(CC BY 4.0) | ISSN 2525-3409 | DOI: http://dx.doi.org/10.33448/rsd-v9i12.10964 Addition of lipids to cookies containing orange-fleshed sweet potatoes increases carotenoid retention¹

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Adição de lipídios em biscoitos contendo batata-doce de polpa alaranjada elevam a retenção de carotenoides

Incremento en la retención del contenido de carotenoides por adición de lípidos en galletas con papas dulces anaranjadas

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Laysa Borges Tomas de Sousa ORCID: https://orcid.org/0000-0002-0800-8256 Universidade Federal do Espírito Santo, Brazil E-mail: laysabts@hotmail.com Mariana Guadagnini Lisboa Soares ORCID: https://orcid.org/0000-0002-4607-6110 Universidade Federal do Espírito Santo, Brazil E-mail: mgulisboa@hotmail.com Adriana Paula da Silva Minguita ORCID: https://orcid.org/0000-0001-9938-8671 Embrapa Agroindústria de Alimentos, Brazil E-mail: adriana.minguita@embrapa.br José Luiz Viana de Carvalho ORCID: https://orcid.org/0000-0002-9734-2087 Embrapa Agroindústria de Alimentos, Brazil E-mail: jose.viana@embrapa.br Érica Aguiar Moraes ORCID: https://orcid.org/0000-0003-1223-1466 Universidade Federal do Espírito Santo, Brazil E-mail: erica.a.moraes@ufes.br

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Erika Madeira Moreira da Silva

ORCID: https://orcid.org/0000-0003-4428-9769 Universidade Federal do Espírito Santo, Brazil E-mail: erika.alimentos@gmail.com José Luis Ramírez Ascheri ORCID: https://orcid.org/0000-0001-7449-8815 Embrapa Agroindústria de Alimentos, Brazil E-mail: jose.ascheri@embrapa.br

Abstract

Sweet potato is one of the seven major staple crops in the world. Although most creamfleshed sweet potatoes are produced locally, orange-fleshed sweet potato (OFSP) is an excellent source of beta-carotene and can be promoted to alleviate vitamin A deficiency. The aim of this study was to evaluate the retention of total carotenoids (TC) and all-trans-betacarotene (ATBC) after processing and storage of cookies with different proportions of OFSP and margarine. Cookies formulated with 18 to 22.23 g/100 g of OFSP and 28.57 to 34.6 g/100 g of margarine were analyzed by spectrophotometry and high-performance liquid chromatography. The formulation containing the highest content of OFSP (22.23 g/100 g) and 32.84 g/100 g of margarine showed greater retention of TC and ATBC after processing. Among the other samples with the same proportion of OFSP, the one containing the highest margarine concentration (34.6 g/100 g) has shown a greater capacity to maintain TC and ATBC during storage (up to 56 days), demonstrating lipids important role in the preservation of these components.

Keywords: Carotenoids; Retention; Processing; New products; Baked goods.

Resumo

A batata-doce é uma das sete principais culturas básicas do mundo. Embora a maioria da batata-doce de polpa cor creme seja produzida localmente, a batata-doce de polpa alaranjada (BDPA) é uma excelente fonte de beta-caroteno e pode ser promovida para aliviar a deficiência de vitamina A. O objetivo deste estudo foi avaliar a retenção de carotenoides totais (CT) e todo-trans-beta-caroteno (TTBC) após o processamento e armazenamento de biscoitos com diferentes proporções de BDPA e margarina. Os biscoitos formulados com 18 a 22,23 g / 100 g de BDPA e 28,57 a 34,6 g / 100 g de margarina foram analisados por espectrofotometria e cromatografia líquida de alta eficiência. A formulação com maior teor de

BDPA (22,23 g / 100 g) e 32,84 g / 100 g de margarina apresentou maior retenção de CT e TTBC após o processamento. Dentre as demais amostras com a mesma proporção de BDPA, a que apresentou maior teor de margarina (34,6 g / 100 g) apresentou maior capacidade de manutenção de CT e TTBC, durante o armazenamento (até 56 dias), conferindo aos lipídios papel importante na preservação desses componentes.

Palavras-chave: Carotenoides; Retenção; Processamento; Novos produtos; Panificação.

