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Physicochemical, antioxidant, rheological, and sensory properties of juice produced with guava pulp and peel flour

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Abstract: Juice formulations containing guava pulp and different amounts (0%, 1%, 3%, and 5%) of its by-product flour (GBF) were developed and evaluated for antioxidant activity, total phenolic compounds, physicochemical, rheological, microbiological and sensorial parameters. The GBF addition to guava juice increased the acidity and altered the color and rheological parameters, especially at higher levels (3% and 5%). There was an increase in the content of dietary fiber, anthocyanins and antioxidant activity with GBF addition, but no changes occurred in the soluble solids, total phenolic compounds, and ascorbic acid content. The guava juice containing 1% GBF received better sensory scores when compared with the 3% and 5% GBF formulations. Storage at 4° C for 21 days affected most of the parameters examined, but the microbiological parameters remained stable. Shelf life of 14 days is recommended to maintain the physicochemical and antioxidants characteristics of guava juice.

Key words: Anthocyanins, ascorbic acid, by-product, fiber, phenolic compounds.

INTRODUCTION

Guava (*Psidium guajava*) is a tropical fruit produced on a large scale in Brazil (578,803 t in 2018) (IBGE 2018), which is considered one of the largest guava producers in the world. The red variety has a pleasant aroma and flavor and high nutritional value (Ninga et al. 2018). It contains substantial amount of minerals and vitamins, with 2–3-fold more vitamin C than fresh orange juice (Akesowan & Choonhahirun 2013).

Guava is commonly described as “superfruit” due to its high levels of polyphenolic antioxidants, like anthocyanins (Flores et al. 2015), which scavenge free radicals and reactive oxygen species involved in the occurrence of some human diseases, like cancer and atherosclerosis. Therefore, studies had focused on the use of antioxidants, especially of naturals

sources, to overcome oxidative damage caused by the unstable molecules (Vyas et al. 2010, Almulaiky et al. 2018).

Guava is perishable, so, it is usually consumed as fresh, or processed into various commercial products, including pulp, squash, nectar, puree, paste, canned slices in syrup, and juice. Among these, the juice has become economically important because of its flavor, aroma and high nutritional value (Almulaiky et al. 2018, Ninga et al. 2018).

In Brazil, it is estimated that 55% of the total guava volume produced is processed, which corresponds to 233,368 t, and after processing, a large amount of waste is generated (about 30% of the raw material), constituted by peels and seeds, corresponding to around 70,000 t of residue (Amaya-Cruz et al. 2015, Lima et al. 2019). In this sense, there is increasing interest in applying

fruit processing by-products as functional food ingredients since they are a source of dietary fiber (DF) and most of the beneficial bioactive compounds remain in the fruit waste (Balasundram et al. 2006). DF has an important role in health due to its beneficial physiological effects, such as lowering cholesterol and obesity, preventing cardiovascular and gastrointestinal diseases, and also helping to minimize the blood glucose level (Mudgil et al. 2012). Furthermore, polyphenol compounds associated with DF can scavenge free radicals, protecting cells against oxidative damage in the intestinal ecosystem (Pérez-Jiménez et al. 2013).

Despite the potential of guava by-products as a functional ingredient (Lima et al. 2019), there is a lack of researches about its addition to food products. One challenge involving the inclusion of DF into different food matrices is the potential to produce undesirable sensory changes, especially concerning color and texture. As a consequence, alternative sources of DF from wastes to obtain high-quality fiber products are being pursued (Pla et al. 2012). It is especially important in the juice production, considering there are few studies in the literature aiming at DF addition to this kind of product (Moreira et al. 2014, Cassani et al. 2016) so, it was not found works that investigated the inclusion of guava by-products in fruit juices or other kinds of beverages. In addition, during processing and storage, fruit juices tend to exhibit increased turbidity, discoloration and degradation of their nutrients and active substances (You et al. 2018), which must be studied.

Therefore, this work aimed to develop juice formulations containing guava pulp and different amounts of its by-product flour, evaluating their physicochemical and antioxidant properties during storage, as well as their sensorial acceptance and rheological parameters.

MATERIALS AND METHODS

Materials

The guava fruits were obtained in the local market from Teodoro Sampaio, São Paulo, Brazil. The following reagents were used in the physicochemical analysis: boric acid (Anidrol), hydrochloric acid (Anidrol), sodium hydroxide (Anidrol), sulfuric acid (Anidrol), protease enzyme (Savinase, Novozymes), MES reagent (2-(N-Morpholino) ethanesulfonic acid) (Sigma-Aldrich Chemical), TRIS reagent (Tris(hydroxymethyl) aminomethane) (Sigma-Aldrich Chemical), ethanol (Anidrol), acetone (Anidrol).

