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Physical, chemical and technological characterization of cakes added with ora-pro-nóbis (*Pereskia aculeata* Miller) flour

Caracterização físico química e tecnológica de bolos adicionados de farinha de ora-pro-nóbis (*Pereskia aculeata* Miller)

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Pereskia aculeata Miller, também conhecida no Brasil como ora-pro-nóbis, possui alto teor de proteínas, fibras e minerais, sendo uma alternativa para alimentos mais nutritivos e saudáveis. O objetivo deste trabalho foi produzir e avaliar bolos com diferentes concentrações (10, 20, 30%) de farinha de ora-pro-nóbis (FO) associadas à redução do nível de óleo (5, 10, 15%), conforme delineamento experimental 2^2. Onde as variáveis independentes são o óleo (X1) e farinha de ora-pro-nóbis (X2), e as variáveis dependentes são as análise físico-químicas. Foram determinadas as propriedades físico-químicas e funcionais dos bolos. A adição de farinha de ora-pro-nóbis aumentou os teores de proteínas, fibras, cinzas, cálcio, sódio, magnésio, potássio e ferro. Também melhorou/aumentou os valores dos parâmetros a* e ΔE no miolo e a elasticidade. Bolos produzidos com 30% de FO apresentaram as melhores propriedades nutricionais. O bolo com adição de 20% e 10% de FO e 5% de redução no teor de óleo apresentou boa aceitabilidade sensorial, demonstrando que a adição de FO é uma alternativa para a elaboração de produtos alimentícios mais nutritivos.
Palavras-chave: *Pereskia aculeata*, bolo, caracterização físico-química.

Pereskia aculeata Miller, also known in Brazil as ora-pro-nobis, has a high content of protein, fiber and minerals, making it an alternative to more nutritious and healthy foods. The objective of this work was to produce and evaluate cakes with different concentrations (10, 20, 30%) of ora-pro-nobis flour (FO) associated with reduction in oil level (5, 10, 15%), according to the experimental design 2^2, where the independent variables are oil (X1) and ora-pro-nóbis flour (X2), and the dependent variables are the physicochemical analysis. The physicochemical and functional properties of the cakes were determined. The addition of ora-pro-nobis flour increased the protein, fiber, ash, calcium, sodium, magnesium, potassium, and iron. It also improved the crumb color (parameter a* and ΔE) and the elasticity of cake products. Cakes produced with up to 30% FO showed the best nutritional properties. The cake with addition of 20% and 10% FO and up 5% of reduction in oil content showed good sensory acceptability, with FO being a nutritious alternative for application in food products.
Keywords: *Pereskia aculeate*, cake, physicochemical characterization.

1. INTRODUCTION

Pereskia aculeata Miller, also known in Brazil as ora-pro-nobis, is a plant belonging to the Cactaceae family that is distributed in different parts of the world. It is a perennial plant that is easy to propagate [1]. The leaves of *Pereskia aculeata* Miller have a desirable nutritional value in terms of the high content of proteins, fibers, and minerals such as calcium, magnesium, phosphorus, iron, and copper [2-4]. The addition of ora-pro-nóbis leaf flour in food products helps to reduce or replace oils and fats, due to the presence of mucilage in its composition, which can act as a thickening, gelling and emulsifying agent [5, 6].

The increased consumption of foods with low nutrients and a high-fat content generates negative consequences for health. Thus, studies with the use of unconventional food plants (UFPs) in the preparation of food products can increase the nutrient content and provide a low-cost diet, rich in natural and healthy compounds [7].

The consumption of cakes is significant around the world. The production of cakes leads to the development of more nutritious products with the addition of healthy ingredients such as oilseeds, linseed, rye, oats, fruits, vegetable flours, grains, and fibers. These ingredients help to reduce the glycemic index, prevent weight gain, and the emergence of pathologies such as systemic arterial hypertension and cardiovascular diseases [8, 9]. Moreover, the inclusion of UFPs, such as flour from the leaves of *Pereskia aculeata* Miller, can be investigated to contribute with bioactive substances and to improve the nutritional content of conventional cakes [10-12]. Therefore, the objective of this work was to evaluate the nutritional and functional quality of cakes made with the addition of different concentrations of *Pereskia aculeata* Miller flour by means to replace wheat flour associated with a reduction in the total oil content of cakes.

2. MATERIAL AND METHODS

2.1 Materials and flour mix preparation

The ingredients for the preparation of the cakes, such as wheat flour type 1, sugar, eggs, milk, cocoa powder, and baking powder, were purchased from local companies. The ora-pro-nobis flour was prepared from the leaves of *Pereskia aculeata* Miller collected at EPAGRI in the city of Chapecó - SC (-27.0900833 S and -52.636389.19 W), selected, washed, and sanitized in sodium hypochlorite solution (15 ppm) for 10 min. Then, the leaves were dried in an oven (Cienlab, Model: CE-205/49) with air circulation at 60 °C for 24 hours. Subsequently, they were ground in a knife mill (MDR302 Cadence, Brazil), stored in vacuum-sealed packages (hermetic and metalized), and kept under refrigeration (5 ± 2 ºC) until use. Ora-pro-nobis flour (FO) added at concentrations of 0% (control), 10% (FO10), 20% (FO20), and 30% (FO30) in combination with wheat flour (WF) were characterized and used to prepare the cakes based on prior tests.

2.2 Formulations of cakes and experimental design

The cakes and the control were produced according to the standard formulation developed by Segundo et al. (2017) [13] with slight modifications. The percentage of ingredients was determined by the total mass in grams of wheat flour, as described below: 100% wheat flour, 91.6% sugar, 100% whole milk, 41 .6% eggs, 66.6% soybean oil, 8.3% baking powder and 30% cocoa powder. This cake formulation was used to prepare the control and the other cake formulations were prepared according to the experimental design. The ingredients were mixed separately according to each formulation and the cakes were baked in an electric oven (Fischer Grill) at 180 ºC for 30 minutes. Then they were cooled at room temperature and stored under refrigeration (8 ± 2 °C) until the time of analyses.

The formulations of cakes with ora-pro-nobis flour followed the Central Composite Rotational Design (CCRD) 2^2, varying the percentage of ora-pro-nobis flour (10%, 20%, and 30%) and the reduction of the soybean oil (5%, 10%, and 15%), using the Protimiza Experimental Design software (http://experimental-design.protimiza.com.br) [14]. The other ingredients used in the production of the cakes remained constant as previously described. The reduction in the amount of oil (X_1) and the percentage of ora-pro-nobis flour (X_2) were established as independent variables and the physicochemical analysis of cakes were established as dependent variables. This resulted in 10 trials, with two repetitions at the central point, according to the experimental design matrix (Table 1).

Table 1. Coded and real levels of independent variables used in Central Composite Rotational Design (CCRD) 2^2 for cake formulations.

Essay	Coded names	X_1 (Soybean oil reduction)		X_2 (Ora-pro-nobis flour addition)	
		Coded	Real (%)*	Coded	Real (%)*
1	CO5FO10	-1	5	-1	10
2	CO15FO10	+1	15	-1	10
3	CO5FO30	-1	5	+1	30
4	CO15FO30	+1	15	+1	30
5	CO3FO20	-1.41	2.95	0	20
6	CO17FO20	+1.41	17.05	0	20
7	CO10FO6	0	10	-1.41	5.9
8	CO10FO34	0	10	+1.41	34.1
9	CO10FO20	0	10	0	20
10	CO10FO20	0	10	0	20

*Wheat flour-based percentages. $-\alpha = -1.41$ e $+\alpha = +1.41$.

2.3 Physicochemical and microbiological analysis of cakes

Physicochemical analysis of dependent variables was performed for the cakes to evaluate moisture, proteins, lipids, carbohydrates, total fibers, calories, ash, minerals, color, and water activity according to official standards [15-17]. All analyses were performed in triplicate. The analyses for crumb color and crust color (L*, a*, b*, and ΔE) were performed in a colorimeter (Minolta ®) by the CIELAB system, according to methodology 14-22 [18]. Water activity was measured in an Aqualab® Pre water activity tester (Decagon Devices, USA). The specific volume ($cm^3.g^{-1}$) was determined through volume displacement, according to method n° 10-05.01 [19]. The yield was determined by the difference in mass before and after cooking, according to Minãrro et al. (2012) [20]. The microbiological analysis for total coliforms, thermotolerant coliforms at 45 ºC, *Salmonella* sp., *Staphylococcus* coagulase-positive, and *Bacillus cereus* were performed for cakes selected to the sensory analysis, according to current standards, RDC nº 12 [21], IN nº 161 [22], and IN nº 313 [23].

2.4 Texture profile of cakes

The texture profile of the samples was evaluated in a texturometer (Brookfield, model CT3), with samples of 30x30x30 mm (length, breadth, height), through double compression tests, using an acrylic cylinder with a diameter of 20 mm and a speed of 2.0 $mm.s^{-1}$, according to the method n° 74-10.02 [19].

2.5 Scanning electron microscopy (SEM)

Scanning electron microscopy was performed for cake formulations selected for the sensory analysis. These formulations were frozen at -86 °C in an ultra-freezer (Indril Ultra Freezer) for 24 hours and then lyophilized (Ihshin Freeze Dryer, TF5503) for 24 hours. After, they were covered with gold and the microscopic images were performed with a magnification of 1000-1500X, an acceleration voltage of 10-15 kV, in a scanning electron microscope (JEOL, JSM6701F).

2.6 Sensory analysis

To carry out the sensory analysis, the work was sent and approved by the Research Ethics Committee - CEP of UDESC under registration CAAE 28653419.9.0000.0118, with opinion number 3.952726, in order to enable sensory analysis with human beings.

The cakes were evaluated by 120 untrained panelists (78.33% female and 21.66% male, with ages between 17 and 56 years) regarding their pleasantness about flavor, texture, appearance, color, and global acceptance through the 9-point hedonic scale ranging from 1 (“dislike very much”) to 9 (“like very much”) and purchase intention, according to Dutcosky (2011) [24].

