

Ultra-refined yerba mate (*Ilex paraguariensis* St. Hil) as a potential naturally colored food ingredient

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ABSTRACT: Ultra-refined yerba mate (URYM), with a final particle size of 5 to 12 microns, is an innovation in the market that aims to diversify and increase the consumption of yerba mate (*Ilex paraguariensis* St. Hil) through its practicality and versatility of preparation. The present work seeks to evaluate the potential for applying naturally colored URYM in food products. The nature of this study was exploratory, adopting a chemical approach (analysis of the antioxidant potential); physical approach (color stability) and sensory perception. URYM is a suitable naturally colored food ingredient with natural antioxidant appeal. Methanolic extraction of URYM (MEUR) was more efficient in scavenging DPPH radicals compared with aqueous extract of URYM (AEUR) (959.5 vs. 638.1 $\mu\text{mol Trolox g}^{-1}$). No differences were found between AEUR and MEUR for total phenolic content in mg GAE g^{-1} (266.4 and 339.0, respectively) and scavenging of ABTS radicals in $\mu\text{mol Trolox g}^{-1}$ (1008.9 and 1053.8, respectively). Water was able to extract phenolic compounds with antioxidant activity. Ice cream, juice, cake, and cookies emerged as the food products in which consumers used URYM in their homemade foods. With good color stability (no difference between L^* , a^* and b^* parameters during the six-day) and good acceptance, cake with URYM added has a profile described as tasty, sweet, soft, and mate flavor. The results also demonstrated consumer interest in food products and ingredients with a healthy appeal.

Keywords: Check-all-that-apply, consumer behavior, natural antioxidant, natural colorant, word association

Introduction

Yerba mate (*Ilex paraguariensis* St. Hil) is a plant with significant economic and cultural importance for certain regions of South America. It is widely consumed as a traditional beverage in its regions of origin: mate (traditional regional hot drink) or tea bag (Bracesco et al., 2011; Mesquita et al., 2021; Pagliosa et al., 2009; Vázquez and Moyna, 1986). The leaves of the yerba mate species contain a complex chemical composition of elements such as phenols, methylxanthines, theobromines, saponins, minerals which confer functional properties on the human organism (Mesquita et al., 2021). Regarding antioxidant properties, phenolic compounds are known for their great antioxidant capacity, direct participation in the regulation of metabolic energy, and anti-inflammatory and anti-tumor properties (Bracesco et al., 2011). Although much is known about the composition of yerba mate and its health benefits, it has a limited market, primarily concentrated in its growing regions. It is worth noting that this finding is not new, as Vázquez and Moyna (1986) had already highlighted the difficulties of innovation in the yerba mate market and attributed this resistance to the deeply rooted habitual use of mate.

Given this background, ultra-refined yerba mate (URYM) emerges as an innovative product due to its versatility of use and its nutraceutical appeal. The production process characterizes the uniqueness that the yerba mate goes through, namely, specific granulometry, which gives it more solubility than the traditional product.

Processing does not compromise its sensory quality. This is an essential factor since it is known that sensory appeal is one of the most critical food choice motivators (Veiga et al., 2021).

Food appearance is of primary importance to consumers and color is considered one of the most significant attributes in food appearance (Bordim et al., 2021; Cömert et al., 2020; König and Renner, 2018; Paakki et al., 2016; Schifferstein et al., 2019). Studies have shown an association between colorful foods and healthy foods (Cömert et al., 2020; König and Renner, 2018), and therefore the effect of food color on eating habits (König and Renner, 2018). Yerba mate is characterized by its green color caused by the presence of chlorophyll compound and its use as an ingredient that creates a green product (Knapp et al., 2019). The present study evaluates the potential for applying naturally colored URYM in food products. The nature of this study was exploratory, adopting a chemical approach (analysis of the antioxidant potential), a physical approach (color stability) and sensory perception.

Materials and Methods

Analysis of the antioxidant potential of URYM

Samples

The URYM (Figure 1) was supplied by a yerba mate producer located in the central-southern region of



Figure 1 – Ultra-refined yerba mate (URYM). Source: @MateTeaBrasil

Paraná, in the municipality of Laranjeiras do Sul – 25°38'52" S; 52°41'39" W; altitude 841 m). URYM processing is based on the collection of mate residue generated in the "soque" (equipment used to crush the mate leaves), followed by sieving, in which there is a reduction in the concentration of sticks down to a maximum of 1 %. Final refining produces a URYM with a final particle size of 5 to 12 microns.

Extraction of antioxidant compounds

The methanolic (MEUR) and aqueous extract (AEUR) of URYM were prepared using 2 g samples and, respectively, 25 mL of a mixture of methanol and water (80 %) (Pagliosa et al., 2010). The extracts were homogenized and subjected to an ultrasound bath for 15 min at 25 °C. The mixture was then filtered and ethanol solvent was removed using a rotary evaporator at 40 °C and approximately 120 mbar. The residual water was freeze-dried. After this, solutions of 1000 mg L⁻¹ from methanolic and aqueous extracts of URYM were produced.

