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# Utilization of mathematical models to evaluate the acceptance and physicochemical parameters for the development of a beverage made from cashew nut

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**Abstract**

The influence of the addition of pineapple juice and sugar (independent variables) over sensory acceptance and physicochemical parameters (dependent variables) was evaluated considering the development of a cashew nut kernel-based beverage by means of the response surface methodology. The effect of the independent variables could only be evaluated in relation to the acceptance of the overall impression, where only sugar concentration influenced, being determined that the beverage formulation that allows for greater acceptance should be added with 35% pineapple juice and 7% sugar. Soluble solids were influenced by both juice and sugar concentrations, where higher concentration of both led to lower sensory acceptance. The juice addition only affected total titratable acidity and pH, but did not interfere in the formulations' acceptance. It was possible to combine the nutritional, functional and sensory characteristics of the used raw materials (cashew nuts, pineapple juice and prebiotic substances) to elaborate a beverage with high nutritional and functional appeal.



## Introduction

Response Surface Methodology (RSM) is a mathematical and statistical approach that has been used for optimization and development of formulations and processes (Villegas *et al.*, 2010; Villarino *et al.*, 2015; Ilyasoglu *et al.*, 2015; Huang and Ma, 2016). Concerning formulations' optimization or development, by means of this methodology, it is possible to analyze the combination effect among the levels of ingredients (independent variables) in one or more product's quality parameters (dependent variable) (Yaakob *et al.*, 2012). Thus, it is possible to determine optimal concentrations of addition of ingredients, as well as to evaluate the possible interaction among them by means of adjusted mathematical models (Wadikar *et al.*, 2008). When used to evaluate consumers' responses, RSM generates prediction equations that will correlate consumer's response with the analyzed independent variables. From the models (equations) generated, it is possible to estimate consumer's response in relation to the combination of levels of not tested variables and, in this way, optimize the product (Mendes *et al.,* 2001).

Population's growing search for healthy food has stimulated industry to research and develop new products to meet this criterion (El-Salam *et al.,* 2011). Results of several clinical trials with tree nuts have demonstrated the favorable effect that their consumption has over lipids and lipoproteins in the blood, main risk factors for cardiovascular diseases. These effects have been evident in different population groups, having several planning and study methods been used (Griel and Kris-Etherton, 2006). These benefits are partly related to the lipid components (Alasalvar and Pelvan, 2011), which contain innumerous bioactive compounds and health promoting components (Alasalvar and Shahidi, 2009; Shin *et al.*, 2010). Cashew nut kernel's lipid fraction corresponds to 44%, 20% of which is composed of saturated fatty acids, and the rest is majorly composed of oleic acid (57%), followed by linoleic acid (22%), a polyunsaturated fatty acid (Robbins *et al.*, 2011).

Cashew nut kernel-based beverages added with fruit juice and prebiotic substances have already demonstrated to be a viable alternative in the field of development of new functional beverages, making it possible to obtain food with excellent nutritional and functional characteristics, besides acceptable sensory quality (Rebouças *et al.*, 2014). Within this context, research on new juices to be used as this beverage's flavoring agents is absolutely valid and widens the possibilities of use for cashew nut kernels.

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Table 1. Experimental design and sensory evaluation results regarding cashew nut-based beverage added with pineapple juice.

