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## PREGELATINIZED RICE BRAN FLOUR AS STABILIZER FOR GUAVA NECTAR

Fernanda Doring Krumreich<sup>1\*</sup>, Ana Paula Antunes Corrêa<sup>2</sup>, Tailise Beatriz Roll Zimmer<sup>1</sup>, Jair Costa Nachtigal<sup>2</sup>, Rui Carlos Zambiazi<sup>1</sup>

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**ABSTRACT** - Rice bran flour is a byproduct of rice processing and can be used as food. The objective of the current study was to use pregelatinized rice bran flour along with hydrocolloids to stabilize guava nectar, as well as to evaluate the stability of bioactive compounds during a storage period of 180 days. For this purpose, the guava fruits were processed in the form of pulp, following the elaboration of the nectars. Four formulations were prepared: one control (pulp, water, and sugar) and three by the addition of xanthan, guar gum, and pregelatinized rice flour. The analyses on guava nectars were performed 24 h after their processing and at every 45-day intervals during the entire 180 days study period. The experiment was done in triplicate and the results were expressed on a wet basis. The physicochemical properties, color, and the bioactive compounds in the guava nectars were influenced mainly in the formulation of pregelatinized rice bran flour & guar gum. The same composition also resulted in greater stability. Despite the absence of considerable differences in the guava nectar formulations for the evaluated parameters, the addition of stabilizers seems to be necessary for a visual quality aspect, since the sedimentation that occurs in this type of nectar depreciates the appearance of this product, thus compromising its competitiveness. The pregelatinized rice bran flour associated with guar gum was found to be the most effective strategy for stabilization of guava nectar, combined with the low cost and ease of obtaining, compared to other hydrocolloids commonly used in the food industry. **Keywords:** nectar, rice bran flour, stability, storage.

## FARINHA DE ARROZ PRÉ-GELATINIZADA COMO ESTABILIZANTE DE NÉCTAR DE GOIABA

**RESUMO** - A farinha de arroz é um subproduto da indústria arrozeira e que pode ser utilizada como alimento. O objetivo desse estudo foi utilizar a farinha de arroz pré-gelatinizada combinada com hidrocolóides para estabilizar néctar de goiaba, bem como, avaliar a estabilidade de compostos bioativos durante o armazenamento por 180 dias. Para isso, os frutos foram processados na forma de polpa, seguindo a elaboração dos néctares. Foram elaborados quatro formulações, incluindo uma controle (polpa, água e açúcar) e as demais pela adição de xantana, goma guar e farinha de arroz pré-gelatinizada. As análises dos néctares de goiaba foram realizadas 24 h após o processamento e a cada 45 dias durante todo o período de estudo de 180 dias. O experimento foi realizado em triplicata e os resultados foram expressos em base úmida. As propriedades físico-químicas, a cor e os compostos bioativos dos néctares de goiaba foram influenciados principalmente na formulação da farinha de arroz pré-gelatinizada e goma guar. A mesma composição também resultou em maior estabilidade. Apesar da ausência de diferenças consideráveis nas formulações de néctar de goiaba para os parâmetros avaliados, a adição de estabilizantes parece ser necessária para um aspecto de qualidade visual, uma vez que a sedimentação que ocorre neste tipo de néctar deprecia a aparência deste produto, comprometendo sua competitividade. A farinha de arroz pré-gelatinizada associada à goma guar mostrou ser a estratégia mais eficaz para estabilização do néctar de goiaba, aliado ao baixo custo e facilidade de obtenção, em relação aos outros hidrocoloides comumente utilizados na indústria de alimentos.

Palavras-chaves: armazenamento, néctar, farinha de arroz pré-gelatinizada, estabilidade.

#### INTRODUCTION

The market for fruit juices and nectars is on the significant rise, due to the several reasons, such as feasibility, high nutritional value, and the desire to acquire healthier foods. As a result, the food industry is increasingly expanding its range of healthy beverages with a variety of flavors. In 2020, according to the Brazilian Association of Soft Drinks and Non-Alcoholic Beverages (ABIR, 2020), the sector sold 31 billion liters of the beverages.