The reagents used in the determination of total phenolic compounds, antioxidant activity, anthocyanins and ascorbic acid were: Folin-Ciocalteu reagent (Sigma-Aldrich Chemical), gallic acid (Sigma-Aldrich Chemical), sodium carbonate (Anidrol), DPPH (2,2-difenil-1-picril-hidrazil, Sigma-Aldrich Chemical), Trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid, Sigma-Aldrich Chemical), sulfuric acid (Quimex), potassium iodide (Synth), starch (Cromoline), potassium iodate (Synth), potassium chloride (Nuclear), sodium acetate (Nuclear), acetic acid (Anidrol).

Production of guava by-product flour and guava juice

Red guava fruit (variety Pedro Sato) at the mature stage were selected based on the color of the skin (yellowish-green) and the ratio of soluble solids (SS) to the acidity of the pulp (15 ± 2). The pulp and the seeds were separated from the peel by cutting. The peels were triturated in a grinder (Cadence, Santa Catarina - Brazil) and dried at 50 °C in a forced ventilation oven (Marconi MA 35/5) until the material reached $10 \pm 2\%$ moisture. The dried material was ground in a knife grinder (Solab, SL 31) and passed through a series of

Tyler sieves (Bertel) of 20 to 100 mesh in a mechanical sieve agitator (Marconi, MA 750). The guava by-product flour (GBF) retained in the 100-mesh sieve (0.149 mm) was used in the juice production.

To juice production, a proportion of 1:1 (m/v) of guava pulp to water was mixed using a blender (Cadence, Santa Catarina, Brazil), followed by sieving to separate the solids. A formulation without GBF addition (control, 0% GBF) and formulations containing 1%, 3%, and 5% (m/v) of GBF, named respectively, 1% GBF, 3% GBF and 5% GBF, were prepared. The juices were pasteurized at 90 °C for 5 min, according to Oliveira et al. (2012). Each formulation was then incorporated with the sweetener stevia (Finn) at 1 g.L⁻¹, packed in 500-ml glass jars, hermetically sealed, and stored at 4 °C until analysis.

Physicochemical, antioxidants and microbiological analysis

The stability test (pH, acidity, soluble solids, color, microbiological analysis and antioxidant compounds and activity) was carried out under cooling temperature (4 °C) and the formulations were analyzed at 0, 7, 14 and 21 days of storage, which was defined in preliminary tests and according to Chakraborty & Athmaselvi (2014).

Measurements of pH were determined electrometrically using a Tecnal TEC 3PMP pHmeter (Piracicaba, Brazil). Soluble solids content was measured using a Tecnal Reichert AR 200 digital refractometer and reported as °Brix. Total titratable acidity was determined by titrating the samples with 0.1 M NaOH to an end point of pH 8.1 and expressed as g of citric acid 100 mL⁻¹.

Coloration was measured based on the CIE-Lab parameters L* (lightness), +a* (red), -a* (green), +b* (yellow) and -b* (blue), using a Konica Minolta Color Reader CR-10 colorimeter (Chiyoda, Japan).

The content of ascorbic acid and anthocyanins were determined according to the recommendations of Instituto Adolf Lutz (2008) and Latado et al. (2008), respectively. The results were expressed in mg 100 mL⁻¹ of guava juice.

The methodology of Singleton et al. (1999) was used for the total phenolic compound's analysis. Absorbance was measured in a spectrophotometer at 760 nm (Femto 700 plus). A calibration curve was prepared using gallic acid (0.1 mM – 0.5 mM), with a regression coefficient (R²) of 0.99. The results were expressed as gallic acid equivalent (GAE) in mg per 100 g of sample.

Antioxidant activity was measured using the DPPH free radical scavenge method according to Brand-Williams et al. (1955). After 30-minute incubation at room temperature, the absorbance was read at 515 nm. Trolox solutions (0.05mM – 0.5 mM) were used to generate the calibration curve (R² of 0.99) and the results were expressed in µmol of Trolox per g of sample.

Microbiological parameters evaluated in the formulations were Coliforms at 35 °C, Coliforms at 45 °C and *Salmonella* sp. according to the recommendation of Brazilian legislation, Resolution RDC n° 12, Jan 02, 2001 of National Health Surveillance Agency (Brasil 2001). The analyses were carried out according to American Public Health Association (APHA 2001).

Fiber content and rheological measurements

The dietary fiber content was determined according to method 991.43 of AOAC (Horwitz & Latimer 2005).