2.7 Statistical analysis

The Central Composite Rotational Design (CCRD) results were validated by ANOVA analysis of variance, at a confidence level of 95%, according to the F test (Fcal > Ftab), using the Protimiza Experimental Design software (http://experimental-design. protimiza.com.br) [14]. The results obtained on sensory analysis were statistically evaluated by the Tukey test, through analysis of variance (ANOVA), using the Statistic ® software (Statistica v. 12, StatSoft, USA) to detect the significant difference between the samples ($p<0.05$), at a confidence level of 95%.

3. RESULTS AND DISCUSSION

3.1 Physicochemical characterization and technological analysis

The independent variable percentage of ora-pro-nobis flour (X_2) showed a significant ($p < 0.05$) a decrease of the dependent variables carbohydrates ($R^2 = 0.6181$), crumb color (parameters L* ($R^2 = 0.7180$) and b* ($R^2 = 0.6981$)) and crust color (parameter a* ($R^2 = 0.5322$)), as it was observed with R^2 value < 0.80 (figure S1). However, there was a negative effect on the cake formulations, but which indicates that the reduction in ora-pro-nobis flour (FO) increased the aforementioned variables. For the water activity parameter ($R^2 = 0.6874$), the reduction in the amount of oil (X_1) showed a negative effect for the cake formulations, which indicates that the reduction in oil increased water activity (Figure S1). The independent variable X_2 positively influenced the protein parameters, which indicates that the increase of FO in the cake formulations increased the fiber, ash, calcium, sodium, magnesium, potassium, iron, zinc, and crumb color (parameter a* and ΔE) and elasticity of cakes. The independent variables X_1 and X_2 showed a negative effect, which indicates that the reduction of oil and increase of FO reduced the hardness, gumminess, and chewiness parameters. All the mentioned dependent variables that obtained $R^2 > 0.80$ and Fcalc > Ftab allowed the establishment of the mathematical model (Table S2 and Figure S2).

The reduction of 5, 10, and 15% of oil in the cake formulations increased potassium and iron content, improved crumb color (parameters a* and ΔE), and reduced hardness, gumminess, and chewability (Table S1). According to Amaral et al. (2018) [25] ora-pro-nobis has mucilage in its composition, which has a high emulsifying capacity and thermal stability at different temperatures [26]. Thus, cake formulations with ora-pro-nobis flour and oil reduction are beneficial to health, as they decrease the fat and total calorie contents, and they can contribute to the prevention and treatment of pathologies.

The CO10FO34 sample showed the highest levels of lipids, proteins, fibers, calories, and ash, and the lowest carbohydrate values, compared to the other samples (Table S1). This is due to the presence of 34.1% of FO in the formulation of this sample, and this ingredient has high levels of lipids, proteins, fibers, calories, and ashes and low content of carbohydrates [27]. However, the control (Table S1) prepared only with FT, presented the lowest values of proteins, fibers, and ashes.

Samples with the addition of FO at levels of 10% (CO5FO10 and CO15FO10), 20% (CO3FO20, CO17FO20, CO10FO20, and CO10FO20), 30% (CO5FO30, CO15FO30) and 34.1% (CO10FO34) showed an increase in protein content of 24; 55 and 75%, in fiber content of 42; 83 and 123% and in ash content of 15; 27 and 36%, respectively, when compared with the control. Thus, the importance of ora-pro-nobis flour is highlighted in the preparation of food products, as it directly contributes to the nutritional quality of the final product.

There was a positive effect for increase of FO on the protein contents, however, decrease in the oil content did not influence the protein values of the cake formulations (Figure 1).

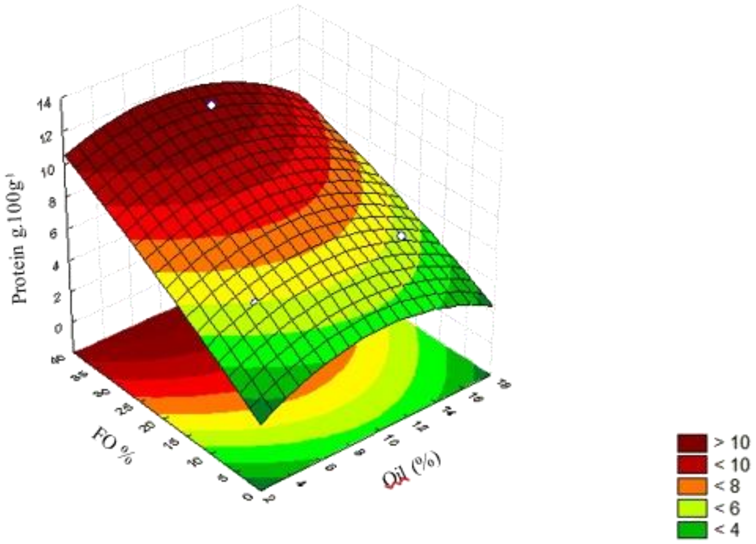

Figure 1. Protein variation of cakes as function of contents of ora-pro-nobis flour (FO) and oil.

For protein contents, it was possible to verify a R_2 value of 0.9268 and the calculated F was 5.50 times greater than tabulated F, according to the mathematical model established by the equation (1).

$$y = 7,68 + 1,78\ X_2 \qquad (1)$$

Where y = protein content (g.100g^{-1}) and X_2 = percentual of ora-pro-nobis flour (FO).

The protein content observed in the cakes formulated with FO (CO10FO34) was about 3 times higher than the control (Table S1). Zem et al. (2017) [28] found protein values of 7.69 g.100g^{-1} in cakes formulated with more than 30% of flour from leaves of ora-pro-nobis, which agrees with the values observed in the present study. A food considered a source of protein must have at least 6 g of protein in 100 g of product, and a food rich in protein at least 12 g in 100g [29]. According to the results obtained in the present study, the cakes with FO are protein sources as they provide between 6.11 to 11.62 g of protein in 100 g.

There was an increase in the total fiber content of the cakes as the concentrations of FO increases (Figure 2).

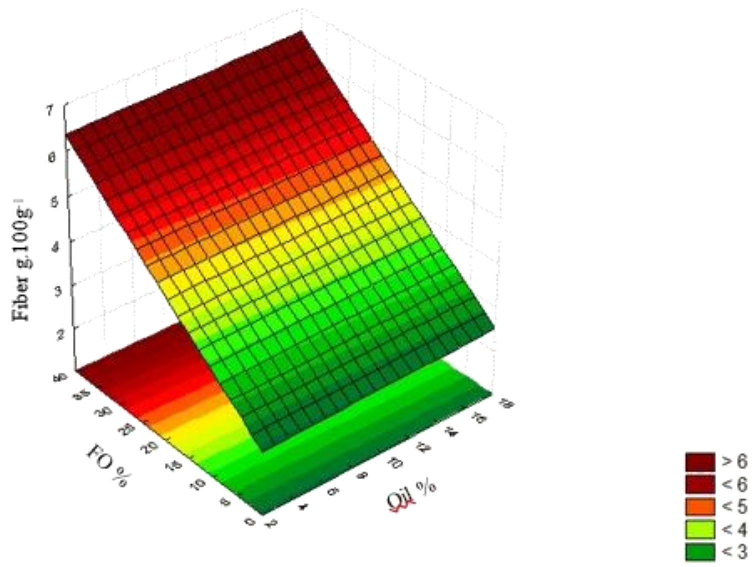

Figure 2. Fiber variation of cakes as function of contents of ora-pro-nobis flour (FO) and oil.

It was possible to verify a positive effect of FO in increase fiber content with a R_2 value of 0.9999 and the calculated F 5.95 times greater than tabulated F, according to the mathematical model established and validated by the equation (2).

$$y = 4{,}43 + 0{,}03\ X_1 + 0{,}99\ X_2 + X_1\ X_2 \qquad (2)$$

Where y = fiber content ($g.100g^{-1}$), X_1 = percentual of reduction in the amount of oil, and X_2 = percentual of ora-pro-nobis flour (FO).

According to the mathematical model, it is important to highlight that the independent variables (X1 and X2) influenced the fiber content individually (0.03 X1 and 0.99 X2) and in a combinated form (X1X2).

The addition of FO in the cakes characterized the product as a source of fiber, as it presented a minimum of 3 g of fiber in 100 g of the product, as required by legislation [29]. Zem et al. (2017) [28] observed an increase in fiber content with the use of dry leaves (5.41 $g.100g^{-1}$) in cakes prepared with percentages superior to 30% of *Pereskia aculeata* flour and these values were higher than those found in the present study. The American Dietetic Association recommends a daily intake of 25 g to 35 g of fiber per day for adults [30], it would be necessary to ingest approximately 500 grams of cake to meet the daily recommendation. Fibers improve intestinal transit speed, improve colon pH, produce by-products with important physiological functions, and prevent intestinal constipation and weight gain [31, 32], and cakes with FO are a source of fiber.

There was an increase in ash content in the cakes with increasing FO concentrations (Figure S2) and the reduction in oil did not affect the ash content (Table S1). The highest content of ash was found for cake CO10FO34 (3.16%) followed by CO5FO30 and CO15FO30 (3.10%) samples and the lowest ash content was found for the control (2.28%) and CO10FO6 (2.43%) (Table S1). The ash content is directly associated with the mineral content in the food. The samples CO10FO34, CO5FO30, and CO15FO30 showed high ash contents (3.16, 3.10, and 3.10 $g.100g^{-1}$, respectively) and high mineral contents (Ca, Mg, K, Na, Fe, and Zn), (Table S1). Rosa et al. (2020) [33] found ash contents of 2.2% in cakes made with 30% fresh leaves of *Pereskia aculeata* Miller, like those found in samples CO15FO10 and CO10FO6, made with 10% and 6% FO.