Fruits are sources of vitamins, minerals, and soluble carbohydrates. Guava (*Psidium guajava* L.) is notable for its high content of vitamins A and B, mainly thiamine (B1), riboflavin (B2), and niacin. Guava also contains a high content of fiber, protein, total sugar and minerals, such as calcium, phosphorus, and potassium. In addition, the fruit also displays high contents of vitamin C, carotenoids, and phenolic compounds. Such characteristics makes the guava complete and balanced in relation to the

<sup>1</sup>Faculty of Agronomy Eliseu Maciel, Universidade Federal de Pelotas (UFPel), Pelotas, RS, Brazil. E-mail: <u>nandaalimentos@gmail.com</u>. \*Corresponding author.

<sup>2</sup>Empresa Brasileira de Pesquisa Agropecuária - Embrapa, Pelotas, RS, Brazil.

nutritive value (CONCEIÇÃO et al., 2012). Thus, in addition to its *in natura* consumption, guavas are also widely used for manufacturing of sweets, preserves, juices, and nectars.

In the processing of guava nectars, the phase separation may occur soon after its production or during the storage period and depreciates the visual appearance of the nectar, thus compromising its competitiveness. In order to stabilize fruit nectar, hydrocolloids and gums are used; however, they also alter the viscosity and texture of the products (SAHA; BATTACHARYA, 2010). The common hydrocolloids used in the food industry are xanthan gum, guar gum, carrageenan, and pectin, due to their thickening, stabilizing, gelling and emulsifying properties (CODEX ALIMENTARIUS, 2021). Rice bran flour is a byproduct obtained from broken rice grains and can be an alternative to hydrocolloids utilization (CARVALHO et al., 2011).

Dors et al. (2006) relate that rice bran flour is not widely used, although it has promising characteristics, such as slightly pronounced taste, no interference with the final taste of other foods, and being devoid of any allergenic compounds. A possible application of the pregelatinized rice bran flours could be the production of instant foods, which are widely rising in the market. Vissoto et al. (2006) relate that pregelatinized rice bran flour can be used in the production of foods with less valuable raw materials and with desirable technological characteristics.

Thus, the present study aimed to evaluate the efficiency of the use of pregelatinized rice bran flour along with hydrocolloids (xanthan and guar) in the stabilization of guava nectar. We also evaluated the stability of bioactive compounds and physicochemical parameters of the nectar during the storage period of 180 days at room temperature.

#### MATERIAL AND METHODS Material

Guavas of the Paluma cultivar were obtained from a rural area in the municipality of Pelotas/RS-Brazil (latitude: 31°29'12"S, longitude: 52°32'57" O and altitude: 226 m) during the harvesting of 2013 and 2014. Pregelatinized rice bran flour and xanthan (Sigma Aldrich) and guar gums (Sigma Aldrich) were used as the stabilizing agents. For determination of antioxidant capacity, 2,2-diphenyl-1-picrylhydrazyl (DPPH) (Sigma Aldrich) was used. The other reagents used were of PA grade.

#### **Processing nectar**

A sample of 20 kg was used to conduct the present experiment. First, the fruits were sanitized in chlorinated water at 500 ppm for 10 min followed by passing through a 0.8-mm mesh stripper. The pulp was packed in polyethylene packages with a capacity of 1 kg and stored in a freezer at -18°C until the nectars were prepared. The base/control formulation of the nectar consisted of 55% water, 35% guava pulp and 10% sugar (sucrose). For the other formulations, hydrocolloids were combined with pregelatinized rice bran flour & xanthan, pregelatinized rice bran flour & guar, and xanthan & guar. The amounts used were 0.05% of each hydrocolloid, i.e., a total of 0.1% 135

hydrocolloid content. The formulations of nectars were prepared in accordance with Ordinance No. 12, dated September 04, 2003, Art. 1 Annex II – Identity and Nectar Quality Standards, which establishes a minimum quantity of 35% of pulp for this beverage. The samples were stored in glass bottles and subjected to a temperature of  $22^{\circ}C \pm 3.6^{\circ}C$  and ambient light for 180 days.