Rheological measurements were performed using a controlled rate Brookfield rheometer model LV DV III Ultra (673 'dyne.cm spring torque) at concentric cylinder arrangement equipped with SC4-13R small sample chamber and SC4-18 spindle. Data of viscosity versus shear rate (the so-called viscosity curves) were recorded at successively higher shear rate ($\dot{\gamma}$) from 0.34 s⁻¹

to 17.00 s^{-1} , in steps of $\dot{\gamma} = 0,17 \text{ s}^{-1}$ every 1.02 s. All viscosity curves were obtained at a temperature of $8.0 \text{ }^{\circ}\text{C}$ and absence of pre-shear. A water bath Brookfield model TC-550, equipped with water jacket Brookfield model SC4- 45Y, has been used for the temperature control. The rheometer was interfaced with a PC computer using the Brookfield Rheocalc version 3.2 operating software. Data analysis was performed by the same software.

The rheological experiments were conducted in triplicates and the viscosity curves were modeling using Ostwald-de-Waele equation (Power Law) calculated by Eq. 1,

$$\eta_a = k\dot{\gamma}^{n-1} \quad (1)$$

Where, (η_a) is the apparent viscosity (mPa.s), $\dot{\gamma}$ is the shear rate (s^{-1}), k is the consistency index (mPa), and n is the flow behavior index (dimensionless).

Sensory analysis

A sensory panel comprising 100 non-trained panelists aged 17–59 years (58% women and 42% men) conducted the sensory evaluation, which occurred in individual booths under controlled temperature and lighting conditions. The juice formulations were prepared, as described in section Materials. All panelists received approximately 40 ml of each sample at a temperature of $7\text{--}8 \text{ }^{\circ}\text{C}$ in disposable plastic cups of 50-ml capacity, coded with three-digit random numbers. Panelists were instructed to perform the test from left to right, following the method of Dutcosky (2011). The sensory test was approved by the Ethics Committee on Research at the State University of Maringá, Maringá, Brazil (CAAE: 84065618.9.0000.0104).

For the sensory acceptance test, each formulation was evaluated for appearance, aroma, flavor, and texture, using a nine-point hedonic nominal scale ranging from 1 (“dislike

extremely”) to 9 (“like extremely”). Purchase intention was evaluated using a five-point scale, ranging from 5 (“would certainly buy”) to 1 (“would certainly not buy”) (Meilgaard et al. 1999).

Data analyses

The experiments and analyses were performed in genuine triplicates, and results were expressed as mean values $\pm$ standard deviation in dry basis. Data collected were analyzed by ANOVA using Excel® 2010 software and Tukey tests (with a 95% confidence interval) to evaluate differences between results.

RESULTS AND DISCUSSION

Physicochemical properties of guava juice formulations during storage

The SS content, tartness (expressed as pH or acidity), and color are important attributes that affect the acceptability of guava juices (Akbar et al. 2016). For the guava juice formulations at 0, 7, 14, and 21 days of storage, these data are shown in Table I.

It was observed that the higher content of GBF (5% versus 0%, 1%, and 3%) used in the guava juice formulations decreased the pH values, except at day 21 of storage when the values did not differ among the formulations ($p > .05$). As expected, the acidity displayed a contrary behavior, such that, the values were typically higher in the juices with 3% and 5% GBF relative to the formulations containing 0% and 1% GBF. The pH and acidity data found in this work were similar to those obtained by Lamo et al. (2019) in pasteurized guava juice (pH ~ 4.1 , acidity of $0.5 \text{ g } 100 \text{ g}^{-1}$).

During storage, the pH values decreased and acidity increased only at day 21, except for the formulation containing 5% GBF that had an increase from 14^o day when compared with the

Table I. Acidity, pH, soluble solids and color parameters (L^* , a^* , b^*) of guava juice formulations containing 0, 1, 3 and 5% of guava by-product (GBF) at 0, 7, 14 and 21 days of storage at 4 °C.

