4 months it was 1,314 mg/100 g. The pasteurized juice stored at room temperature presented an initial ascorbic acid content of 988 mg/100 g, and at the end of 4 months of storage the content was 673 mg/100 g.

Table 1. Ascorbic acid content displayed on the label of beverages *versus* the content determined experimentally

Samples	Type	Ascorbic acid – label – (mg/100 g)	Ascorbic acid – experimental – (mg/100 g)	Significance (p-level)
1	Powder juice	214.29	85.85 ± 0.98	0.007
2	Powder juice	166.67	75.67 ± 1.23	0.021
3	Powder juice	214.29	86.47 ± 1.77	0.004
4	Powder juice	272.00	179.96 ± 0.40	0.031
5	Powder juice	272.00	153.68 ± 0.44	0.009
6	Powder juice	ND	7.80 ± 0.06	NA
7	Powder juice	111.67	70.82 ± 1.54	0.006
8	Powder juice	111.67	64.42 ± 1.49	0.004
9	Powder juice	111.67	67.59 ± 1.56	0.005
10	Powder juice	214.29	270.61 ± 7.99	0.054
11	Powder juice	200.00	119.54 ± 6.80	0.043
12	Powder juice	471.43	240.43 ± 11.63	0.013
13	Powder juice	200.00	104.86 ± 8.66	0.012
14	Powder juice	200.00	97.07 ± 8.54	0.008
15	Powder juice	471.43	205.98 ± 23.57	0.001
16	Powder juice	200.00	119.64 ± 10.70	0.006
17	Powder juice	166.67	85.00 ± 9.14	0.005
18	Powder juice	166.67	99.95 ± 8.50	0.008
19	Powder juice	166.67	87.73 ± 7.76	0.006
20	Powder juice	166.67	76.61 ± 5.68	0.004
21	Powder juice	166.67	75.02 ± 4.46	0.013
22	Powder juice	166.67	77.66 ± 5.20	0.011
23	Powder juice	155.56	281.85 ± 14.67	0.004
24	Powder juice	155.56	288.35 ± 12.38	0.019
25	Powder juice	155.56	338.43 ± 21.45	0.004
26	Powder juice	111.67	84.10 ± 8.23	0.068
27	Powder juice	111.67	66.42 ± 4. 37	0.039
28	Powder juice	111.67	80.41 ± 2.89	0.017
29	Powder juice	166.67	66.23 ± 3.98	0.002
30	Powder juice	214.29	101.14 ± 4.67	0.011
31	Powder juice	272.00	164.34 ± 6.82	0.003
32	Powder juice	100.00	21.87 ± 1.49	0.015
33	Powder juice	562.50	108.51 ± 10.77	0.008
34	Powder juice	366.67	92.23 ± 7.69	0.003
35	Powder juice	366.67	61.35 ± 4.84	0.002
36	Powder juice	366.67	76.49 ± 5.42	0.005
37	Powder juice	471.43	256.24 ± 15.25	0.001
38	Powder juice	200.00	123.46 ± 10.53	0.035
39	Powder juice	111.67	78.90 ± 6.79	0.024
40	Powder juice	366.67	84.78 ± 8.44	0.004
41	Powder juice	366.67	88.92 ± 9.28	0.004
42	Liquid	7.58	39.75 ± 1.77	< 0.001
43	Liquid	9.00	7.63 ± 0.21	0.070
44	Liquid	100,00	7.04 ± 0.33	< 0.001
45	Liquid	ND	1.01 ± 0.15	NA
46	Liquid	ND	0.50 ± 0.09	NA
47	Liquid	ND	0.50 ± 0.07	NA

Samples	Type	Ascorbic acid – label – (mg/100 g)	Ascorbic acid – experimental – (mg/100 g)	Significance (p-level)
48	Liquid	ND	0.50 ± 0.03	NA
49	Liquid	ND	1.38 ± 0.07	NA
50	Liquid	ND	1.01 ± 0.02	NA
51	Liquid	23.00	9.98 ± 0.13	0.029
52	Liquid	7.50	7.42 ± 0.05	0.644
53	Liquid	12.00	5.64 ± 0.05	0.001
54	Liquid	12.00	7.75 ± 0.04	0.001
55	Liquid	4.25	5.14 ± 0.31	0.104
56	Liquid	4.25	7.04 ± 0.03	0.005
57	Liquid	ND	0.75 ± 0.05	NA
58	Liquid	3.00	5.03 ± 0.21	0.006
59	Liquid	23.50	14.75 ± 0.44	0.045
60	Liquid	9.00	7.04 ± 0.06	0.076

Note: ND = not declared on the label.

