Functional characterization of biofortified sweet potato flour dried at different temperatures

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Abstract

Sweet potato (Ipomoea batatas (L.) Lam.) is an abundant source of carotenoids, and the processing of its tubers as flour is an alternative to add value and expand its use. The objective was to characterize flour from the pulp of biofortified sweet potato dried at different drying temperatures. Biofortified sweet potatoes (accession CNPH1210) were dried in a forced air ventilation oven, under four temperature conditions: 45, 55, 65 and 75 °C. Drying continued until the sliced roots reached moisture content of 14 % (d.b.). Fibers, phenolic compounds, carotenoids, β-carotene, antioxidant capacity by ABT and DPPH, scanning electron microscopy, high-field magnetic resonance imaging, infrared absorption spectrometry were evaluated. Flours obtained from sliced roots dried at 45, 55, 65 and 75 ºC showed stability in their morphological characteristics. The temperature of 45 °C was the most indicated for the production of biofortified sweet potato flours, as the produced flours had higher contents of bioactive compounds, being a significant source of carotenoids.

Keywords: Dehydration; Phenolic Compounds; β-carotene; Antioxidant Capacity; Magnetic Resonance.

1. Introduction

Sweet potato (*Ipomoea batatas*) is a rustic tuberous vegetable of wide adaptability, cultivated in virtually the entire Brazilian territory (Silva et al., 2004). It is a crop of great economic and social importance, with the advantages of low production cost, easy cultivation, prolonged harvesting and resistance to pests and diseases (Silva et al., 2008).

The roots of orange-fleshed sweet potatoes have stood out due to their nutritional and functional value, especially due to the high concentration of β -carotene, provitamin A (Van Jaarsvel et al., 2005; Bovell-Benjamin, 2007; Donado-Pestana et al., 2012), as well as antioxidants (Islam et al., 2016), carbohydrates (Zhu & Wang, 2014), fibers (Mullin, 1994), minerals and vitamins (Suarez et al., 2016).

In Brazil, sweet potatoes is commercialized almost entirely in the form of raw roots. The access to this vegetable by consumers and its acquisition are still limited, either by the low production (IBGE, 2020), by the prices offered, which vary greatly over the years (CEAGES, 2020), or by eating habits.

In this context, sweet potato flour can be an alternative to increase consumption and marketing, because this vegetable in the form of flour, besides having an increased shelf life, can be introduced as food in regions with less access to the fresh root or that do not have the habit to consume it. Flour can be incorporated into several other products and can even partially replace wheat flour in manufacture of breads (Nzamwita et al., 2017), cakes, biscuits and other products (Rodrigues-Amaya et al., 2011).

Studies have demonstrated that sweet potato flour can add nutritional value to processed products due to its high content of carotenoids. When quantifying bioactive compounds in biofortified sweet potato flour, Trancoso-Reyes et al. (2016) observed carotenoid content of 137.49 μg g^{-1}, as well as Rodrigues et al. (2016), who obtained carotenoid content of 196.6 μg g^{-1} in biofortified sweet potato flour.

Nunes et al. (2016) reported that breads enriched with biofortified sweet potato showed good sensory acceptance. Infante et al. (2017) demonstrated an increase in the nutritional and sensory quality of sorghum biscuit formulations enriched with biofortified sweet potato flour. Studies conducted by Pereira et al. (2019)

demonstrated positive effects replacing wheat flour with biofortified sweet potato flour in panettone doughs, including the reduction of fermentation time, predominance of yellow color and presence of new volatile compounds. Gomes et al. (2017), studying the sorghum extrusion process associated with biofortified sweet potatoes, found that the addition of biofortified sweet potatoes led to an increase in iron bioavailability due to the presence of β-carotene.

However, the drying conditions of sweet potato in oven, such as temperature and exposure time, can affect the quality of the flour produced. Some studies have already been conducted evaluating the impact of temperature and drying time on food composition. Miranda et al. (2010) evaluated the effect of drying temperature (40, 50, 60, 70 and 80 °C) on phenolic content and antioxidant capacity of quinoa seeds, Multari et al. (2018) studied the influence of drying temperature (60 and 70 ºC) on the contents of phenolic compounds and carotenoids in quinoa seeds, and Liu et al. (2019) assessed heat treatment (65, 80 and 95 °C) on the physicochemical composition of potato.