	Time (days)	0%GBF	1% GBF	3% GBF	5% GBF
Acidity (g citric acid 100 g ⁻¹)	0	0.18±0.01 ^{bA}	0.18±0.01 ^{bA}	0.24±0.03 ^{aA}	0.25±0.01 ^{aA}
	7	0.18±0.02 ^{bA}	0.19±0.01 ^{bA}	0.24±0.01 ^{abA}	0.28±0.02 ^{aAB}
	14	0.19±0.01 ^{cA}	0.20±0.01 ^{cA}	0.25±0.00 ^{bA}	0.30±0.02 ^{aBC}
	21	0.27±0.00 ^{bB}	0.29±0.01 ^{bB}	0.33±0.01 ^{aB}	0.34±0.01 ^{aC}
pH	0	4.16±0.01 ^{aA}	4.12±0.01 ^{bA}	4.10±0.02 ^{bcA}	4.08±0.01 ^{cA}
	7	4.18±0.01 ^{aA}	4.14±0.01 ^{bA}	4.10±0.01 ^{cA}	4.07±0.02 ^{dA}
	14	4.19±0.03 ^{aA}	4.13±0.03 ^{abA}	4.08±0.02 ^{bcA}	4.05±0.02 ^{cA}
	21	3.97±0.02 ^{aB}	3.96±0.01 ^{aB}	3.90±0.03 ^{aB}	3.87±0.12 ^{aB}
Soluble solids (°Brix)	0	20.67±0.58 ^{aA}	20.67±0.58 ^{aA}	20.67±0.58 ^{aA}	20.33±0.58 ^{aA}
	7	20.33±0.58 ^{aA}	20.33±1.15 ^{aA}	20.67±0.58 ^{aA}	20.00±1.00 ^{aA}
	14	20.00±1.00 ^{aA}	20.33±1.15 ^{aA}	20.33±0.58 ^{aA}	19.67±1.15 ^{aA}
	21	20.33±1.15 ^{aA}	20.67±0.58 ^{aA}	20.00±1.00 ^{aA}	19.67±0.58 ^{aA}
L^*	0	50.49±0.17 ^{aA}	50.20±0.60 ^{aA}	45.85±0.34 ^{cA}	47.88±0.63 ^{bA}
	7	45.84±1.21 ^{aB}	47.07±1.19 ^{aB}	44.31±0.90 ^{aB}	44.33±0.27 ^{aB}
	14	42.42±1.38 ^{aB}	42.84±1.09 ^{aC}	41.73±0.13 ^{aC}	41.80±0.15 ^{aC}
	21	41.03±0.68 ^{aB}	41.74±0.68 ^{aC}	41.56±0.34 ^{aC}	41.83±0.26 ^{aC}
a^*	0	8.49±0.49 ^{aB}	8.69±0.39 ^{aA}	7.79±0.33 ^{bA}	6.91±0.29 ^{bB}
	7	9.92±0.54 ^{aA}	8.96±0.11 ^{bA}	5.76±0.18 ^{dB}	7.80±0.07 ^{cA}
	14	9.95±0.45 ^{aA}	7.81±0.15 ^{bB}	7.08±0.57 ^{bA}	6.79±0.29 ^{bB}
	21	9.99±0.35 ^{aA}	7.91±0.15 ^{bB}	6.70±0.73 ^{cAB}	6.88±0.08 ^{bcB}
b^*	0	7.26±0.65 ^{bA}	8.42±0.29 ^{bBC}	11.90±0.42 ^{aA}	10.33±0.95 ^{aAB}
	7	7.74±0.58 ^{cA}	9.91±0.25 ^{bA}	10.38±0.26 ^{bAB}	11.59±0.57 ^{aA}
	14	7.70±0.23 ^{bA}	7.90±0.06 ^{bC}	10.11±0.95 ^{aAB}	9.61±0.28 ^{aB}
	21	8.61±0.79 ^{aA}	8.73±0.37 ^{aB}	9.56±1.22 ^{aB}	9.51±0.21 ^{aB}

Means followed by the same lowercase letter (in each line) and same uppercase letter (in each column) were not different ($p > .05$).

initial time. Before 21 days, the similar acidity and pH-values of most of formulations was related to the low activity of some acid-producing bacteria (Kang et al. 2003). At 21 days, this activity must be increased due to the multiplication of the remaining bacteria (even with the pasteurization process) which uses mainly the soluble sugars in the fermentative process, thus, there was no

influence of GBF addition in pH values at this time of evaluation.

The SS content did not range with the GBF addition because the sucrose present in the guava pulp is most responsible for the SS value, while the guava by-product did not contribute to increasing SS content, because it is formed by high dietary fiber content (69.1 g 100 g⁻¹)

mainly insoluble (57.7 g 100 g⁻¹) as reported by Martínez et al. (2012). The values observed in this work (around 20°Brix) were higher than those reported by Sinchaipanit et al. (2015) in guava juices (12.0–12.2 °Brix), which can be due to the differences in processing conditions and maturation grade of the fruit. For all samples, the SS content was similar at the different time points evaluated, indicating the stability of this parameter during storage.

Regarding luminosity (L^*), the formulations containing 0% GBF and 1% GBF had higher initial values in comparison to the other samples, suggesting that the higher amounts of fiber addition (3% and 5%) caused a darkening effect. The chromaticity results showed that the incorporation of GBF into the juices lowered the redness (a^*) values and increased the yellowness (b^*) values. During the drying process of guava peel, the Maillard reaction could have occurred, as mentioned by Karam et al. (2016), reflecting in a darker color of GBF (brownish color) that altered the coloration of the juice formulations. The values obtained for the instrumental color parameters were similar to those found by Silva et al. (2016) using different preservatives in guava juice.