Minerals have an important physiological function, they improve the immune system and are essential for healthy [34]. The average daily recommendation (DRI) of minerals for adults is 1000mg for calcium, 400 mg (men) and 320 mg (women) of magnesium, 3500 mg of potassium, 2000 mg for sodium, 8 mg (men), and 18 mg (women) of iron, 11 mg (men) and 8 mg (women) of zinc daily [35-37]. The cakes showed a significant increase in mineral content with an increase in FO concentration when compared to the control (Table S1). The cakes made with the addition of 30% or more of FO (CO10FO34, CO15FO30, and CO5FO30) presented the highest levels of Ca, Mg, K, Na, Fe, and Zn.

Calcium contents increased with increasing FO and remained constant with reducing oil in the cake formulations (Table S1 and Figure S2). It is observed that the cakes CO10FO34 and CO5FO30 presented the highest concentrations of calcium with average values of 3.77 $g.kg^{-1}$. The consumption of 100 g of cake with 30% FO would supply 3 times more than the daily recommendation of 1000 mg per day of calcium, which is a micronutrient essential for bone growth, nerve impulses, and muscle contraction [38, 39]. Calcium is abundant in dark green leafy vegetables, and these vegetables are food options to provide an adequate daily consumption of calcium [38]. Zem et al. (2017) [28] found 5 $g.kg^{-1}$ of calcium in cakes prepared with more than 30% of ora-pro-nobis dry leaf flour, a value higher than that found in the present study.

The concentrations of 30% and 34.1% of FO and the reduction of 5% and 10% of oil increased the potassium contents in the formulations CO10FO34 and CO5FO30 (2.38 $g.kg^{-1}$) concerning the control (1.73 $g\ .kg^{-1}$), (Table S1 and Figure S2). The consumption of 100 g of cakes with 30% FO is equivalent to half the daily recommendation of 3500 mg of potassium, a micronutrient essential for muscle contraction, nerve conduction, heart rate, energy production, and synthesis of nucleic acids and proteins [40]. Zem et al. (2017) [28] found 3.25 $g.kg^{-1}$ of potassium in the cake prepared with more than 30% of ora-pro-nobis dry leaf flour, values similar to those found in the present study.

There was no variation in sodium content for the cake formulations tested with different concentrations of FO and oil (Table S1 and Figure S2). The control presented 0.025 $g.kg^{-1}$ and CO5FO30 with 30% FO obtained 0.033 $g.kg^{-1}$ of sodium. Zem et al. (2017) [28] found 0.59 $g.kg^{-1}$ of sodium in cakes with ora-pro-nobis leaf flour. The World Health Organization recommends a maximum daily sodium intake of 2000 mg [37]. All cakes evaluated in the present study showed sodium values below this recommendation and can be included in regular diets.

Magnesium concentrations increased with the increase of FO in the cakes. The reduction of oil showed no influence on magnesium contents in the different formulations (Table S1 and Figure S2). The control showed a content of 0.24 $g.kg^{-1}$ of magnesium and the sample CO10FO34 showed 0.39 $g.kg^{-1}$. Magnesium acts in the body to regulate blood sugar, nerve, and muscle conduction, improve heart function and reduce blood pressure [41]. The magnesium values found in the present study were close to those found by Zem et al. (2017) [28] with 0.18 $g.kg^{-1}$ for the control and 0.69 $g.kg^{-1}$ for the cake with more than 30% ora-pro-nobis leaf flour.

Iron content increased with the addition of 30% and 34.1% of FO and with the decrease of 5% and 10% of oil in the cake formulations (Table S1 and Figure S2). The iron content of 0.013 $g.kg^{-1}$ was found for the control and 0.021 $g.kg^{-1}$ for the CO10FO34 cake, which characterizes the product as a source of iron. A high iron content (0.12 $g.kg^{-1}$) was found for cakes prepared with more than 30% of ora-pro-nobis dry leaf flour in the study conducted by Zem et al. (2017) [28]. This mineral is one of the essential nutrients for life, as it acts mainly in the synthesis of proteins, including enzymes and hemoglobin essential for the transport of oxygen to all cells of the body [42]. The leaves of *Pereskia aculeata* Miller have non-heme iron, which is the most consumed form found in different concentrations in all foods of plant origin, especially in dark greens [43]. However, non-heme iron, present in ora-pro-nobis, has an absorption of only 5% of the total ingested content [44].

As shown in Figure S2, the concentrations of FO and oil showed no influence on zinc content in the different formulations tested (Table S1). There was no statistical difference in zinc content between the evaluated samples. The highest value found for this mineral was 0.0030 $g.kg^{-1}$ for CO5FO30 and CO10FO34. This mineral plays an important role in the physiological body's processes, acting on the synthesis and degradation of carbohydrates, lipids, and proteins, the maintenance of normal growth and development, the functioning of the immune system, antioxidant defense, neurosensory function, transcription and translation of polynucleotides [45, 46].

The cakes showed a significant increase in mineral content (Ca, Mg, K, Na, Fe) with increasing FO concentration concerning the control. Cakes with an addition of 30% and 34.1% of FO are considered rich in Ca, K, Mg, and Fe because they presented more than 30% of the DRI concerning the control. Cakes with an addition of 20% of FO are considered a source of Ca, K, and Mg, as they presented more than 15% of the DRI concerning the control [29]. The specific volume and yield (Table S1) were not directly influenced by the independent variables of reduction in the amount of oil (X_1) and the percentage of ora-pro-nobis flour (X_2) (Figure 3).

Figure 3. Cakes prepared with different concentrations of ora-pro-nobis flour (%) and oil (%). Control: 0% ora-pro-nobis flour + 66.60% oil + 100% wheat flour; CO5FO10: 10% ora-pro-nobis flour + 63.27% oil (5% reduction) + 90% wheat flour; CO15FO10: 10% ora-pro-nobis flour + 85% oil + 90% wheat flour; CO5FO30: 30% ora-pro-nobis flour + 63.27% oil (5% reduction) + 70% wheat flour; CO15FO30: 30% ora-pro-nobis flour + 85% oil + 70% wheat flour; CO3FO20: 20% ora-pro-nobis flour + 64.63% oil (2,95% reduction) + 80% wheat flour; CO17FO20: 20% ora-pro-nobis flour + 82.95% oil + 80% wheat flour; CO10FO6: 5.9% ora-pro-nobis flour + 90% oil + wheat flour 94.1%; CO10FO34: 34.1% ora-pro-nobis flour + 90% oil + wheat flour 65.9%; CO10FO20: 20% ora-pro-nobis flour + 90% oil + 70% wheat flour; CO10FO20: 20% ora-pro-nobis flour + 90% oil + 70% wheat flour.

The microbiological analysis of the cakes showed that all samples selected for sensorial analysis were under the standards established by legislation that regulates microbiological standards for food [21-23].

3.2 Texture and color profile

The reduction of 10% of oil and the addition of 5.9% of FO influenced the color parameter a*, increasing the tendency towards a red hue. The reduction of 5% and 10% of oil and the addition of 30% and 34.1% of FO in the cake formulations modified the ΔE with a small change in the color of the cakes concerning the control (Table S1 and Figure S2).

The increase in the concentration of FO added to the cakes reduced the lightness parameter (L*). All samples with more than 20% FO addition were considered darker than the control, as they presented values close to zero [47]. The high L* values in cakes with a light color or the presence of flour and starch in the crust can be due of their great light reflectance [48]. The reduction in luminosity is also due to the presence of cocoa in the formulations and the presence of sugars and eggs, which, subject to heat treatment, accelerate the caramelization and Maillard reactions, with progressive darkening of the crust and crumb [49-51].

The increase in the percentage of FO used in the preparation of the cakes increased the dark green shade (Table S1 and Figure S2). According to Padilha et al. (2010) [50], the combination of coordinates being positive between a* and b* resulted in the brown tone of the cakes, which was observed in the cakes with lower FO content and in the control. Another factor that may have interfered with the darker color of the cakes is the reduction of fat in the formulations. According to Kim et al. (2001) [52], the replacement of fat in sponge cakes promoted a reduction in L*, a*, and b* values. It is important to highlight that the visual acceptability of a food product is influenced by its color and uniformity [53].

Cake samples elaborated with addition of FO showed a tendency to increase the parameters of hardness, gumminess, and chewiness and to decrease the cohesiveness and elasticity. There was an increase in the hardness of cakes directly associated with the increase of 20%, 30%, and 34.1% in the concentration of FO and a reduction of 10% and 15% of oil, as observed in the samples CO15FO30 and CO10FO34 (Table S1 and Figure S2). According to Esteller et al. (2006) [54], the amount of fat, sugar, and moisture exert a softening effect on the dough, maintaining lower hardness values. This can be attributed to the dilution of gluten due to the interaction that occurs between polysaccharides and wheat flour proteins and the interaction between fibers present in the FO [55]. According to Tuncel et al. (2014) [56], ora-pro-nobis flour could cause discontinuation of the protein network through the fibers present and consequently increase the hardness of cakes or other baked goods.

Concentrations of FO lower than 20% increased the cake's elasticity (Figure S2) and there was no influence on oil reduction for this parameter (Table S1). The elasticity parameter evaluates how the product returns to its initial shape after deformation [57]. The reduced elasticity of cakes made with FO can be attributed to the low presence of glutenin, a gluten protein, due to the reduced concentration of wheat flour in their preparation. According to Gutkoski et al. (2011) [58], glutenin is responsible for dough elasticity and resistance to extension, so products with higher wheat contents have more elasticity than others with low wheat content [59].

The gumminess and chewiness parameters were influenced by the decrease of 10% and 15% of oil and by the variations in the FO concentrations in the different cake formulations (Table S1 and Figure 1). Regarding the decrease in gumminess, according to Paz et al. (2015) [60], this parameter is one of the most affected, due to the increase in fiber concentration. The increase in chewability values of cakes made with FO can be attributed to the increase in fiber content and reduction in oil, compared to the control. Esteller et al. (2004) [61] observed that foods with higher fiber content require greater salivation and a greater number of chews before swallowing, so there is an increase in mechanical work and chewing.