Thus, the objective of the present study was to determine the best drying condition of biofortified sweet potato pulp for the maintenance of its nutrients.

2. Material and Methods

2.1. Preparation of sweet potato flours

The roots of biofortified sweet potato (genotype CNPH1210) were developed by Embrapa Vegetables (Brasília, DF, Brazil) and cultivated in experimental field of the Federal Institute of Goiás (Rio Verde, GO, Brazil). The sweet potato used for the experiment had moisture of 74% (d.b.). The roots were washed and peeled manually, and then the pulp was sliced into chips with approximate dimensions of 4.6 x 4.0 x 0.3 cm (length, width and thickness) (Borges et al., 2008), using a domestic grater. The slices were dried in an oven with air circulation, with velocity of 1.0 m s^{-1} and four different temperatures: T1: 45 °C for 7 h, T2: 55 °C for 6 h, T3: 65 °C for 5 h and T4: 75 °C for 3 h, and the moisture content was standardized by the breakage percentage calculation to 14%. Each sample, composed of 0.3 kg of roots, was evenly distributed in rectangular aluminum trays (25 x 10 cm), without perforation. After drying, the samples were ground in a mill (Tecnal TE-650, Piracicaba, SP, Brazil) and placed in polypropylene plastic bags, sealed and stored at 4 ºC in B.O.D. type chambers in the absence of light until the analyses.

2.2. Dietary fiber

The total, soluble and insoluble dietary fiber contents of the biofortified sweet potato flours were determined by the gravimetric-enzymatic method described by AOAC (1995), adapted by Freitas et al. (2008). Due to the low lipid content of the flours, it was not necessary to degrease them. The samples were hydrolyzed by thermoresistant α-amylase (A-3306 Sigma), protease (P-3910 Sigma) and amyloglucosidase (A-9913 Sigma) to remove protein and starch. After enzymatic hydrolysis, insoluble fiber was filtered and separated, and soluble fiber was precipitated using four volumes of ethanol (95%). The alcoholic solution was then filtered and the precipitated residues were washed with ethanol (95%) and acetone, dried and weighed. The total dietary fiber was calculated from the sum of insoluble dietary fiber and soluble dietary fiber.

2.3. Total phenolic compounds

The spectrophotometric determination of phenolic compounds was performed according to the methodology described by Singleton (1965), using the Folin-Ciocalteu reagent. Extracts of sweet potato flour were prepared using 1 g of flour and 20 mL of 70% ethanol. The solution was homogenized for 15 min and then filtered through Whatman filter paper nº 4. Absorbance was read at 765 nm in spectrophotometer (Biospectro SP-220, São Paulo, SP, Brazil).

2.4. Antioxidant activity: tests with DPPH and ABTS

The antioxidant capacity by the stable radical 2,2-diphenyl-1-picryl-hydrazyl (DPPH) was evaluated following the procedure adapted from Brand-Williams et al. (1995). A volume of 100 µL of extracts previously prepared for phenolic compound analysis and 3.9 mL of DPPH solution were added. Then, the solution was homogenized in a tube shaker (Merse M81, Taquara, RJ, Brazil). Readings were performed in a spectrophotometer (Biospectro SP-220, São Paulo, SP, Brazil) at 517 nm, 30 min after the beginning of the reaction.

To determine the antioxidant capacity by ABTS (2,2-azinobis (3-ethyl-benzothiazoline-6-sulfonic acid), a 30-µL aliquot of the extract was transferred to test tubes with 3.0 mL of the ABTS radical and homogenized. The samples were read in a spectrophotometer at 734 nm, after 6 min of the mixture. The ability to scavenge the radical ABTS and DPPH was determined using a standard curve obtained with Trolox (0 to 300 µmol L^{-1}) and ABTS·+ and DPPH. The results were expressed in Trolox (µmol 100 g^{-1}).

