Scientific
Research

Using Response Surface Methodology and High-Intensity Sweeteners' Positive Synergy to Optimize Peach Nectar Acceptability

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ABSTRACT

Combining the high-intensity sweeteners' positive synergy with response surface methodology (RSM), it is possible to optimize food and beverage products liking, masking undesired sensory attributes and reducing costs and consumers' additives intake (up to 50%). A RSM based on a five-level, two variable central composite rotatable designs (CCRD) was applied, varying aspartame and acesulfame-K concentrations in order to optimize consumers' sensory liking (n = 118) for peach nectar in terms of appearance, aroma, flavor, and overall liking. Statistically valid models were found for flavor and overall liking attributes and highest liking values were found for samples combining the two sweeteners (therefore overcoming acesulfame-K's sensory limitations) in lower concentrations, confirming the synergetic effect and allowing cost reduction and less additives intake, without losing sensory quality.

Keywords: Sweetener Blends; Aspartame; Acesulfame-K; CCRD; RSM; Sensory Evaluation

1. Introduction

Nowadays, consumers expect to obtain pleasure from food and require that this food has good sensory qualities; therefore, food and drink manufacturers are continuously working to improve the sensory quality of foods to respond to consumers' expectations [1]. On the other hand, consumers wish products with low percentage of fats, sugar and calories, in order to increase or maintain their health and well being. Beverage manufacturers have been using sweetener blends instead of single sweeteners in reduced-calorie beverages for some time now, with many successful products well established in the marketplace [2].

The beverage industry frequently uses sweetener blends to overcome the sensory limitations of individual sweeteners [3]. Several additives with the same function, as high-intensity sweeteners, can show synergistic effects, which provide an improved end result and also permit the reduction of the amounts needed for each individual component. That is particularly important when it comes to safe additive levels intake [4]. The synergism of

blends among different sweeteners makes possible the reduction of their individual quantities, turning their use safer and more desirable than the isolated ones. These mixtures, in different proportions, can also allow the improvement of functional and sensorial characteristics desired for a product [5].

Combinations of sweeteners have multiple benefits because they provide synergistic taste enhancement (positive synergy) often associated with a perceived improvement of the taste profile. The use of sweeteners blends makes possible to increase the stability of sweetness and sweetening power, to improve the sweetness quality, to reduce costs and also to increase the low-calorie product choices for the consumer [6,7].

For instance, acesulfame-K in peach nectar was reported with high grades in the sensory attributes bitterness and residual bitterness-unpleasant attributes that may negatively influence its consumers' acceptance [8]. A significant reduction of the non-sweet tastes and after-tastes usually associated with acesulfame-K and cyclamate was observed when they were used in blends [1]. Noble *et al.* [9] reported no non-sweet side tastes for the aspartame-acesulfame-K blend in aqueous solution, and a

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reduction of the lingering sweetness of aspartame was also observed in the fruit-flavored aqueous solutions. The sensory profiles of aspartame, aspartame/acesulfame-K blend and sucralose were the most similar to 10% high-fructose corn syrup [3].

Response surface methodology (RSM) is an important tool in process and product improvement [10] (Altan *et al.*, 2008). RSM consists of a group of mathematical and statistical procedures that can be used to study relationships between one or more responses (dependent variables) and a number of factors (independent variables) [11]. In the last decade, RSM has been successfully used to adjust formulations [12-22] and process conditions [10,23-31], reducing the number of trials and providing multiple regression approach to achieve optimization [18]. In addition, for each product, the sweeteners' potencies and synergies are unique, because they depend on the dispersion matrix in which sweeteners are found. Thus one must evaluate the replacement of sucrose by sweeteners in each food separately [32]. However, to the best of our knowledge, there are no studies using response surface methodology to explore the positive synergistic power of high-intensity sweeteners, optimizing the liking of a food or beverage product in relation to its sweetener blend proportions.

Therefore, the objective of the current exploratory study was to optimize peach nectar liking with regards to proportions of aspartame/acesulfame-K used in blends, using response surface methodology (RSM) and aiming to obtain the product with the most possible sensory appealing. As a consequence, it would be possible to reduce costs and additives intake with less high-intensity sweetener ingredients.

2. Material and Methods

2.1. Materials

Peach nectar samples were prepared with unsweetened concentrated juice (Del Valle, Americana-SP, Brazil). The samples were sweetened with sucrose and high-intensity sweeteners aspartame (Nutrasweet, São Paulo-SP, Brazil) and acesulfame-K (Steviafarma, Maringá-PR, Brazil), and their blends.

