

## Sensory Properties Evaluation of Pine Nut (*Araucaria angustifolia*) Cereal Bars Using Response Surface Methodology

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The objective of this work was to study the effect of crystal sugar/glucose concentration (SG - 40/60% wt to 60/40% wt) and Pine nut concentration (Pn - 0 to 20% wt in substitution of the oat bran) on the main sensory properties of cereal bars, following a 2<sup>2</sup> central composite rotational design (CCRD). The results were analyzed using response surface methodology (RSM). Samples of cereal bars were prepared with oat bran, rice flakes and dehydrated Pine nut as ingredients. In addition, was added syrup crystal sugar/glucose with 86°Brix which were heated for 2 minutes at 105 °C, to produce the dough (0,8 Kg). The responses evaluated were the sensory acceptance test for appearance, aroma, flavor, texture and overall acceptance, and purchase intention. In the sensory acceptance test, all samples presented acceptable scores (>5) which were considered good scores. Increasing crystal sugar concentration reduced all the sensory properties of Pine nut cereal bars. According to the results the samples 6 and central points were considered the best Pine nut cereal bar.

Key words: Pine nut; Cereal bars; Syrup crystal sugar/glucose.

### 1. Introduction

Currently, consumers and food companies have become increasingly concerned about healthy and practice feed. This fact has driven the consumption of products such as cereal bars, mainly due to the consumption's trend of foods with high dietary fiber and low fat (Izzo and Niness, 2001; Kahlon and Keagy, 2003). The main aspects considered in the cereal bar preparation include the choice of cereal (oats, wheat, rice, barley, corn), selecting the appropriate carbohydrate to maintain the balance between the sensory characteristics and shelf-life, enrichment with various nutrients, stability in the processing and the use of dietary fiber (Gutkoski et al., 2007).

The Pine nut is the seed of the pine *Araucaria angustifolia* (Betortoloni, Otto Kuntze), it is considered to be an excellent source of nutrients. According to Cordenunsi et al. (2004), the Pine nut presents carbohydrates (36 %), dietary fiber (5 %), protein (3 %), fat (1 %) and soluble sugars (2.4 %) in the raw material, being also a source of copper and magnesium. In addition, Gama (2006) cited that the Pine nut can present 15-20 % of dietary fiber in its composition after cooking or roasted. Its use also has an environmental appeal, involving the preservation of a species that has been devastated for many years in South America (Conto et al., 2011).

The purpose of this study was to evaluate the influence of the addition of Pine nut and crystal sugar/glucose concentration on the sensory properties of Pine nut cereal bars, following a Central Composite Rotational Design (CCRD) and analyzing the results by the Response Surface Methodology (RSM).

## 2. Materials and method

### 2.1 Pine nut preparation

The cooked, peeled and milled Pine nuts (5 Kg), *Araucaria angustifolia* var. *angustifolia*, were purchased in May 2013, in Lages (SC), Brazil, and stored at -18 °C, to be used during the development of the work. The dehydration of the cooked, peeled and milled Pine nut was performed in an oven air at 115 °C until constant weight. Later, the separation of particle size (< 0.5 cm) was made and subsequent packaging of "grits" was held in plastic bags and storage for further processing of the cereal bars.

### 2.2 Cereal bars formulation

The cereal bars presented as basic formulation dry ingredients (oat bran – 35 %, and rice flakes – 15 %) and agglutinating (crystal sugar/glucose syrup), according to the methodology proposed by Gutkoski et al. (2007) and Freitas and Moretti, (2006). The percentages of Pine nut and crystal sugar/glucose were calculated by total dough weight multiplied by the concentration determined by the experimental design (Table 1). The percentages of Pine nut were utilized in substitution of oat bran.

The cereal bars were produced in the pilot plant of cereal bars of Cereal-Chocotec in the Institute of Food Technology (ITAL) of São Paulo, in Campinas/SP, in July 2014.

All ingredients were weighed in a semi-analytical balance. To compose the binding phase or syrup of agglutination, the crystal sugar and glucose syrup were mixed, solubilized in water and heated over low heat in a stainless pan mixer coupled with automatic ignition stove from *Braslaer Indústria e Comércio de Máquinas Ltda.* (Limeira, SP). The control of the binder phase was followed by measuring the temperature (105 °C) by use of a digital thermometer GULTERM 180 (*Gulton Instrumentos de Medição e Automação Indústria e Comércio Ltda.*, São Paulo, SP) and total soluble solids (86 °Brix) by digital refractometer model RDA8600 (ACATEC, São Paulo, SP). The dry ingredients (500 g) were homogenized and added of binder phase (300 g ± 20 g). The dough was laminated in manual laminator table (*Braslaer Indústria e Comércio de Máquinas Ltda.*, Limeira, SP). The cooling was carried out at room temperature. After cooling, the cut of the cereal bars in a rectangular shape was made using the cutting table semi-automatic composed by rotating knives (*Braslaer Indústria e Comércio de Máquinas Ltda.*, Limeira, SP), with dimensions: 10 cm (length) x 3 cm (width) x 1.5 cm (height), which presented a mean weight of 20 g. Bars were packed in polyethylene adhesives bags and frozen.