| | Experimental design | | Sensory evaluation | | | | | |
|---|---|---|---|---|---|---|---|---|
| Formulations | 20(-1) | 4(-1) | Color | Aroma | Flavor | Sweetness | Thickness | Overall Impression |
| F1 | 20(-1) | 8(+1) | $6.2^{de}$ | $5.6^{bc}$ | $5.4^{cd}$ | $5.5^{cd}$ | $5.7^{bcd}$ | $5.5^{bc}$ |
| F2 | 40(+1) | 4(-1) | $6.4^{bcde}$ | $5.8^{abc}$ | $5.6^{bcd}$ | $5.5^{cd}$ | $5.5^{cd}$ | $5.5^{bc}$ |
| F3 | 40(+1) | 8(+1) | $6.3^{cde}$ | $6.3^{ab}$ | $6.1^{abc}$ | $6.4^{ab}$ | $6.1^{abc}$ | $6.0^{ab}$ |
| F4 | 16(-1.41) | 6(0) | $6.4^{bcde}$ | $6.5^{a}$ | $6.4^{ab}$ | $6.1^{abc}$ | $6.7^{a}$ | $6.4^{a}$ |
| F5 | 44(+1.41) | 6(0) | $5.9^{e}$ | $5.8^{abc}$ | $6.0^{abc}$ | $6.2^{abc}$ | $6.1^{abc}$ | $6.1^{ab}$ |
| F6 | 30(0) | 3(-1.41) | $6.8^{ab}$ | $5.9^{abc}$ | $5.9^{abc}$ | $5.7^{bcd}$ | $6.0^{abc}$ | $6.0^{ab}$ |
| F7 | 30(0) | 9(+1.41) | $6.1^{de}$ | $5.2^{c}$ | $4.9^{d}$ | $5.0^{d}$ | $5.1^{d}$ | $4.9^{c}$ |
| F8 | 30(0) | 6(0) | $6.0^{e}$ | $6.2^{ab}$ | $6.3^{abc}$ | $6.1^{abc}$ | $6.0^{abc}$ | $6.1^{ab}$ |
| F9 | 30(0) | 6(0) | $6.6^{abcd}$ | $6.1^{ab}$ | $6.2^{abc}$ | $6.2^{abc}$ | $6.2^{abc}$ | $6.4^{a}$ |
| F10 | 30(0) | 6(0) | $6.6^{abcd}$ | $6.4^{a}$ | $6.2^{abc}$ | $6.3^{abc}$ | $6.1^{abc}$ | $6.4^{a}$ |
| F11 | 30(0) | 6(0) | $6.7^{abc}$ | $6.3^{ab}$ | $6.2^{abc}$ | $6.4^{ab}$ | $6.4^{ab}$ | $6.4^{a}$ |
| F12 | 30(0) | 6(0) | $6.4^{bcde}$ | $6.5^{a}$ | $5.9^{abc}$ | $5.9^{abc}$ | $6.0^{abc}$ | $6.1^{ab}$ |
| F13 | 20(-1) | 4(-1) | $7.0^{a}$ | $6.5^{a}$ | $6.5^{a}$ | $6.5^{a}$ | $6.7^{a}$ | $6.6^{a}$ |

[ab] Different letters in the same column represent significant difference according to Tukey's test ($p < 0.05$).

Pineapple is a tropical fruit heavily consumed in several countries in the form of various products (Hossain and Rahman, 2011). This fruit's juice is highly popular due to its sensory characteristics of aroma and flavor being appreciated, as well as for its nutritional composition (Laorko *et al.*, 2013), being rich in vitamins A, B and C and minerals such as calcium, iron and phosphorus (Hossain and Rahman, 2011), besides a huge amount of phenolic compounds and antioxidants that are beneficial to health (Laorko *et al*., 2013).

These factors combined make the pineapple juice a raw material with potential to be used in a new product with high sensory and nutritional appeal. In this context, this study aimed to evaluate the influence of the addition of pineapple juice and sugar on sensory acceptance and physicochemical parameters considering the development of a cashew nut kernel-based beverage through the response surface methodology.

## Materials and Methods

### *Experimental design and beverages formulation*

The beverages were formulated using hydrosoluble extract of cashew nut kernels (HEK) (3.46g carbohydrates, 6.22g lipids, 3.41g proteins, 0.35g ashes, 3.6°Brix, pH 6.56), concentrated pineapple juice (pH 3,39; 11,6ºBrix), commercial sugar and the prebiotic substances inulin (polymerization degree ≥ 10, Orafti GR, São Paulo - BRA) and oligofructose (2 to 8 monomers, Orafti P95, São Paulo - BRA). The HEK was obtained following the methodology described by Rebouças *et al*. (2014).