#### Physico-chemical analysis

The analyses on guava nectars were performed 24 h after their processing and at every 45-day intervals during the entire 180 days study period. The pH value was measured in a pH meter Digimed DM 20, the total titratable acidity was expressed as % citric acid (IAL, 2008), and the total soluble solids content was measured by rephratometer (Atago Palette PR-32). The color was measured according to the C.I.E. L\* a\* b\* system, in colorimeter Minolta (CR-300), with illuminant D 65, 8 mm-illumination area, and L\* values (brightness) ranging from black (0) to white (100); a\* values from green (-a) to red (+a), and b\* values ranging from blue (-b) to yellow (+b). The phenolic compounds followed the method described by Swain and Hillis (1959) expressed in mg EAG.100 g<sup>-1</sup>, the carotenoid content was expressed in mg of lycopene 100  $g^{-1}$  (AOAC 970.64), the antioxidant capacity was determined by the ability of the compounds to sequester the radical DPPH (2,2- difenil-1picrilhidrazila) following the method described by Brand-Williams et al. (1995). L-ascorbic acid content was quantified using the titrimetric method of Lorenz-Steves, expressed as mg of L-ascorbic acid per 100g sample (ZAMBIAZI, 2010), and the sedimentation was done by the clarified phase measurement of nectars performed twice a week for a period of 90 days. The experiment was done in triplicate. All the results were expressed on a wet basis.

#### Statistical analysis

The data were analyzed by Tukey's and Dunnett's average comparison tests with 5% significance level using the statistical program SAS v8. The results were expressed as mean  $\pm$  standard deviation.

#### **RESULTS AND DISCUSSION**

## Influence of the addition of hydrocolloids on guava nectars - pH, soluble solids and titratable acidity (AT)

According to Zambiazi (2010), each microorganism has a minimum and maximum optimum pH range of growth and most of the microbes grow at a pH close to neutrality (6.6-7.5) while a few have the capacity to develop at a pH less than 4.0. The formulated nectars showed a pH in the range of 3.85 to 4.29, where a pathogenic microorganism can hardly grow. The pH of the formulations of the nectars with xanthan & guar and rice bran flour & xanthan differed significantly from the control, presenting higher pH values (mean  $4.08 \pm 0.04$ ) (Figure 1A). On the other hand, the formulation of the nectar with pregelatinized rice bran flour & guar presented a lower pH value (3.85  $\pm$  0.01). This could have occurred due to the acceleration of food decomposition by hydrolysis increasing the hydrogen concentration (QUEIROZ et al.,

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2010). Chaikham and Apichartsrangkoon (2012) observed a reduction in pH when they added xanthan gum in longan juice. During the storage period (Figure 1A), a significant increase in pH in the nectar formulations was observed, except for nectar containing pregelatinized rice bran flour and guar gum.

The content of soluble solids in the guava nectar formulations was significantly higher  $(14.03 \pm 0.12)$  than that in the control (Figure 1 B). Throughout the storage,

only the formulated xanthan & guar nectar presented variations in the total soluble solids content, differing from the other formulations, in which the soluble solids content remained almost constant. According to the Ministry of Agriculture, Livestock and Food Supply (MAPA), the minimum soluble solids content for guava nectars should be 10° Brix (BRASIL, 2003); therefore, all the formulations of guava nectars complied with the minimum limit established by the legislation until the end of the storage period.