3.3 Scanning electron microscopy (SEM)

The microstructure analysis of the cakes at 1000x magnification showed the influence of different ingredient content on the formation of the gluten network and in the structure of the doughs compared to the control. A matrix with folds was observed in the cake microstructure of the control and CO5FO30. The control sample appears to have more defined granules, which could be starch granules. Samples CO5FO10 and CO3FO20 showed a more homogeneous structure, with smaller folds and they were more compactness than the control (Figure 4).

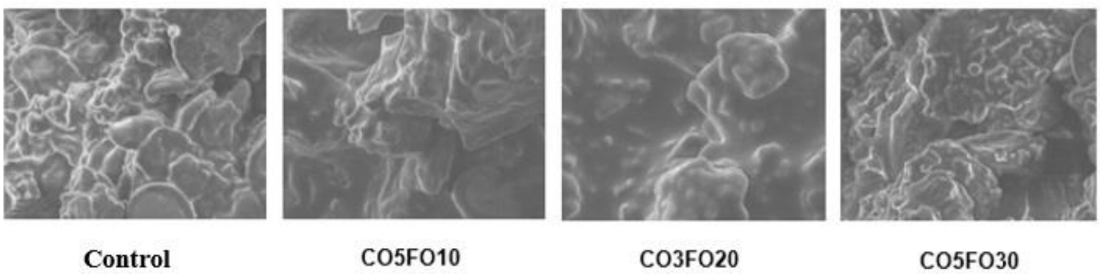

Figure 4. Scanning electron microscopy at 1000x magnification. Control: 0% ora-pro-nobis flour + 66.60% oil + 100% wheat flour; CO5FO10: 10% ora-pro-nobis flour + 95% oil + 90% wheat flour; CO3FO20: 20% ora-pro-nobis flour + 97.05% oil + 80% wheat flour; CO5FO30: 30% ora-pro-nobis flour + 95% oil + 70% wheat flour.

In the literature, no studies were found about the structure of cakes or baked goods using ora-pro-nobis flour. According to Li et al. (2020) [62], with the decrease in the gluten network due to the presence of other ingredients, a discontinuity occurs around the gas cells, which modifies the product structure. It was observed in the microstructures (Figure 4) and especially in the CFO30O95 formulation, which has a greater number of folds than the control, with a rough appearance and irregular air alveoli. The presence of a higher concentration of FO, an ingredient rich in fiber and gluten-free, can change the formation of the gluten network and destabilize the air retention of the dough, causing deformities [13, 63]. According to He and Hoseney (1991) [64], flours with lower quality and lower gluten content incorporate more air during cooking. However, there is a reduction in volume due to the loss of carbon dioxide during fermentation in the initial stage of dough cooking [62].

3.4 Sensory analysis of cakes

For the sensory analysis, the control and the other three formulations of cakes containing 10%, 20%, and 30% of FO and with a reduction of 3% to 5% of oil were chosen considering the desirable effects on the physicochemical and technological parameters of the cake formulations studied.

The addition of 10% FO (CO5FO10) in replacement of wheat flour maintained the acceptability of the cakes. A significant difference ($p<0.05$) was observed for control and the formulations evaluated with 20% (CO3FO20) and 30% (CO5FO30) of FO (Table 2).

Table 2. Sensory analysis of cakes made with reduced oil and different concentrations of ora-pro-nobis flour.

Formulation	Color	Flavor	Texture	Appearance	Overall acceptance
Control	7.82 ± 0.93^{a}	7.81 ± 1.08^{a}	7.56 ± 1.53^{a}	7.88 ± 1.05^{a}	7.86 ± 0.99^{a}
CO5FO10	7.78 ± 1.05^{ab}	7.61 ± 1.38^{a}	7.63 ± 1.32^{a}	7.84 ± 1.09^{a}	7.71 ± 1.01^{a}
CO3FO20	7.58 ± 1.14^{bc}	6.83 ± 1.50^{b}	7.18 ± 1.35^{b}	7.52 ± 1.28^{b}	7.13 ± 1.43^{b}
CO5FO30	7.42 ± 1.35^{c}	6.27 ± 1.84^{c}	7.03 ± 1.45^{b}	7.43 ± 1.41^{b}	6.76 ± 1.67^{c}

Control: 0% ora-pro-nobis flour + 66.60% oil + 100% wheat flour; CO5FO10: 10% ora-pro-nobis flour + 95% oil + 90% wheat flour; CO3FO20: 20% ora-pro-nobis flour + 97.05% oil + 80% wheat flour; CO5FO30: 30% ora-pro-nobis flour + 95% oil + 70% wheat flour. Results are expressed as mean ± standard deviation. In each column, means followed by the same letters did not differ significantly from each other ($p > 0.05$), $n = 120$ panelists.

All means obtained correspond to score 7 – "like moderately", except for the flavor of the CO3FO20 and CO5FO30 formulations, and overall acceptance for CO5FO30 (Table 2). Barroso et al. (2019) [65] and Miranda et al. (2013) [66] also studied the use of alternative flours to partially replace wheat flour in the preparation of bakery products and found averages greater than 7 at a 9-point hedonic scale for the parameters evaluated on the sensory analysis. In the present study, lower percentages of replacement (10% and 20%) obtained better acceptance by the panelists (Table 4).

The purchase intention of cakes was also evaluated, and the panelists indicated it on a scale from 1 - "certainly would not buy" to 5 - "certainly would buy". A product must have a minimum acceptability index (AI) of 70% to be considered accepted by consumers [67]. In the present study, all formulations presented AI above this value, being: 98.31% for the control, 94.11% for the CO5FO10 formulation, 84.87% for the CO3FO20, and 71.42% for the CO5FO30. The cakes also received scores with an average higher than the score 3 "Maybe would buy/maybe wouldn't buy", indicating that the formulations have quality attributes that please the consumer and can lead him to purchase them (Figure 5).

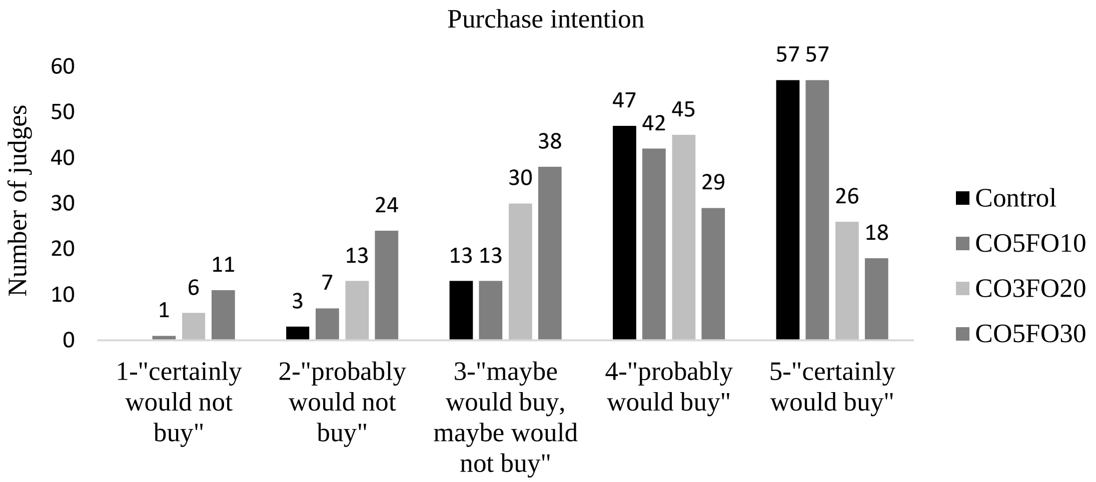

Figure 5. Profile of the panelists regarding the purchase intention. Control: 0% ora-pro-nobis flour + 66.60% oil + 100% wheat flour; CO5FO10: 10% ora-pro-nobis flour + 95% oil + 90% wheat flour; CO3FO20: 20% ora-pro-nobis flour + 97.05% oil + 80% wheat flour; CO5FO30: 30% ora-pro-nobis flour + 95% oil + 70% wheat flour.

Similar rates of AI >70%, were found by Marcelino et al. (2018) [68] and Silva et al. (2014) [69], who analyzed cakes with concentrations of baru husk flour and pulp (AI of 74 to 84%) and gluten-free carob cake (AI of 92.8%), respectively. The higher the AI of the product is, the higher the expectation of purchasing the product. Stringheta et al. (2007) [70] emphasize that the addition of nutritionally beneficial ingredients in products can reduce public health costs, a fact that can enhance their commercialization and acquisition [68].

Figure 6 shows the very similar color, development, and characteristics of crumbs for all cake samples evaluated on the sensory analysis.

Figure 6. Effect of ora-pro-nobis flour concentrations (%) and oil reduction (%) on the structure of cakes submitted to sensory analysis. Control: 0% ora-pro-nobis flour + 66.60% oil + 100% wheat flour; CO5FO10: 10% ora-pro-nobis flour + 63.27% oil (5% reduction) + 90% wheat flour; CO3FO20: 20% ora-pro-nobis flour + 64.63% oil (2,95% reduction) + 80% wheat flour; CO5FO30: 30% ora-pro-nobis flour + 63.27% oil (5% reduction) + 70% wheat flour.

4. CONCLUSION

The use of ora-pro-nobis flour as an ingredient in the preparation of cakes to replace conventional ingredients makes it possible to increase the nutritional value since this raw material is a source of protein, fiber, and minerals.

The addition of 30% of ora-pro-nobis flour resulted in products considered rich in calcium, magnesium, potassium, and iron. In addition, it improved the crumb color (parameter a* and ΔE) and the elasticity of the formulations. All formulations showed an acceptability index greater than 70% and the formulation with 5% oil reduction and 10% addition of ora-pro-nobis flour (CO5FO10) obtained the best sensory and average acceptance for purchase intention with excellent nutritional value.

Cakes made with ora-pro-nobis flour are an alternative food with functional properties and, considering this vegetable is rich in proteins, minerals, and fiber, it improves the nutritional value and brings beneficial effects to the health of consumers.

