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Yellow Mombin Nectar Enriched with Encapsulated Green Tea (*Camellia Sinensis* Var Assamica): Physical-chemical, Rheological and Sensory Aspects

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ABSTRACT

This research aimed to evaluate the feasibility of applying green tea extract encapsulated in yellow mombin (*Spondias mombin* L.) nectar. Two formulations were developed: with microencapsulated green tea extract (NM) and control (NC). NM was the variant concentration of green tea microencapsulated (4.6 g of green tea microcapsules for each 240 mL of beverage). The samples were subjected to physical-chemical and bioactive compounds analysis, such as the study of rheological behavior and sensory analysis. The addition of microcapsules resulted in changes in all of the parameters evaluated, highlighting the contribution in antioxidant activity and total phenolic compounds. The inclusion of microcapsules did not influence the rheological behavior. For the sensory analysis, it was found that NC presented better sensory acceptance. The inclusion of microcapsules of green tea extract in yellow mombin nectar presents itself as a viable alternative for the development of a new product.

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Microcapsules; bioactive compounds; food development

Introduction

Motivated by the population's lack of free time, the food industry increasingly seeks to develop new products that bring consumers the combination of practicality and health benefits, such as fruit-based products. In this way, the growing interest in new functional foods with special characteristics and health properties has led to the development of new beverages (Maia et al., 2019).

In addition, some consumers expect beverages to offer health benefits beyond their natural nutritional content, which has led the beverage industry to enrich these products with ingredients that bring additional health benefits (Kasapoğlu, Daşkaya-Dikmen, Yavuz-Düzgün, Karaça, & Özçelik, 2019). A challenge found in the development of these products is to maintain the natural flavor of the beverages and to protect the integrity of the additives until the end of processing, which requires strategies like microencapsulation.

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Microencapsulation is a promising technology to protect and deliver compounds (Speranza et a., 2017) and it has been used in beverages, such as fish oil microcapsules added to pomegranate juice (Habibi, Keramat, Hojjatoleslamy, & Tamjidi, 2017) and norbixin microcapsules in an isotonic tangerine soft drink (Tupuna-Yerovi et al., 2020).

The microcapsules containing green tea (*Camellia sinensis*) extract have the potential to be included in beverages, considering that they are quite costly and easy to obtain, have a high content of bioactive compounds and are not considered toxic (Silva et al., 2018).

The leaves of *Camellia sinensis* have a high amount of catechins, reaching about 30% of its dry weight. Catechins are colorless, water-soluble and responsible for the bitterness and astringency of green tea (Camargo, 2011; Lima, Mazzafera, Moraes, & Silva, 2009).

The characteristic bitterness of these leaves often makes it impossible for some people to consume this tea. Sensory characteristics of functional beverages have been improved using encapsulation technology creating innovative products (Ozdal, Yolci-Omeroglu, & Tamer, 2020)

Thus, the encapsulation of green tea possibly could mask this sensory characteristic and the inclusion of these capsules in a drink can further facilitate consumption and thus the supply of bioactive substances present.

Yellow mombin (*Spondias mombin L.*), commonly grown in Nigeria, Brazil, and several other tropical forests (Yadav, Srilekha, Barbhai Mrunal, & Uma Maheswari, 2018), presents in its constitution several bioactive compounds (Silva et al., 2014), such as vitamin C, phenolic compounds (Freitas et al., 2020), carotenoids (Costa & Mercadante, 2018), and flavonoids (Sousa et al., 2019). Several health benefits have been associated with the consumption of yellow mombin pulp, such as cardiac remodeling after myocardial infarction (Pereira et al., 2020), healing of the chronic ulcer, regeneration of the gastric mucosa, and restoration of mucus levels in glandular cells (Brito et al., 2018), and may also have benefits for kidneys and liver (Coolborn, Chinedu, Abiola, & Olowatosin, 2018).

Thus, the development of a drink containing microcapsules of green tea extract will combine the benefits associated with the consumption of green tea and the benefits associated with the consumption of yellow mombin, with the possibility of synergism between the compounds present, in addition to masking the flavor of green tea, often considered unpleasant and making consumption by some people unfeasible.

For the development of food products, it is necessary to study the characteristics of the food, highlighting several aspects, such as physical-chemical, sensory and study of rheological behavior. Thus, this work aimed to evaluate whether the addition of cashew gum and maltodextrin microcapsules added with green tea extract in yellow mombin nectar results in changes in the product's acceptability, composition and rheological behavior.