2.5. Total carotenoids

Total carotenoids were determined using 1 g of biofortified sweet potato flour and 50 mL of acetone and, after that, the mixture was homogenized (Marconi MA102, Piracicaba, SP, Brazil) for 1 min. This extract was vacuum-filtered in Büchner funnel with Whatman filter paper nº 4 and the filtrate was collected in Kitasato flask. The extraction was repeated until the residue was white. Then, 40 mL of oil ether were added; then, 250 mL of distilled water were added and, after separation of the lower aqueous phase, the washing was repeated three more times to remove all acetone. The ethereal phase was collected in a 50-mL volumetric flask and the readings were performed in a spectrophotometer at 450 nm (Rodrigues-Amaya, 2001). The results were expressed in µg g^{-1}.

2.6. β-carotene

β-carotene determination was performed according to Rodrigues-Amaya (2001), using the extracts previously prepared for the analysis of total carotenoids. A 5-mL aliquot of extract dried in liquid nitrogen was collected and resuspended in 5 mL of acetone. β-carotene was quantified in a high-performance liquid chromatograph (HPLC), SPD-M20A (Shimadzu Co., Kyoto, Japan), with C18 column (ODS Hypersil 150 × 4.6 mm, 5 µm), mobile phase with acetonitrile, methanol and ethyl acetate (ratio ranging from 10 to 80% in 22.5 min), flow rate of 0.8 mL min^{-1}, photodiode array detector (PDA) with scanning from 300 to 550 nm, column temperature of 22 °C and external standardization. The identification of β-carotene was performed by comparing the retention time of the peaks found for the samples with those obtained from a commercial standard (Sigma-Aldrich, São Paulo, SP, Brazil). The results were expressed in µg g^{-1}. Data

were collected using *Labsolution* software (version 5.57).

2.7. Scanning electron microscopy

Microstructural analysis was used to evaluate the morphology of flour particles, mainly starch granules, under scanning electron microscope (JSM-6610 Jeol®), equipped with EDS, ThermoScientific NSS Spectral Imaging. The samples were previously degreased using the Soxhlet method (AOAC, 2000), placed in aluminum stubs with double-sided tape, covered by an ultra-thin gold film (electrically conductive material), allowing the principle of operation of the SEM, i.e. emission of electron beams with 5 kV acceleration voltage by a tungsten filament. The micrographs were performed with magnifications of 500x and 1000x.

2.8. High-field high-resolution solid-state NMR

The high-resolution nuclear magnetic resonance (NMR) analysis of carbon 13 (^{13}C) of starch (solid samples) was performed using the cross-polarization pulse sequence with magic angle spinning (spinning of 10 KHz) and high-power decoupling known as CP-MAS. Hexamethylbenzene, the methyl line referenced at 17.3 ppm, was used to reference the chemical displacement. The instrument used was an Avance III HD 400 MHz spectrometer (Bruker, Berlin, Germany). The acquisition parameters used for the analyses were waiting time of 5 sec, scan number 1024 and contact time 1 ms. Samples (approximately 200 mg) were placed in 4-mm-diameter cylindrical zirconium oxide rotors (Schiller et al., 2001).

2.9. Infrared absorption spectrometry (FTIR)

Fourier transform infrared absorption spectroscopy (FTIR) analyses were performed on Vertex 70 spectrometer (Bruker, Berlin, Germany). The samples were analyzed by attenuated reflectance (ATR) using a diamond crystal as support. The spectra were acquired with 32 scans and with a resolution of 4 cm^{-1}.

2.10. Statistical analysis

A completely randomized design (CRD) was used, with one sweet potato genotype, four drying temperatures and five replicates. The data were assessed by analysis of variance - ANOVA (GLM-ANOVA) of the SAS® Statistical Analysis System v. 8.0 (SAS Institute Inc., 1999). The means were compared by Tukey test at 5% significance level.

3. Results and discussion

The contents of total, soluble and insoluble fibers (Figure 1) of the samples dried at 75 ºC were lower than those found in samples dried at 45 ºC. Thermal processing reduces fiber content due to solubilization and subsequent degradation of dietary fiber components (Arrigoni et al., 1986). Similar values were reported in a study on biofortified sweet potatoes, 4.47 g 100 g^{-1} for soluble fibers and 4.60 g 100 g^{-1} for insoluble fibers (Mullin, 1994).