5. ACKNOWLEDGMENT

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6. REFERENCES

1. Silva DO, Primio EM, Botelho FT, Gularte MA. Nutritional composition and sensory analysis of bread rolls added *Pereskia aculeata*. Demetra. 2014 Mar;9(4):1027-40. doi: 10.12957/demetra.2014.11119
2. Paula MC, Oliveira RB, Felipe DF, Magrine ICO, Sartor CFP. Processamento de bolo com a planta P*ereskia Aculeata mill*. (ora-pro-nóbis). RBPA. 2016 June;18(2):167-74. doi: 10.15871/1517-8595%2Frbpa.v18n2p167-174
3. Tofanelli MBD, Resende SG. Sistemas de condução na produção de folhas de ora-pro-nóbis. Pesqui Agropecu Trop. 2011 Sep;41(3):466-9. doi: 10.5216/pat.v41i3.12497
4. Takeiti CY, Antonio GC, Motta EMP, Collares-Queiroz FP, Park KJ. Nutritive evaluation of a non-conventional leafy vegetable (*Pereskia aculeata* Miller). Int J Food Sci Nutr. 2009 May;60(1):148-60. doi: 10.1080/09637480802534509
5. Lago AMT, Neves ICO, Oliveira NL, Botrel DA, Minim LA, Resende JV. Ultrasound-assisted oil-in-water nanoemulsion produced from *Pereskia aculeata* Miller mucilage. Ultrason Sonochem. 2019 Jan;50:339-53. doi: 10.1016/j.ultsonch.2018.09.036
6. Oliveira NL, Rodrigues AA, Neves ICO, Lago AMT, Borges SV, Resende JV. Development and characterization of biodegradable films based on *Pereskia aculeata* Miller mucilage. Ind Crops Prod. 2019 Apr;130:499-510. doi: 10.1016/j.indcrop.2019.01.014
7. Pereira GIS, Pereira RGFA, Barcelos MFP, Morais AR. Avaliação química da folha de cenoura visando ao seu aproveitamento na alimentação humana. Ciênc Agrotec. 2003 Ago;27(4):852-7. doi: 10.1590/S1413-70542003000400017
8. Jun Y, Bae IY, Lee S, Lee HG. Utilisation of preharvest dropped apple peels as a flour substitute for a lower glycaemic index and higher fibre cake. Int J Food Sci Nutr. 2013 Feb;1(7):62-8. doi: 10.3109/09637486.2013.830083
9. Psimouli V, Oreopoulou V. The effect of alternative sweeteners on bater rheology and cake properties. J Sci Food Agric. 2012 Jan;92(1):99-105. doi: 10.1002/jsfa.4547
10. Seifried HE, Anderson DE, Fisher EI, Milner JA. A review of the interaction among dietary antioxidants and reactive oxygen species. J Nutr Biochem. 2007 Sep;18(9):567-79. doi: 10.1016/j.jnutbio.2006.10.007
11. Santos AQ, Santos RX, Marisco G. Atividades biológicas, toxicológicas e parâmetros nutricionais da *Pereskia aculeata* Miller: uma revisão bibliográfica. Sci Amazon. 2018;7(2):1-16.
12. Araújo WMC, Montebello NP, Botelho RBA, Borgo LA. Alquimia dos alimentos. 3. ed. Brasília (DF): Senac; 2016.
13. Segundo C, Román L, Gómez M, Martínez MM. Mechanically fractionated flour isolated from green bananas (*M. cavendishii* var. *nanica*) as a tool to increase the dietary fiber and phytochemical bioactivity of layer and sponge cakes. Food Chem. 2017 Mar;219:240-8. doi: 10.1016/j.foodchem.2016.09.143
14. Rodrigues MI, Iemma AF. Planejamento de experimentos e otimização de processos. 3. ed. Campinas (SP): Caritá; 2014.
15. Association of Official Agricultural Chemists. Total dietary fiber in foods – Enzimatic-gravimetric method – First action. J Assoc Anal Chem. 2005;68(2):399.
16. Association of Official Analytical Chemists (AOAC). Official methods of analysis of Association of Official Analytical Chemists. 20. ed. Gaithersburg: AOAC; 2016.
17. Association of Official Agricultural Chemists (AOAC). Total dietary fiber in foods – Enzimatic-gravimetric method – First action. J AOAC. 2005;68(2):399.
18. American Association of Cereal Chemists (AACC). Approved methods of the AACC. 10. ed. São Paulo (SP): AACC; 2000.
19. American Association of Cereal Chemists International (AACCI). Approved methods. 11. ed. São Paulo (SP): AACCI; 2010.

20. Minãrro B, Albanell E, Aguilar N, Guamis B, Capellas M. Effect of legume flours on baking characteristics of gluten-free bread. J Cereal Sci. 2012;56(2):476-81.
21. Brasil. Ministério da Saúde, Agência Nacional de Vigilância Sanitária. Resolução RDC nº 12, de 02 de janeiro de 2001. Aprova o regulamento técnico sobre os padrões microbiológicos para alimentos. Brasília (DF): Diário Oficial da União; 2001. Available from: https://bvsms.saude.gov.br/bvs/saudelegis/anvisa/2001/res0012_02_01_2001.html
22. Brasil. Ministério da Saúde, Agência Nacional de Vigilância Sanitária. Instrução Normativa - IN nº 161, de 1 de julho de 2022. Estabelece os padrões microbiológicos dos alimentos. Brasília (DF): Diário Oficial da União; 2022. Available from: https://www.in.gov.br/en/web/dou/-/instrucao-normativa-in-n-161-de-1-de-julho-de-2022-413366880
23. Brasil. Ministério da Saúde, Agência Nacional de Vigilância Sanitária. Instrução Normativa - IN nº 313, de 4 de setembro de 2024. Altera a Instrução Normativa - IN nº 161, de 1º de julho de 2022, que estabelece os padrões microbiológicos dos alimentos. Brasília (DF): Diário Oficial da União; 2024. Available from: https://www.in.gov.br/en/web/dou/-/instrucao-normativa-n-313-de-4-de-setembro-de-2024-582631446
24. Dutcosky SD. Análise sensorial de alimentos. 3. ed. Curitiba (PR): Champagnat; 2011.
25. Amaral TN, Junqueira LA, Prado MET, Cirillo MA, Abreu LR, Costa FF, et al. Blends of *Pereskia aculeata* Miller mucilage, guar gum, and gum Arabic added to fermented milk beverages. Food Hydrocoll. 2018 Jun;79:331-42. doi: 10.1016/j.foodhyd.2018.01.009
26. Lima Junior FA, Conceição MC, Resende JV, Junqueira LA, Pereira CG, Prado MET. Response surface methodology for optimization of the mucilage extraction process from *Pereskia aculeata* Miller. Food Hydrocoll. 2013 Aug;33(1):38-47. doi: 10.1016/j.foodhyd.2013.02.012.
27. Trentin MM, Dinin AZ, Bampi M. Nutritional and rheological evaluation of blends based on ora-pro-nóbis (*Pereskia aculeata* Miller) and wheat flour. RSD. 2020 Nov;9(11):1-19. doi: 10.33448/rsd-v9i11.9728
28. Zem LM, Helm CV, Zuffellato-Ribas KC, Koehler HS. Centesimal and mineral analysis of cupcakes base meal of leaves and stems of ora-pro-nobis (*Pereskia aculeata*). Rev Eletr Cient. UERGS. 2017 Aug;3(2):428-46. doi: 10.21674/2448-0479.32.428-446
29. Agência Nacional de Vigilância Sanitária. Resolução da Diretoria Colegiada – RDC nº 54, de 12 de novembro de 2012. Dispõe sobre o regulamento técnico sobre informação nutricional complementar. Brasília (DF): Diário Oficial da União; 2012. Available from: https://bvsms.saude.gov.br/bvs/saudelegis/anvisa/2012/rdc0054_12_11_2012.html
30. American Dietetic Association. Position of the American Dietetic Association: health implications of dietary fiber. J Am Diet Assoc. 2002;102:993-1000.
31. Bernaud FSR, Rodrigues TC. Dietary fiber – Adequate intake and effects on metabolism health. Arq Bras Endocrinol Metab. 2013 Aug;57(6):397-405. doi: 10.1590/S0004-27302013000600001
32. Vitolo MR, Campagnolo PDB, Gama CM. Fatores associados ao risco de consumo insuficiente de fibra alimentar entre adolescentes. J Pediatr. 2007 Feb;83(1):47-52. doi: 10.1590/S0021-75572007000100009
33. Rosa L, Queiroz CRAA, Melo CMT. Fresh leaves of ora-pro-nóbis in cakes prepared from commercial pre-mixture. Biosci J. 2020 Mar/Apr; 36(2):376-82. doi: 10.14393/BJ-v36n2a2020-42319
34. Gondim JAM, Moura MFV, Dantas AS, Medeiros RLS, Santos KM. Centesimal and mineral composition in fruit peels. Food Sci Technol. 2005 Dec;25(4):825-7. doi: 10.1590/S0101-20612005000400032
35. Padovani RM, Amaya-Farfán J, Colugnati FAB, Domene SMÁ. Dietary reference intakes: application of tables in nutritional studies. Rev Nutr. 2006 Dec;19(6):741-60. doi: 10.1590/S1415-52732006000600010
36. Institute of Medicine. Dietary reference intakes the essential guide to nutrient requirements. Washington (DC): National Academy Press; 2006.
37. Word Health Organization (WHO). Reducing salt intake populations: report of a WHO forum and technical meeting. Paris: WHO; 2006.
38. Bueno AL, Czepielewski MA. A importância do consumo dietético de cálcio e vitamina D no crescimento. J. Pediatr. 2008 Oct;84(5):386-94. doi: 10.2223/JPED.1816
39. Pereira GAP, Genaro PS, Pinheiro MM, Szejnfeld VL, Martini LA. Cálcio dietético: estratégias para otimizar o consumo. Rev Bras Reumatol. 2009 Apr;49(2):164-80. doi: 10.1590/S0482-50042009000200008
40. Fonseca HAR, Zamith TP, Machado VA. Relationship between dietary potassium and blood pressure. Rev Bras Hipertens. 2015 Jan;22(1):9-12.
41. Cunha AR, Umbelino B, Correia ML, Neves MF. Efeitos do magnésio sobre a estrutura e função vascular. Rev HUPE. 2011 Jul;10(3):39-45.