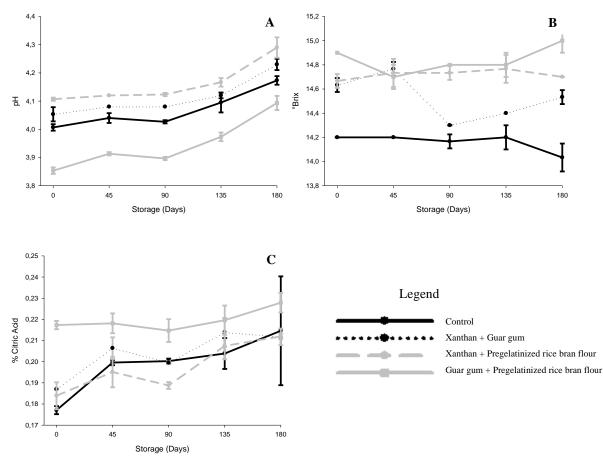


FIGURE 1 - Effect of hydrocolloid addition on pH (A), total soluble solids (B) and titratable acidity (C) of guava nectars during storage.

The total soluble solids contents of 11.4 to  $13.8^{\circ}$ Brix and pH of 3.3 to 3.5 for different acerola nectars from commercial propolis extract have been reported in the literature (NEVES et al. 2010). Carvalho and Guerra (1995), while evaluating the stability of pasteurized fresh acerola juice packed in glass bottles, found that the pH of the samples remained constant at 3.3 throughout 150 days of storage at room temperature ( $28^{\circ}C\pm 2^{\circ}C$ ). In the present study, a small increase in pH was observed only after 135 days of storage. In relation to the soluble solids, Krumreich et al. (2018), in their study on pasteurized guava nectar packed in glass bottles and kept at room temperature, observe variations along the storage, whereas in the present study, some variations were observed mainly in the formulation containing xanthan & guar.

Several formulations of nectars have shown an increase in the titratable total acidity values after

processing, differing significantly from the control. It was observed that the titratable total acidity remained practically stable for all the formulations during the 180 days of storage with values between 0.18% and 0.22%. The titratable acidity remained stable for all formulations during 180 days of storage (KRUMREICH et al., 2018). Chaikham and Apichartsrangkoon (2012), for longan juice added with xanthan gum, found 0.22% of the titratable acidity values. These values are close to those found in our study. According to the legislation (PIQ), the minimum value of acidity allowed for nectars is 0.10% (BRASIL, 2003); therefore, all the formulations tested met this requirement, since they presented values above the stipulated minimum.

#### Color analysis

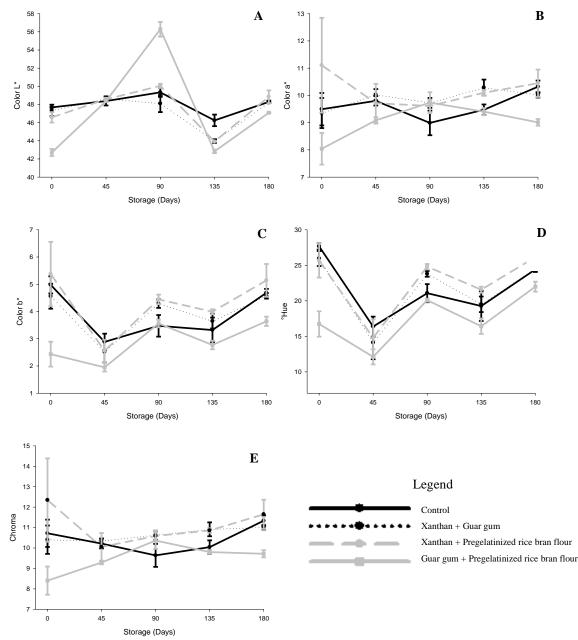
Color is a foremost attribute of food quality. In the case of fruits, the color change may occur due to thermal

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processing, chemical reactions and/or the degradation of bioactive compounds, such as carotenoids, vitamin C, and phenolic compounds. Therefore, monitoring of this parameter during evaluation of the quality of a food or drink is important.