During storage, all formulations exhibited a reduction in luminosity (L^*) from day 0 to 7 ($p < .05$), and after this time there were no differences ($p > .05$) in this parameter among the formulations. Otherwise, color variations (a^* and b^*) were noted only in the samples with GBF addition and became more relevant from day 14 of storage. Campoli et al. (2018) analyzed guava juices processed by ultrasound and also found slight alterations in the color parameters during the shelf life of 14 days. Although heat treatment during juice processing can inhibit enzymatic browning, the non-enzymatic browning reaction and anthocyanin degradation can still occur during the storage. Ascorbic acid (AA) also

contributes to darkening in citrus juice, as its degradation has been significantly correlated with the development of browning (Paravisini & Peterson 2019).

Microbiological parameters during storage presented in accordance with the legislation, being all results were < 3 MPN mL⁻¹ for Coliforms at 35° and 45° C and for *Salmonella* analysis the results were absence of all formulations.

Antioxidant compounds and activity

AA, anthocyanins, and phenolic compounds are known as substances with antioxidant potential, and these components were present at varying levels in the guava juice formulations (Table II).

In general, the GBF addition did not interfere in the AA content since only at time 0 this value was lower in the sample with 5% GBF in relation to the formulations with 0% GBF and 1% GBF ($p < .05$). The AA levels detected in this work were similar to those determined by Ani & Abel (2018) for grape juice (26.36 mg 100 mL⁻¹), and by Cunha et al. (2014) for orange juice (34.87 mg 100 mL⁻¹).

AA is an essential micronutrient found in food and, due to its sensitivity to moisture, temperature, light, oxygen, and pH, it is highly unstable during the shelf life of the product (Sanchez et al. 2018), which explains the storage-induced decrease in the AA content of all the formulations produced. AA degradation can occur aerobically or anaerobically. Under aerobic conditions, AA is oxidized to dehydroascorbic acid and irreversibly converted into 2,3-diketogulonic acid and other degradation products, such as furfural, xylosone, and reductones. Anaerobically, AA is degraded into furfural via various reaction steps (Buvé et al. 2018). The decrease in AA and the consequent production of the degradation compounds might corroborate with the darkening of the samples during storage.

Table II. Ascorbic acid, anthocyanins, total phenolic compounds (TPC) and antioxidant activity by DPPH method of guava juice formulations containing 0, 1, 3 and 5% of guava by-product (GBF) at 0, 7, 14 and 21 days of storage at 4 °C .

	Time (days)	0%GBF	1% GBF	3% GBF	5% GBF
Ascorbic acid (mg 100 mL ⁻¹)	0	34.05±2.03 ^{aA}	34.05±2.03 ^{aA}	30.53±1.02 ^{abA}	28.18±1.76 ^{bA}
	7	28.18±1.76 ^{aB}	27.01±1.02 ^{aB}	27.01±2.03 ^{aAB}	26.42±1.76 ^{aAB}
	14	28.77±2.69 ^{aAB}	28.77±2.03 ^{aB}	24.66±1.76 ^{aB}	26.38±1.76 ^{aAB}
	21	23.48±2.03 ^{aB}	24.36±0.51 ^{aC}	20.55±1.02 ^{aC}	22.31±2.03 ^{aB}
Anthocyanins (mg 100 mL ⁻¹)	0	0.41±0.02 ^{bAB}	0.32±0.12 ^{bA}	1.81±0.08 ^{aA}	1.91±0.13 ^{aA}
	7	0.32±0.01 ^{dAB}	0.63±0.05 ^{cA}	1.12±0.03 ^{bA}	1.73±0.11 ^{aAB}
	14	0.51±0.03 ^{bA}	0.44±0.02 ^{bA}	1.11±0.01 ^{aA}	1.05±0.09 ^{aB}
	21	0.15±0.01 ^{bB}	0.26±0.01 ^{bA}	0.91±0.01 ^{aA}	1.01±0.08 ^{aB}
TPC (µg GAE 100 mL ⁻¹)	0	48.81±3.33 ^{aA}	46.34±5.53 ^{aA}	50.78±7.21 ^{aA}	46.29±0.34 ^{aA}
	7	28.06±1.42 ^{bCB}	34.65±2.96 ^{abB}	36.37±3.62 ^{aB}	24.21±2.02 ^{cB}
	14	31.35±3.49 ^{aB}	35.74±5.78 ^{aB}	35.74±5.78 ^{aB}	24.41±14.09 ^{aB}
	21	25.84±0.18 ^{CC}	35.70±0.16 ^{aB}	28.19±0.44 ^{bB}	26.73±0.29 ^{bCB}
DPPH (µmol de Trolox 100 mL ⁻¹).	0	1.67±0.13 ^{bA}	1.77±0.04 ^{bA}	2.12±0.10 ^{aA}	1.85±0.14 ^{abA}
	7	1.63±0.23 ^{bA}	1.74±0.16 ^{bA}	2.26±0.01 ^{aA}	1.93±0.05 ^{abA}
	14	1.40±0.03 ^{bB}	1.23±0.04 ^{cB}	1.29±0.11 ^{cB}	1.66±0.22 ^{aB}
	21	1.42±0.03 ^{bB}	1.42±0.12 ^{bB}	1.53±0.20 ^{bB}	1.77±0.23 ^{aB}