42. Bortolini GA, Fisberg M. Nutritional guidelines for patients with iron deficiency. Rev Bras Hematol Hemoter. 2010 Jun;32(2):105-13. doi: 10.1590/S1516-84842010005000070
43. Almeida MEF, Corrêa AD. Utilization of cacti of the genus *Pereskia* in the human diet in a municipality of Minas Gerais. Rev Cienc Rural. 2012 Apr;42(4):751-56. doi: 10.1590/S0103-84782012000400029
44. Gallagher ML. Os nutrientes e seu metabolismo. In: Mahan LK, Escott-Stump S, editors. Alimentos, nutrição e dietoterapia. 12. ed. Rio de Janeiro (RJ): Elsevier; 2010. p. 39-143.
45. Sena KCM, Pedrosa LFC. Zinc supplementation and its effects on growth, immune system, and diabetes. Rev Nutr. 2005 Apr;18(2):251-9. doi: 10.1590/S1415-52732005000200009
46. Salgueiro MJ, Zubillaga M, Lysionek A, Sarabia M, Caro R, Paoli T, et al. Zinc as an essencial micronutrient: a review. Nutr Res. 2000 May;20(5):737-55. doi: 10.1016/S0271-5317(00)00163-9
47. Cohen KO, Jackix MNH. Estudo do liquor de cupuaçu. Food Sci Technol. 2005 Mar;25(1):182-90. doi: 10.1590/S0101-20612005000100030
48. Gutkoski LC, Teixeira DMF, Durigon A, Ganzer AG, Bertolin TE, Colla LM. Influence of oat and fat levels in technological and functional characteristics of cakes. Food Sci Technol. 2009 Jun;29(2):254-61.
49. Meral R, Dogan, IS. Grape seed as a functional food ingredient in bread-making. Int J Food Sci Nutr. 2013 May;64(3):372-9. doi: 10.3109/09637486.2012.738650
50. Padilha VM, Rolim PM, Salgado SM, Livera AS, Andrade SAC, Guerra NB. Sensorial profile of chocolate cakes formulated with yacon flour (*Smallanthus sonchifolius*). Ciênc Tecnol Aliment. 2010 Sep;30(3):735-40. doi: 10.1590/S0101-20612010000300026
51. Giese J. Color measurement in food as a quality parameter. Food Technol. 2000;54(2):62-3.
52. Kim HYL, Yeom HW, Lim HS, Lim S. Replacement of shortening in yellow layer cakes by corn dextrins. Cereal Chem. 2001 May;78(3):267-71. doi: 10.1094/CCHEM.2001.78.3.267
53. Dias NAA, Lara SB, Miranda LS, Pires ISC, Pires CV, Halboth NV. Influence of color on acceptance and identification of flavor of foods by adults. Ciênc Tecnol Aliment. 2012 Apr;32(2):296-301. doi: 10.1590/S0101-20612012005000059
54. Esteller MS, Zancanaro Júnior O, Lannes SCS. Bolo de “chocolate” produzido com pó de cupuaçu e kefir. Braz J Pharm Sci. 2006 Jul;42(3):447-54.
55. Aziah AAN, Min WL, Rajeev B. Nutritional and sensory quality evaluation of sponge cake prepared by incorporation of high dietary fiber containing mango (*Mangifera indica* var. Chokanan) pulp and peel flours. Int J Food Sci Nutr. 2011 Sep;62(6):559-67. doi: 10.3109/09637486.2011.562883
56. Tuncel NB, Yilmaz N, Kocabiyik H, Uygur A. The effect of infrared stabilized rice bran substitution on B vitamins, minerals, and phytic acid content of pan breads: part II. J Cereal Sci. 2014 Mar;59(2):162-6. doi: 10.1016%2Fj.jcs.2013.12.005
57. Szczesniak AS. Texture is a sensory property. Food Qual Prefer. 2002 Jun;13(4):215-25. doi: 10.1016/S0950-3293(01)00039-8
58. Gutkoski LC, Durigon A, Mazzutti S, Cezare K, Colla LM. Influence of the type of wheat flour in the elaboration of english-type cake. Braz. J Food Technol. 2011 Dec;14(4):275-82. doi: 10.4260/BJFT2011140400033
59. Hoseney RC, Wade P, Finley JW. Soft wheat products. In: Pomeranz Y, editor. Wheat: Chemistry and technology. 3rd ed. Gaithersburg (MD): AACC; 1988. p. 407-56.
60. Paz MF, Marques RV, Schumann C, Corrêa LB, Corrêa ÉK. Technological characteristics of bread prepared with defatted rice bran. Braz J Food Technol. 2015 Oct;18(2):128-36. doi: 10.1590
61. Esteller MS, Amaral RL, Lannes SCS. Effect of sugar and fat replacers on the texture of baked goods. J Texture Studies. 2004 Oct;35(4):383-93. doi: 10.1111/j.1745-4603.2004.tb00602.x
62. Li Q-M, Li Y, Zou J-H, Guo S-Y, Wang F, Yu P, Su X-J. Influence of adding chinese yam (*Dioscorea opposita* Thunb.) flour on dough rheology, gluten structure, baking performance, and antioxidant properties of bread. Foods. 2020;9(3):1-15.
63. Andrade BA, Perius DB, Mattos NV, Luvielmo MM, Mellado MS. Produção de farinha de banana verde (Musa spp) para aplicação em pão de trigo integral. Braz. J Food Technol. 2018;21:1-10. doi: 10.1590/1981-6723.5516
64. He H, Hoseney RC. Gas retention of different cereal flours. Cereal Chem. 1991;68(4):334-6.
65. Barroso AJR, Silva HA, Almeida FAC, Silva SN, Silva PB, Brito KD, et al. Use of residue of peanut exchange in the preparation of gluten free cake. Braz J Develop. 2019 Mar;5(4):3327-40. doi: 10.34117/bjdv5n4-1476
66. Miranda AA, Caixeta ACA, Flávio EF, Pinho L. Desenvolvimento e análise de bolos enriquecidos com farinha da casca do maracujá (*Passiflora edulis*) como fonte de fibras. Alim Nutr. 2013;24(2):225-32.
67. Teixeira E, Meinert E, Barbeta PA. Análise sensorial dos alimentos. Florianópolis (SC): UFSC; 1987.

68. Marcelino G, Coleta IT, Candido CJ, Santos EF. Characterization and sensorial analysis of cakes made with different concentrations of bark flour and baru pulp (*Dipteryx alata* Vog.). Multitemas. 2018;23(54):265-81.
69. Silva BC, Carpenedo E, Silva LO, Lavinas FC, Ribeiro-Alves MA. Elaboração de bolos de alfarroba isento de glúten. Alman Multid Pesq. 2014;1(1):79-93.
70. Stringheta PC, Oliveira TT, Gomes RC. Políticas de saúde e alegações de propriedades funcionais e de saúde para alimentos no Brasil. Rev Bras Cienc Farm. 2007 Jun;43(2):181-94. doi: 10.1590/S1516-93322007000200004

Appendix A

Table S1. Physicochemical and technological characteristics of cakes elaborated with different concentrations of ora-pro-nobis flour (n=3). (CONTINUOUS)

			Dependent Variables							
Essay	**X1 (Soybean Oil)**	**X2 (Ora-pro-nobis flour)**	**Y25 (Proteins)**	**Y26 (Fibers)**	**Y11 (Ash)**	**Y12 (Calcium)**	**Y13 (Potassium)**	**Y14 (Sodium)**	**Y15 (Magnesium)**	**Y16 (Iron)**
1	-1 (5%)	-1 (10%)	5.37 ± 0.05	3.41 ± 0.09	2.64 ± 0.07	2.30 ± 0.09	1.89 ± 0.04	0.027 ± 0.005	0.290 ± 0.005	0.016 ± 0.005
2	+1 (15%)	-1 (10%)	5.45 ± 0.06	3.47 ± 0.02	2.51 ± 0.05	2.93 ± 0.11	1.85 ± 0.05	0.026 ± 0.005	0.260 ± 0.004	0.014 ± 0.004
3	-1 (5%)	+1 (30%)	5.77 ± 0.17	5.38 ± 0.06	3.10 ± 0.04	3.78 ± 0.08	2.38 ± 0.03	0.033 ± 0.005	0.350 ± 0.001	0.019 ± 0.005
4	+1 (15%)	+1(30%)	5.85 ± 0.05	5.45 ± 0.05	3.10 ± 0.07	3.50 ± 0.10	2.21 ± 0.05	0.031 ± 0.005	0.370 ± 0.006	0.017 ± 0.005
5	-1.41 (2.95%)	0 (20%)	5.56 ± 0.04	4.40 ± 0.10	2.87 ± 0.07	3.55 ± 0.08	2.24 ± 0.03	0.031 ± 0.005	0.310 ± 0.005	0.018 ± 0.001
6	+1.41(17.05%)	0 (20%)	5.67 ± 0.02	4.48 ± 0.06	2.94 ± 0.06	3.40 ± 0.02	2.15 ± 0.01	0.030 ± 0.005	0.300 ± 0.001	0.014 ± 0.005
7	0 (10%)	-1.41 (5.9%)	5.35 ± 0.05	3.05 ± 0.04	2.43 ± 0.09	2.92 ± 0.08	1.81 ± 0.04	0.026 ± 0.005	0.250 ± 0.005	0.014 ± 0.005
8	0 (10%)	+1.41 (34.1%)	5.90 ± 0.01	5.84 ± 0.05	3.16 ± 0.02	3.76 ± 0.03	2.38 ± 0.01	0.033 ± 0.001	0.390 ± 0.013	0.021 ± 0.005
9	0 (10)	0 (20%)	5.62 ± 0.07	4.43 ± 0.07	2.89 ± 0.003	2.65 ± 0.04	2.07 ± 0.02	0.024 ± 0.005	0.330 ± 0.003	0.016 ± 0.003
10	0 (10)	0 (20%)	5.61 ± 0.10	4.43 ± 0.08	2.88 ± 0.06	2.56 ± 0.07	2.02 ± 0.03	0.022 ± 0.005	0.320 ± 0.004	0.017 ± 0.003
Control	-	-	4.93 ± 0.53	2.42 ± 0.07	2.28 ± 0.01	2.75 ± 0.01	1.73 ± 0.05	0.025 ± 0.001	0.240 ± 0.001	0.013 ± 0.001