### 2.3 Experimental design

The cereal bars were produced with varying concentrations of crystal sugar/glucose syrup (S/G) and Pine nut concentration (Pn), according to the levels presented in Table 1. The 2<sup>2</sup> central composite rotational design (CCRD) described in Table 1 was followed, with 4 factorial points, 4 axial points and 3 central points, totalizing 11 trials.

### 2.4 Pine nut "grits" characterization

The moisture content of the Pine nut "grits" was determined according to AACC method 44-15.02 (2010). Lipid content was determined according to AACC method 30-25.01 (2010). Protein content was conducted using the Kjeldahl method and expressed using the conversion factor N × 5.7, according to AACC method 46-13.01 (2010). Ash content was determined using the approved method 08-01.01 (AACC, 2010). Dietary fiber was determined according to AACC Method 32-21.01 (2010), utilizing analytical proceed K-TDFR 06/14 (Megazyme, 2014). Carbohydrates were determined by difference. All results were made in triplicate and the results were expressed in dry base.

### 2.5 Sensory analysis of cereal bars

The sensory acceptance test (appearance, aroma, flavor, texture and overall acceptance) was carried out using a 9-point hedonic scale (1 = "disliked extremely", 9 = "liked extremely"), according to Stone and Sidel (1985), and the purchase intention, using 5-point scale (1 = "certainly would buy" and 5 = "certainly would not buy"). The sensory analysis counted with 54 untrained panelists, over than 18 years old, who were recruited among students, staff and professors of the Federal Institute of Santa Catarina (IFSC) – Lages, SC. Half of Pine nut cereal bar of each test was presented monadically, on plastic plates coded with three digits, in individual booths.

## 2.6 Statistical analysis

The evaluation of the effects of the different concentrations of Pn and S/G on sensory characteristics of cereal bars was done using the STATISTICA 7.0 software (StatSoft Inc., Tulsa, OK, USA), verifying the possibility of analysis of results by the Response Surface Methodology. The same program was used for the mean comparison test, by analysis of variance (ANOVA) and the Tukey test, at a significance level of 0.95 ( $p \leq 0.05$ ).

Table 1: Effect of the concentration of Pine nut (Pn) and crystal sugar/glucose (S/G) on the sensory characteristics of cereal bars.