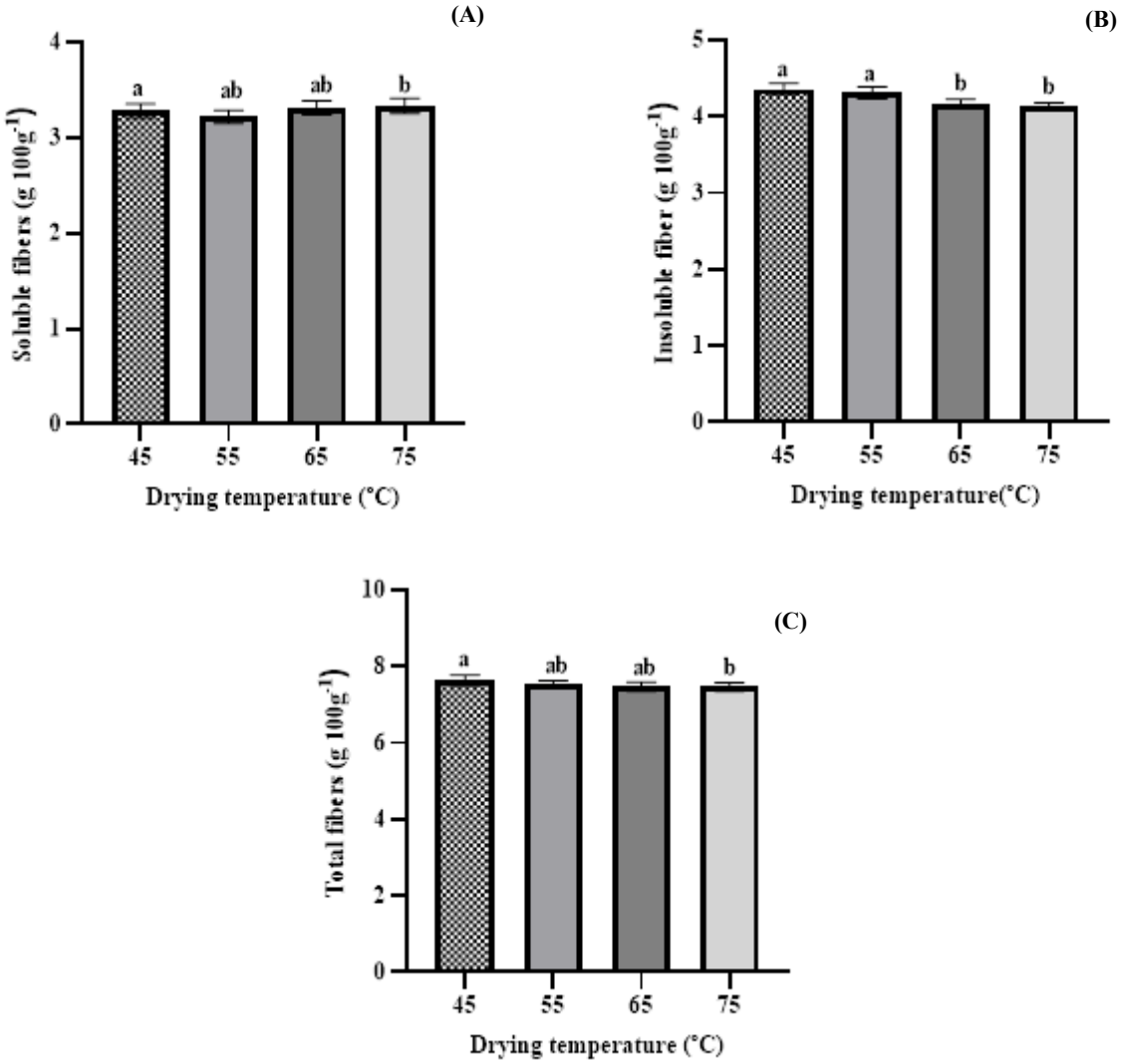

Figure 1. (A) soluble fibers, (B) insoluble fibers and (C) total fibers (g 100 g^{-1}) of biofortified sweet potato flours, dried at 45, 55, 65 and 75 °C.