Reducing capacity of the Folin-Ciocalteu reagent (FCR)

Tests were performed using 500 µL of the methanolic (MEUR) and aqueous (AEUR) extracts of URYM (500 µg mL⁻¹), 2.0 mL of Na₂CO₃ 4 %, and 2.5 mL of Folin-Ciocalteu reagent diluted ten times (Singleton et al., 1999). The test tubes were incubated for 2 h without light, and absorbance was measured at 740 nm on a spectrophotometer. As a standard for the calibration curve, phenolic compound gallic acid was used, and the final results were expressed as mg gallic acid equivalent (GAE) g⁻¹ of yerba mate.

Antioxidant activity by scavenger of 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical

To perform the assay, 300 µL of DPPH radical solution (0.5 mmol L⁻¹) was incubated without light for 1 h with

3 mL of ethanol and 500 µL of the MEUR and AEUR at a concentration of 500 µg mL⁻¹ (Brand-Williams et al., 1995). Trolox was used as a standard, and the results were expressed as µmol Trolox g⁻¹ of yerba mate.

Antioxidant activity by scavenger of 2,2'-azino-bis (3-ethylbenzotiazolin) 6-sulfonic acid (ABTS) radical

The assay was performed according to the methodology suggested by Rufino et al. (2007). First, 3.0 mL of the ABTS radical was mixed with 30 µL of MEUR and AEUR at 500 µg mL⁻¹. The standard Trolox was used as a reference, and absorbance of samples and calibration curve were read at 734 nm after 6 min of reaction. The final results were expressed as µmol of Trolox equivalent antioxidant capacity TEAC g⁻¹ of yerba mate.

Sensory analyses

Participants

The test participants were students and lecturers randomly selected at the Universidade Tecnológica Federal do Paraná. The participants were aged between 20 and 55 years old. The tests were performed in a sensory analysis laboratory according to ISO 8589 (ISO, 2007). Participants were provided with an informed consent form at the beginning of the questionnaire. The Ethics Committee for Human Research of Universidade Tecnológica Federal do Paraná approved the study (CAAE number 18936719.9.0000.5547).

Preference ranking test and frequency of yerba mate consumption

A sheet with images of eight food products with green color (supposedly with added URYM) (Figure 2) was presented to 67 participants. The products were: macaroon, spaghetti, bread, cookie, flan, ice cream, cake, and juice. Participants were asked to rank the samples in ascending order (from least preferred to most preferred) according to preference or interest in preparing the food product at home, adding URYM as an ingredient in the recipe. The ranking test was carried out according to ISO (2006). Based on ranking test results for the following tests, one food product was chosen to add URYM as an ingredient to the recipe.

"How frequently do you consume yerba mate?" was also a question that was asked of this group of participants and was measured on a 7-point scale: (7) Daily- (6) several times a week- (5) weekly- (4) several times a month- (3) monthly- (2) less than monthly- (1) never. Additionally, the participants were asked to indicate the way they consume (if they consume) yerba mate: mate (a traditional regional hot drink); tea bag; or other.



Figure 2 – Sheet with images of green colored food products. The images are distributed with a Non-commercial attribution 3.0 Unported (CC BY-NC-ND 3.0) Licence: <https://creativecommons.org/licenses/by-nc/3.0/>. Source: PowerPoint Images Copyright. Numbers: code of three-digit random number.

Preparation of the food product with added URYM

The cake was the product chosen. The recipe for the sponge cake with added URYM was based on the concentrations suggested by the producer Mate Laranjeiras (one teaspoon of URYM to 50 g of wheat flour). The ingredients used in the cake formulation were: wheat flour (24 %); egg (9.7 %); whole milk (23.66 %); soybean oil (5.17 %); sugar (30.78 %); baking powder (2.37 %) and URYM (4.32 %). The sponge cake preparation was based on the Köten (2021) method. All the dry ingredients were mixed until homogeneous before being added to the cake batter (made with milk, soybean oil and egg). The batter was placed in baking pans greased with oil and baked for 40 min at 230 ± 3 °C. Next, the cakes were left to cool in the baking pans for 10 min. They were then removed from the pans and cooled at room temperature for 30 min on cooling racks. Until the analysis they were kept in the dark polyethylene pots at room temperature (25 ± 1 °C). Sponge cake samples were prepared 24 h before sensory tests.

Color stability assessment of the food product with added URYM

The color stability of the food product with added URYM was evaluated by the colorimeter (KONICA MINOLTA®), model CR-400, using the D65 illuminant. The cake was cut into pieces with dimensions of 6 × 6

× 6 cm and kept in closed dark polyethylene containers at room temperature (25 ± 1 °C). Over six days, they were periodically analyzed for the colorimetric parameters L* (Lightness), a* (green to red coordinate), *b (blue to yellow coordinate), Hue (hue angle) and C* (Chroma). Values were measured in six places, with the instrument sensor resting on the central part of the cake. Additionally, values of ΔE (total color difference) were calculated using Eq. (1) (Mitterer-Daltoé et al., 2012):

Word association to secure a holistic perception of the cake with URYM added and hedonic test

To assess the holistic and hedonic perception of the cake with added URYM, the Word Association (WA) and facial hedonic scale techniques were applied. Fifty consumers (Bordim et al., 2021; Zen et al., 2020) participated in the cake evaluations. Before sensory analysis, the samples were sliced into equally sized pieces (2 cm thick) and served on plastic dishes coded with a 3-digit random number.