Means followed by the same lowercase letter (in each line) and same uppercase letter (in each column) were not different ($p > .05$).

According to the Institute of Medicine (2006), adequate intake of AA for men and women is 90 and 75 mg day⁻¹ respectively. As the content of AA was similar ($p > .05$) among the formulations, the amount of guava juice with GBF addition required to achieve the Recommended Dietary Allowance (RDA), an average, is 280 mL for men and 235 mL for women, at 0 days of storage; but at the end of storage (21 days), these formulations have a reduction of RDA, 23 and 27%, respectively.

In general, the content of total phenolic compounds (TPC) was not influenced by the addition of guava by-product. Inada et al. (2017) described a higher TPC value in jabuticaba juice (913 µg gallic acid equivalents [GAE] 100 mL⁻¹) in comparison to the formulations in this study (29.84–50.78 µg GAE 100 mL⁻¹). Ninga et al.

(2018) noticed only slight variations in the TPC contents among guava juice samples treated with different enzymatic concentrations (44.90–47.20 µg GAE 100 mL⁻¹). Haida et al. (2015) reported that the TPC content could be influenced by several factors, such as maturity, species, type of crop, geographical origin, growth level, harvest conditions, and storage process.

It was noted that after 7 days of storage, that the TPC content decreased in all formulations but then plateaued until the end of storage (21 days), except for the control sample. It infers that the addition of the guava by-product aided in the maintenance of the TPC content, probably due to its composition. A hypothesis is related to the phenolic profile of GBF that could be more resistant to the storage losses than those found

in guava pulp. Amaya-Cruz et al. (2015) reported that guava by-product has a high content of polyphenols (7.5 mg g^{-1}), composed mainly of *p*-hydroxybenzoic acid, chlorogenic acid, ferulic acid, ellagic acid and sinapic acid.

The higher levels of GBF (3% and 5%) added to guava juice promoted higher anthocyanins contents during the time evaluated, probably because of the concentration of this pigment during the drying process of guava by-product. The amounts detected were comparable to those present in concentrated blackcurrant juice (*Ribes nigrum* L.) of 0.33 to $1.88 \text{ mg } 100 \text{ mL}^{-1}$ (Bánvölgyi et al. 2009).

During storage, it was verified that the guava juices containing 1% and 3% GBF presented no alteration in the anthocyanins content ($p > .05$), while the formulations 0% GBF and 5% GBF exhibited a decrease at day 21 and day 14, respectively. These results indicate that anthocyanins had more storage stability than AA and TPC in guava juices. Furthermore, at day 21, the formulations containing 3% and 5% GBF had six to seven times more anthocyanins than the control juice. As mentioned by Hellstrom et al. (2013), anthocyanins are highly susceptible to degradation, which can cause a decrease in the redness of the samples and this might explain the decrease in a^* values, as discussed in section Physicochemical properties of guava juice formulations during storage.

Antioxidants have the function of inhibiting or attenuating free radical reactions and retarding or inhibiting cellular damage (Thébault 2017). At the initial storage times (0 and 7 days), the samples containing 3% GBF and 5% GBF had higher antioxidant potential than the other formulations. After this period, the highest antioxidant potential was noted in the 5% GBF formulation. The 2,2-diphenyl-1-picrylhydrazyl (DPPH) antioxidant activities recorded in this study (1.23 to $2.12 \text{ } \mu\text{mol Trolox } 100 \text{ mL}^{-1}$) was lower

than the values observed in orange juice (5.3 – $12.6 \text{ } \mu\text{mol Trolox } 100 \text{ mL}^{-1}$) (Navarro et al. 2011).

At day 14, the antioxidant potential of all formulations decreased, and this was maintained throughout storage. This decrease was more expressive in the formulations containing 1 and 3% of GBF addition and it is related decrease in the content of antioxidant compounds, ascorbic acid, anthocyanins and phenolics, in the same period of storage. Cassani et al. (2016) observed that all the strawberry juice samples enriched with prebiotic fibers exhibited variations in the DPPH antioxidant capacity during storage, decreasing gradually.