Materials and methods

Pulps and microcapsules

For this research, yellow mombin pulp was used without the addition of preservatives, obtained from a local market in Fortaleza (CE-Brazil). The pulps were stored in a freezer at −18 ± 1°C until they were used for nectar development.

The microcapsules were obtained following the methodology described by Silva et al. (2018), containing 20% cashew gum, 10% maltodextrin and 7.5% dry green tea extract. The cashew gum used was obtained from the exudate of dwarf cashew trees (*Anacardium occidentale* L.) grown in the Experimental Field of Pacajus, Embrapa Agroindustry Tropical (CE-Brazil). The maltodextrin used in the experiments was obtained from a local market in Fortaleza-CE, from the brand "Cargill" and equivalent dextrose under 20%.

Formulations

The yellow mombin pulps were unfrozen in the original product packaging in a refrigerator (5°C/18 hours). The pulps went through a mass balance so that drinking water and sugar were added to obtain a product that meets the identity and quality standards stipulated by BRASIL.Ministry of Agriculture and Supply. Normative Instruction no. 12 (2003), with a minimum of 25% of yellow mombin pulp.

Two formulations of yellow mombin nectar were prepared: yellow mombin nectar with microencapsulated green tea extract (NM) and yellow mombin nectar without the extract (NC).

The treatment (NM) was the variant concentration of green tea microencapsulated. Due to the lack of studies that prove the minimum and maximum amount of green tea that should be consumed per day, the added amounts of microencapsulated green tea extract were based on recommendations found in the literature. In this work, the recommendation of the American Dietetic Association was taken as reference, which suggests the consumption of 4 to 6 cups of green tea per day, as well as the recommendation given by Lamarão and Fialho (2009) who say that the daily dose should be equivalent to about 240 mg of catechins.

According to the European Food Safety Authority (EFSA), consumption of up to 400 mg of caffeine a day (about 4 cups of coffee) is recommended for adult individuals weighing 70 kg on average and who are not pregnant, consequently not representing significant health risks. For pregnant or lactating women, the supposedly safe value would be 200 mg of caffeine a day, since consumption in greater quantities may hinder intrauterine brain development and represent a risk factor for diseases such as epilepsy (Toledo, 2015).

Given the above and the concentrations of catechins and caffeine found according to Silva et al. (2018), to obtain 240 mg of catechins from the studied green tea dry extract, it was necessary to add 4.6 g of green tea microcapsules for each 240 mL (1 cup) of yellow mombin nectar. Consequently, the total amount of caffeine contained in the added capsules was 205.8 mg, not exceeding the recommended daily amount for adults and non-pregnant women.

Nectar characterization

Based on instrumental analyses of microcapsules (Silva et al., 2018), aiming to protect bioactive compounds from green tea extract, as well as the viable use of cashew gum as a wall material, only one microcapsule sample was chosen to be used as an ingredient that promotes the increase of antioxidant capacity and bioactive compounds in conventional foods, using yellow mombin nectar as a model system. The formulations of nectars, yellow mombin nectar with microencapsulated green tea extract (NM) and yellow mombin nectar (NC), were characterized by physical-chemical analysis to evaluate their functional compounds and rheological behavior. All analyses were performed in triplicate, expressed as mean ± SD (n = 3).

Chemical and physicochemical analysis

The formulations were submitted to the following analysis: pH using a pH meter (Hanna Instruments, Villafranca, Padovana, Italy); titratable acidity, according to AOAC (1995), expressed as "g citric acid/100 ml nectar"; soluble solids (°Brix), measured at 20°C using an automatic Atago PR-101 digital refractometer (Atago, Norfolk, VA, USA); reducing and total sugars, according to Miller (1959), by the 3,5-dinitrosalicylic acid (DNS) method.

Total phenolic compounds were determined according to the Folin-Ciocalteu method (Larrauri, Rupérez, & Saura-Calixto, 1997). In this method, the values were calculated from a calibration curve obtained with five different concentrations of gallic acid.