2.2. Sample Preparation

Samples were prepared in laboratory following the procedure used by the manufacturer:

1) Hydration of the concentrated base, according to the manufacturer's recommendations: each 181.46 g of concentrated base should be hydrated up to 1000 g, total solution.

2) Homogenizing at 2500 psi the mixture.

3) Following peach nectar producer' instructions, pas-

teurization in a microwave oven to 98.0˚C, hot-filling in bottles, cooling in water (20˚C) to room temperature. A maximum of 1 L was pasteurized at a time, for 6 minutes, in order to avoid the loss of aroma volatiles as much as possible. More information on sample preparation can be provided contacting authors and under peach nectar manufacturer approval.

2.3. Response Surface Methodology

Response surface methodology (RSM) was based on a five-level ($-\alpha$, -1, 0, 1, and α), two variable central composite rotatable design (CCRD). The independent variables were aspartame (X_1) and acesulfame-K (X_2) concentrations and the dependent variable was consumer liking (relating to appearance, aroma, flavor, and overall liking). Actual values of variation levels and experimental design for the experiment are shown in **Table 1** [8,33]. Expecting possibly synergistic effect, the central point for each high-intensity sweetener has been chosen using a value (0.04%) smaller than aspartame (0.054%) or acesulfame-K (0.053%) when sweetening peach nectar separately, both sweetness equivalent to 10% sucrose (the ideal sweetness) [33].

The results were analyzed by a multiple linear regression method which describes the effects of variables in first- and second-order polynomial models. Experimental data were fitted to the selected models and regression coefficients obtained.

2.4. Consumer Testing

Participants ("ready-to-drink" fruit nectars consumers—

Table 1. Central composite rotatable design with coded levels and actual variable levels.

Samples	Coded levels		Actual levels	
	X_1	X_2	Aspartame (%)	Acesulfame-K (%)
1	−1	−1	0.0116	0.0116
2	1	−1	0.0684	0.0116
3	−1	1	0.0116	0.0684
4	1	1	0.0684	0.0684
5	−1.41	0	0	0.04
6	1.41	0	0.08	0.04
7	0	−1.41	0.04	0
8	0	1.41	0.04	0.08
9	0	0	0.04	0.04
10	0	0	0.04	0.04
11	0	0	0.04	0.04

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at least once a week, recruited amongst faculty staff and students) evaluated peach nectar samples to determine liking of appearance, aroma, flavor, and overall liking. A total of 118 consumers, 52 male and 66 female (44% male and 56% female) with age varying from 18 to 54 years old, have participated in this study. Samples, presented under refrigeration (6˚C), covered the aspartame/acesulfame-K blends from the central composite rotatable design (CCRD) (see previous section) and samples sweetened only with aspartame (0.054%), acesulfame-K (0.053%), or sucrose (10%) (sucrose at ideal sweetness and aspartame and acesulfame-K at equi-sweet concentrations in peach nectar, see more details in Cardoso & Bolini [33].

Consumer affective testing was carried out using a 9-cm unstructured line scale with anchors "dislike extremely" and "like extremely". According to Greene *et al.* [34], there is evidence of no decrease in sensitivity and reliability with regards to consumer perception using line scales instead of 9-point hedonic scale. Samples (20 mL) coded with three-digit numbers were presented monadically in a balanced block design [35] to 118 consumers on disposable plastic glasses. Sensory tests were carried out in individual air-conditioned booths. Taste-free water was provided for palate cleansing. In addition, there was a 2-minute break after each sample and a 10-minute break after the seventh sample to minimize sensory fatigue. Consumers' decisions were based solely on the sensory characteristics of the samples, since product information and formulation were not provided.

2.5. Statistical Analyses

The liking results were analyzed by ANOVA, using two factors (consumer-random, and sample-fixed), and Tukey's HSD average test. ANOVA, Tukey's HSD average test and response surface regressions of liking data (including ANOVA of the model) were performed using the software STATISTICA 7.0 (StatSoft, Inc.). Internal preference mapping [36] based on overall liking data was also performed.

3. Results and Discussion

Table 2 shows the results for flavor and overall liking for the central composite rotatable design (samples 1 to 11) and for the other three peach nectar samples sweetened with only one sweetener (samples 12 to 14). Results for appearance and aroma will not be presented since it was not possible to fit statistically valid models for these sensory liking attributes. Importantly, **Table 2** shows that consumers did not discriminate on appearance and very low discrimination on aroma, which was confirmed by

Table 2. Central composite rotatable design (samples 1 to 11), including three more samples sweetened with only one sweetener (samples 12 to 14).