Beverages' formulations followed a 22 central composite rotatable design with five repetitions at the central point, where the independent variables studied were pineapple juice and sugar concentrations (Table 1). The product was developed with the objective to be a cashew nut based beverage, the addition of pineapple juice had the objective to give a better taste, besides favoring its nutritional and functional characteristics. Due this fact, the addiction of juice is not superior to 50% of the final composition of the beverage. In relation to the addition of sugar, this is considered an important factor since the addition of different amounts of pineapple juice in the beverage generates different acidity degrees. This acidity must be corrected, however the amount of sugar necessary varies in accordance with the acidity presented, thus the amount of sugar must be an independent variable in this study.

The addition of HEK in the formulations was done in a way to complement, along with the juice, the 100% liquid volume. The amount of sugar (w / v) added to a mixture of HEK and juice followed the experimental design, plus 3% (w / v) mixture of inulin and oligofructose in a 50:50 proportion. The ingredients were homogenized at a rotation of 900rpm and then stored into polystyrene bottles and subjected to thermal treatment at 65°C for 2 minutes. The samples were cooled and kept under refrigeration until the analyses were carried out.

### *Sensory analysis*

The acceptance of different formulations was measured by the 9-point structured hedonic scale test (9 = 'like extremely'; 5 = 'neither like nor dislike'; 1 = 'dislike extremely'), where the following sensory attributes were analyzed: color, aroma, flavor, sweetness, thickness and overall impression. The test was conducted with 130 consumers, mostly composed of females (72.31%), aging between 18 and 25 years (84.62%) and undergraduate students

(80.77%).

Sensory evaluation was conducted in individual booths under artificial daylight illumination. A 25 mL portion was served in a monadic way in cups codified with three random digits according to an incomplete blocks design (Cochran and Cox, 1992). This study was approved by the Comitê de Ética em Pesquisa which regulates research involving humans, with the number 873.769.

*Physicochemical analysis*

The physicochemical analysis performed were pH, using a pH meter model 3505 (Jenway, Staffordshire, UK), total titratable acidity (TTA), determined by the reaction with NaOH solution and expressed in citric acid percentage, and soluble solids (SS) by refractometer PAL-1 (Atago). All determinations followed a methodology described by Instituto Adolfo Lutz (IAL, 2004).

*Data analysis*

Mathematical models adjusted to a quadratic equation were used to evaluate the effect of sugar and pineapple concentrations over the acceptance of sensory attributes and physicochemical determinations. The models were subjected to ANOVA in order to evaluate the adjustment quality and the significance of the effects (linear, quadratic and interaction). Based on the coefficient of determination ($R^2$), coefficient of determination adjusted ($R^2_{adjusted}$) and on the analysis of lack of fit, the quality of the models obtained was evaluated so, then, the response surface graphs could be generated. ANOVA and Tukey's test were used in models that presented either a significant lack of fit ($\alpha = 0.05$) or low coefficients of determination. The Pearson correlation coefficients (r) were calculated in order to verify the associations between the sensory attributes analyzed. All the analyses were performed using the statistical software Statistica version 7.0.

## Results

*Sensory analysis*

The formulations reached satisfactory acceptance attaining hedonic scores equal or close to 6.0 (like slightly) and 7.0 (like moderately) (Table 1), only formulation 7, to which the lowest sugar concentration was added, reached an average around 4.0 (disliked slightly) on flavor and overall impression. The average comparison test applied showed that most of formulations did not differ significantly in most evaluated attributes, except for formulations 1, 2 and 7, which, generally, differed from the others.

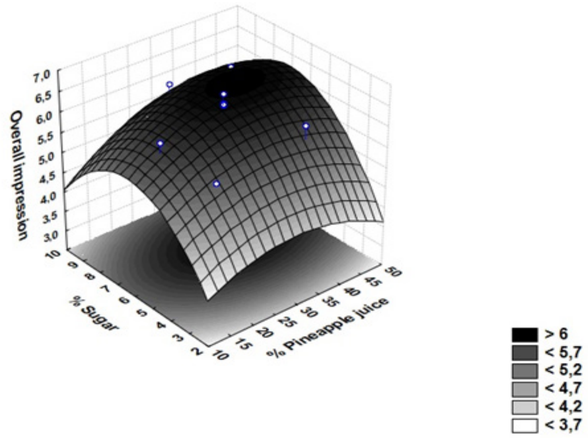


Figure 1. Effect of sugar and pineapple juice concentration on overall impression.