Resumen

La papa dulce anaranjada (PDA) es uno de los siete cultivos básicos más importantes del mundo. Aunque la mayor parte de la PDA es producida en América latina, la PDA es una excelente fuente de betacaroteno y puede promoverse para aliviar la deficiencia de vitamina A. El objetivo de este estudio fue evaluar la retención de carotenoides totales (TC) y todo-trans-beta-caroteno (ATBC) después del procesamiento y almacenamiento de galletas con diferentes proporciones de PDA y margarina. Las galletas formuladas con 18 a 22.23 g/100 g de PDA y 28.57 a 34.6 g/100 g de margarina se analizaron mediante espectrofotometría y cromatografía líquida de alta resolución. La formulación con el mayor contenido de PDA (22.23 g/100 g) y 32.84 g/100 g de margarina mostró una mayor retención de TC y ATBC después del procesamiento. Entre las otras muestras con la misma proporción de PDA, la que contenía el mayor contenido de margarina (34.6 g/100 g) mostró mayor capacidad para retener TC y ATBC, durante el almacenamiento (hasta 56 días), otorgando a los lípidos un papel importante en la conservación de estos componentes.

Palabras clave: Carotenoides; Retención; Procesamiento; Nuevos productos; Panadería.

1. Introduction

Sweet potato is one of the seven major world staple crops and is being cultivated in more than 100 countries. The crop plays an important role in developing countries due to its hardiness, low input requirements and versatility (Laurie, Faber, Adebola, & Belete, 2015).

The sweet potato has technological properties that allow its use in the production of baked goods (Rodriguez-Amaya, Nutti, & Carvalho, 2011). Among the varieties of baked goods, cookies stand out as a highly consumed food worldwide (ABIMAPI, 2019), as they are well accepted, highly stable, have a long shelf life, are easily transported, do not require preparation intermediaries and are rapidly consumed.

Although most cream-fleshed sweet potatoes are more commonly found, the orange-

fleshed sweet potato is an excellent source of beta-carotene and can be promoted to supplement vitamin A deficiency (Burri, 2011). Only 64 % of children in need are receiving the life-saving benefits of vitamin A supplementation, and more than 140 million children are being left behind, in countries that have been prioritizing national-level vitamin A supplementation programs (UNICEF, 2018).

Beta-carotene is also an important biocompound with several well-established molecular and biological properties (Islam, Nusrat, Begum, & Ahsan, 2016). Beauregard sweet potato, which is a rich source of beta-carotene, other bioactive compounds and carbohydrates (Niu et al., 2019; Zhu, & Wang, 2014), is a potential raw-material for new edible products development.

During cooking, carotenoid retention in OFSP is between 70% and 95% (Rautenbach, Faber, Laurie, & Laurie, 2010). In the formulation of new products containing OFSP, the presence of fat and some additives (e.g., emulsifiers) has a positive effect on the incorporation of all-trans-beta-carotene into micelles, increasing the retention of these nutrients (Bengtsson, Brackmann, Enejder, Alminger, & Svanberg, 2010).

Although carotenoid retention is a known process, information about carotenoid loss in packaged products during storage is scarce and, at times, conflicting (Lesková et al., 2006). Therefore, it is important to develop technological solutions for greater conservation of micronutrients, through the appropriate choice of the packaging type for processed and ultraprocessed products.

Studies that evaluated the impact of processing and packaging on the content of carotenoids in biofortified products have pointed out that packaging with a high barrier to oxygen, nitrogen, luminosity and moisture results in a longer product shelf life and greater maintenance of nutrients such as carotenoids and all-trans-beta-carotene (Taleon, Mugode, Cabrera-Soto, & Palacios-Rojas, 2017; Júnior, Ito, Ribeiro, Da Silva, & Alves, 2018; Nogueira et al., 2018).

The aim of this study was to evaluate the retention of total carotenoids and all-transbeta-carotene during the processing and quantification of these components during the storage of cookies formulated with different proportions of orange-fleshed sweet potato and margarine.

2. Methodology

This is an experimental quantitative study (Pereira, Shitsuka, Parreira, & Shitsuka, 2018) carried out at the Federal University of Espirito Santo, Health Sciences Center – Vitória, ES – Brazil.

2.1 Raw material and cookie formulation

For the cookies, the OFSP cultivar Beauregard, provided by Embrapa Hortaliças (Brasília - DF), was used. Other ingredients [corn starch, refined sugar, margarine (interesterified fats and soy lecithin)] were purchased from the local commerce in Vitória, ES.

After cleaning, OFSP was sliced and submitted to microwave cooking for 10 minutes, peeled and kneaded manually. The ingredients were mixed, and the dough was homogenized and molded into a 4-cm-diameter standard shape. Afterwards, the cookies were placed in an electric oven at 160 °C for 25 minutes.