The parameter L\* defines the color clarity, in which the value 0 indicates complete black color and 100 complete white. The chromaticity coordinate  $a^*$ , when positive, indicates the existence of a higher content of red pigments, whereas a negative value indicates the existence of green pigments. The value of b\* refers to the yellow tint when positive and blue when negative. When the values of the scales  $a^*$  and  $b^*$  are close to zero, the sample has a color close to neutrality (PATHARE et al., 2013).

The color parameters (L\*, a\* and b\*) of most nectars, at each evaluated period, remained very close to the control formulation and did not show any significant variation (Figure 2A), except for the rice flour formulation. The pregelatinized rice & guar gum formulation showed some variations only at 45 and 135 days. At 90 days of storage, a more significant increase in the L\* parameter was observed in the nectar with pregelatinized rice bran flour & guar gum, but at the end of the storage period (180 days), it was decreased even in the other formulations. Tribst et al. (2009), in his study with mango nectar processing, attributed the increased luminosity that persisted until the end of storage to the heat treatment by which the nectar was processed.



**FIGURE 2** - Influence of the addition of hydrocolloids in color parameters  $L^*(A)$ ,  $a^*(B)$ ,  $b^*(C)$ , Hue (D) and Chroma (E) of guava nectars during storage.

Both the variables a\* (Figure 2B) and b\* (Figure 2C) showed a trend toward increasing values for all the guava nectars during storage. This tendency can be explained by the carotenoid content of the nectars, as the higher values of a\* and b\* indicate the red and yellow tones, respectively. According to the observations, the different formulations of the nectars induced color values similar to the control.

The hue angle expresses the intensity of the tone or the color itself, and by definition, the starting point at the +a  $(0^{\circ})$  axis corresponds to red; thus, the nectars obtained from the red pulp of the guava have a hue angle of near  $0^{\circ}$ . The formulations of the nectar with xanthan & guar gum and pregelatinized rice bran flour & xanthan differed from the control at 90 days of storage, while those with pregelatinized rice flour & guar gum differed at 0,45, and 180 days of storage (Figure 2D). Regarding the storage time, there was no significant variation in the hue angle until 45 days. Thus, it can be observed that guava nectars retained their characteristic red color during the studied storage period.

The chroma expresses the association between the values of a\* and b\*, which indicates the intensity or saturation of a color. The different clarifying materials added in the nectars did not result in any color change immediately after the processing with only minor variations occurring at 90 and 135 days of storage (Figure 2E). Among the formulations of nectar, the one that differed the most in relation to the control was the nectar elaborated with pregelatinized rice bran flour & guar gum, which presented a lower value of 9.72 (Figure 2E). For the other formulations, the color intensity increased only slightly during the storage period.

# Bioactive compounds and antioxidant capacity Phenolic Compounds

All the nectar formulations showed very similar phenolic content in comparison to the control immediately after the processing (Figure 3A). A significant degradation in the phenolic compounds of all the formulations was recorded during the storage period, except for 90 days of storage. The pasteurization process might be associated with the degradation of phenolic compounds during the storage, which was approximately 65% after the 90 days storage. Chaikham and Apichartsrangkoon (2012) evaluated the role of pasteurization and pressurization in triggering the loss of bioactive compounds in longan juice and observed a reduction of 38% in the content of phenolic compounds in the pasteurized juices. Igual et al. (2010) observed that the phenolic compounds in the pasteurized grapefruit juice  $(80 \pm 2.5^{\circ}C \text{ for } 11 \text{ s})$  were reduced by 14.64% in relation to fresh juice. Klimczak et al. (2007) and Krumreich et al. (2018) also found a loss of phenolic content in citrus juices during a 4-month storage period.