Samples	% Aspartame (coded levels)	% Acesulfame-K (coded levels)	Sucrose (%)	Apperance (SD)	Aroma (SD)	Flavor (SD)	Overall liking[a] (SD)
1	0.0116 (−1)	0.0116 (−1)	-	6.87[a] (1.77)	6.53[a] (1.87)	6.31[a,b] (1.63)	6.47[a,b] (1.52)
2	0.0684 (1)	0.0116 (−1)	-	7.02[a] (1.68)	6.46[a,b] (1.91)	5.15[c,d] (2.41)	5.77[b,c,d] (1.97)
3	0.0116 (−1)	0.0684 (1)	-	7.08[a] (1.64)	6.46[a] (1.72)	4.90[d,e] (2.47)	5.63[c,d,e] (2.08)
4	0.0684 (1)	0.0684 (1)	-	6.96[a] (1.68)	6.38[a,b] (1.85)	4.12[e,f] (2.63)	4.96[e] (2.15)
5	0 (−1.41)	0.04 (0)	-	7.02[a] (1.66)	6.49[a] (1.79)	5.92[b,c] (1.89)	6.19[a,b,c] (1.76)
6	0.08 (1.41)	0.04 (0)	-	7.04[a] (1.68)	6.25[a,b] (1.94)	4.42[d,e] (2.36)	5.21[d,e] (2.15)
7	0.04 (0)	0 (−1.41)	-	7.03[a] (1.69)	6.58[a] (1.82)	6.36[a,b] (2.07)	6.44[a,b] (1.89)
8	0.04 (0)	0.08 (1.41)	-	6.75[a] (1.96)	6.51[a] (2.08)	4.32[d,e] (2.68)	5.21[d,e] (2.29)
9	0.04 (0)	0.04 (0)	-	6.47[a] (1.99)	6.31[a,b] (2.04)	4.83[d,e] (2.46)	5.37[d,e] (2.16)
10	0.04 (0)	0.04 (0)	-	6.73[a] (1.87)	6.42[a,b] (1.97)	5.01[d,e] (2.36)	5.62[c,d,e] (2.06)
11	0.04 (0)	0.04 (0)	-	6.66[a] (2.03)	6.42[a,b] (2.04)	4.86[d,e] (2.52)	5.50[c,d,e] (2.09)
12	-	-	10.0	6.78[a] (1.73)	6.39[a,b] (1.92)	6.09[a] (1.79)	6.75[a] (1.70)
13	0.054	-	-	6.58[a] (1.86)	6.01[a,b] (2.02)	5.22[c,d] (2.26)	5.48[c,d,e] (1.87)
14	-	0.053	-	6.46[a] (1.78)	5.70[b] (2.03)	3.33[f] (2.33)	3.94[f] (2.08)
MSD	-	-	-	0.656	0.763	0.909	0.778

[a]Different letters on a column indicate that means are different (Tukey's HSD average test, p < 0.05).

their not statistically valid models. In addition, a high Pearson correlation (0.88) was found for consumer's individual dataset between flavor and overall appearance.

Dependent variables flavor and overall liking were analyzed by multivariate regressions and significance of the models was tested [37] by Analysis of Variance (ANOVA), providing *F*-test values (72.3 for flavor and 101.2 for overall liking) higher than the critical value of 4.53 (d.f. = 4; $p < 0.001$; R^2 of 0.98 for flavor and 0.99 for overall liking). Thus, it was possible to present statistically valid models (Equations (1) and (2)) for these sensory liking attributes (only statistically significant—p < 0.05-model terms are shown):

$$\text{Flavor} = 4.90 - 0.509X_1 - 0.665X_2 + 0.188X_2^2$$

$$\text{p-values} : < 0.000 \text{ (Intercept)}; < 0.000\left(X_1\right); \quad (1)$$

$$< 0.000\left(X_2\right); 0.020\left(X_2^2\right)$$

$$\text{Overall liking} = 5.50 - 0.344X_1 - 0.424X_2 + 0.149X_2^2$$

$$\text{p-values} : < 0.000 \text{ (Intercept)}; < 0.000\left(X_1\right); \quad (2)$$

$$< 0.000\left(X_2\right); 0.009\left(X_2^2\right)$$

(where X_1 is percentage of aspartame and X_2 is percentage of acesulfame-K).