Degradation of bioactive compounds was higher with the increase in drying temperature (Figure 2). The higher the drying air temperature and relative humidity, the faster the chemical reactions inside the food (Lajolo and Mercadante, 2018).

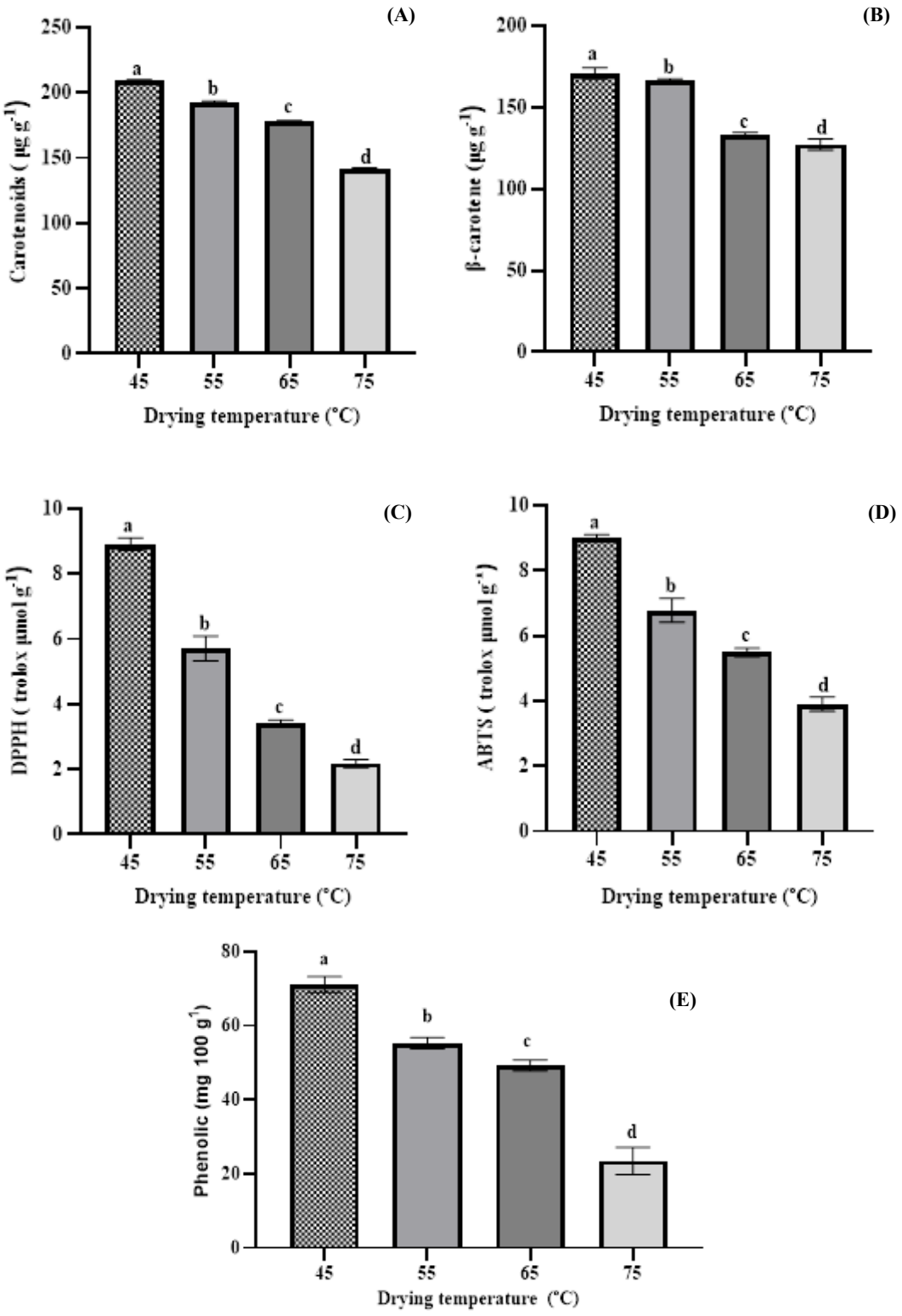

Figure 2. (A) total carotenoids (µg g^{-1}), (B) β-carotene (µg g^{-1}), antioxidant activity by (C) DPPH (trolox µmol g^{-1}), (D) ABTS (trolox µmol g^{-1}) and (E) phenolic compounds (mg 100 g^{-1}) of biofortified sweet potato flours, dried at 45, 55, 65 and 75 °C.