Except for sweetness, which presented a significant lack of fit ($p < 0.05$), it was possible to obtain a mathematical model ($p < 0.05$) for all the other evaluated attributes, which means that the error within the model and in its replicates (central points) was low and that the model can be used for prediction purposes. Nonetheless, for the attributes color ($R^2 = 0.64$; $R^2_{adjust.} = 0.39$; P-value (lack of fit) = 0.42), aroma ($R^2 = 0.73$; $R^2_{adjust.} = 0.54$; P-value (lack of fit) = 0.07), flavor ($R^2 = 0.68$; $R^2_{adjust.} = 0.46$; P-value (lack of fit) = 0.08) and thickness ($R^2 = 0.65$; $R^2_{adjust.} = 0.40$; P-value (lack of fit) = 0.23), the coefficients $R^2$ and R2adjust of the models were very low, not being safe to use them for results interpretation. Due to the lack of a significant fit, it was not possible to evaluate the effect of variables and their different levels on the acceptance of sweetness (lack of fit: p = 0.048).

Regarding overall impression, this was shown to be influenced only by sugar concentration (linear and quadratic effects) (Table 2). The surface response obtained (Figure 1) through equation 1 shows that, the positive linear effect and the negative quadratic made the elevation in the sugar concentration lead to an increase of acceptance up to a maximum where, then, there was a response decrease. Through the graph, it is possible to view a region of maximum acceptance by means of sugar addition between 6 and 7% and juice, approximately, 30 and 44%.

$$\text{Overall impression} = 0.92 + 0.086\text{Juice} + 1.15\text{Sugar} - 0.001\text{Juice}^2 - 0.097\ \text{Sugar}^2 - 0.004\ \text{JuiceSugar}$$ ($R^2 = 0.79$; $R^2_{adjust.} = 0.64$; P-value (lack of fit) = 0.14) (1)

*Physicochemical analyses*

The mathematical models obtained for the determinations of soluble solids, pH and total titratable acidity were satisfactory ($p < 0.05$), with an insignificant lack of fit ($p > 0.05$). The results of

Table 2. Regression coefficients of the fitted equation obtained to evaluate the overall impression, soluble solids, pH and total titratable acidity.

| | Parameters | Estimated coefficient | Standard error | p-value |
|---|---|---|---|---|
| Overall Impression | Mean | 0,925478 | 1,364149 | 0,534721 |
| | Juice (L) | 0,086357 | 0,057105 | 0,205010 |
| | Juice (Q) | -0,001600 | 0,000793 | 0,113652 |
| | Sugar (L) | 1,159315 | 0,264866 | 0,011903 |
| | Sugar (Q) | -0,096089 | 0,017728 | 0,005616 |
| | Interaction | 0,004375 | 0,005127 | 0,441589 |
| Soluble solids | Mean | 10,43669 | 0,499427 | 0,000031 |
| | Juice (L) | 0,05008 | 0,011773 | 0,013115 |
| | Sugar (L) | 0,71569 | 0,056822 | 0,000229 |
| pH | Mean | 5,925573 | 0,168726 | 6,394032 |
| | Juice (L) | -0,068230 | 0,011546 | -0,036173 |
| | Juice (Q) | 0,000642 | 0,000190 | 0,001170 |
| TTA | Mean | 0,137221 | 0,016019 | 0,001020 |
| | Juice (L) | 0,008968 | 0,000517 | 0,000065 |

TTA: Total titratable acidity; (L): Linear; (Q): Quadratic

the determinations of soluble solids performed in the formulations varied between 13.6 (Formulation 1) and 18.5°Brix (Formulation 4) (Table 3), obtaining the highest values when there was a combination between higher contents of juice and sugar. The beverages suffered influence from both sugar and juice concentrations (Table 2), both with a positive linear effect (Equation 2). The increase in the concentration of these variables led to a higher content of soluble solids as can be seen in Figure 2.

