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SODIUM CHLORIDE REDUCTION IN BREAD: CHEMICAL AND SENSORY CHARACTERIZATION AMONG TEENAGERS

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ABSTRACT

The research's purpose was to verify the sensory acceptability of bread made with different levels of sodium chloride (NaCl), potassium chloride (KCl) and monosodium glutamate (MSG) among teenagers and evaluate the product's chemical composition. In Step 1, five samples were prepared with NaCl and KCl (0 to 0.60%). Step 2 used the same percentages that were used in Step 1, with 0.30% of MSG, though. The samples with KCl (0.45% and 0.60%) were the less accepted for flavour in comparison with the standard formulation, while the one with 0.60% of KCl got lower scores for aftertaste, overall acceptance and purchase intent in comparison with the standard formulation. The addition of MSG promotes the products' acceptability increase, enabling the addition of up to 0.45% of KCl. The bread presented the following levels of moisture, ash, protein, lipid, carbohydrate and energy, respectively: 24.47 $g.100g^{-1}$, 0.91 $g.100g^{-1}$, 8.91 $g.100g^{-1}$, 8.50 $g.100g^{-1}$, 57.18 $g.100g^{-1}$, 340.73 $kcal.100g^{-1}$. The formulation F4, with greater addition level of KCl and whose sensory acceptance was similar to the standard formulation, presented contents of 338.70 and 375.60 mg/day of sodium (Setps 1 and 2, respectively). We can infer that it is possible to restrain the NaCl level in breads, especially through the joint use of NaCl (0.15%), KCl (0.45%) and MSG (0.30%) to improve their acceptability among teenagers.

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INTRODUCTION

Sodium chloride (NaCl), or salt, is chemically constituted by sodium (40%) and chloride (60%) and it is the greatest sodium source in our diet (90%) (Lopes *et al.*, 2017). In our body, sodium is essential for the maintenance of several physiological functions, such as nervous transmissions, muscle contractions, blood pressure maintenance and fluid balance. It may be naturally found in several *in natura* foods, such as maize, wild cabbages, broccolis, cucumbers, beets, carrots and passion fruits. As for chloride, it is usually linked to another element, such as sodium and potassium and it is naturally found in water. Chloride is not toxic for human beings and, combined with sodium, maintains the fluid balance in our organism (Stanley *et al.*, 2017). Although the NaCl level found in nature is enough to replace our daily metabolic losses, nowadays we can see a disturbing intake of this mineral by people several ages. According to the World Health Organization (WHO), around 9 to 12 g of NaCl are consumed daily, while the recommended amount is a maximum of 5 g/day (2.000 mg of sodium) for adults. This amount is even smaller for children and teenagers (1.5 g of NaCl/day), since they belong to a part of the population that is more vulnerable to several pathologies (WHO, 2011). We must highlight that an intake reduction of this mineral can reduce the development risk of cardiac diseases in adulthood (Bibbins-Domingo *et al.*, 2010), since its excessive intake is directly linked to blood pressure increase and consequently to systemic hypertension

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(He *et al*., 2011). Systemic hypertension is defined as a clinical condition in which the systolic pressure is equal or greater than 140 mmHg and/or a diastolic pressure equal or greater than 90 mmHg (Rolim *et al*., 2017). Simultaneously, it may compromise our blood vessels, heart, brain, eyes and kidneys (Widimsky, 2016). In Brazil, systemic hypertension cases affect around 20.8 to 22% of the adult population (Theme Filha *et al*., 2015) and from 6.2 to 10.5% of the teenagers (Magliano *et al*., 2013). Among the factors related to systemic hypertension development are obesity, a sedentary lifestyle, physical inactivity and improper dietary habits (Almeida *et al*., 2017). Considering this aspect, several public policies have being developed to restrain the NaCl intake by the population (He *et al*., 2011).

Considering a world tendency for a lower salt intake, the Brazilian government, since 2010, with the Ministry of Health, the National Sanitary Surveillance Agency, and other institutions and private organizations have been promoting actions to change this situation. Among them are a greater offer of healthy foods and guidance activities for the population, health professionals and food handlers on a proper reading of the nutritional label (Nilson *et al*., 2012). Additionally, the subjects of encouraging the intake of *in natura* foods and new formulations of processed foods are receiving more attention, with the purpose of adding less fat, sugar and sodium in commercial products (Monteiro *et al*., 2011) because 77% of the sodium of our diet comes from processed foods (Stall, 2013). Baked goods, such as breads, for example, represent 30% of this amount (Lynch *et al*., 2009).