A better understanding of the influence of the sensory attributes on the peach nectar samples sweetened with high-intensity sweetener blends would be achieved applying a descriptive technique. A previous investigation has studied the sensory descriptive profile of peach nectar sweetened with high-intensity sweeteners applied separately and might bring initial highlights [8]. For instance, Equations (1) and (2) showed that the aspartame concentration decreased flavor and overall liking less than the acesulfame-K concentration did in the linear model terms, which is consistent with descriptive studies that reported bitterness (an undesired characteristic in

peach nectar) when adding acesulfame-K to peach nectar [8]. However, acesulfame-K concentration increased flavor and overall liking in the quadratic model terms.

In order to define sweeteners' influence on peach nectars' liking, response surfaces and contour diagrams were also generated for flavor and overall liking (**Figures 1** and **2**), showing higher liking values on the lowest sweeteners concentrations regions.

Samples 1, 5, and 7 (**Table 2**), containing lower sweetener blend concentrations, presented liking at least equivalent to liking of peach nectar samples with only aspartame (sample 13) or acesulfame-K (sample 14) at concentrations equivalent to sucrose at ideal sweetness (10%) [33]. It is important to highlight that sample 1 (with 0.0232% of sweeteners, combining 0.0116% of aspartame and 0.0116% of acesulfame-K) presented overall liking statistically higher (p < 0.05) than the samples sweetened only with aspartame or acesulfame-K in higher concentrations, equivalent to the ideal sweetness (0.054% and 0.053%, respectively), which confirms the positive synergy between the high-intensity sweeteners and brings sweetener addition and intake reductions higher than 50%. Even considering that aspartame presents less sensory disadvantages than acesulfame-K [38], using a blend of both is strongly economically interesting —without losing sensory quality as proved in our study—to reduce the total amount of added sweeteners.

Figure 3 shows internal preference mapping with the 118 consumers. Preference mapping is in accordance with Tukey's results, showing that most consumers preferred peach nectar samples 1, 5, 7, and 12 (see **Table 2**) and that sample 14 (with only acesulfame-K at 0.053%) was the least preferred, demonstrating once again acesulfame-K's sensory limitations. As an exploratory technique, the internal preference mapping confirmed patterns found with ANOVA and Tukey's HSD average test.
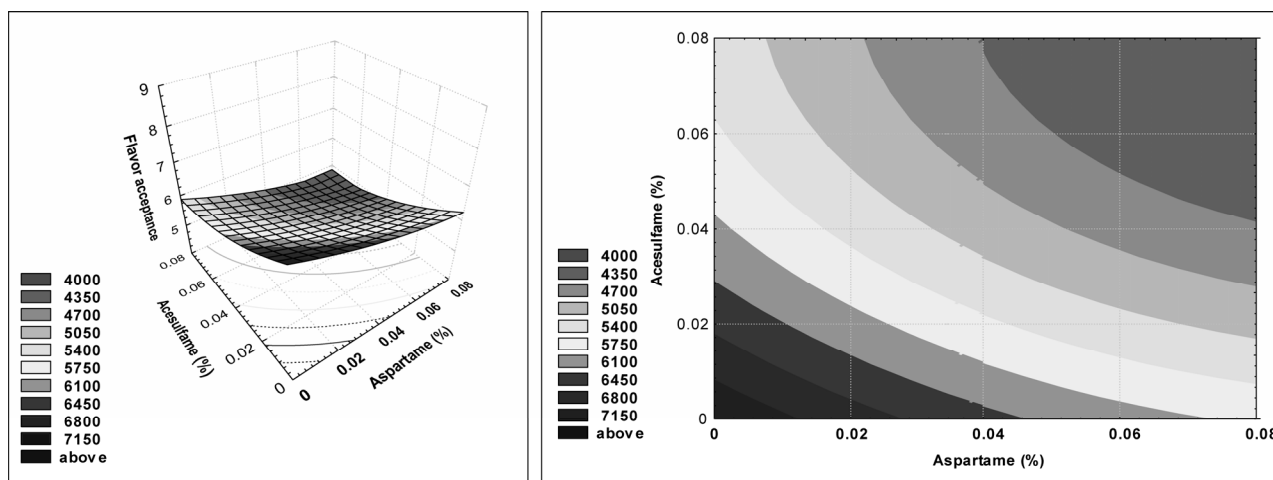