The ability to scavenge free radicals from extracts was determined by Ferric Reducing Antioxidant Power (FRAP) and the scavenging ability of the cation free radical derived from 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) ($ABTS^{\bullet+}$). The determination of total antioxidant capacity through FRAP and ABTS followed the methodology described by Benzie and Strain (1999) and adapted by Rufino et al. (2010). 6-hydroxy-2,5,7,8-tetramethyl-chroman-2-carboxylic acid (Trolox) was used as the standard antioxidant to plot calibration curve in ABTS analyses.

Rheological behavior

The rheological behavior of yellow mombin nectars was determined using a concentric cylinder rheometer (Brookfield Searle, model R/S SST Plus 2000, Brookfield Co., USA). Determinations were carried out at 8°C provided by a thermostatic bath (Brookfield, model TC-502) connected to the equipment. Results of shear stress and shear rate were recorded using the RHEO V2.8 software (Brookfield Co., USA), according Silva et al. (2018).

The rheological behavior was measured with a shear rate ranging from 0 to 500 s^{-1} (upward curve) and from 500 to 0 s^{-1} (downward curve) for 1 min with 25 readings for each curve. Readings were carried out in triplicate and, for each measurement, a new sample was used. The Ostwald-de Waele, Herschel-Bulkley and Casson models were used to fit the rheological behavior of yellow mombin nectar due to its simplicity and good fit. The experimental results were fitted to the Ostwald-de Waele, Casson and Herschel Bulkley model using SAS Software version 9.1 (Silva et al., 2012).

Sensory analysis

Juice consumers were recruited among employees and collaborators of Embrapa Agroindustry Tropical, Fortaleza-CE, as specified by Meilgaard, CARR, and Civille (2006) and Stone and Sidel (1993). The public, consisting of 58 untrained individuals, was characterized in terms of gender and age and asked to sign a Free and Informed Consent Form (ICF). The protocols for sensory tests were previously approved by the Research Ethics Committee of the Federal University of Ceará, under n° 147.279.

Sensory tests were performed at the Sensory Analysis Laboratory of Embrapa Agroindustry Tropical, in Fortaleza – CE. Affective tests of acceptance and intensity of strange flavor were applied with the two formulations of nectars (NC and NM). The analysis of the drinks was performed in individual air-conditioned booths (24°C), under controlled lighting (white, fluorescent light) and equipped with computer terminals for automatic data registration/collection using the FIZZ software. About 30 mL of each sample was served in 100 mL glass bowls, encoded with random three-digit numbers, capped with a watch glass and presented in a balanced and monadic manner (Macfie et al., 1989) to minimize the effects of position of the samples. A glass of mineral water was offered between the samples to eliminate the residual taste from the subject's mouth.

The affective tests included the analysis of global acceptance and acceptance of the appearance, aroma and flavor attributes, using the 9-point verbal hedonic scale, ranging from "I liked it a lot" to "I disliked it a lot" (Peryam & Pilgrim, 1957). In the evaluation of the intensity of strange flavor, a structured 9-point scale was used, anchored at its ends with terms referring to the intensity (0 = none and 9 = Strong).

For the purpose of statistical analysis, the categories of the hedonic scale were associated with numerical values, 9 = "Like extremely," 8 = "Like very much," 7 = "Like moderately," 6 = "Like slightly," 5 = "Neither like or dislike," 4 = " Dislike slightly," 3 = "Disliked moderately," 2 = " Dislike very much" and 1 = "Disliked extremely." The data from the acceptance tests, as well as the intensity of the strange flavor, were submitted to analysis of variance (ANOVA) and F test with a level of significance of 5% for comparison of means, using the statistical program Statistical Analytical Systems – SAS User's Guide: Version 6.11.

Results and discussion

Chemical and physicochemical analysis

The inclusion of microcapsules in yellow mombin nectar resulted in changes in all parameters evaluated (Table 1). The addition of the microcapsules to the yellow mombin nectar did not cause a great variation for the pH values (Table 1), which were 3.14 for NM and 3.08 for NC, despite having a significant difference with the pH values being below 4.5.

Lower pH values are preferred by the industry, since pathogenic bacteria do not find favorable conditions to develop, thus being a product of easier conservation (Hoffmann, 2001). Further research involving the inclusion of bioactive compounds encapsulated in fruit juices has been carried out. Dima et al. (2020) found pH close to 4, greater than that obtained in this research, which is justified by the difference between fruits. The authors also found no differences in pH after the inclusion of the particles.