Control: 0% ora-pro-nobis flour + 66.60% oil + 100% wheat flour; CO5FO10: 10% ora-pro-nobis flour + 63.27% oil (5% reduction) + 90% wheat flour; CO15FO10: 10% ora-pro-nobis flour + 85% oil + 90% wheat flour; CO5FO30: 30% ora-pro-nobis flour + 63.27% oil (5% reduction) + 70% wheat flour; CO15FO30: 30% ora-pro-nobis flour + 85% oil + 70% wheat flour; CO3FO20: 20% ora-pro-nobis flour + 64.63% oil (2,95% reduction) + 80% wheat flour; CO17FO20: 20% ora-pro-nobis flour + 82.95% oil + 80% wheat flour; CO10FO6: 5.9% ora-pro-nobis flour + 90% oil + wheat flour 94.1%; CO10FO34: 34.1% ora-pro-nobis flour + 90% oil + wheat flour 65.9%; CO10FO20: 20% ora-pro-nobis flour + 90% oil + 70% wheat flour; CO10FO20: 20% ora-pro-nobis flour + 90% oil + 70% wheat flour.

Table S1. Physicochemical and technological characteristics of cakes elaborated with different concentrations of ora-pro-nobis flour (n=3).

			Dependent Variables						
Essay	**X1 (Soybean oil)**	**X2 (Ora-pro-nobis flour)**	**Y17 (Zinc)**	**Y2 (Crumb color a*)**	**Y29 (Crumb color ΔE)**	**Y10 (Hardness)**	**Y11 (Elasticity)**	**Y12 (Gumminess)**	**Y13 (Chewiness)**
1	-1 (5%)	-1 (10)	0.002 ± 0.001	2.19 ± 0.22	2.54 ± 0.68	3.81 ± 0.53	12.06 ± 0.81	1.21 ± 0.19	11.86 ± 0.76
2	+1 (15%)	-1 (10)	0.002 ± 0.003	3.34 ± 0.10	1.94 ± 0.13	3.77 ± 0.40	11.87 ± 0.64	1.40 ± 0.15	20.71 ± 0.67
3	-1 (5%)	+1 (30)	0.003 ± 0.006	0.68 ± 0.05	4.15 ± 0.34	2.47 ± 0.14	8.31 ± 0.25	0.62 ± 0.11	5.65 ± 0.17
4	+1 (15%)	+1(30)	0.003 ± 0.004	0.80 ± 0.12	3.00 ± 0.22	6.12 ± 0.20	9.49 ± 0.29	1.56 ± 0.62	17.57 ± 0.79
5	-1.41 (2.95%)	0 (20%)	0.003 ± 0.006	2.20 ± 0.09	2.62 ± 0.30	2.44 ± 0.16	8.7 ± 0.95	0.61 ± 0.11	5.94 ± 0.69
6	+1.41(17.05%)	0 (20%)	0.003 ± 0.001	2.24 ± 0.22	2.33 ± 0.65	4.71 ± 0.32	9.95 ± 0.98	1.34 ± 0.20	13.88 ± 0.62
7	0 (10%)	-1.41 (5.9%)	0.002 ± 0.007	5.15 ± 0.23	2.95 ± 0.26	3.82 ± 0.36	11.1 ± 0.37	1.17 ± 0.22	13.65 ± 0.56
8	0 (10%)	+1.41 (34.1%)	0.0030 ± 0.003	-0.73 ± 0.15	5.73 ± 0.17	6.3 ± 0.91	9.30 ± 0.42	2.15 ± 0.29	15.92 ± 0.92
9	0 (10)	0 (20%)	0.002 ± 0.003	1.80 ± 0.08	3.28 ± 0.24	5.35 ± 0.57	10.10 ± 0.17	1.80 ± 0.18	19.73 ± 0.51
10	0 (10)	0 (20%)	0.002 ± 0.005	1.09 ± 0.20	3.81 ± 0.51	5.22 ± 0.51	10.78 ± 0.96	1.80 ± 0.20	19.04 ± 0.99
Control	-	-	0.002 ± 0.001	4.31 ± 0.31	-	1.97 ± 0.20	10.54 ± 0.50	0.67 ± 0.12	7.28 ± 0.88

Control: 0% ora-pro-nobis flour + 66.60% oil + 100% wheat flour; CO5FO10: 10% ora-pro-nobis flour + 63.27% oil (5% reduction) + 90% wheat flour; CO15FO10: 10% ora-pro-nobis flour + 85% oil + 90% wheat flour; CO5FO30: 30% ora-pro-nobis flour + 63.27% oil (5% reduction) + 70% wheat flour; CO15FO30: 30% ora-pro-nobis flour + 85% oil + 70% wheat flour; CO3FO20: 20% ora-pro-nobis flour + 64.63% oil (2,95% reduction) + 80% wheat flour; CO17FO20: 20% ora-pro-nobis flour + 82.95% oil + 80% wheat flour; CO10FO6: 5.9% ora-pro-nobis flour + 90% oil + wheat flour 94.1%; CO10FO34: 34.1% ora-pro-nobis flour + 90% oil + wheat flour 65.9%; CO10FO20: 20% ora-pro-nobis flour + 90% oil + 70% wheat flour; CO10FO20: 20% ora-pro-nobis flour + 90% oil + 70% wheat flour.

Table S2. R² values, calculated F, tabulated F, and mathematical model for the response variables of the formulated cakes.

Variable	**R^2**	**F_{calc}**	**F_{tab}**	**Mathematical model**
Proteins	0.9268	29.30	5.32	$Y_{25} = 7.68 + 1.78\ X_2$
Fiber	0.9999	28.21	4.74	$Y_{26} = 4.43 + 0.03\ X_1 + 0.99\ X_2 + X_1\ X_2$
Ash	0.9878	11.66	5.32	$Y_{11} = 2.85 + 0.26\ X_2$
Cálcium	0.8843	6.30	4.74	$Y_{12} = 2.95 + 0.24\ X1^2 + 0.40\ X_2$
Potassium	0.9812	61.40	5.19	$Y_{13} = 16.49 – 0.63\ X_1 + 2.70\ X_1^2 + 1.78\ X_2 + 2.29\ X_2^2$
Sodium	0.9818	63.30	5.19	$Y_{14} = 23.77 – 0.98\ X_1 + 3.90\ X_1^2 + 2.55\ X_2 + 3.27\ X_2^2$
Magnesium	0.9808	60.50	5.19	$Y_{15} = 2.31 – 0.10\ X_1 + 0.38\ X_1^2 + 0.25\ X_2 + 0.32\ X_2^2$
Iron	0.9822	62.90	5.19	$Y_{16} = 13.17 – 0.53\ X_1 + 2.16\ X_1^2 + 1.41\ X_2 + 1.88\ X_2^2$
Zinc	0.9825	65.00	5.19	$Y_{17} = 2.15 – 0.09\ X_1 + 0.36\ X_1^2 + 0.23\ X_2 + 0.30\ X_2^2$
Crumb color a*	0.8435	37.40	5.32	$Y_2 = 1.88 – 1.55\ X_2$
Crumb color ΔE	0.9023	20.48	4.74	$Y_{29} = 0.33 + 0.53*X_1 - 0.03*X_1^2 + 0.001*X_2 + 0.002*X_2^2 - 0.002*X_1*X_2$
Hardness	0.9427	17.70	5.19	$Y_{18} = 5.08 + 0.85\ X_1 - 0.85\ X_1^2 + 0.57\ X_2 + 0.92\ X_1\ X_2$
Elasticity	0.8081	14.60	5.32	$Y_{20} = 10.17 – 1.09\ X_2$
Gumminess	0.8011	5.40	5.32	$Y_{21} = 1.69 – 0.40\ X_1^2$
Chewiness	0.8584	11.50	4.74	$Y_{22} = 17.21 + 4\ X_1 – 3.52\ X_1^2$

R^2 = coefficient of determination; F_{calc}= calculated F; F_{tab} = tabulated.