Increase in drying temperature caused greater losses of carotenoids (Figure 2A). Higher temperature and the presence of oxygen in the drying process in the oven provided a more favorable environment for the

occurrence of oxidative reactions. Carotenoid degradation comprises several simultaneous transformations: isomerization with the formation of oxidative products and volatile compounds (Zepka et al., 2009; Zepka & Mercadante, 2009; Zepka et al., 2014).

Hagenimana et al. (1999) observed that the drying of sweet potato slices in an oven at 6 ºC for 12 h reduced the total content of carotenoids by 30%. Bechoff et al. (2009) reported that β-carotene losses, after sweet potato drying, ranged between 16 and 34%.

It was observed that the increment in the drying temperatures led to the greatest losses of these phenolic compounds. Similarly, Donado-Pestana et al. (2012) reported that thermal processing reduced the contents of phenolic compounds in biofortified sweet potato flour. Other authors have also reported losses of these compounds due to thermal degradation (Miranda et al., 2009; Pedreschi et al., 2011).

Thermal processing negatively affects the contents of phenolic compounds, causing the softening and rupture of cells and consequently facilitating their hydrolysis (Arruda et al., 2005).

The antioxidant capacity of sweet potato flour samples decreased significantly with the increase in drying temperature (Table 3C). This may have occurred due to the reduction of bioactive compounds in the samples caused by thermal degradation (Tang et al., 2015). Studies have reported lower antioxidant capacity with increased temperature during drying (Roy et al., 2007; Negri et al., 2009; Garcia-Perez et al., 2010; Djendobi et al., 2012).

Figure 3 presents the microstructures of the samples of biofortified sweet potato flour. Protein bodies (P) usually have spherical shapes, starch granules (S) may show oval shapes with lipids (L) adhered to their surface (Damodaran et al., 2010), and fibers (F) are porous and irregular structures (Santana and Gasparetto, 2009). These characteristics can be identified in the images obtained from sweet potato flours (Figure 3).