#### Carotenoids

The formulations of nectars differed from the control in relation to carotenoid content, especially at 0, 90, and 135 days of storage. At 90 days of storage, the

maximum reduction in carotenoids was observed (Figure 3B). Xu et al. (2008), while evaluating fifteen different citrus juices, found a variation from 0.06 to 10.02 mg L<sup>-1</sup> of  $\beta$ -carotene among juices. Ordóñez-Santos et al. (2017) evaluated the differences in the retention of carotenoids by the method employed for processing of gooseberry juice using pasteurization and ultrasound for 10, 20 and 40 min. The authors observed that there was an increase from 24.84% to 90.20% in  $\beta$ -carotene content in the ultrasound treated juices. Abid et al. (2014), Guerrouj et al. (2016), and Santhirasegaram et al. (2013) also reported an increase in the carotenoid content in mango, apple, and orange juices treated with ultrasound, the increase could be ascribed to the mechanical rupture of the cell walls, thereby allowing the release of these compounds.

#### L-ascorbic acid (vitamin C)

As vitamin C degrades easily, it is generally used as a reference in several industrial processes as a measure of high nutritional quality in the final product (COZZOLINO, 2012).

The content of ascorbic acid (vitamin C) varied significantly in the different guava nectars relative to the control nectar, with the exception only in the final storage period (Figure 3C). There was a gradual decrease in L-ascorbic acid in all the formulations of guava nectars after 90 days of storage, and the most significant loss was observed at 135 days of storage. A greater protection of L-ascorbic acid in the nectars was observed by addition of xanthan hydrocolloid. The formulation containing pregelatinized rice bran flour & guar gum contained 15.13 mg 100g<sup>-1</sup> of L-ascorbic acid till 135 days of storage.

The Recommended Daily Intake (RDI) of vitamin C for adults is 45 mg (BRASIL, 2005); therefore, until 90 days of storage, the formulations containing pregelatinized rice bran flour meet this recommendation. The reduction in the vitamin C content of the nectar during the storage may be attributed to the oxidation reactions due to the oxygen present inside the package as well as the dissolved one in the beverage, as the nectar formulations were processed for deaeration. According to Carvalho et al. (2005), the storage temperature and the incidence of light in the transparent glass packaging may also contribute to the reduction in the vitamin C content. The pasteurization, according to Fellows (2009), besides being a relatively mild heat treatment, also alters the nutritional value of foods, especially of vitamin C, since it is heat labile. The chemical oxidation of vitamin C and/or thermal degradation as a consequence of bleaching, cooking, pasteurization, sterilization, dehydration, and freezing has already been reported in the literature (COZZOLINO, 2012).

Hansen (2011), when pasteurizing tropical mango juice at 90°C for 1 min., also observed a marked reduction in the ascorbic acid content from 95.63 mg 100 g<sup>-1</sup> in the pulp to 45 mg 100 g<sup>-1</sup> in the juice.

#### Antioxidant capacity

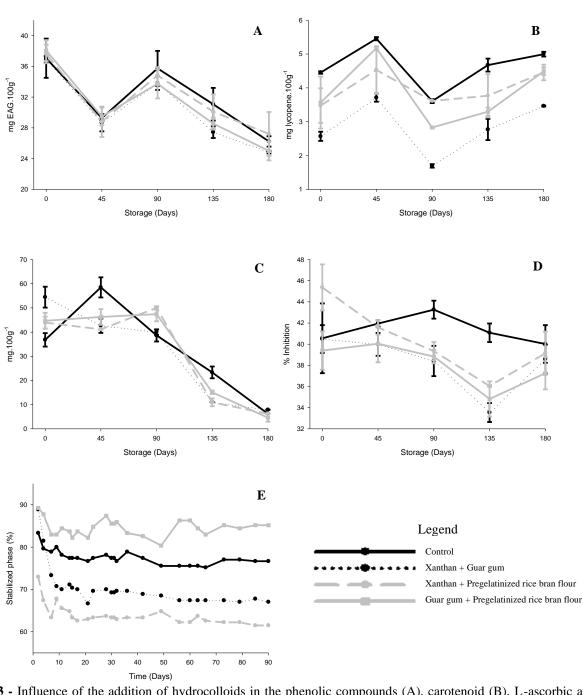
The antioxidant capacity is related to the composition and concentration of bioactive compounds,

# such as vitamins, polyphenols, carotenoids, and flavonoids (SUCUPIRA et al., 2012). A significant difference was observed in the values of the antioxidant capacity between the control and the other formulations of the nectars at 90 and 135 days of storage (Figure 3D). During storage, the value of the antioxidant capacity remained practically constant for all the formulations.