Dietary fiber content and rheological parameters

The juice industry produces large amounts of by-products, which could be used for the development of nutraceutical products due to the high fiber content present in these residues (Russo et al. 2015), as exemplified by the guava peel flour used in this study ($72.21 \pm 2.21 \text{ g fiber } 100 \text{ g}^{-1}$). The consumption of a fiber source, such as GBF, is important in the prevention of obesity and its complications. DF is reported to exert an anti-obesogenic effect associated with the high proportion of insoluble fiber, which constitutes about 94% of the total DF (Amaya-Cruz et al. 2015). In the juice formulations (Table III), the higher the GBF addition, the higher was the DF content, presenting a two- to five-fold increase above the control. All the formulations are considered as a source of fiber, according to Brazilian legislation (Brasil 2012).

Figure 1 shows the viscosity curves of the guava juice formulations. For all formulations, the apparent viscosity decreased with increasing shear rate ($p < 0.5$), indicating a non-Newtonian flow behavior (Rigoto et al. 2018). At low shear rates, the viscosity is high because of the high forces required in breaking agglomerated

molecules (pectinaceous substances), whereas, at high shear rates, the low viscosity is a result of molecules orientation and alignment, reducing friction between molecules (Abdullah et al. 2017). Table III shows the consistency index (k) and flow rate (n) data, both derived from Eq. (2). All formulations displayed a pseudoplastic behavior ($n < 1$), similarly to guava juice (Diniz et al. 2014).

The apparent viscosity of the formulations increased with the addition of guava by-product. The presence of fibers, such as pectin, in the guava by-product might have aided in increasing

the viscosity, considering that the increment in hydrated molecules and hydrogen bonding with the hydroxyl groups, provides high water holding capacity and results in the development of a cohesive network structure (Sharoba & Ramadan 2011, Sharma et al. 2014, Abdullah et al. 2017).

No significant alterations in the DF content and rheological parameters occurred during the storage of guava juice formulations (data not shown).

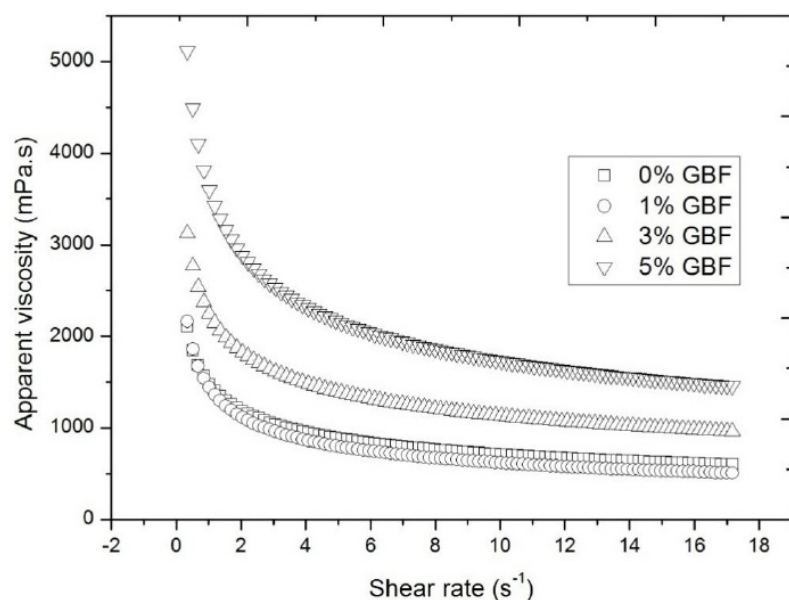


Figure 1. Apparent viscosity (η_a) as a function of shear rate (viscosity curve) of the different formulations of guava juice containing by-product (GBF) described in the Table III.

Table III. Dietary fiber content and rheological parameters of guava juice formulations containing 0, 1, 3 and 5% of guava by-product (GBF).

Formulations	Dietary fiber (g 100g ⁻¹)	$\eta(\dot{\gamma} = 16.32 \text{ s}^{-1})$ [mPa.s]	K [mPa.s]	n	R^2
0% GBF	1.02 ± 0.30 ^d	221.83 ± 1.34 ^d	1491 ± 0.89 ^a	0.32 ± 0.01 ^{ab}	94.6
1% GBF	2.02 ± 0.01 ^c	243.17 ± 1.94 ^c	1453 ± 1.16 ^c	0.37 ± 0.02 ^a	92.0
3% GBF	4.00 ± 0.37 ^b	316.18 ± 1.64 ^b	2262 ± 1.17 ^b	0.30 ± 0.01 ^b	95.8
5% GBF	4.92 ± 0.51 ^a	524.89 ± 5.20 ^a	3620 ± 3.62 ^a	0.32 ± 0.03 ^{ab}	89.5

Means followed by the same lowercase letter in each column were not different ($p > .05$).