Table S3. Analysis of variance for response variables. (CONTINUOUS)

		Lipids			Calories			Crost color L*			Crost color b*		
Parameter	**df**	**Sum of squares (SS)**	**Value p**	**Value f**	**Sum of squares (SS)**	**Value p**	**Value f**	**Sum of squares (SS)**	**Value p**	**Value f**	**Sum of squares (SS)**	**Value p**	**Value f**
Soybean Oil linear (L)	1	2.12187	0.72159	0.14625	186.539	0.5939	0.3348	0.9155	0.6728	0.2069	0.7125	0.3094	1.3529
Soybean Oil quadratic (Q)	1	2.61446	0.69303	0.18020	3.708	0.9389	0.0066	0.7802	0.6961	0.1763	0.0071	0.9132	0.0134
Ora-pro-nobis flour linear (L)	1	11.6227	0.42135	0.89112	12.849	0.8866	0.0231	0.8867	0.6776	0.2004	0.2913	0.4983	0.5532
Ora-pro-nobis flour quadratic (Q)	1	0.22886	0.90610	0.01577	37.736	0.8075	0.0677	0.2353	0.8289	0.0532	0.3410	0.4661	0.6475
Soybean Oil (L) x Ora-pro-nobis flour (L)	1	0.00040	0.99606	0.00003	200.222	0.58116	0.3594	0.0002	0.9946	0.00005	0.3844	0.4411	0.7299
Error	4	58.03214			2228.582			17.6980			2.1065		
Total of SS	9	76.07244			2666.541			21.1942			3.9729		

Table S3. Analysis of variance for response variables. (CONTINUOUS)

		ΔE Crost			Volume			Yield			Moisture		
Parameter	**df**	**Sum of squares (SS)**	**Value p**	**Value f**	**Sum of squares (SS)**	**Value p**	**Value f**	**Sum of squares (SS)**	**Value p**	**Value f**	**Sum of squares (SS)**	**Value p**	**Value f**
Soybean Oil linear (L)	1	0.2053	0.5384	0.4515	0.0001	0.9488	0.0046	0.1212	0.8512	0.040	6.8855	0.0698	6.0422
Soybean Oil quadratic (Q)	1	0.0078	0.9022	0.0171	0.0098	0.5624	0.3978	1.9650	0.4656	0.6491	7.1571	0.0663	6.2806
Ora-pro-nobis flour linear (L)	1	0.1090	0.6500	0.2397	0.0021	0.7834	0.0863	0.2535	0.7866	0.0837	0.0289	0.8812	0.0253
Ora-pro-nobis flour quadratic (Q)	1	0.0108	0.8846	0.0239	0.0044	0.6919	0.1816	0.2843	0.7745	0.0939	1.0045	0.4009	0.8815
Soybean Oil (L) x Ora-pro-nobis flour (L)	1	0.0380	0.7868	0.0836	0.0225	0.3928	0.9154	0.0182	0.9419	0.0060	4.0602	0.1321	3.5629
Error	4	1.8187			0.0983			12.108			4.5582		
Total of SS	9	2.1842			0.1335			16.041			22.7155		

Table S3. Analysis of variance for response variables.

		Cohesiveness			Adhesiveness		
Parameter	**Df**	**Sum of squares (SS)**	**Value p**	**Value f**	**Sum of squares (SS)**	**Value p**	**Value f**
Soybean Oil linear (L)	1	0.0017	0.4764	0.6123	0.0131	0.2329	1.9723
Soybean Oil quadratic (Q)	1	0.0044	0.2734	1.6094	0.0054	0.4173	0.8163
Ora-pro-nobis flour linear (L)	1	0.0042	0.2856	1.5162	0.0019	0.6170	0.2930
Ora-pro-nobis flour quadratic (Q)	1	0.0002	0.8122	0.0644	0.0005	0.7939	0.0779
Soybean Oil (L) x Ora-pro-nobis flour (L)	1	0.0001	0.8586	0.0360	0.0030	0.5361	0.4571
Error	4	0.0111			0.02647		
Total of SS	9	0.0218			0.0535		

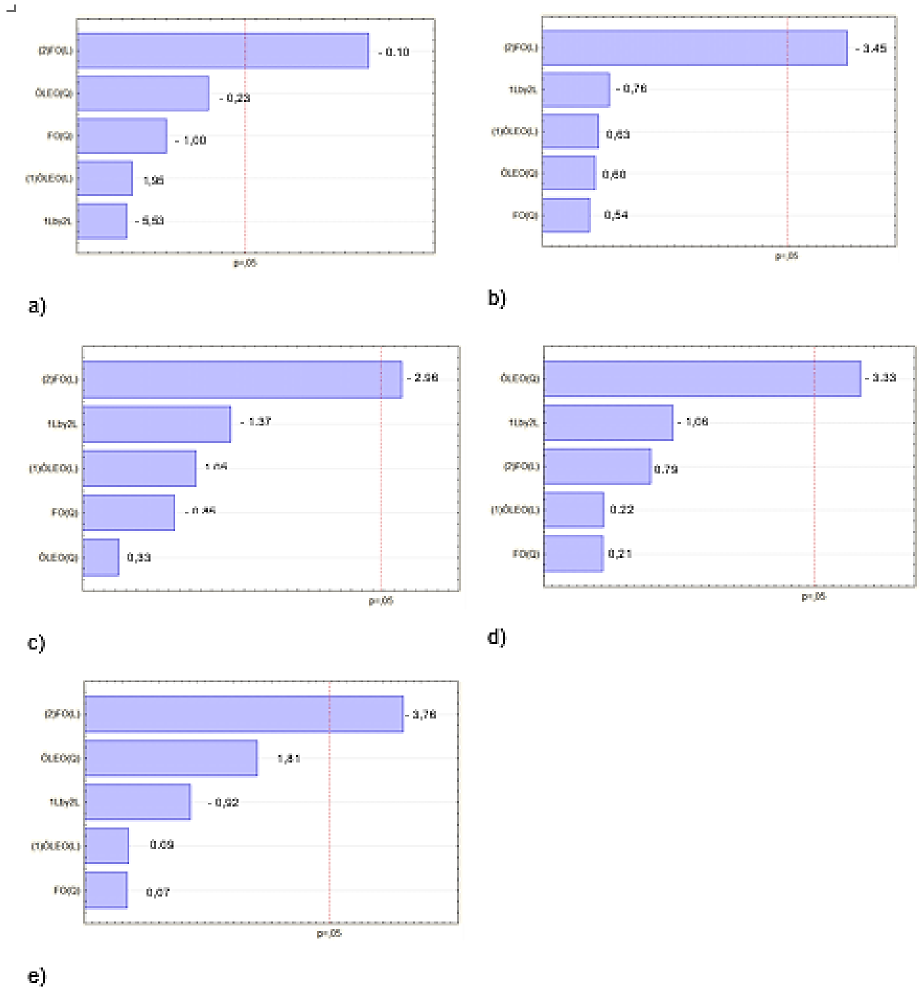

Figure S1. Pareto for the dependent variables of crumb color parameter L (a), parameter b* (b) of crost color parameter a* (c), water activity (d), and carbohydrates (e) of cakes.*

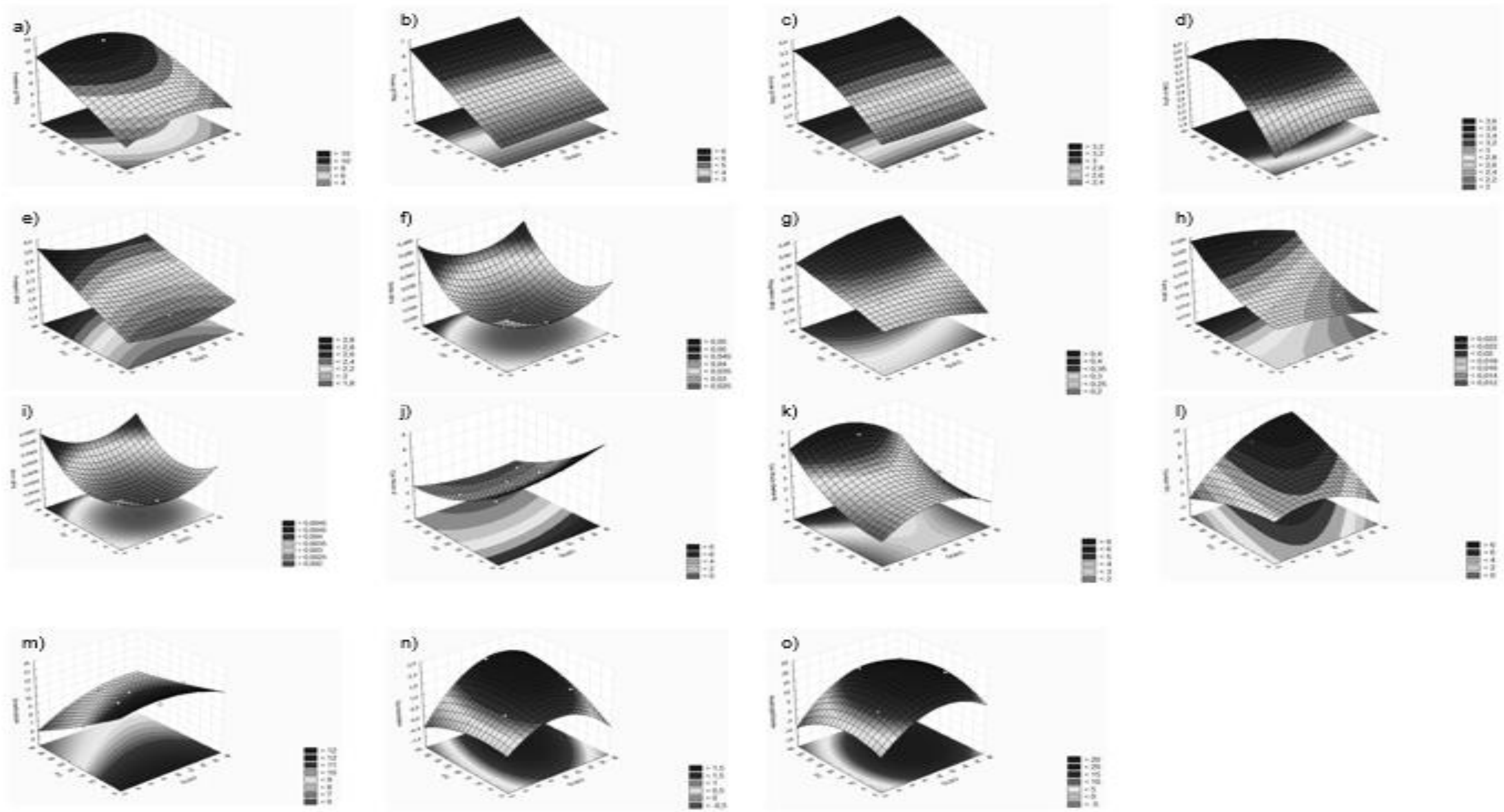

Figure S2. Response surface for dependent variables of proteins (g.100g^{-1}), fibers (g.100g^{-1}), ashes (g.100g^{-1}), minerals (g.kg), color, and texture. a) proteins; b) fibers; c) ashes; d) calcium; e) potassium; f) sodium; g) magnesium; h) iron; i) zinc, j) crumb color a; k) crumb color ΔE, l) hardness; m) elasticity; n) gumminess, o) chewiness.*