Word association was first applied according to Bordim et al. (2021), Latorres et al. (2016) and Mitterer-Daltoé et al. (2021). After the participants received the cake sample (stimulus), they were asked to "Write the first four words, sensations or feelings that come to your mind when you taste the cake with ultra-refined yerba mate added". Next, the hedonic test was applied using a facial 'smiley' scale with five points. Each consumer was asked to mark with an X the emoji that identified their level of overall acceptance of the tested product.

Check-all-that-apply (CATA) to obtain consumers' expectations of health issues involved in the URYM product

To assess perceptions and expectations of health issues involved in the URYM product, a CATA question was applied to 50 participants (Zen et al., 2020). For each statement, an opposite sentence was presented to obtain reliable results. Participants who marked the statement and its opposite were eliminated. The sentences were presented in a randomized way (Belusso et al., 2016).

Data analysis

Results from the ranking test were evaluated by Friedman's test and significant mean difference (ISO, 2006). The Tukey test was applied to evaluate the difference between the color parameters over six-days. Word Association data analysis was based on Latorres et al. (2016). Associations were grouped into different categories. The researchers independently created the groupings and the final categories were discussed and defined by the researchers. Only those categories which were mentioned by more than 5 % of participants were included in the final analysis.

To better understand and visualize the categories that represent cake with added URYM, a word cloud map, generated by the Orange 3.23 software program, was applied

The acceptance index (AI) was calculated from the mean value obtained, where the maximum value on the scale (five) was 100 %. Multinomial logistic regression analysis was applied to evaluate the effects of the variables of age and gender on the acceptance of the cake (Latorres et al. (2016)). The Z test analyzed the CATA data for two proportions carried out in all statements with a 5 % significance level ($\alpha = 0.05$). The data were analyzed using Statistica 12.7.

Results

Antioxidant potential of URYM

The antioxidant activity obtained by scavenging the synthetic radicals DPPH and ABTS (Figure 3A) and reducing the capacity of the Folin-Ciocalteu reagent (FCR) (Figure 3B) obtained for methanolic (MEUR) and aqueous extracts (AEUR) of URYM was observed.

The results obtained for antioxidant activity indicated MEUR was more efficient in scavenging DPPH radicals and that there were no differences between AEUR and MEUR in terms of total phenolic content and scavenging of ABTS radicals. The differences between assays result from differences in method and experimental conditions. The FCR and DPPH methods were based mainly on a Single Electron Transfer (SET), while scavenging of ABTS using non-physiological radical was based on both SET and HAT (Hydrogen Atom Transfer)

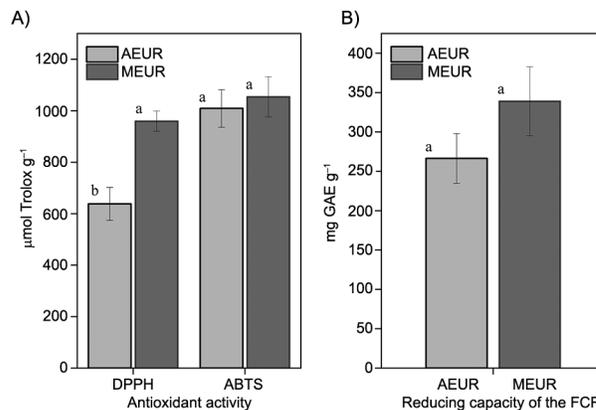


Figure 3 – A) Antioxidant activity. B) Reducing capacity of the Folin-Ciocalteu reagent (FCR). Data are expressed as mean \pm standard deviation. Means followed by different letters are statistically different ($p \leq 0.05$), using the t-test. MEUR (methanolic extract of URYM) and AEUR (aqueous extract of URYM). URYM = ultra-refined yerba mate; DPPH = 2,2-diphenyl-1-picrylhydrazyl. ABTS = 2,2'-azino-bis (3-ethylbenzotiazolin) 6-sulfonic acid.

mechanisms (Prasniewski et al., 2021; Prior et al., 2005). The DPPH methodology is most suitable for hydrophilic systems, whereas ABTS assay is recommended for both lipophilic and hydrophilic systems. This could explain the similarities between AEUR and MEUR in the ABTS assay.