George Institute for Global Health's (2010) study, which evaluated the contents of breads in Australia, reported an average of 436 mg of sodium/100 g of the product. In Brazil, a slightly higher amount were reported: nearly 475 mg of sodium/100 g (Brasil, 2013). Therefore, considering the elevated world and national bread consumption, new strategies are fundamental to reduce the amount of sodium in these foods to cooperate for a lesser risk of non-communicable chronic diseases (Lynch *et al.,* 2009).

Breads are defined as products made by the cooking of dough, which may be fermented or not, and can be made with several kinds of flour and water, among other ingredients (Lopes *et al*., 2017). In this kind of food, salt is a very important ingredient, since it contributes for its flavour and interferes in its technological properties, such as in the development of its gluten network and to strengthen the dough's viscoelastic properties. It also contributes to inhibit the yeast's activity in fermentation and to control the water's activity in the finished product (Mueller *et al*., 2016). Therefore, reducing the amount of sodium in breads is a great challenge for the food industry. One of the strategies that have been studied to replace NaCl in breads is the use of potassium chloride (KCl), since its action on fermentation and on the dough's rheological properties is very similar to NaCl (Mueller *et al*., 2016). However, we must consider its negative effects on foods, especially the resulting colour, texture and metallic, bitter and/or astringent taste that the addition of KCl may cause (Aliño *et al*., 2009). In spite of that, these effects may be mitigated with the use of flavour enhancers, such as monosodium glutamate (MSG) (Henney *et al*., 2010), which is the glutamic acid's sodic salt-an non-essential amino acid naturally found in protein food sources, such as wheat (Masic *et al*., 2017). This salt is often used in products to enhance the salty tastes naturally present in foods (Rangan and Barceloux, 2009).

One of the fundamental tools to evaluate products that present changes in their basic formulation is the sensory analyses. This technique's purpose is to identify the acceptance or rejection of a given food product, through the study of its sensory features, using specific methods that test the human senses (Liem, 2017). Another evaluation of great importance in foods are chemical analyses, which help us to identify possible frauds or falsifications, physical-chemical and sensory changes, quantification of food additives, adequacy to the Standard of Identity and Quality and nutritional label (Pedersen *et al*., 2016), which help us to ensure a product's quality. Therefore, it is fundamental to apply these analyses to consider a possible market commercialization of new products. The purpose hereof was to verify the sensory acceptability of bread made with different levels of NaCl, KCl and MSG among teenagers and evaluate the product's chemical composition.

MATERIALS AND METHODS

Raw material

The ingredients that were used in the formulations were bought in supermarkets of the city of Guarapuava, Paraná, Brazil. However, the salts were donated by partner companies and had the following classification: NaCl (for analysis) - molecular weight of 58.44; KCl (for analysis) - molecular weight of 74.55. The MSG used in the research was a commercial product, containing 12.300 mg of sodium/100 g.

Formulations

The following ingredients were used for the breads' basic formulation: wheat flour (49.55%), lukewarm water (23.78%), oil (11.30%), sugar (2.68) and biological backing powder (0.99%). In Step 1, five bread formulations were made with different NaCl and KCl levels, which were: F1 (0.60% NaCl - 100%), F2 (0.45% NaCl - 75% and 0.15% KCl - 25%), F3 (0.30% NaCl - 50% and 0.30% KCl - 50%), F4 (0.15% NaCl - 25% and 0.45% KCl - 75%) and F5 (0.60% KCl - 100%). In Step 2, we used the same salt addition percentages used in Step 1, but each sample received MSG in the percentage of 0.30%, totalizing five formulations. The salt addition levels established in Steps 1 and 2 were defined through preliminary sensory tests made with the product.

To prepare the formulations, initially, the backing powder was dissolved in lukewarm water (80 °C) and then we added oil, sugar and salt. Then, we progressively added wheat flour to obtain a consistent dough. The breads were kneaded and placed on aluminum baking pans (21.5 x 10 x 5 cm), previously greased with oil and covered with wheat flour and baked in a preheated (220 °C) domestic oven (Atlas®, Brazil) for approximately 40 minutes.