2.2 Experimental design

A second-order central composite rotational design (Box, Hunter, & Hunter, 1978) was used to study the effect of different proportions of OFSP (11.98 g/100 g to 24 g/100 g) and margarine (22.58 g/100 g to 34.6 g/100 g), totaling 10 treatments (four factorial points, four axial points and two central points, using $\alpha = 1.41$ as the rotability). This experimental design was applied to submit those treatments to a sensory analysis (acceptance test using a nine-point hedonic scale – Minim, 2013) in order to choose the most accepted samples (unpublished data). Only selected samples were submitted to carotenoid and all-trans-beta-carotene determinations. For this stage, a control sample was included (with cream-fleshed sweet potato, using the proportions of sweet potato and margarine referring to the central point of the design – cream-fleshed sweet potato: 18 g/100 g and margarine: 28.57 g/100 g).

2.3 Sample selection criteria

According to Dutcosky (2019), an acceptance rate of 70% or more is considered good. However, for more rigorous selection, the criteria adopted were the presentation of a higher average, as well as a higher acceptance rate (>80%), for all evaluated items (color, aroma,

flavor, texture, appearance and purchase intention), also taking into account the highest possible level of OFSP in the samples.

2.4 Extraction and determination of total carotenoids and all-trans-beta-carotene

For total carotenoid analysis, 3 g of cookies were weighed and macerated with celite. The carotenoids were extracted with acetone, and the ketone extract was partitioned in petroleum ether (Rodriguez-Amaya, 2001). Total carotenoids were quantified with a spectrophotometer (model UV-1800, Shimadzu - Tokyo, Japan) at 450 nm. The concentration of total carotenoids (micrograms per 100 grams of sample) was determined according to equation 1 (Pacheco, 2009).

 $\textit{Concentration} \; (\mu g/100g) = \; \frac{abs.x\; dil.x\; vol.x\; 10000}{2592\; x\, sm.}$

(Eq. 1)

Where:

abs. = absorbance

dil. = extract dilution (when used in the linear range)

vol. = volume of the volumetric flask used (mL)

sm. = sample mass (g)

The analysis of the extract for quantification of all-trans-beta-carotene was performed with a high-performance liquid chromatograph (HPLC) W600 (Waters, Milford, USA) equipped with an automatic injector 717 plus, YMC column (Waters®) C30 and a W26996 photodiode array detector, at 450 nm. The oven temperature was adjusted to 35 °C. The flow rate of the mobile phase was 0.8 mL/min, and the injection volume was 15 μ L for the standard extracts produced in the laboratory and sample extracts. The mobile phase consisted of methanol (A) and methyl tert-butyl ether (B). An elution gradient was employed with 80 mL A/20 mL B, 0.0–0.5 min; 75 ml A/25 ml B, 0.5–15.0 min; 15 ml A/85 ml B, 15.0–15.5 min; 10 ml A/90 ml B, 15.5–16.5 min; and finally, 80 ml A/20 ml B, up to 28 min (Pacheco, 2009). The R² of the calibration curve was 0.9973, according to the equation (β-carotene: Y = 5.17*105.X - 6.45*104). The detection limits were 0.012 µg/mL, and the quantification limit was 0.5 µg/mL.

2.5 Total carotenoids and all-trans-beta-carotene retention

The retention of total carotenoids and all-trans-beta-carotene was evaluated before and after cookie processing. The identification of all-trans-beta-carotene was performed by comparing the retention time obtained between the standard and the selected treatments, under the same conditions. The conversion of total carotenoids and all-trans-beta-carotene values into true retention (TR) was calculated according to Murphy, Criner and Gray (1975), according to equation 2.

 $\% TR = \frac{\text{carotenoid content in the processed sample (g)x post processing sample weight (g)}{\text{carotenoid content in raw sample (g)x sample weight before processing (g)}} x 100$

(Eq. 2)

2.6 Cookies storage

Cookies were packaged in metallized bioriented polypropylene packaging (BOPP/met) and sealed (Sulpack model SP400T, Caxias do Sul, RS, Brasil). The samples were kept at room temperature (±27 °C), stored for 84 days and analyzed every 28 days.

2.7 Data analysis

The data distribution was tested using the Kolmogorov–Smirnov test. The results of carotenoid analysis were subjected to analysis of variance (one-way ANOVA) followed by the Tukey test. A probability of 5% was considered for all evaluations. All statistical analysis were performed using Statistica 10.0 software.