The solvent extraction capacity is related to the solubility of the analytes in the solvent and the interactions of the analytes with other sample constituents (Oldoni et al., 2019). Ethanol and methanol solvents are more efficient in dissolute phenols and flavonoids than water (Rodríguez-Pérez et al., 2015). Since methanol has a dipole moment (1.70 D) lower than water (1.85 D), a variation in the composition and content of phenolic compounds extracted is expected, which can also affect the antioxidant activity. URYM is a good source of phenolic compounds. The wide variety of chemical structures in this group lends different polarities to this class of phytochemicals (Prior et al., 2005) which can be affected by the solvent used in extraction. Thus, our results (Figure 3A and B), indicate that water was able to extract phenolic compounds with antioxidant activity. Our study reported 52 and 70 mg GAE g^{-1} of phenolic compounds in *I. paraguariensis* leaves from methanolic and aqueous extracts, respectively (6.5 and 3.8 times lower than results obtained in this study) (Pagliosa et al., 2010). Other authors obtained 319 and 300 $\mu\text{mol Trolox } g^{-1}$ in the DPPH assay and 273 and 240 $\mu\text{mol Trolox } g^{-1}$ for methanolic and aqueous extract from yerba mate leaves, respectively (Vieira et al., 2010). A comparison with our results indicates approximately two and four times higher antioxidant activity by the DPPH and ABTS methodologies, respectively, for URYM.

These results also suggested that URYM shows a high antioxidant potential that can be explained by its lower granulometry (5 to 12 microns) which provides superior extraction of bioactive compounds. Particle

size reduction is expected to increase the extraction yield since it enhances the accessible superficial area for mass transfer. Additionally, URYM can be considered a product with significant market and nutritional potential since it shows a high content of phenolic compounds and antioxidant activity (Rajha et al., 2014).

Preference ranking test and frequency of yerba mate consumption

Results of the preference ranking test of food products with green color (supposedly with added URYM) noted that food products such as ice cream, juice, cake and cookie showed a greater preference for applying URYM as an ingredient in recipes for preparation at home. Macaroon and flan were the food products with the lowest scores (Table 1).

Cake, although showing no statistical difference from cookies, bread, juice and ice cream, was the product chosen. Cake, although not a staple food like bread, has acquired value and importance in the Brazilian market as it is a food product that is easy to prepare and consume (Moscatto et al., 2004). Additionally, as mentioned by Köten (2021), bakery products, such as cakes, play an increasingly significant role in today's eating habits. Consequently, cakes can be an essential vehicle for added healthy components.

The results related to the frequency and manner of consumption of yerba mate revealed that the consumption frequency is 71.4 %, which corresponds to the scale (using a maximum scale value of 8 for 100%), a frequency of consumption between several times a week to weekly.

Additionally, the primary manner of consumption is through mate, the traditional regional hot drink. Fifty-two (77.6 %) of the 67 participants mentioned consuming this traditional drink. Consumption in tea bag form was by 20.9 % of participants and 1.5 % via other forms of consumption.

Color stability assessment of sponge cake with added URYM

Colorimetric results evaluating the color stability of sponge cake with added URYM over a six-days are shown in Table 2. Individual analysis of the results of the L^* , a^* , and b^* parameters showed no difference over the six-days suggesting good color stability of the cake when URYM is an added ingredient.

Although the color parameters, individually, have not shown differences during storage, the URYM reflection on cake color stability can be better visualized by tan analysis of total color difference (ΔE). The ΔE parameter measures the total color change by accounting for combined changes in L^* , a^* and b^* (Mancini and Hunt, 2005). The delta values (ΔL^* , Δa^* and Δb^*) indicate how the sample differs from standard L^* , a^* and b^* values. The delta values are often used in quality control and formulation adjustments (Mitterer-Daltoé et al., 2012).

ΔE revealed there was a noticeable difference to the untrained human eye (> 3.0) (Mokrzycki and Tatol, 2011) between the first and the second day of storage (Figure 4), suggesting that the color of sponge cake with added URYM is most visually affected during the first 24 h of storage.

Table 1 – Difference between the total sum of orders for the preference of food products with green color.

	217 macaroon	285 spaghetti	317 bread	295 cookie	218 flan	332 ice cream	379 cake	369 juice
217 macaroon	0							
285 spaghetti	68	0						
317 bread	100*	32	0					
295 cookie	78	10	22	0				
218 flan	1	67	99*	77	0			
332 ice cream	115*	47	15	37	114*	0		
379 cake	162*	94*	62	8	161*	47	0	
369 juice	152*	84	52	74	151*	37	10	0

*Significant difference for minimum significant difference ≥ 85.93 .

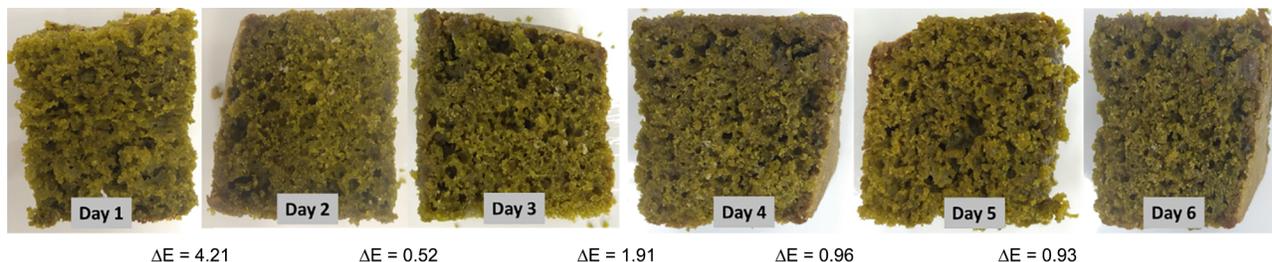


Figure 4 – Total color difference between each day within a six-day period.