Sensory Analysis

Two hundred and forty untrained teenagers, enrolled in public schools of Guarapuava, Paraná, took part in the sensory analysis (60 subjects in each one of the four sensory tests that were applied). The consumers comprised teenagers of both genders between 14 and 17 years old. The sensory evaluation was made in a classroom, where cabin-like individual booths were arranged, under natural light. The evaluated features were appearance, aroma, flavour, aftertaste, texture, colour and overall acceptance. We used a 9-point structured hedonic scale, varying from dislike extremely (1) until like extremely (9). In purchase intent test, a 5-point attitude structured scale was used, varying from 1 (definitely would not buy it) to 5 (definitely would definitely buy it) (Meilgaard *et al.*, 2016). Each sample was served to consumers in white plates coded with randomly selected 3-digit numbers in monadic form and using balanced design (Macfe *et al.*, 1989). Sensory evaluations were performed by consumers under fluorescence lighting. After consuming each sample, consumer was instructed to drink water for palate cleansing. Samples were evaluated in triplicate in separate session.

A ranking test was applied to compare the samples' differences in the specific feature of their salty flavor, since the breads' sodium was reduced. The consumers classified the samples in a salty flavor ascending order, from less salty to more salty (Meilgaard *et al.*, 2016). The sensory Acceptability Index (AI) was calculated by multiplying the average score reported by consumers to the product by 100, dividing the result by the maximum average score given to the product within the hedonic scale for 9.0 points.

Chemical composition

The chemical composition was evaluated in triplicate in the bread standard formulation. All results were expressed in weight base. The moisture and ash levels were determined in oven (105 °C) and muffle (550 °C) (AOAC, 2011), respectively. The lipid contents were assessed by the cold extraction method (Bligh and Dyer, 1959). The protein quantification was made through the sample's total nitrogen level, Kjeldahl method (semimicro level with a nitrogen conversion factor of 6.25) (AOAC, 2011). The carbohydrate level was theoretically calculated (by difference) in the triplicates' results, according to the formula: *% Carbohydrate = 100 – (% moisture + % protein + % lipid + % ash + % dietary fiber*). For the total caloric theoretical calculation, we used the conversion values for lipids (8.37 kcal/g), proteins (3.87 kcal/g) and carbohydrates (4.11 kcal/g) (Merrill and Watt, 1973). The chemical composition related to the sodium and potassium levels was evaluated in triplicate in the five bread formulations (Steps 1 and 2). The samples were initially digested with HNO_3 and H_2O_2. The quantification was determined by optical emission spectrometry with inductively coupled plasma (ICP OES) (Thermo Fisher Scientific®, iCAP 6300 Duo model, England). The Daily Reference Values (DRV) was calculated for 50 g of the sample (2 slices), based

Table 1. Sensory scores (mean ± standard deviation) and acceptability index (AI) of the bread formulations with the addition of several levels of sodium chloride (NaCl) and potassium chloride (KCl), and with (Step 1) or without (Step 2) addition of monosodium glutamate