3. Results and discussion

3.1 Selected samples

The Beauregard orange-fleshed sweet potato as well as the most accepted treatments (cookies) according to sensory analysis are available in Figures 1 and 2, respectively.

Figure 1 - Internal and external aspect of orange-fleshed sweet potato, Beauregard cultivar.



Source: Portal EMBRAPA.

Figure 2 - Cookies selected from the sensory analysis and control treatment. OFSP: orangefleshed sweet potato; CFSP: cream-fleshed sweet potato; CT: control treatment.



Source: Authors.

Figure 1 shows the internal and external aspects of OFSP Beauregard and its intense orange color. In Figure 2, the control sample was included for comparison purposes. It is noted that color is the first attribute that stands out among the samples, with the sample containing the highest content of OFSP having a more evident yellow-orange color. Considering other aspects, it can be noticed that there are no visible differences in the cookies even with different proportions of OFSP and margarine, which can be good for the sensory acceptance.

3.2 Total carotenoids and all-trans-beta-carotene retention

The retention of TC and ATBC was greater in the sample containing the highest content of OFSP (T3) (Table 1). The other samples did not differ in terms of retention

(approximately 40%). However, among the other samples with the same OFSP content (T8 and T9), the one containing the highest margarine content showed greater retention of ATBC. These nutrients were not detected in the control sample.

Table 1 - Total carotenoids and all-trans-beta-carotene retention in selected cookies made

 with orange-fleshed sweet potato (OFSP) and the control treatment (CT).

Treatments	Total carotenoids (%)	All- trans-beta-carotene (%)
T3	$89.46 \pm 1.02^{\text{a}}$	108.77 ± 1.02^{a}
Τ8	$43.01 \pm 1.40^{\text{b}}$	50.34 ± 1.40^{b}
T9	39.22 ± 0.64^{b}	$30.63 \pm 1.49^{\circ}$
TC	ND	ND

- Data are expressed as means and standard deviations for three replicates. Means with the same letters in the same column, for each treatment, are not significant, considering a 95% confidence interval (Tukey test). OFSP: margarine (g/100 g): T3 = 22.23: 32.84; T8 = 18: 34.6; T9 = 18: 28.57. Cream-fleshed sweet potato: margarine TC = 18: 28.57. ND: not detected. Source: Authors.

The composition of the cookie formulation and the amount of heat applied during processing are important factors in the retention of carotenoid compounds.

In the formulation of cookies, the presence of soy lecithin can act in the formation of colloidal solutions called micelles, since this element acts as an emulsifier. Thus, the greater presence of lipids in the dough may aid in the penetration of carotenoids into these micelles, which are naturally attracted by margarine due to their lipophilic nature. The micelles then act as a protector to the carotenoids from possible degradation, increasing their retention (Bechoff et al., 2015; Bengtsson et al., 2010).

The retention and bioaccessibility of beta-carotene determine its bioavailability (Bechoff et al., 2011). Studies that evaluate the effect of heat treatment on carotenoids suggest that it may increase their activity by inactivating oxidative enzymes and denaturing carotenoid-protein complexes in plant cells. Therefore, cooking in an electric oven, steaming, frying, and microwaving, with few exceptions, increase true retention when compared with raw food (Sultana, Anwar, & Iqbal, 2008; Natella, Belelli, Ramberti, & Scaccini, 2010; Mazzeo et al, 2011; Vimala, Nambisan, & Hariprakash, 2011). In addition, the destruction of the cell matrix during the cutting and preparation of OFSP (heat treatment followed by kneading) increases the bioaccessibility of carotenoids (Bengtsson et al., 2010). On the other hand, during thermal processing there may be a conversion from the trans isomeric form to the cis form, with less power for conversion to retinol (Rodriguez-Amaya et al., 2011).

Therefore, dietary lipid intake is necessary to increase bioavailability through the absorption and conversion of beta-carotene to retinol (Lemmens et al., 2014).

3.3 Determination of total carotenoids and all-trans-beta-carotene during storage

The carotenoid contents in all treatments decreased during storage up to 84 days (Table 2). Until the end of this period, the average reduction in carotenoid content varied by approximately 50%-60%. However, upon evaluation of samples following 56 days of storage, it was observed that sample T8, which contained the highest margarine content, showed 36.9% and 39% reductions in the contents of TC and ATBC, respectively. This reduction was smaller than those observed in other samples (T3 = reduction of 50% and 50.3% and T9 = reduction of 44.4% and 49.3%, of TC and ATBC, respectively).