Word association of holistic perception of cake with added URYM and hedonic test

The word association technique determines the consumer's cognitive perception of a particular product. This method assigns a stimulus to the consumer and asks him or her to associate the perceptions that come to mind freely. This method provides unrestricted access to mental representations in relation to the stimulus provided (Bordim et al., 2021; Mitterer-Daltoé et al., 2012). Results fell into several categories emerging from the responses in the word association task (Table 3). The participants spontaneously mentioned 155 terms in response to the stimulus employed. The three researchers that participated in the data analysis process

created nine categories by consensus. Words with similar meanings were grouped in the same category (Varela and Fiszman, 2013). From categories obtained for cake with added URYM, the holistic description of the product by consumers was ascertained. The categories that emerged involved hedonic, sensory, mood, food and market terms.

"Tasty" was the category most mentioned by 80 % of the consumers. The other responses are categorized in Table 4. These results reveal not only that "tasty" is the most popular term used and, therefore, the one that best describes consumer perception of the product (Antmann et al., 2011), but also the implication of good acceptance of the cake.

"Sweet", "soft" and "mate" were also categories strongly associated with the cake with added URYM. These categories, plus "tasty" correspond to 72.95 % of the terms cited to describe the cake.

"Color" was another sensory attribute that emerged as having importance. Although green could be considered an unfamiliar color for a cake, it did not strongly impact on consumers' perception of the appearance since only 16 % mentioned this attribute possibly, due to consumers being aware of the addition of URYM in the cake.

Awareness of the addition of URYM in the cake can also be visualized in the categories "bitter", "mate" and "innovation". The category "bitter" drew attention which was probably cited due to the influence of the knowledge of the presence of yerba mate. Although it is known that the bitter taste is related to phenolic compounds with low-molecular-weight (Drewnowski and Gomez-Carneros, 2000), and yerba mate is an important source of these compounds (Santetti et al., 2021), it is expected that the concentration added in the formulation of URYM in the cake would not produce a perception of bitterness. It is inferred that a high amount of sugar suppresses bitter taste (Keast and Breslin, 2003). This can be seen when 18 % of the consumers mentioned "bitter" to describe the cake, while 52 % mentioned "sweet" to describe it.

To better understand the categories used to represent cake, a word cloud map was applied with all terms generated by word association technique (Figure

Table 2 – Color parameters of sponge cake with URYM added within a six-day period.

Days	L*	a*	b*
1	41.91 ^a ± 0.31	-6.28 ^a ± 0.09	32.79 ^a ± 0.78
2	42.76 ^a ± 2.48	-6.04 ^a ± 0.53	34.11 ^a ± 2.05
3	42.20 ^a ± 3.71	-5.49 ^a ± 0.84	33.62 ^a ± 3.18
4	43.84 ^a ± 2.06	-5.54 ^a ± 0.29	32.85 ^a ± 0.93
5	40.47 ^a ± 3.67	-5.38 ^a ± 0.51	31.7 ^a ± 3.57
6	43.07 ^a ± 2.96	-5.77 ^a ± 0.33	34.62 ^a ± 1.48

Values in the table shown in the column with different letters are statistically different by Tukey test ($p \leq 0.05$). URYM = ultra-refined yerba mate. L* = Luminosity; a* = (+) red and (-) green; b* = (+) yellow and (-) blue.

Table 3 – Frequency of categories mentioned for the cake with URYM added.

Category	N° of mentions
Tasty	40
Mild	6
Bitter	9
Sweet	26
Soft	26
Color	8
Happiness	7
Mate	24
Innovation	9

URYM = ultra-refined yerba mate.

Table 4 – Calculated values for the z test.

	Tasty	Mild	Bitter	Sweet	Soft	Color	Happiness	Mate*	Innovation
Tasty	-								
Mild	6.82*	-							
Bitter	6.20*	0.84	-						
Sweet	2.95*	4.28*	3.56*	-					
Soft	2.95*	4.28*	3.56*	0	-				
Color	6.40*	0.57	0.26	3.79*	3.79*	-			
Happiness	6.61*	0.29	0.54	4.04*	4.04*	0.28	-		
Mate	3.33*	3.09*	3.19*	0.4	0.4	3.42*	3.67*	-	
Innovation	6.20*	0.84	0	3.59*	3.56*	0.26	0.54	3.19*	-

* $p < 0.05$.