Sensory analysis

In relation to the sensorial results of the formulations (Table IV), those with higher levels of GBF received lower scores for overall appearance and color ($p < .05$), which can be associated with the brown coloration of the flour that visibly altered the characteristics of the guava juice. During the drying process of guava peel, the degradation of chlorophyll by the action of the chlorophyllase enzyme could have occurred, as mentioned by Palharini et al. (2016), and reflected in the color change of the juice formulations.

For aroma and flavor attributes, there were no differences ($p > .05$) between samples containing 1% and 3% GBF, however, the scores obtained in these samples were lower than the values found in the control ($p < .05$). It was observed that the 5% GBF formulation had the lowest acceptability, according to the attributes mentioned above. Cassani et al. (2016) mentioned that most of the nutraceutical compounds have a natural bitter or astringent flavor, which can cause consumers to reject the product. The scores for texture were similar between the formulations 0% GBF and 1% GBF ($p > .05$), while the 5% GBF formulation received the lowest acceptance for this attribute. This can be attributed to the presence of fragments associated with the guava peel by-product,

which possibly caused a decrease in the product acceptance.

Among the formulations with guava by-product addition, 1% GBF and 3% GBF were considered more accepted by the tasters, with scores ranging from 6 (“slightly liked”) to 7 (“moderately liked”), whereas the control formulation (0% GBF) scored between 7 and 8 (“really liked”). Moreira et al. (2014) observed a decrease in the acceptance of pure watermelon juice (6.08) when sorghum and soybean were incorporated (3.08 and 5.36, respectively), receiving much lower scores than those assigned in the current study. Likewise, Sun-Waterhouse et al. (2010) observed that the increasing addition of apple fiber to smoothies decreased their sensory acceptability.

For the purchase intention test, the most accepted juice was the control (3.52), followed by 1% GBF (2.97) and 3% GBF (2.66). The closest values of 4 (control) and 3 (1% and 3% GBF) correspond to “probably buy” and “maybe buy/maybe not buy”, respectively. Although these results indicate a lower acceptance after the source of fiber addition (GBF), recent consumer trends are oriented towards the consumption of healthier products produced with natural ingredients and processes that align with the sustainability and preservation of the environment (Paravisini & Peterson 2019).

Table IV. Sensorial attributes of guava juice formulations containing 0, 1, 3 and 5% of guava by-product (GBF).

Attributes	0% GBF	1% GBF	3% GBF	5% GBF
Overall appearance	7.55±1.40 ^a	6.86±1.52 ^b	6.05±1.64 ^c	4.58±2.05 ^d
Color	7.87±1.35 ^a	6.88±1.49 ^b	5.97±1.76 ^c	4.29±2.14 ^d
Aroma	7.67±1.39 ^a	6.99±1.55 ^b	6.72±1.87 ^b	5.91±2.11 ^c
Flavor	7.29±1.78 ^a	6.51±1.82 ^b	6.20±1.81 ^b	5.23±2.01 ^c
Texture	7.18±1.40 ^a	6.84±1.37 ^{ab}	6.27±1.77 ^{bc}	5.72±1.88 ^c
Purchase intention	3.52±1.10 ^c	2.97±1.23 ^b	2.66±1.07 ^b	2.22±1.19 ^a

Means followed by the same lowercase letter (in each line) and same uppercase letter (in each column) were not different ($p > .05$).

CONCLUSIONS

The addition of 3% and 5% of guava by-product to juice formulations increased the acidity and caused alterations to the instrumental color parameters (decreased L^* and a^*) when compared with the formulations 0% GBF and 1% GBF. The contents of SS, AA, and TPC were similar among the formulations produced, while the anthocyanins, DPPH antioxidant activity, DF content, and apparent viscosity increased with the increasing addition of GBF. Storage altered some of the parameters evaluated and indicated 14 days as the recommended shelf life to maintain the physicochemical, and antioxidant attributes of the guava juice. The guava juice containing 1% GBF scored higher in the sensory analysis than the formulations 3% GBF and 5% GBF. Therefore, the addition of guava by-product showed promising results to enhance the nutritional quality of guava juice.

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The authors declare that there is no conflict of interest regarding the publication of this article. Thairla V.B da Silva carried out the experiments, Isabela J. Iwassa performed characterization analysis, Anderson R. Sampaio was responsible for rheological analysis, Suelen P. Ruiz and Beatriz Cervejeira Bolanho Barros supervised the findings of this work. All authors discussed the results and contributed to the final manuscript.