Igual et al. (2010), while evaluating the antioxidant capacity of pasteurized grapefruit juice at  $80\pm2.5$  °C for 11 sec., found that the antioxidant activity decreased

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significantly by 40% compared to fresh juice. Klimczak et al. (2007) in their studies also verified the loss of antioxidant activity in citrus juices as a function of the applied heat treatment. Xu et al. (2008) evaluated fifteen different citrus juices and reported variation in the percentage of DPPH inhibition from 23.69% to 61.62%. In general, the antioxidant activity in guava nectars is high, since guava is rich in phenolic compounds, carotenoids, and vitamin C, which are well-known antioxidants.



**FIGURE 3** - Influence of the addition of hydrocolloids in the phenolic compounds (A), carotenoid (B), L-ascorbic acid (C) antioxidant (D) and sediment (E) nectars guava during storage.

#### **Testing of stabilization of nectars**

The guava nectars demonstrated more rapid sedimentation (Figure 3 E) at the beginning of the storage process, more specifically in the first fifteen days, followed by a slow rise in the precipitate formation until a stable height was reached (at 90 days). The formulations with xanthan & guar gum and xanthan & pregelatinized rice bran flour presented a behavior inferior to the control in relation to the stabilization of the nectars. An interesting behavior was recorded for the formulation of guar gum & pregelatinized rice bran flour, which presented lower sedimentation than the control, maintaining 95% of the volume of the nectar in stable form at the end of 90 days. Sinchaipanit et al. (2013), while evaluating the stability of carrot juice using various hydrocolloids, found that guar gum was the most effective in preventing sedimentation, which corroborates with the present study, where the formulation of nectar containing guar gum together with pregelatinized rice flour also showed the best result in relation to sedimentation.

For the passion fruit nectar, the hydrocolloids that presented best results as stabilizers were carrageenan and guar gum among the studied ones (high and low methoxylation pectins, xanthan, guar gum and carrageenan). Similar to the case of other fruits, the control sample contained no stabilizers (COELHO et al., 2017).

In the present study, xanthan gum was not effective in stabilizing guava nectar at 0.05% concentration, and the same was observed in the study by Coelho et al. (2017). Chaikham and Apichartsrangkoon (2012) pointed out that addition of xanthan gum at 0.15% was ideal to stabilize the longan juice. In order to achieve stabilization of apple juice, Genovese and Lozano (2000) used a concentration of 0.4– 0.5% of xanthan, a concentration that does not comply with Brazilian legislation (ANVISA, 2021), allowing a maximum of 0.2% of xanthan in nectars.

New studies are suggested, with different concentrations of pregelatinized rice flour as a guava nectar stabilizer so that in the near future it can be used as a stabilizer in the food industry, more specifically in nectar processing.

#### CONCLUSION(S)

The physicochemical properties, color, and the bioactive compounds in the guava nectars were influenced mainly in the formulation of pregelatinized rice bran flour & guar gum. The same composition also resulted in greater stability.

Despite the absence of considerable differences in the guava nectar formulations for the evaluated parameters, the addition of stabilizers seems to be necessary for a visual quality aspect, since the sedimentation that occurs in this type of nectar depreciates the appearance of this product, thus compromising its competitiveness.

The pregelatinized rice bran flour associated with guar gum was found to be the most effective strategy for stabilization of guava nectar, demonstrating low cost and ease of obtaining the raw material.

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