5). Word clouds are graphical maps that show words in relation to their frequency of use (Jin, 2017; Mitterer-Daltoé et al., 2021). As the font size represents the number of mentions by consumers, a profile can be seen for cake with URYM added, described as tasty, sweet, soft and mate flavor. Antioxidant, Culture, Tradition, Grandmother and Childhood, although cited by less than 5 % of the participants, were also found relating to cake categorization.

The category "tasty", as cited above, was the most often mentioned by WA task, which suggested a good acceptance of the cake with URYM added. Results from the hedonic test corroborated this hypothesis. With an acceptance rate of 87.6 % and an average of 4.38 (maximum value of scale five) the cake with URYM added proved to have good acceptance.

Aiming to better explore the acceptance data, a multinomial logistic regression analysis was applied to correlate demographic data (age and gender) and acceptability. A multivariate analysis technique is frequently used to evaluate sensory and consumer data to detect product and consumer aspects related to product acceptance (Symoneaux et al., 2012). Understanding this relationship is extremely important for implementing strategies to introduce new products to the market (Mitterer-Daltoé et al., 2012). Multinomial logistic regression is a suitable model in situations where the dependent variable is qualitative. When the logistic coefficient is statistically significant, its interpretation relates to how it affects the dependent variable (González et al., 2011).

Results show the demographic characteristics of the participants and the statistical significance of each coefficient of each independent variable (Table 5). From the multinomial logistic regression data, it was noted that there was no age, gender nor age*gender interaction effects ($p \geq 0.05$) on the acceptance of the cake with URYM added. These results suggest that there is no target audience related to gender and age in relation to the use of URYM in food recipes, specifically cake. In

other words, there is a potential for using URYM equally by men and women of any age group.

Check-all-that-apply (CATA) to ascertain consumers' expectations of health issues involved in the URYM product

Respondents answered the CATA questionnaire that contained sentences to assess perceptions and expectations of health issues involved in the URYM product. Results show the number of times consumers marked each statement (Table 6). Test Z presents the significance of the difference between each statement (Table 7). The most marked statements were 1, 6, 9, 11, 14 and 16. The data shows that the statements with the highest frequencies differ from their opposite statement, revealing no ambiguity regarding the characteristics of interest. What could be seen from these results is that for this group of consumers, there is interest in URYM, which contains natural antioxidants which are thermogenic, sugar-free and without preservatives. This revealed a food product profile with natural appeal, healthy concept and functional properties.

Discussion

URYM emerged as a new food ingredient with the potential for use as a naturally colored ingredient to be used in homes, restaurants and food industries.

Under the chemical approach, the results also revealed URYM as a potential naturally colored antioxidant ingredient, giving the ingredient an important nutraceutical appeal which consumers value, especially when the nutraceuticals had scientific evidence, including safety and effectiveness (Teoh et al., 2021). Water was able to extract phenolic compounds with antioxidant activity. Additionally, because it is a safer solvent when compared to methanol, it could be an appropriate choice for producing extracts rich in phenolic compounds with the capacity for scavenging



Figure 5 – Word cloud from the categories that appeared from the stimulus cake with URYM added. URYM = ultra-refined yerba mate.

Table 5 – Demographic characteristics of the participants and statistical significance of each variable.

Variables	n = 50	
Gender	Female	33
	Male	17
Age	18-25	37
	26-50	13
Acceptance	1	0
	2	0
	3	5
	4	21
	5	24
Effect	Wald	p
Age	1.055	0.589
Gender	0.656	0.720
Age × Gender	3.111	0.211

Table 6 – Results of *check-all- that-apply* questionnaire. Frequencies for each sentence.

n	Questionnaire	Total
1	I prefer food products without preservatives.	32
2	I'm not interested in thermogenic foods, that is, foods that help with weight loss.	13
3	I'm not interested in sweetener-free food products.	13
4	When I think of ultra-refined yerba mate, I expect it to be without colorant.	23
5	I wouldn't mind if the food product didn't have in its composition natural antioxidants.	19
6	I find sugar-free ultra-refined yerba mate interesting.	37
7	When I think of ultra-refined yerba mate, I don't imagine a homemade product.	20
8	I'm interested in gluten-free ultra-refined yerba mate.	13
9	I don't care if a food product contains sweetener.	26
10	Whenever I think of ultra-refined yerba mate, I hope the product is homemade.	12
11	I'm interested in food products that contain natural antioxidants.	36
12	I'm not interested in gluten-free ultra-refined yerba mate.	7
13	I don't care if there are preservatives in food products.	13
14	If offered in the supermarket, I would buy a thermogenic product, that is, one that helps with weight loss.	30
15	When I think of ultra-refined yerba mate, I don't care if it contains colorant.	16
16	I don't care if the ultra-refined yerba mate contains sugar in its composition.	27

Table 7 – Calculated values for the z test.