These findings need to be seriously considered by governmental agencies such as the National Health Surveillance Agency (ANVISA), once they suggest that the food industry has not paid sufficient attention to the ascorbic acid content added to the products or even to measure accurately this substance in such products before and after the unitary operations used to their development. Another hypothesis to be taken into consideration is that the food industry may have added unstable chemical forms of ascorbic acid to the products, which may oxidize throughout transportation and shelf-life.

Ascorbic acid is easily oxidized as compared to other vitamins. It is known that AA is stable only in acidic medium, in the absence of light, oxygen and heat, and the factors favoring its degradation are the presence of alkali, oxygen, heat, light, metals, and some enzymes[17]. Taking into consideration the type of packaging, the thermal processing and other unit operations, it is possible to assume that all these factors may also be responsible for the differences between experimental values and those declared on the labels. It is known that the air incorporation during the processing steps and the storage temperatures enhance aerobic degradation reactions[18]. In the case of hermetically sealed anaerobic conditions, ascorbic acid decomposes to 2,5-dihydro-2-furanoic acid to become carbon dioxide and furfural. The furfural undergoes polymerization as an active aldehyde and can be combined with amino acids, contributing to the juice darkening[19].

Losses may also occur due to different types of equipments used during the process and / or by chemical oxidation of ascorbic acid and/or thermal degradation through blanching, cooking, pasteurization and dehydration[20]. Freitas et al.[21] evaluated the ascorbic acid stability in acerola juices produced by the hot fill (glass bottles) and aseptic (carton packaging) processes during 350 days of storage in similar conditions of marketing (25 ± 2 °C). The authors observed a reduction in ascorbic acid content of 23.61% for the hot fill process and 35.95% for the aseptic process.

A study conducted by Kabasakalis, Siopidou and Moshatou[22] concluded that the AA content in different commercial fruit juices marketed in Greece ranged from 2.4 to 43 mg/100 mL. The storage of such products in closed containers at room temperature for a period of 4 months resulted in ascorbic acid losses ranging from 29% to 41%. Commercial orange juice, when stored in open containers in the refrigerator for 31 days, lost 60% to 67% of its ascorbic acid content while fresh orange juice lost between 7% and 13% of the AA content. Open containers of commercial fruit juice, when stored outside the refrigerator for 10 days, lost 12.5% of their ascorbic acid content, while the refrigerated juice for the same period, lost up to 9% of AA.

Although ANVISA allows a variation range of ± 20% in relation to the mean value of the nutrient displayed on the label, we believe that this value is extremely high for beverages due to the main reasons: 1) this high variance range allows the industry not to monitor suitably each lot of the final products; 2) a less intense development of new packaging systems to prevent deterioration of chemical constituents such as vitamins, sterols, phenolic compounds, carotenoids, among others; 3) the higher the variance range, the lesser attention to quality control is paid by food industries; 4) with less attention paid to quality control, consumers do not get what they believe they actually buy, and implications on satisfaction, trust, and loyalty to brand get compromised. Thus, ANVISA should monitor such products evaluated in this study and possibly reduce the acceptable range for the values of nutrients, including ascorbic acid, displayed on the label. This action would prevent eventual relapses on quality control and more attention on food processing would be paid in the beverage industry.

CONCLUSIONS

The study showed a high variability between the vitamin C content experimentally determined and the one declared by the manufacturer. Among the 60 samples analyzed in this study, 76.7% of the products had their vitamin C content statistically different ($p < 0.05$) from the amount displayed on the container label, with a content ranging from 12 to 90% lower than the value declared on the label. These results indicate it is necessary to perform a stricter quality control in powdered juices and ready-to-drink beverages.