Table 2	- Total	carotenoids	and	all-trans-beta-	-carotene	during	storage of	of cookies	containing
orange-	fleshed	sweet potato	and t	the control tre	atment.				

Treatments	Days	Total carotenoids	All-trans-beta-carotene
		(µg/100 g)	(µg/100 g)
	1	2700.50 ± 97.28^{a}	1949.00 ± 77.78^{a}
Т3	28	2044.50 ± 68.58^{b}	$1581.00 \pm 7.07^{\rm b}$
15	56	1352.50 ± 45.96^{c}	$968.50 \pm 42.42^{\rm c}$
	84	$1187.00 \pm 48.08^{\circ}$	$962.00 \pm 48.79^{\circ}$
	1	2001.00 ± 11.31^{a}	1339.50 ± 31.81^{a}
ΤQ	28	1894.50 ± 30.40^{a}	$1234.50\pm2.12^{\mathrm{a}}$
10	56	1261.00 ± 32.52^{b}	816.00 ± 42.42^{b}
	84	$780.00\pm32.52^{\circ}$	$507.00 \pm 67.88^{\rm c}$
	1	2093.00 ± 48.79^{a}	$1665.60 \pm 17.67^{\rm a}$
TO	28	1646.50 ± 9.19^{b}	$1322.00\pm4.24^{\text{b}}$
19	56	$1163.50 \pm 23.33^{\circ}$	$843.50 \pm 19.09^{\rm c}$
	84	964.50 ± 10.60^{c}	$789.00\pm9.89^{\circ}$
	1	<ql< td=""><td>ND</td></ql<>	ND
TC	28	<ql< td=""><td>ND</td></ql<>	ND
IC IC	56	<ql< td=""><td>ND</td></ql<>	ND
	84	<ql< td=""><td>ND</td></ql<>	ND

- Data are expressed as means and standard deviations for three replicates. Means with the same letters in the same column, for each treatment, are not significant, considering a 95% confidence interval (Tukey test). QL: spectrophotometer readings below the quantification limit. ND: not detected. OFSP:

margarine (g/100 g): T3 = 22.23 : 32.84; T8 = 18 : 34.6; T9 = 18 : 28.57. Cream-fleshed sweet potato: margarine TC = 18 : 28.57. Source: Authors.

It has suggested that a higher fat content has a positive effect on the ATBC micellarization, which may have contributed to protection against its deterioration (Bengtsson et al., 2010; Hadnađev, Hadnađev, Pojić, Rakita, & Krstonošić, 2015). A study carried out with the use of different packaging in the conservation of all-trans-beta-carotene in BDPA chips demonstrated that the use of BOPP/met did not significantly alter the content of this micronutrient for up to 62 days (Júnior et al., 2018).

The loss of carotenoids during storage can occur through isomerization (from the trans to the cis form) and non-enzymatic oxidation, as they are heat-treated foods. Of these, oxidation is the major cause of carotenoid loss and depends, in this case, on the availability of oxygen and the level of light passing through the package, in addition to the chemical structure of the carotenoid (Rodrigues-Amaya et al., 2011). Variations in room temperature can also cause changes in the structure of carotenoids, reducing their capacity for provitamin A (Bechoff, Taleon, Carvalho, Carvalho, & Boy, 2017).

4. Final Considerations

Cookies with a higher content of OFSP (22.23 g/100 g) and 32.84 g/100 g of margarine showed higher retention of total carotenoids and all-trans-beta-carotene after processing. Among the other samples with the same OFSP content, the one containing the highest margarine content (34.6 g/100 g) showed a higher capacity to maintain total carotenoids and all-trans-beta-carotene levels (up to 56 days of storage), pointing lipids important role in maintaining these components.

In-depth studies that aim to evaluate the physicochemical and nutritional characteristics of the cookies under other storage conditions considering modified atmosphere and vacuum conditions are suggested. Also, the evaluation of lipid peroxidation, the formation of micelles as a protective factor against carotenoids, the *in vivo* contribution of the ingestion of cookies to the requirements of vitamin A and sensory tests for risk groups.

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Percentage of contribution of each author in the manuscript

Laysa Borges Tomas de Sousa – 20 % Mariana Guadagnini Lisboa Soares – 8 % Adriana Paula da Silva Minguita – 6 % José Luiz Viana de Carvalho – 6 % Érica Aguiar Moraes – 20 % Erika Madeira Moreira da Silva – 30 % José Luis Ramírez Ascheri – 10 %