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
1	-															
2	3.81*	-														
3	3.81*	0	-													
4	1.80	2.08*	2.08*	-												
5	2.60*	1.28	1.28	0.81	-											
6	1.08	4.8*	4.8*	2.85*	3.62*	-										
7	2.40*	1.48	1.48	0.60	0.2	3.43*	-									
8	3.81*	0	0	2.08*	1.28	4.8*	1.48	-								
9	1.21	2.66*	2.66*	0.60	1.40	2.27*	1.20	2.6*	-							
10	4.02*	0.23	0.23	2.30*	1.51	5.00*	1.71	0.23	2.88*	-						
11	0.85	4.60*	4.60*	2.64*	3.41*	0.22	3.22*	4.60*	2.06*	4.80*	-					
12	5.12*	2.31*	1.5	3.49*	2.73*	6.04*	2.92*	1.5	4.04*	1.27	5.85*	-				
13	3.81*	0	0	2.08*	1.28	4.8*	1.48	0	2.66*	0.23	4.60*	1.5	-			
14	0.41	3.43*	3.43*	1.40	2.20*	1.48	2.00*	3.43*	0.80	3.64*	1.26	4.76*	3.43*	-		
15	3.20*	0.66	0.66	1.43	0.62	4.20*	0.83	0.66	2.02*	0.89	4.00*	2.13*	0.66	2.80*	-	
16	1.01	2.85*	2.85*	0.8	1.60	2.08*	1.40	2.85*	0.20	3.07*	1.86	4.22*	2.85*	0.60	2.22*	-

* $p < 0.05$.

radicals, mainly ABTS from URYM. In addition, extracts produced with water can be considered safer for use as natural antioxidants in the food industry.

Natural food antioxidants or foods rich in antioxidants are a prominent subject in much research for food preservation and the inhibition of the oxidation process in human metabolism (Cömert and Gökmen, 2018). The importance given by consumers to natural antioxidants was verified by Mitterer-Daltoé et al. (2021), in which researchers explored consumers' perception of food antioxidants (Synthetic vs. Natural). The results showed a consumer perception of positive aspects of health and food safety, indicating a potential use of natural food antioxidants by the food sector.

Another study also focused on the potential use of natural antioxidants in the food industry, but now exploring the influences of color on different naturally

colored antioxidants added to different food product categories. Food products did not negatively impacted consumers with naturally colored antioxidants; their awareness of the presence of natural antioxidants positively influenced their perception and acceptance (Bordim et al., 2021).

In the sensory approaches, the present study also explored consumers' preferences, perceptions, and acceptance of food products or a product with naturally colored antioxidant ingredient added, URYM in this case. Ranking test results revealed not only those foods with potential for the addition of URYM by consumers, but also the foods they are used to preparing at home, since the question was directed towards this. Participants were asked to rank the samples in ascending order according to their preference or interest in preparing the food product at home, adding URYM as an ingredient in the

recipe. In this regard, ice cream, juice, cake, and cookies emerged as the food products in which consumers found themselves using URYM in their homemade recipes, which makes these products also potential vehicles for nutraceutical ingredients. As expected, macaroon and flan were the food products with the lowest scores; that is, this group of consumers does not intend to make them at home by adding URYM, probably, for cultural reasons, due to the low consumption of these foods by Brazilians.

The cake was chosen as a food product that is easy to prepare and consume and has increasing significance in today's eating habits. It is noteworthy that the ranking test is a sensory analysis test that induces forced choice. Therefore, the fact that consumers preferred the cake as one of the food products to be prepared with URYM added does not mean that it is a product with good acceptance. To explore this issue, the word association technique was applied to evaluate the holistic perception of the cake with URYM added. The hedonic test was applied to evaluate acceptance of the cake. These two tests showed good acceptance of the cake with URYM added. Good acceptance was verified by many citations for the "Tasty" category and by an acceptance rate of 87.6 %. It is also worth noting that neither gender nor age influenced ($p > 0.05$) acceptance of the cake. This result was expected since, as mentioned by Kaur and Kaur (2018), cake is a food that consumers of all ages easily accept due to its sweet taste, soft texture, ready-to-eat nature, variety, and affordable price.

Good acceptance is possibly related to the high rates of consumption of yerba mate. The typical frequency of consumption is between several times a week to weekly. What is known is that this habit has an impact on both intention and consumption behavior (Verbeke and Vackier, 2005). Therefore, the high consumption of yerba mate significantly affects the behavior or the intention to consume or, in this case, acceptance of the cake.

The predominant means of consumption is through mate, the traditional regional hot drink. This drink has been prepared in a sui generis manner by South American populations for centuries, having a social, stimulating, and healthy (detoxifying properties) appeal (Bracesco et al., 2011; Mesquita et al., 2021; Vázquez and Moyna, 1986). The traditional regional hot drink, called "chimarrão" is prepared in a dried porongo, called a "cuia". Yerba mate is placed in the porongo, filling 2/3 of the internal capacity and the rest is added to hot water. An infusion is formed and the resulting aqueous extract is drunk with a metal straw called a "bomba" (Mesquita et al., 2021; Vázquez and Moyna, 1986).