The levels of total soluble solids found were 17.3 and 11.0 °Brix for NM and NC, respectively, as proposed by the Brazilian legislation for yellow mombin nectar, which recommends a minimum value of 11 °Brix (BRASIL.Ministry of Agriculture and Supply. Normative Instruction no. 12, 2003) and 20 °Brix (FAO, 2013). In studies with yellow mombin nectar formulations, De Souza Filho et al. (2000) found values that varied between 12.2 and 14 ° Brix and a value of 16.3 ° Brix for araçá-boi nectar, similar values to those obtained in this study. Melo, Dantas, de Oliveira, da Costa, and de Medeiros Santos (2020) found soluble solids values between 6.4 and 14.4 for yellow mombin pulp.

Table 1. Physical and chemical characteristics of nectars (NM and NC).

Sample	pH	Soluble solids ° Brix (20°C)	Total acidity (%)	Total sugars (g/100 g)	Reducing sugars (g/100 g)
NC	3,08 ± 0,006 [b]	11,0 ± 0,1 [b]	0,21 ± 0,0007 [b]	7,46 ± 0,46 [b]	5,06 ± 0,13 [b]
NM	3,14 ± 0,01 [a]	17,3 ± 0,06 [a]	0,28 ± 0,02 [a]	11,12 ± 0,14 [a]	6,85 ± 0,44 [a]

* NC = Yellow mombin control nectar and NM = Yellow mombin nectar with microencapsulated green tea extract.
Means followed by equal letters in the same column do not differ significantly for the F test ($p > 0.05$).

The values found for titratable acidity in citric acid was 0.28% and 0.21% for NM and NC, respectively. These values are in accordance with the Identity and Quality Standard for yellow mombin nectar (BRASIL.Ministry of Agriculture and Supply. Normative Instruction no. 12, 2003), which establishes a minimum value of 0.20% for total acidity in citric acid. Similar values to those presented in this study were obtained by De Castro et al. (2014) who found titratable acidity values around 0.25% in mixed pineapple and buttercup nectar formulations.

The percentage of total sugar was 11.12% and 7.46% for NM and NC, respectively. These results are also in accordance with the Identity and Quality Standard for yellow mombin nectar (BRASIL, 2003), which establishes a minimum of 7.0% of total sugars. The reducing sugars (% of glucose) were 6.85% for NM and 5.06% for NC. Damiani et al. (2011), in a study with mixed nectar of yellow mombin-mango and mint, found values of 14.24 and 6.62% for total and reducing sugars, respectively. The composition in plants varies with species, variety, physiological conditions, soil, climate, and agricultural practices, among other factors (Medina-Medrano et al., 2015), therefore justifying the differences found for sugars.

The addition of the microencapsulated green tea extract to the yellow mombin nectar resulted in a significant increase in the levels of all the functional compounds evaluated in the samples (Table 2).

The values obtained for total extractable polyphenols were 302.56 ± 4.20 for NM and 159.60 ± 4.47 mg/100 g for NC. We observed that the value obtained for the NM sample practically doubled when compared to that determined for the NC, which demonstrates that the addition of the microcapsules to the nectar is a good strategy to make those compounds available in beverages.

The bioactivity exerted by green tea phenolic compounds has been associated with reduced risk of inducing severe illnesses such as cancer, cardiovascular and neurodegenerative diseases (Lorenzo & Munekata, 2016). Besides, yellow mombin is recognized for having yellow flavonoids, anthocyanin, phenolic compounds (Silva et al., 2014) and dietary fiber (Schiassi, de Souza, Lago, Campos, & Queiroz, 2018), that also presents health benefits.

Abreu et al. (2011) developed a mixed fruit beverage using yellow mombin, cashew apple and passion fruit pulps and found phenolic compounds ranging from 51.70 to 62.59 mg GAE*/100 g, values lower than those obtained in this

Table 2. Total Extractable Polyphenols (TEP), antioxidant activity obtained for NM and NC nectars.

Samples	TEP(mg GAE*/100 g)	Antioxidant activity/ $ABTS^{\bullet+}$ (μM of Trolox.g^{-1})	Antioxidant activity/ FRAP (μM of Ferrous Sulfate.g^{-1})
NC	159.60 ± 4.47	2.97 ± 0.19 ^{b}	11.03 ± 1.02 ^{b}
NM	302.56 ± 4.20	10.90 ± 1.68 ^{a}	17.63 ± 1.19 ^{a}

* NC = Yellow mombin control nectar and NM = Yellow mombin nectar with microencapsulated green tea extract.
Means followed by equal letters in the same column do not differ significantly for the F test (p > 0.05).

research, even though the authors' formulation contained cashew apple pulp, which has a high phenolic content (5286 mg AGE/100 g pulp dry basis) (Silva et al., 2014).