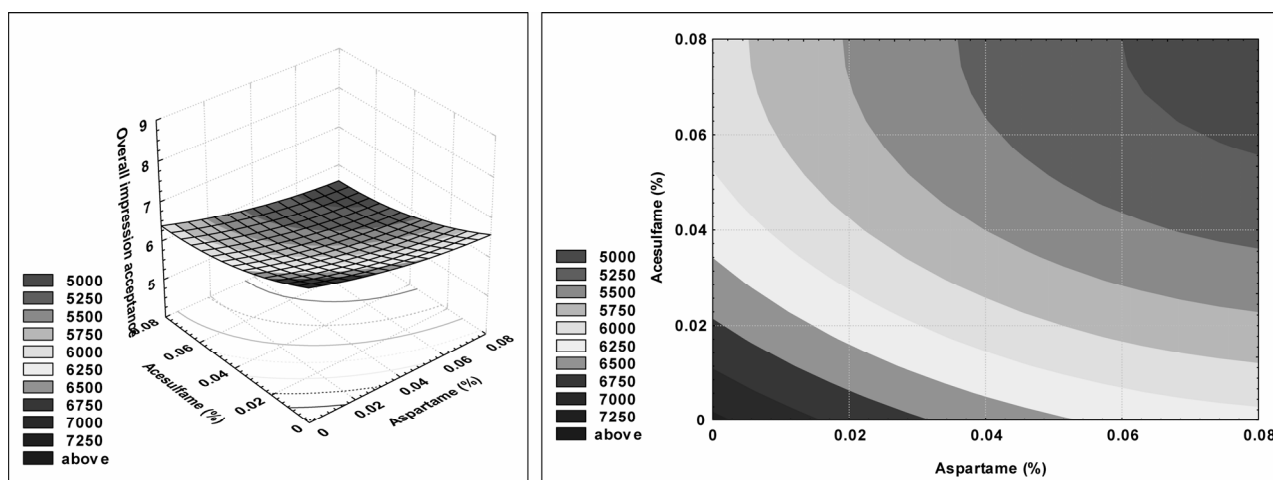**Figure 1. Response surface and contour diagram for flavor acceptability of peach nectar.**

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Figure 2. Response surface and contour diagram for overall liking acceptability of peach nectar.
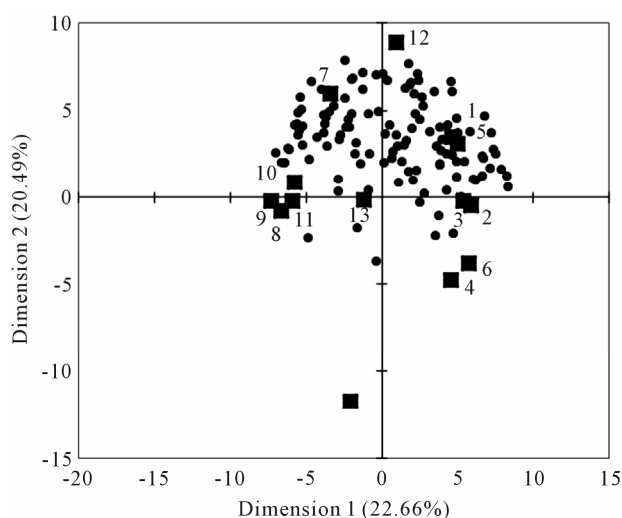

Figure 3. Internal preference mapping indicating the position of the consumers (n = 118) and the peach nectar samples (see Table 2).

Clusters were found but any statistical difference was found between them, using all collected variables (data not shown).

All differences showed in mean separation tests and suggested by the internal preference mapping might be explained by descriptive attributes. From other studies with peach nectar using aspartame and acesulfame-K separately, sensory attributes bitterness and residual bitterness were found, especially with acesulfame-K and other off-flavors (e.g. metallic) might also be involved. Further sensory descriptive analyses on samples with sweetener blends are needed to bring new highlights on consumers' preferences. It is important to highlight as a possible limitation of this study that these 118 consumers could not represent the Brazilian population socio-demographics, therefore the results about the influence of the sweeteners on consumer liking cannot be generalized.

4. Conclusion

Results showed the importance of the synergistic effect of aspartame/acesulfame-K blend on cost reduction and healthy (up to 50% less additives intake—with 0.0232% of sweeteners, combining 0.0116% of aspartame and 0.0116% of acesulfame-K compared to 0.054% or 0.053 when used separately). Even though acesulfame-K presents sensory limitations, its use combined with aspartame can lead to the positive synergy, reducing costs, additives intake and increasing liking. In this sense, response surface methodology (RSM) showed to be a powerful technique, reducing the number of trials. That is extremely important especially considering that our results are applicable for the studied high-intensity sweeteners—aspartame and acesulfame-K—in peach nectar, given that the sweetness power depends on the substance and on the matrix where the substance is inserted. Same approach can be used during product development and improvement to optimize liking of food and beverages that contain sweeteners, taking advantage of their positive synergy and reducing costs and consumers' additives intake.

5. Acknowledgements

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