This aqueous extract is characterized by a bitter taste (Godoy et al., 2020; Pagliosa et al., 2009). This is probably the main reason for describing the cake with URYM added being placed in the "bitter" category. This was mentioned by 18 % of the participants. It is

suggested that the bitter taste arose from the sensory memory of the mate's bitterness rather than from the actual perception of the cake. This is because the sugar added in the recipe was expected to suppress the bitter taste produced by adding URYM to the cake (Keast and Breslin, 2003). More evidence of this was the high acceptance rate and the large number of mentions in the "Tasty" category.

Independent of the above, the category "Bitter" cannot be overlooked, and what is suggested is that the use of URYM as an ingredient in food products be accompanied by sugar or salt, which are recognized bitter taste suppressors (Keast and Breslin, 2003). The bitter taste as a drawback provided by an ingredient was explored with the same technique, word association, in the study by Bordim et al. (2021). The authors added three different natural antioxidants to three distinct categories of food products. One of the natural antioxidants, the lyophilized extract of propolis, presented an off-flavor (bitter taste) when used in breads. It is noteworthy that the same natural antioxidant was added to patés and yogurts and that, for these products, the bitter category was not mentioned. Consequently, other ingredients added to formulations, such as salt, sugar and seasonings, suppressed bitter taste perception.

The color produced by adding URYM could be another drawback. Plant ingredients have pigmented compounds that provide specific colors (Cömert et al., 2020). Pigments that provide color are considered a challenge for the food sector. Their use could be limited in specific food industries. Depending on consumer perception, the coloring potential may or may not be desirable, depending on consumer perception (Bordim et al., 2021). Meanwhile, for the present study, the green color did not seem to be a problem for the consumer. The color was mentioned by only 16 % of participants, possibly due to the awareness of the presence of yerba mate in the cake recipe.

Color stability was also explored, since the importance of color for gastronomy or food science occur not only when the food is ready for consumption but also during its shelf life. In this respect, results suggested good color stability, with no difference between the L^* , a^* and b^* parameters during the six-days. By ΔE results, the color of the cake with URYM added could be visually affected in the first 24 h of storage, achieving color stability after this period.

Results from CATA exposed the consumer interest in food products and ingredients with a healthy appeal. More specifically, consumers were interested in issues related to thermogenic properties, sugar-free, preservative-free and natural antioxidants. URYM meets all these requirements. Yerba mate is a food rich in caffeine (Marx et al., 2003; Pagliosa et al., 2009) and is recognized as a thermogenic food. Its digestion uses a large amount of energy and is considered a functional food that could help preserve a positive energy balance and prevent obesity (Westerterp-Plantenga et al., 2006).

URYM with sugar added is not desired by consumers. This question was asked, since it is common to find yerba mate with sucrose added in the supermarket. This result may suggest that of people interested in the consumption of URYM are also interested in low sugar products. This fact is relevant since there is an attempt to reduce sugar consumption in Brazil (Veiga et al., 2021) and globally (WHO, 2015). Worldwide, over the past 50 years, sugar consumption has increased significantly, which is related to a rise in several chronic non-communicable diseases.

CATA also showed that consumers are interested in food products containing natural antioxidants and without preservatives, probably indicating an aversion to synthetic additives. Studies have analyzed responses toward food ingredients or additives (Mitterer-Daltoé et al., 2021; Varela and Fiszman, 2013). The following conclusion was commonly noted: consumers are exceptionally concerned about additives and chemicals in their diet, and consumer knowledge and awareness affect the perceived safety of additives. Further, the essential conclusion to be noted is the affirmative insights into natural ingredients (Varela and Fiszman, 2013). In this regard, URYM emerges as a potential ingredient to be used with natural/safety appeal. Meanwhile, it is also important to point out that although participants demonstrated a rejection of synthetic preservatives and antioxidants, they did not show concern about colorings and sweeteners. This behavior may suggest a non-relationship between colorants and sweeteners with synthetic additives.

Conclusions

URYM has emerged as a suitable naturally colored food ingredient with natural antioxidant appeal. Ice cream, juice, cake, and cookies emerged as the food products in which consumers found themselves using URYM in their homemade foods. Cake was the food product chosen for the following tests. With good color stability and acceptance, cake with URYM added has a profile that can be described as tasty, sweet, soft and mate flavor. Results from CATA showed consumer interest in food products and ingredients with a healthy appeal, especially in issues related to thermogenic properties, sugar-free, preservative-free and natural antioxidants. In this context, the potential of URYM as a naturally colored food ingredient was verified.

Based on the present results, future studies exploring the stability of color and antioxidants in different food classes with URYM added deserve to be explored. In addition, a detailed study of formulation definitions based on URYM concentrations also emerges as a need for future work.

Concerning sensory tests, future studies involving different targets emerge as an essential strategy for a better understanding of consumer's perceptions

of URYM as a food ingredient. A study involving a population from a region that does not consume yerba mate may provide meaningful answers.

Nevertheless, the results discovered by the sensorial approach revealed that the category Bitter, although mentioned by only a small group of participants, arose as a possible drawback in using URYM as a food ingredient. Therefore, future works focusing on the bitter taste from URYM also deserve to be explored.

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