Parameter	F1	F2	F3	F4	F5
Step 1					
Appearance	7.55±0.15^{a}	7.45±0.17^{a}	6.85±0.19^{a}	7.22±0.21^{a}	7.00±0.33^{a}
AI (%)	83.89	82.78	76.11	80.22	77.78
Aroma	7.37±0.16^{a}	7.05±0.18^{a}	6.93±0.21^{a}	7.38±0.16^{a}	7.23±0.19^{a}
AI (%)	81.89	78.33	77.00	82.00	80.33
Flavour	7.22±0.25^{a}	6.88±0.22^{a}	6.25±0.27^{ab}	5.32±0.36^{bc}	4.98±0.37^{c}
AI (%)	80.22	76.44	69.44	59.11	55.33
Aftertaste	6.50±0.27^{a}	5.55±0.32^{ab}	5.40±0.33^{ab}	5.42±0.34^{ab}	4.92±0.35^{b}
AI (%)	72.22	61.67	60.00	60.22	54.67
Texture	6.32±0.28^{a}	5.83±0.28^{a}	5.47±0.30^{a}	5.75±0.29^{a}	5.57±0.32^{a}
AI (%)	70.22	64.78	60.78	63.89	61.89
Colour	7.32±0.24^{a}	7.02±0.28^{a}	6.67±0.28^{a}	6.78±0.30^{a}	6.50±0.31^{a}
AI (%)	81.33	78.00	74.11	75.33	72.22
Overall acceptance	6.92±0.18^{a}	6.85±0.12^{a}	6.90±0.18^{a}	6.40±0.26^{ab}	6.05±0.30^{b}
AI (%)	76.89	76.11	76.67	71.11	67.22
Purchase intent	3.73±0.15^{a}	3.40±0.17^{a}	3.22±0.15^{ab}	3.23±0.19^{ab}	2.65±0.19^{b}
AI (%)	74.60	68.00	64.40	64.60	53.00
Notes sum*	256^{a}	217^{b}	213^{b}	112^{c}	102^{c}
Step 2					
Appearance	7.20±0.20^{a}	7.08±0.20^{a}	6.76±0.19^{a}	6.83±0.20^{a}	6.76±0.21^{a}
AI (%)	80.00	78.67	75.11	75.89	75.11
Aroma	7.24±0.15^{a}	7.05±0.14^{a}	7.02±0.18^{a}	7.09±0.17^{a}	7.21±0.18^{a}
AI (%)	80.44	78.33	78.00	78.78	80.11
Flavour	7.46±0.18^{a}	7.30±0.17^{a}	6.96±0.17^{a}	6.85±0.23^{a}	6.72±0.23^{b}
AI (%)	82.89	81.11	77.33	76.11	74.67
Aftertaste	6.33±0.24^{a}	5.86±0.30^{a}	5.65±0.21^{a}	6.23±0.26^{a}	5.66±0.26^{a}
AI (%)	70.33	65.11	62.78	69.22	62.89
Texture	6.53±0.22^{a}	6.28±0.21^{a}	6.22±0.22^{a}	6.17±0.29^{a}	6.00±0.24^{a}
AI (%)	72.56	69.78	69.11	68.56	66.67
Colour	7.35±0.20^{a}	7.09±0.20^{a}	7.07±0.21^{a}	7.07±0.21^{a}	6.77±0.20^{a}
AI (%)	81.66	78.78	78.56	78.56	75.22
Overall acceptance	7.09±0.20^{a}	7.26±0.18^{a}	6.64±0.17^{a}	6.30±0.15^{a}	6.69±0.09^{b}
AI (%)	78.78	80.67	73.78	70.00	74.33
Purchase intent	3.79±0.13^{a}	3.79±0.14^{a}	3.40±0.13^{a}	3.36±0.13^{a}	2.71±0.15^{b}
AI (%)	75.80	75.80	68.00	67.20	54.20
Notes sum*	226^{a}	202^{ab}	178^{bc}	150^{bc}	149^{c}

Distinct letters in row indicate significant difference by Tukey's test ($p<0.05$); Values are mean of three replicates. Step 1: F1 (0.60% NaCl), F2 (0.45% NaCl and 0.15% KCl), F3 (0.30% NaCl and 0.30% KCl), F4 (0.15% NaCl and 0.45% KCl) and F5 (0.60% KCl); Step 2: Addition of 0.30% monosodium glutamate in each of the formulations; *Least significant difference (LSD) of ≥ 34 between the samples present statistic difference ($p<0.05$), according to Christensen's Table (Christensen *et al.*, 2006) for 60 judgments and 5 samples.

on the mean values recommended for teenagers (14 to 17 years old) (DRI, 2005), resulting in: 1.901 kcal/day, 259 g/day of carbohydrate, 66.50 g/day of protein, 68.70 g/day of lipid and 12.80 g/day of dietary fiber. The DRV for the micronutrients sodium and potassium was calculated for 2.000 mg/day of sodium (WHO, 2011) and 2.120 mg/day of potassium (DRI, 2005).

Statistical analysis

The data were evaluated through an analysis of variance (ANOVA), using Tukey's test. In the sensory analysis, we also used the Friedman test and Christensen' Table (Christensen *et al*., 2006), which indicated a least significant difference (LSD) between the samples and the number of opinions obtained in the ranking test. All tests were analyzed with a 5% level of significance, with the assistance of the Statgraphics plus® software, version 5.1.