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REFERENCES

1. Sahoo PK, Mukherjee SC. Immunomodulation by dietary vitamin C in healthy and aflatoxin B1-induced immunocompromised rohu (*Labeo rohita*). Comp. Immun. Microb. Infect. Dis. 2003;26(1):65-76.
2. Chao JCJ, Huang CH, Wu SJ, Yang SC, Chang NC, Shieh MJ, et al. Effects of β-carotene, vitamin C and E on antioxidant status in hyperlipidemic smokers. J Nutrit Biochem. 2002;13(7):427-34.
3. Frank J, Flaccus A, Schwarz C, Lambert C, Biesalski HK. Ascorbic acid suppresses cell death in rat DS-sarcoma cancer cells induced by 5-aminolevulinic acid-based photodynamic therapy. Free Radic Biol Med. 2006;40(5):827-36.
4. Davis MB, Austin J, Partridge D. *Vitamin C: its chemistry and biochemistry*. Cambridge: Royal Society of Chemistry; 1991.
5. Campos FM, Ribeiro SMR, Lucia CMD, Pinheiro-Sant'ana HM, Stringheta PC. Optimization of methodology to analyze ascorbic and dehydroascorbic acid in vegetables. Quím Nova. 2009;32(1):87-91.
6. Andrade RSG, Diniz MCT, Neves EA, Nóbrega JA. Determinação e distribuição de ácido ascórbico em três frutos tropicais. Eclet Quím. 2006;27(1):1-6.
7. Brasil. Portaria SVS/MS nº 31, de 13 de janeiro de 1998. Regulamento técnico referente a alimentos adicionados de nutrientes essenciais. Diário Oficial da União, Brasília, DF, 16 jan 1998.
8. Brasil. Ministério da Saúde. Secretaria Nacional de Vigilância Sanitária. Resolução RDC nº 269, de 22 de setembro de 2005: regulamento técnico sobre a ingestão diária recomendada (IDR) de proteína, vitaminas e minerais. Diário Oficial da União, Brasília, DF.
9. Instituto Adolfo Lutz. *Normas Analíticas do Instituto Adolfo Lutz: métodos químicos e físicos para análise de alimentos*. 4. ed. Brasília; 2005.
10. Suntornsuk L, Gritsanapun W, Nilkamhank S, Paochom A. Quantitation of vitamin C content in herbal juice using direct titration. J Pharmac Biomed Anal. 2002;28:849-55.
11. Quirós ARB, Fernández-Arias M, López-Hernández J. A screening method for the determination of ascorbic acid in fruit juices and soft drinks. Food Chem. 2009;116(2):509-12.
12. José AB, Souza JM, Lavinas FC. Avaliação do teor de ácido ascórbico e dos parâmetros físico-químicos de amostras de preparados sólidos para refresco, sabor limão. Saúde & Amb Ver. 2009;4(1):11-2.
13. Silva PT, Filho E, Lopes MLM, Valente-Mesquita VL. Sucos de laranja industrializados e preparados sólidos para refrescos: estabilidade química e físico-química. Ciênc Tecnol Aliment. 2005;25(3):597-602.
14. Abreu CRA, Pinheiro AM, Maia GA, Carvalho JM, Sousa PHM. Avaliação química e físico-química de bebidas de soja com frutas tropicais. Aliment Nutri. 2007;18(3):291-6.
15. Santos E, Batista E, Carvalho L, Salgado M, Souza P. Avaliação dos teores de vitamina c em néctares de manga. IX ENPPG / IX ENICIT / III SIMPIT. Instituto Federal de Educação, Ciência e Tecnologia do Ceará. Fortaleza, CE. 2009.
16. Yamashita F, Benassi MT, Tonzar AC, Moriya S, Fernandes JG. Produtos de acerola: estudo da estabilidade de vitamina C. Ciênc Tecnol Aliment. 2003;23(1):92-4.
17. Oliveira MEB, Bastos MSR, Feitosa T, Branco MAAC, Silva MGG. Avaliação de parâmetros de qualidade físico-químicos de polpas congeladas de acerola, cajá e caju. Ciênc Tecnol Aliment. 1999;19(3):326-32.
18. Lima VLAG, Mélo EA, Lima LS. Avaliação da qualidade de suco de laranja industrializado. Bol CEPPA. 2000;18(1):95-104.
19. Shaw PE, Nagy S, Rouseff RL. The shelf life of citrus products. *In*: Charalambous G. Shelf life studies of foods and beverages: chemical, biological, physical and nutricional aspects. Amsterdam: Elsevier; 1993.
20. Maia GA, Souza PHM, Santos GM, Silva DS, Fernandes AG, Prado GM. Efeito do processamento sobre componentes do suco de acerola. Ciênc Tecnol Aliment. 2007;27(1):130-4.
21. Freitas CAS, Maia GA, Costa JMC, Figueiredo RW, Sousa PHM, Fernandes AG. Estabilidade dos carotenoides, antocianinas e vitamina C presentes no suco tropical de acerola (*Malpighia emarginata* DC.) adoçado envasado pelos processos *hot-fill* e asséptico. Ciênc Agrotecnol. 2006;30(5):942-9.
22. Kabasakalis V, Siopidou D, Moshatou E. Ascorbic acid content of commercial fruit juices and its rate of loss upon storage. Food Chem. 2000;70(3):325-8.