Trials	Coded variables <sup>A</sup>		Real variables <sup>B</sup>		Appearance <sup>C</sup>	Aroma <sup>C</sup>	Flavor <sup>C</sup>	Texture <sup>C</sup>	Overall acceptance <sup>C</sup>	Purchase <sup>C</sup>
	X <sub>1</sub>	X <sub>2</sub>	S/G (%)	Pn (%)						
1	-1	-1	57/43	3	6.72 <sup>a</sup> ±1.44	6.22 <sup>a</sup> ±1.63	6.23 <sup>cd</sup> ±1.82	5.73 <sup>o</sup> ±2.02	6.28 <sup>bcd</sup> ±1.77	2.97 <sup>o</sup> ±1.22
2	1	-1	43/57	3	7.28 <sup>a</sup> ±1.45	6.65 <sup>a</sup> ±1.68	7.18 <sup>abc</sup> ±1.44	7.43 <sup>a</sup> ±1.33	7.18 <sup>ab</sup> ±1.43	2.02 <sup>a</sup> ±0.97
3	-1	1	57/43	17	7.25 <sup>a</sup> ±1.53	6.22 <sup>a</sup> ±1.74	6.48 <sup>abcd</sup> ±2.00	5.93 <sup>bo</sup> ±2.24	6.52 <sup>abcd</sup> ±1.87	2.58 <sup>abc</sup> ±1.27
4	1	1	43/57	17	6.90 <sup>a</sup> ±1.82	6.05 <sup>a</sup> ±1.70	6.35 <sup>bcd</sup> ±2.05	5.97 <sup>bo</sup> ±2.39	6.18 <sup>cd</sup> ±2.07	2.82 <sup>bo</sup> ±1.17
5	-α	0	60/40	10	6.77 <sup>a</sup> ±1.51	6.40 <sup>a</sup> ±1.66	5.82 <sup>d</sup> ±2.17	5.60 <sup>o</sup> ±2.15	5.90 <sup>d</sup> ±2.07	3.18 <sup>o</sup> ±1.28
6	+α	0	40/60	10	7.20 <sup>a</sup> ±1.36	6.57 <sup>a</sup> ±1.45	7.23 <sup>bc</sup> ±1.70	7.30 <sup>a</sup> ±1.42	7.30 <sup>a</sup> ±1.38	2.07 <sup>a</sup> ±1.02
7	0	-α	50/50	0	7.25 <sup>a</sup> ±1.43	6.40 <sup>a</sup> ±1.56	7.37 <sup>a</sup> ±1.45	7.57 <sup>a</sup> ±1.51	7.22 <sup>a</sup> ±1.71	1.98 <sup>a</sup> ±1.02
8	0	+α	50/50	20	6.95 <sup>a</sup> ±1.77	6.33 <sup>a</sup> ±1.83	6.78 <sup>abcd</sup> ±1.93	5.87 <sup>bo</sup> ±2.23	6.42 <sup>abcd</sup> ±1.95	2.72 <sup>bo</sup> ±1.24
9	0	0	50/50	10	7.23 <sup>a</sup> ±1.61	6.50 <sup>a</sup> ±1.61	7.42 <sup>a</sup> ±1.37	7.18 <sup>a</sup> ±1.80	7.35 <sup>a</sup> ±1.35	1.97 <sup>a</sup> ±1.04
10	0	0	50/50	10	6.92 <sup>a</sup> ±1.75	6.62 <sup>a</sup> ±1.73	7.10 <sup>abc</sup> ±1.53	6.57 <sup>abo</sup> ±1.78	6.87 <sup>abc</sup> ±1.64	2.52 <sup>ab</sup> ±1.20
11	0	0	50/50	10	7.18 <sup>a</sup> ±1.46	6.73 <sup>a</sup> ±1.54	7.17 <sup>abc</sup> ±1.40	6.88 <sup>ab</sup> ±1.60	6.98 <sup>abc</sup> ±1.61	2.20 <sup>ab</sup> ±1.09

<sup>A</sup>  $\alpha = (2^n)^{1/4}$ , where n = number of independent variables, in this case  $\alpha = 1.41$ ; X<sub>1</sub> = Crystal sugar/glucose concentration, X<sub>2</sub> = Pine nut concentration.

<sup>B</sup> Crystal sugar/glucose (S/G); Pine nut (Pn).

<sup>C</sup> The results represent the averages of determinations ± standard deviation. Samples followed by same letters do not differ ( $p > 0.05$ ) by the Tukey test.

## 3. Results and discussion

### 3.1 Pine nut “grits” characterization

The results for moisture, ash, lipid, protein, carbohydrate and dietary fiber obtained in the Pine nut “grits” characterization after 1 month of processing are presented in Table 2, where it can be observed that Pine nut “grits” was rich in carbohydrates and dietary fibers. These results are higher than those found by Gama (2006) which concluded that cooked and roasted Pine nut can present 15-20 % of dietary fiber in its composition. The differences found can be explained by preparation type utilized in this work; Pine nut was cooked milled and roasted at 115 °C.

Table 2: Centesimal composition of Pine nut “grits”.

Trial	Moisture (%)	Ash (%)	Lipid (%)	Protein (%)	Carbohydrate (%)	Dietary fiber (%)
Pine nut “grits” <sup>A</sup>	7.88 ±0.03	2.84 ±0.02	1.23 ±0.07	6.56 ±0.14	89.37 ±0.01	24.44 ±0.85

<sup>A</sup>The results represent the averages of three determinations ± standard deviation.

According to Gutkoski et al. (2007) the oat bran presents between 7.1 and 12.1% of dietary fiber, therefore the present study nutritionally enriched the cereal bars by the substitution of oat bran by Pine nut “grits” (24.44 % of dietary fiber).

Dietary fibers are not considered essential nutrients; nevertheless these have shown a great importance and have attracted interest from experts in nutrition and health. Functional fiber consists of isolated, non-digestible carbohydrates that have beneficial physiologic effects in humans. Furthermore, the dietary reference intake committee set an adequate intake for total fiber in foods of 38 g and 25 g/d for young men and women, respectively, based on intake levels observed to protect against coronary heart disease (Slavin, 2004, Slavin 2005).