For the antioxidant activity obtained from the ABTS$^{\bullet +}$ method, values of 10.90 and 2.97 μM of Trolox/g were found, respectively, for NM and NC, showing that the addition of microcapsules to the juice increased significantly the content of antioxidant compounds to yellow mombin nectar.

Silva et al. (2018) found values ranging from 12.24 to 28.80 μM Trolox.g^{-1} in beverages based on açaí, acerola and mombin pulp, being the value similar to NM.

According to Table 15, the values obtained for the antioxidant activity determined from the reduction of iron were 17.63 μM Fe^{2+}/g for NM and 11.03 μM Fe^{2+}/g for NC. A higher value for NC was also observed, going according to the results obtained by ABTS.

Cheuczuk and Rocha (2014) found a value of 9.43 μM Fe^{2+}/ g in samples of cashew-mango pulp.

Rheological behavior

The nectars were characterized as non-Newtonian fluids, for they presented a behavior index lower than the unit (Figure 1, Table 3). The data showed suitable adjustments to the Ostwald-de Waele and Casson models, with

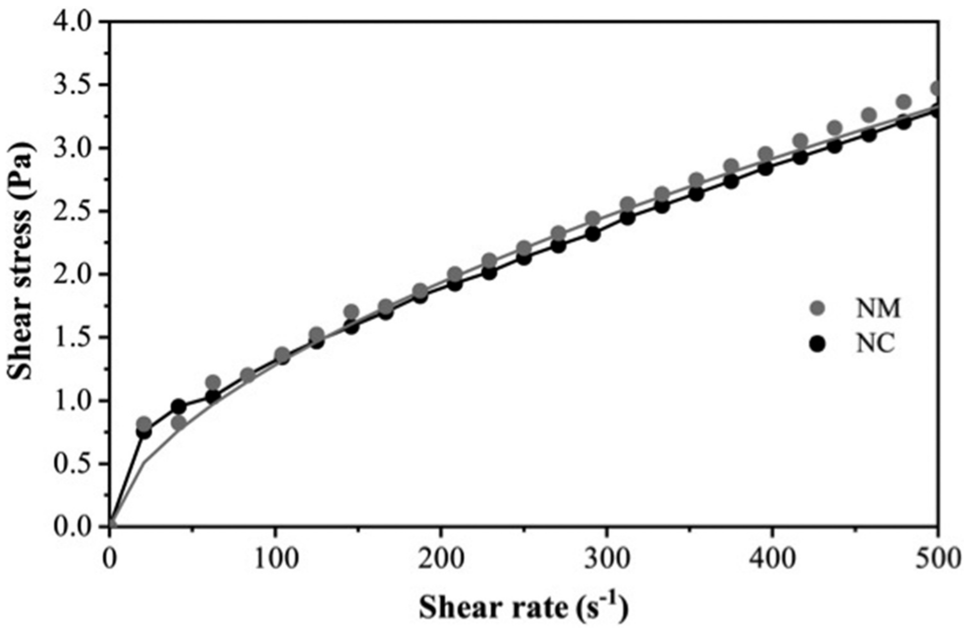

Figure 1. Relationship between shear stress and strain rate for the yellow mombin nectar for different rheological models. points represent the experimental data obtained, and the solid line represents the behavior expected of the model. * NC = Yellow mombin control nectar; and NM = Yellow mombin nectar with microencapsulated green tea extract.

Table 3. Parameters of the Ostwald-de Waele, Herschel-Bulkley and Casson rheological models for yellow mombin nectar added with green tea microcapsules.