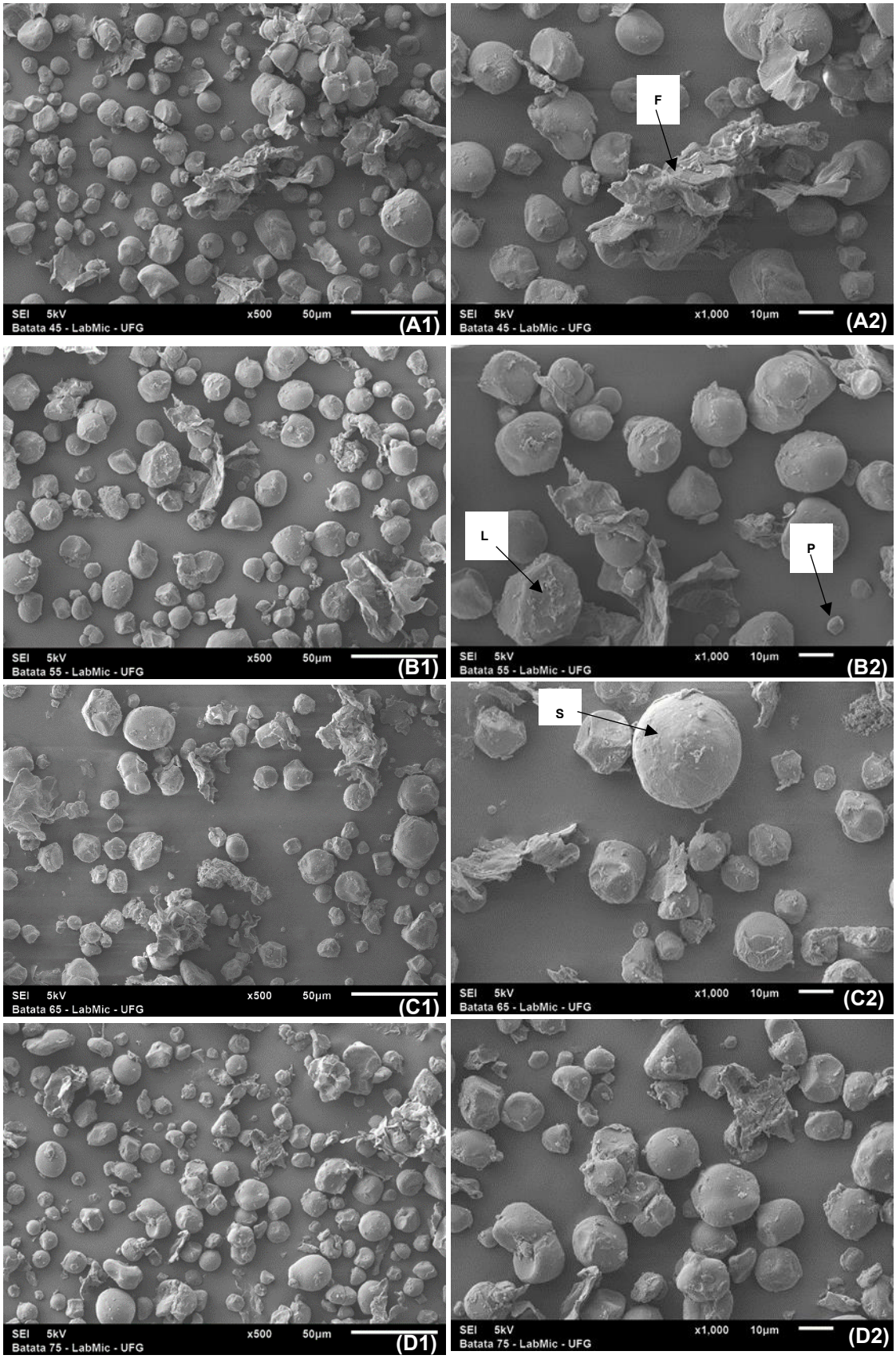

Figure 3. Scanning electron microstructure of biofortified sweet potato flours, dried at (A1) 45 °C magnified 500X, (A2) 45 °C magnified 1000X; (B1) 55 °C magnified 500X, (B2) 55 °C magnified 1000X; (C1) 65 °C magnified 500X, (C2) 65 °C magnified 1000X; (D1) 75 °C magnified 500X, (D2) 75 °C 1000X increase. In the images, the highlighted letters refer to the following structures: S - starch; F

- fiber; L - lipids; and P - protein.

There was no morphological change in the material with the temperature variation between 45 and 75 °C. It can be noted that the shape, apparent size distribution and types of structures found were similar. Gonçalves et al. (2009), when analyzing the effect of heat treatment on sweet potato starch, observed that there were no changes in starch structures. The authors report that the granules showed oval and polygonal shapes, with smooth surfaces, with no evidence of ruptures or cracks after heat treatment at 90 ºC in an oven for 16 h and microwave for 1 h.

Figure 4 shows the Fourier-transform infrared (FTIR) absorption spectra of the samples of biofortified sweet potato flour, which are characteristic of starch (Craig et al., 2015).

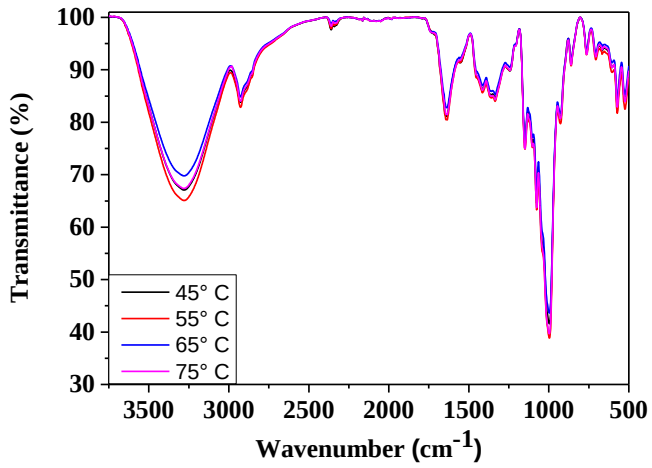

Figure 4. Fourier-transform infrared (FTIR) absorption spectra of samples of biofortified sweet potato flour dried at 45, 55, 65 and 75 °C.