### 3.2 Sensory analysis

Table 1 presents the results obtained in the sensory acceptance test (appearance, aroma, flavor, texture and overall acceptance) and in the purchase intention test of the Pine nut cereal bars, conducted with 60 untrained panelists.

All samples, when evaluated with respect to appearance acceptance, had sensory scores exceeding 6, classified between “liked slightly – score 6” and “liked very much – score 8”. These data corroborate with Gutkosky et al. (2007) study. These authors studied the effect of dietary fiber concentration and sugar in cereal bars made of oats with a high dietary fiber content and obtained samples classified between “liked moderately – score 7” and “liked very much – score 8” for appearance acceptance, their samples did not differ statistically.

It was not possible to obtain a response surface for the dependent variable appearance acceptance, due to the coefficient of determination ( $R^2$ ) being less than 0.74 and all trials did not differ statistically ( $p > 0.05$ ). This fact shows no effect on the appearance acceptance caused by the addition of Pine nut “grits”.

Regarding aroma acceptance of cereal bars, all the averages ranged from “liked slightly – score 6” to “liked very much – score 8”. It was not possible to obtain a response surface for the dependent variable aroma acceptance, due to the coefficient of determination ( $R^2$ ) being less than 0.78 and all trials did not differ statistically ( $p > 0.05$ ). Therefore, no effect was observed on the aroma acceptance caused by the addition of Pine nut “grits”.

The mean scores for flavor acceptance ranged from “neither liked nor disliked – score 5” to “liked very much – score 8”. The mathematical model ( $R^2 = 0.88$ ; F test calculated/F value established = 1.40) for the dependent variable flavor acceptance is shown in Eq (1).

$$\text{Flavor} = y_i = 7.23 + 0.71 x_1 - 0.82 x_1^2 - 0.35 x_2 - 0.27 x_2^2 - 0.54 x_1 x_2 \quad (1)$$

It is possible to observe that increasing the concentration of glucose syrup and decreasing the Pine nut concentration the scores of flavor acceptance raised within the ranges studied with S/G, having a more pronounced effect (Figure 1). However, the central points did not differ statistically ( $p > 0.05$ ) of sample 7 (without Pine nut “grits”).

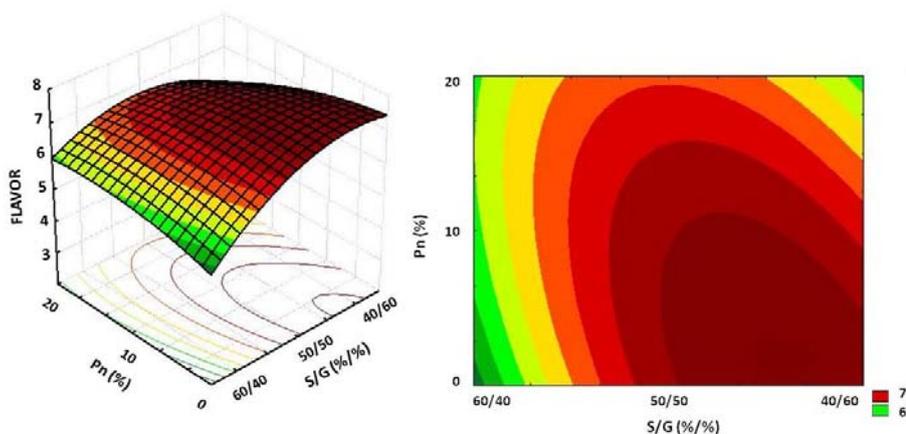


Figure 1 - Response surface and contour curve obtained for the dependent variable flavour acceptance.

where: Pn = Pine nut concentration; S/G = crystal sugar/glucose syrup concentration.

The mean scores for texture acceptance ranged from “neither liked nor disliked” to “liked very much”.

The mathematical model ( $R^2 = 0.89$ ; F test calculated/F value established = 1.64) for the dependent variable texture acceptance is shown in Eq (2).

$$\text{Texture} = y_i = 6.88 + 1.03 x_1 - 0.59 x_1^2 - 0.92 x_2 - 0.32 x_2^2 - 0.83 x_1 x_2 \quad (2)$$

Figure 2 shows the response surfaces and contour curves obtained for texture acceptance, which showed that the effects of the binders (S/G) concentration and the Pine nut concentrations (Pn) presented significant effects. Again, it is possible to observe that increasing the concentration of glucose syrup and decreasing the Pine nut concentration caused an increase in the scores of texture acceptance, within the ranges studied, with S/G having a more pronounced effect. Nevertheless, samples processed with 10% or less Pine nut and the same or greater than 50 % of glucose syrup like binder not differ statistically ( $p > 0.05$ ).