Model		NM	NC
Ostwald-de Waele	**K**	0.102 ± 0.005	0.084 ± 0.003
	n	0.545 ± 0.008	0.592 ± 0.006
	$\mathbf{R^2}$	0.99	0.99
	QME	0.008	0.003
Herschel-Bulkley	$\mathbf{t_0}$	0.223 ± 0.045	0.332 ± 0.036
	$\mathbf{K_0}$	0.054 ± 0.008	0.034 ± 0.004
	n	0.635 ± 0.022	0.729 ± 0.019
	$\mathbf{R^2}$	0.99	0.99
	QME	0.007	0.002
Casson	$\mathbf{K_1}$	0.569 ± 0.011	0.542 ± 0.008
	$\mathbf{K_2}$	0.053 ± 0.000	0.058 ± 0.000
	$\mathbf{R^2}$	0.99	0.99
	QME	0.008	0.003

a coefficient of determination close to the unit and mean square error close to zero. The nectar behavior was similar to that found for yellow mombin pulp, according to Melo et al. (2020). This behavior presents industrial interest, considering that the same pumping conditions used for the fruit pulps can be used for the studied nectars, without industrial adjustments.

The addition of the microcapsules did not result in drastic changes in the rheological behavior of the yellow mombin nectar, however, higher initial stress was found for the sample containing the microcapsules, according to the Herschel-Bulkley model. This behavior can be attributed to the additional interaction of the microcapsules with the components of the beverage, making it difficult to start the flow. However, the amount of material added was not sufficient to cause major changes in the rheological profile. All the evaluated models can be used to describe the rheological behavior of the yellow mombin nectar; and the addition of green tea extract microcapsules, in the concentration used, can be performed without changing the rheological characteristics of the fluid.

Silva et al. (2012) evaluated the rheological behavior of mixed beverages based on yellow mombin and mango pulp. The samples showed non-Newtonian behavior with a pseudoplastic character, and the Herschel-Bulkley model presented a better fit for the data obtained. For this research, the three models evaluated presented similar adjustments to the data. Liu et al. (2019) studied lily pulps' flow behavior and also found good data fitted to the Herschel–Bulkley rheological model ($R^2 > 0.995$).

Sensory analysis

Table 4 shows the results of the sensory analysis obtained for the samples of yellow mombin nectar evaluated. It can be observed that the statistical treatment revealed significant differences ($p \leq .05$) between the samples concerning

Table 4. Averages of sensory results from the yellow mombin nectar (NC and NM) samples.

Sample	Strange taste	Global acceptance	Appearance acceptance	Aroma acceptance	Flavor acceptance
NC	1.06^{b}	7.93^{a}	7.89^{a}	7.53^{a}	8.01^{a}
NM	4.42^{a}	6.27^{b}	7.32^{b}	6.48^{b}	5.79^{b}

* NC = Yellow mombin control nectar; and NM = Yellow mombin nectar with microencapsulated green tea extract.
Means followed by equal letters in the same column do not differ significantly for the F test (p > 0.05).

the intensity of the strange flavor; and the yellow mombin nectar with microencapsulated green tea extract (NM) showed greater intensity than the nectar without the extract (1.06). This demonstrates that the addition of green tea extract, even microencapsulated, gave a strange flavor to the yellow mombin nectar, which can be classified as moderate, since its average value (4.42) was in the middle of the scale used (9 cm).

The presence of the strange flavor impaired the acceptability of the nectar that contains green tea microcapsules (NM), because, concerning the "global acceptance," this sample obtained a lower average than the sample without the extract (NC). However, this was not a criterion for the rejection of the nectar, since the sample reached an average of 6.27, a hedonic value corresponding to "slightly liked," and therefore, it is in the region of acceptance on the hedonic scale.

Mamede, Kalschne, Santos, and Benassi (2015) obtained an average of 6.4 for global acceptance of the yellow mombin nectar, a similar result to that obtained in this study for the sample that contained the green tea extract microcapsules.

The attribute acceptance test shows which sensory attributes were most affected by the addition of green tea extract. Although this difference was statistically significant, the product formulated with the microcapsules lost only 0.57 points in its acceptability, indicating an excellent acceptance of the nectars by the tasters. The aroma, on the other hand, suffered a little more on the hedonic scale.

However, in terms of flavor, a more pronounced drop in the acceptance of the NM sample by the tasters (5.79) was observed, compared to the yellow mombin nectar not added with the microcapsules (8.01), indicating that this was the attribute most influenced by the green tea extract in the yellow mombin nectar, also impacting the product's global acceptability.

Conclusions

The model system of the yellow mombin nectar, used to add green tea microcapsules, proves its ability to turn a conventional food into a potential source of antioxidants and phenolic compounds without causing rejection of the product. Thus, there is a potential for the industry to use microparticles based on dry green tea extract as a food ingredient.

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