The wide signal between 3000 and 3500 cm^{-1} is due to the symmetrical stretches of O-H bonds, the signal between 2960 and 2860 cm^{-1} is due to the symmetrical and asymmetric stretches of C-H bonds, the signal between 1680 and 1600 cm^{-1} is due to the asymmetric stretch of O-H groups, and the intense signal between 1160 and 900 cm^{-1} is related to the symmetrical stretch of C-O bonds.

Since all spectra were virtually identical, there were no changes between the samples dried at different temperatures, as verified in the electron scanning microscopy analyses.

Figure 5A shows the NMR signals of ^{13}C obtained with the CP-MAS method for the different samples of biofortified sweet potato flours, Figure 6b presents the expansion of the spectra between 55 and 90 ppm. In Figure 6A, the signal at about 102 ppm is the signal of carbon 1 of the glucose molecules, the signal at about 82.5 ppm is of carbon 4, the signal between 77 and 67 ppm is related to carbons 2, 3 and 5, and the signal at about 63 ppm is due to carbon 6.

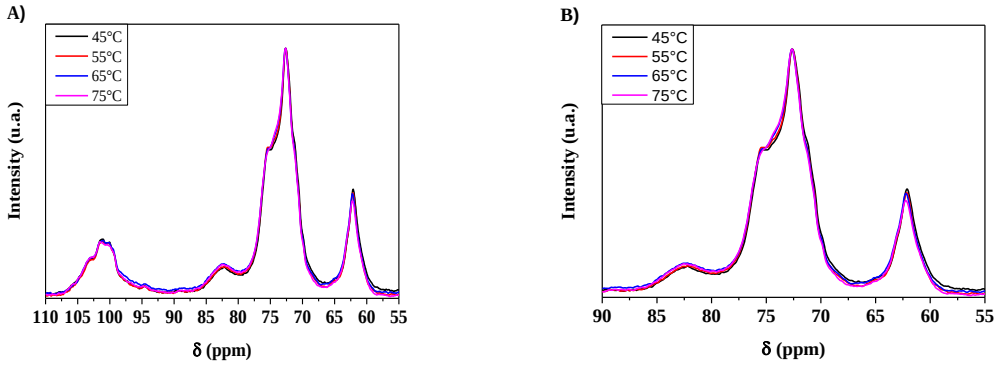

Figure 5. Signals obtained by the CP-MAS method of biofortified sweet potato flours dried at 45, 55, 65 and 75 °C.

In Figure 5B it is possible to observe that there is a small difference in the most intense signal at about 75 ppm, which is an indication that at temperatures of 65 and 75 °C there may have been changes in the crystalline/amorphous ratio of the material, compared to samples dehydrated at temperatures of 45 and 55 °C, in which the material absorbs more water, which in turn has greater mobility.

When starch molecules are heated, part of the structure loses crystallinity, and water molecules form hydrogen bridges between amylose and amylopectin, originating the amorphous regions (Singh et al., 2003).

In samples dried at 65 and 75 °C, there may have been starch retrogradation with formation of more crystalline zones. In the process of starch retrogradation, molecules that have been heated form a gel and, when it is cooled, hydrogen bonds are formed, originating the crystalline structure (Yu et al., 2009; Jiamjariyatam et al., 2015).

4. Conclusion

Drying temperatures did not alter the morphological structures and infrared spectra of the samples dried at 45, 55, 65 and 75 °C. Phenolic compounds, antioxidant capacity, carotenoids, β-carotene decreased with the increase in drying temperature, and in nuclear magnetic resonance signals there was a small change in the crystalline/amorphous ratio of the samples dried at 65 and 75 °C. The temperature of 45 °C was the most indicated for the production of flours, since the produced flours had higher contents of bioactive compounds, being an important source of carotenoids.

5. Acknowledgement

To IF Goiano, Embrapa Vegetables, Embrapa Instrumentation, Labmic, CAPES, FAPEG, FINEP and CNPq, for the indispensable financial support to conduct this study.

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