Ethical issues

This paper was approved by the Research Ethics Committee of Midwest State University, ruling no. 345.569/2013. The following factors were considered as exclusion criteria: subjects over 17 and under 14 years old, not being a student of the evaluated school, or fail to deliver a Term of Free and Clarified Consent signed by his/hers legal guardian.

Table 2. Physicochemical composition of sodium (Na) and potassium (K), and Daily Reference Values (DRV)* (average portion of 50 grams) of the bread formulations'

	F1	DRV (%)*	F2	DRV (%)*	F3	DRV (%)*	F4	DRV (%)*	F5	DRV (%)*
Step 1										
Na (mg.100g^{-1})	248.46^{a}	6.21	188.35^{b}	4.71	128.54^{c}	3.21	68.36^{d}	1.71	8.50^{e}	0.21
% reduction			24.19		48.26		72.49		96.58	
K (mg.100g^{-1})	81.07^{e}	1.91	159.60^{d}	3.76	238.12^{c}	5.62	316.65^{b}	7.47	395.17^{a}	9.32
% increase			101.62		200.83		300.03		399.23	
Step 2										
Na (mg.100g^{-1})	285.36^{a}	7.13	225.31^{b}	5.63	135.28^{c}	3.38	105.22^{d}	2.63	45.27^{e}	1.13
% increase/reduction	14.85$^{\mu}$		9.32$^{\beta}$		45.55$^{\beta}$		57.65$^{\beta}$		81.78$^{\beta}$	

Distinct letters in row indicate significant difference by Tukey's test ($p<0.05$); Values are mean of three replicates; *DRV: nutrients evaluated by the DRI mean (DRI, 2005); $^{\mu}$% increase in comparison with F1 (Step 1); $^{\beta}$% reduction in comparison with F1 (Step 1); Step 1: F1 (0.60% NaCl), F2 (0.45% NaCl and 0.15% KCl), F3 (0.30% NaCl and 0.30% KCl), F4 (0.15% NaCl and 0.45% KCl) and F5 (0.60% KCl); Step 2: Addition of 0.30% monosodium glutamate in each of the formulations.

RESULTS AND DISCUSSION

Sensory analysis

The sensory test results of the bread made with a NaCl level reduction (Step 1) and MSG addition (Step 2) are described in Table 1. There was no significant difference ($p>0.05$) between Step 1's formulations for the features appearance, aroma, texture and colour. Similar results, among adults, were seen by Mueller *et al*. (2016), who evaluated the acceptability of pizza dough with a substitution of up to 25% of NaCl for KCl (30%). This effect may be explained due to the fact that sodium has little or no influence on features like the product's appearance, aroma and colour (Fouladkhah *et al*., 2015). Lower scores were attributed for F4 and F5 regarding flavour (Step 1) in comparison with F1 and F2. Also, F5 had lower scores than F3 ($p<0.05$). Similar results were seen by Mueller *et al*. (2016), in which the pizza crust with a 30% substitution of NaCl for KCl had an acceptance similar to the standard formulation. For aftertaste, greater scores were registered for F1 in comparison with F5, without a significant difference between all other formulations ($p>0.05$).

Generally, a total substitution of NaCl by KCl is not the best technological choice for baked goods. In spite of the fact that potassium performs the same function of sodium in the activation of baking powder and on the rheological properties of the dough, this mineral may negatively interfere in flavour, resulting in a metallic taste in substitution levels above 10-20%. The salty taste is caused by cation ions (positive charge), which can be inhibited by anions (negative charge). However, chloride (anion) has a low inhibiting power and it is exempt of aftertaste, which gives to NaCl an intense salty taste. As for potassium, its bitter aftertaste is related to the diameter sum of the generated ions; therefore, the less we add KCl to the product, the less its diameter is and the fainter its bitter sensation will be. Considering this aspect, these minerals' combination is the best strategy for their use in foods (Israr *et al*., 2016), which was confirmed in present study.

In overall acceptance, F1, F2 and F3 got higher scores than F5, while there was no significant difference between all other formulations. In the purchase intent test, F1 and F2 were more desired than F5 and as for the other samples, there was no difference between them, which agrees with Bernklau *et al.* (2017). We should highlight that the salty taste intensity sensed by the consumers is directly related to their preferences, which is greatly influenced by their individual habits than by their genetic features.