Soluble Solids = 10.43 + 0.050Juice + 0.715Sugar ($R^2$= 0.91; $R^2_{adjust.}$= 0.89; P-value (lack of fit) = 0.23) (2)

Regarding pH, the highest value (5.01) was obtained by the formulation with the lowest juice concentration (Formulation 5) and the lowest value (4.15) was obtained by the formulations with higher content (Formulation 6) (Table 3). The beverage's pH was shown to be influenced only by juice concentration in its linear and quadratic effects (Table 2). As can be observed in equation 3, the negative linear effect of juice concentration was a lot more influent than the positive quadratic effect, this fact is reflected in the surface response obtained (Figure 2), where diminishing juice content leads to a pH increase and vice versa.

pH = 5.92 – 0.068Juice + 0.0006Juice$^2$ ($R^2$= 0.98; $R^2_{adjust.}$= 0.97; P-value (lack of fit) = 0.83) (3)

As obtained in pH determination, also regarding titratable acidity, it was expected that the least acid formulations were added with lower juice concentrations (Table 3). The beverages were

Table 3. Physicochemical analysis results for each formulation.

| Formulations | Juice (%) | Sugar (%) | SS (ºBrix) | pH | TTA (% citric acid) |
|---|---|---|---|---|---|
| F1 | 20 (-1) | 4 (-1) | 13.6 | 4.78 | 0.33 |
| F2 | 20 (-1) | 8 (+1) | 17.2 | 4.84 | 0.27 |
| F3 | 40 (+1) | 4 (-1) | 15.5 | 4.26 | 0.47 |
| F4 | 40 (+1) | 8 (+1) | 18.5 | 4.21 | 0.49 |
| F5 | 16 (-1,41) | 6 (0) | 16.2 | 5.01 | 0.28 |
| F6 | 44 (+1,41) | 6 (0) | 16.7 | 4.15 | 0.53 |
| F7 | 30 (0) | 3 (-1,41) | 14.3 | 4.48 | 0.42 |
| F8 | 30 (0) | 9 (+1,41) | 17.9 | 4.49 | 0.40 |
| F9 (PC) | 30 (0) | 6 (0) | 16.5 | 4.49 | 0.41 |
| F10 (PC) | 30 (0) | 6 (0) | 15.7 | 4.49 | 0.40 |
| F11 (PC) | 30 (0) | 6 (0) | 16.4 | 4.37 | 0.44 |
| F12 (PC) | 30 (0) | 6 (0) | 16.0 | 4.44 | 0.42 |
| F13 (PC) | 30 (0) | 6 (0) | 16.2 | 4.43 | 0.42 |

SS: Soluble Solids; TTA: Total titratable acidity

influenced only by the variable juice in its linear term (Table 2). As expected, the elevation of juice concentration led to a linear increase of acidity, as can be observed in surface response (Figure 2) obtained by means of equation 4.

Titratable acidity = 0.137 + 0.0089Juice ($R^2$= 0.94; $R^2_{adjust.}$= 0.93; P-value (lack of fit) = 0.21) (4)

## Discussion

Regardless the level of the independent variables, the acceptance of the formulations was fairly similar and, possibly, this was one of the facts that caused the low indexes of correlation obtained ($R^2$ and $R^2_{adjusted}$). According to Capitani *et al.* (2009), there must be a variation between the results of the different treatments for a surface response to be modeled. In general, the formulations added with more than 30% juice and sugar equal or superior to 4% obtained sensory acceptance (averages around 6.0 'like slightly') in all analyzed attributes. When one evaluates the acceptance of sensory attributes in food, due to the subjectivity of the analysis, it is not always possible to obtain an adequate mathematical model that explains the experimental data (Nikzadeh and Sedaghat, 2008). Despites it has were possible to obtain significant models, the coefficients $R^2$ and $R^2_{adjusted}$ were very low, indicating that the independent variables do not totally explain the acceptance of the attributes. Thus, it was not considered adequate for the analysis of these results a value for $R^2$ lower