According to Izzo and Niness (2001), the addition of fiber in the formulation increases the hardness of the cereal bars. In addition to the sugars, gelatin and other ingredients such as emulsifiers can be employed in the formulation to reduce the undesirable sensory effects due to the addition of fibers.

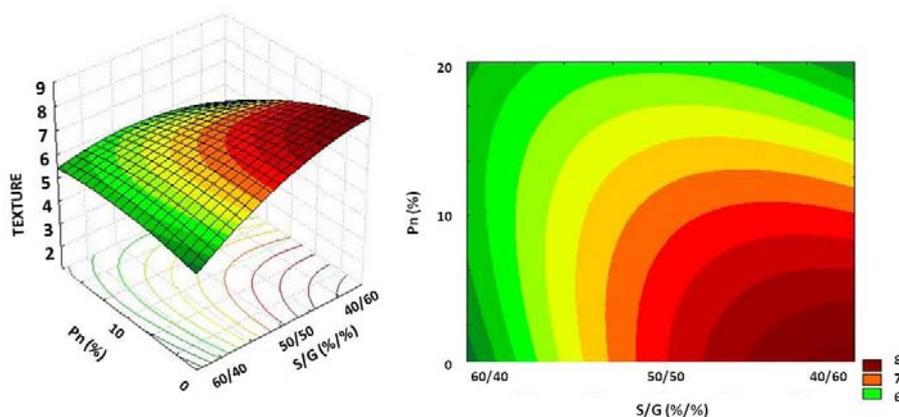


Figure 2 - Response surface and contour curve obtained for the dependent variable flavour acceptance.

The mean scores for overall acceptance ranged from “neither liked nor disliked – score 5” to “liked very much – score 8”. Samples 1, 4 and 5 (in general, with higher concentrations of crystal sugar) statistically differed ( $p \leq 0.05$ ) from the highest score observed (sample 9).

It was not possible to obtain a response surfaces for the dependents variable flavor acceptance and purchase intention, due to the coefficient of determination ( $R^2$ ) being less than 0.84 and the ratio calculated F/tables F being lower than 1 for both, indicating a relevant lack of fit in the analysis of variance of the regressions.

Only the score of sample 5 (sample processed with 60 % crystal sugar) was between 3 and 4 (“would possibly buy/would possibly not buy” and “would probably not buy”), also being the one which presented the lowest scores for flavour, texture and overall acceptance. The other samples had average scores for purchase intention between 1 and 3 (“certainly would buy” and “would possibly buy/would possibly not buy”).

Mourão (2008), studying cashew-plum cereal bars with functional ingredients obtained sensory scores between 5=have doubts and 7=like moderately, such results corroborate with data shown in this study.

Carvalho et al. (2011), evaluating cereal-bars made with almonds of chichá, sapucaia and gurguéia nuts, had lower acceptance for aroma (5.9 to 6.9), the flavor (5.9 to 7.0), texture (6.6 to 7.3) and overall impression (6.2 to 7.1), their results were considered similar to the values presented here. Another relevant study, which also corroborates with this study was present by Peuckert et al. (2010). These authors studied the sensory acceptance of cereal bars added soy protein and *camu-camu*, the values ranging from moderately liked (score 5) and very much liked (score 7) for color, flavor, texture and appearance were observed.

According to the results, the cereal bars produced with 10 % or less of Pine nut concentration and 50 % glucose syrup concentration in binder phase can be considered the best formulations. Therefore, the sample 2 (43/57 % crystal sugar/ glucose syrup and 3 % Pine nut), sample 6 (40/60 % crystal sugar/ glucose syrup and 10 % Pine nut) and the central points (50/50 % Crystal sugar/ glucose syrup and 10 % Pine nut) were considered the best Pine nut cereal bar. Good results also were observed to sample 7, but this sample did not present Pine nut in its composition, thus it was preferred utilized it just to compare the results statistically.

#### 4. Conclusions

The results obtained in this study showed that the addition of Pine nut besides 10 % and more than 50 % crystal sugar caused negative effects on most of the sensory properties of Pine nut cereal bars. However, all samples presented acceptable scores > 5 which were considered good scores. Analyzing the results, the samples 6 and central points were considered the best Pine nut cereal bars. The high acceptance rate obtained by the cereal bars reflects in a large potential purchase of such product. The nutritional appeal because the high amounts of fiber as well as the preservation of the *Araucaria angustifolia* are attractive to consolidates the sale of this cereal bar, especially in the Brazilian market.

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