Additionally, our palate is more exposed to low-level sodium diets, it develops a greater sensibility to the salty taste and vice versa. Therefore, since we know that the teenager public consumes a high sodium level, it has the tendency of acquiring foods with greater amounts of this mineral (Fouladkhah *et al*., 2015). In Step 2 there was no significant difference ($p>0.05$) between the samples in the features appearance, aroma, aftertaste, texture and colour. However, lower scores were attributed to F5 in the flavour feature and for overall acceptance and purchase intent, with no difference between the other samples ($p>0.05$). These results show that after the addition of MSG in the breads, there was an acceptability of the products increased. It happened because MSG gives a different flavour to foods called "umami," which means "tasty" in Japanese. When it touches the flavour receptors, MSG also cooperates to reduce the bitter flavour caused by KCl (Choudhuri *et al*., 2016), which was proved herein. However, in spite of the scores for F5 (0% of sodium), adding the enhancer was not enough to improve this particular formulation's acceptance. After the addition of MSG in the formulations (Step 2), there was a general increase of every sample and feature's AI, while the features of aftertaste, texture and purchase intent remained with results under 70% in this classification. Therefore, the bread formulations with lower sodium level and addition of KCl and MSG were proven viable for the teenager public, favoring good dietary habits and contributing with public campaigns whose purpose is to reduce sodium consumption (Costa *et al*., 2012; Nilson *et al*., 2012).

In the ordering test (Step 1), we notice that F1 was reported as the most salty formulation, while F2 and F3 got better scores than F4 and F5 ($p<0.05$). This result shows that greater KCl levels cause a significant reduction in the products' salty taste. It happens because sodium is the only element that can activate the transduction mechanisms of the epithelial channels, which are responsible for the identification of the salty taste. It also has the property of enhancing the flavour of given ingredients, hiding the bitter taste caused by other elements (Aliño *et al*., 2009). In Step 2, F1 got higher scores than F3, F4 and F5, similarly to F2, while F5 was indicated as the less salty in comparison with F1 and F2 ($p<0.05$), with no statistical difference between the other samples ($p>0.05$). These results indicate an improvement of the salty taste after the addition of MSG. According to Mcgough *et al*. (2012), adding MSG in food may increase the salty taste sensation, improving its palatability. This fact may be explained by the necessary proportion of 2:1 of the enhancer (MSG/NaCl) to keep the product's salty taste (Reyes *et al.,* 2011), which happened only up to the formulation F4.

Chemical composition

The bread was evaluated in its centesimal composition. Its levels of moisture ($24.47 g.100g^{-1}$), ash ($0.91\ g.100g^{-1}$), proteins ($8.91\ g.100g^{-1}$), lipids ($8.50\ g.100g^{-1}$), carbohydrates ($57.18\ g.100g^{-1}$) and energy ($340.73\ kcal.100g^{-1}$) were similar to the ones reported in the literature (Spina *et al*., 2015). The DRV of an average portion of 50 g of bread represents 6.69% of protein, 6.19% of lipid, 11.04% of carbohydrate and 8.96% of energy. The mean levels of sodium and potassium of the five bread formulations are presented in Table 2.

Since we know that F4 (Step 1 and 2) got a better sensory acceptance (Table 1), that is, a greater KCl addition and an acceptance similar to the standard formulation, we managed to do an average reduction of 72.49% of sodium in comparison with the standard formulation. Superior results to what was seen in other studies, which reduced it between 25% (Bernklau *et al*., 2017) and 30% (Mueller *et al*., 2016). Considering NaCl's relevant importance as a raw material of breads (Doyle *et al*., 2010), its reduction becomes a great challenge to the food industry, since this strategy may directly interfere in these products' sensory and technological characteristics. In spite of these adversities, lower sodium consumption may contribute to a lower risk of systemic hypertension in all life stages (Liem *et al*., 2011).

Conclusion

We can reduce the level of NaCl (50%) with the addition of KCl (0.30%) in bread, without interfering in the teenager public's sensory acceptability. The addition of MSG increases the bread's acceptability, reducing the amount of NaCl added to the product in up to 75%. Reducing the NaCl level in bread promotes a healthier diet and cooperates with the world actions related to the prevention and control of chronic diseases. We suggest other studies that may cooperate to a lower use of sodium in foods.

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