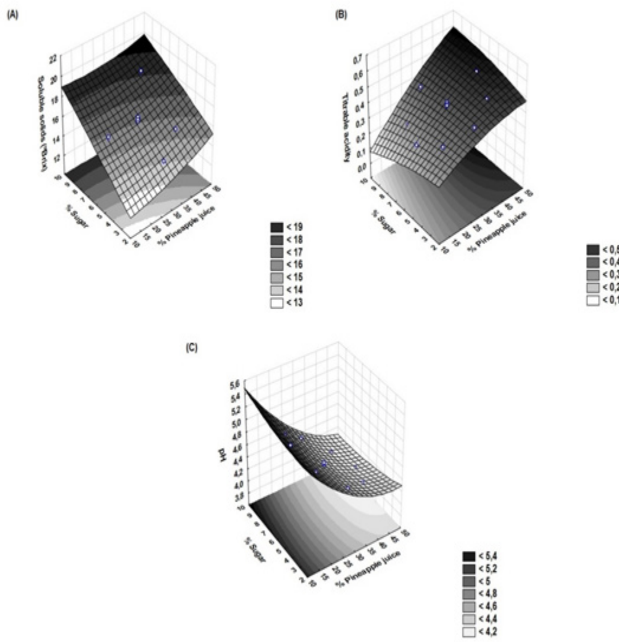

Figure 2. Effect of the addition of sugar and pineapple juice on the concentration of soluble solids (A), titratable acidity (B) and pH (C).

than 0.70, therefore, only the overall impression was analyzed to determine the formulation that allows for greater acceptance. It is important to highlight that, the correlation analysis showed a strong and significant association of all the evaluated attributes (aroma: r = 0.88; flavor: r = 0.97; sweetness: r = 0.91; thickness: r = 0.93), except for color, with the overall impression, which reinforces its importance for the determination of the formulation. Thus, taking this criterion into consideration, the formulation that allows for the achievement of a satisfactory acceptance should be added with 35% pineapple juice and 7% sugar.

The present content and kind of sugars and organic acids influence directly the beverage's sensory properties and acceptance (Kelebek and Selli, 2011). Fruit juices are composed, most of them, of sugars, organic acids and minerals, which correspond to these products' SS (Andrés *et al*., 2015). However, it is noteworthy that, in the case of the evaluated beverages, the determined contents of solids also correspond to the presence of proteins, commercial sugar and prebiotic substances from the other raw materials composing the formulation. In this way, it was expected that, the higher the addition of sugar and juice, the higher the SS content. Several sensory characteristics present in food can be influenced by soluble solid content (King *et al*., 2006), among them, flavor, which can have its perception affected by differences of even one degree Brix between formulations. In general, a lower acceptance of sensory attributes was obtained in low values of SS. In contrast, higher averages were achieved from the combination of 30% juice and 6% sugar (F9, F10, F11, F12 and F13 – central points) and the ones added with 40% sugar and 4 or 8% sugar (Formulations 3 and 4, respectively). Like the SS, the amount of organic acids also influences the beverages' sensory characteristics, mainly in relation to flavor and sweetness (Huang *et al.*, 2009). Regarding pH and TTA measures, formulations showed to be influenced only by the juice content, which was expected, considering that, among the studies variables, only the addition of juice could contribute significantly for the acidity of the beverage. Thus, a higher addition of juice contributed to the product acidification diminishing its pH and increasing its TTA, which is advantageous from the industrial point of view, making it less prone to microbial deterioration. It is worth highlighting that, despite the elevated acidity, it did not interfere in the product's acceptance, which obtained formulations with satisfactory acceptance.

## Conclusion

It was possible to combine the nutritional, functional and sensory characteristics of the used raw materials (cashew nuts, pineapple juice and prebiotic substances) to elaborate a beverage with high nutritional and functional appeal.

The effect of the independent variables could only be evaluated in relation to the acceptance of the overall impression, where only the sugar concentration influenced, determining that the beverage formulation that allows for greater acceptance should be added with 35% pineapple juice and 7% sugar. Soluble solids were influenced by both juice and sugar concentrations, where higher concentration of both led to a lower sensory acceptance. Total titratable acidity and pH suffered influence from the addition of juice only, not interfering in the formulations' acceptance.

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