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Development of a gluten- and lactose-free bakery product with nutritional and functional potential

Desarrollo de un producto de panadería sin gluten y sin lactosa con potencial nutricional y funcional

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Abstract

An increase in world population, malnutrition, overweight, and lack of micronutrients are some of the current challenges of the food industry. The new food products must cover nutritional requirements and help maintain a healthy life. Using functional and super foods as ingredients in a well-accepted product could be a strategy to solve these challenges and cover the market needs. This study evaluated incorporating ingredients recognized as superfoods and unconventional to design a gluten- and lactose-free bakery product. It also included microencapsulated blackberry juice to fortify it with antioxidant properties. A microencapsulation method, proximal chemical, sensory, texture, and microbiological analysis were used to determine the impact of the ingredients on the muffin design. Significant findings regarding nutritional, sensory, and textural quality were found, although crucial components in the muffin preparation, such as milk and wheat flour, were not used. The ingredients and the form of their incorporation significantly affected the textural properties and composition of the muffin. Incorporating elements with a high lipid content (pecan nut and chia) reduced hardness and gumminess. On the contrary, fibrous ingredients with high sugar (apple and chia jelly) promoted low moisture content. Moreover, incorporating encapsulated blackberry juice suggests the contribution of functions related to antioxidant activity since it is an extract rich in polyphenols. Therefore, this research determined that formulations 13 and 14 had the best characteristics as fortified bakery products.

Keywords: Dairy-free, Gluten-free, Muffin, Texture, Superfoods.

Resumen

Los desafíos de la industria alimentaria están relacionados con el aumento de la población, la desnutrición y el sobrepeso. En este contexto, los nuevos productos alimenticios no solo deben enfocarse en los requerimientos nutricionales sino también en contribuir en una vida más saludable. Por lo que la innovación y el diseño de nuevos productos se han centrado en el uso de los superalimentos y de ingredientes funcionales como una estrategia para resolver estos desafíos alimentarios y cubrir las necesidades del mercado. Este estudio evaluó la incorporación de ingredientes reconocidos como superalimentos para diseñar un producto de panadería libre de gluten y lactosa; también contenía jugo de mora encapsulado para fortalecerlo con propiedades antioxidantes. Se utilizó un método de microencapsulación por secado por aspersión, análisis químico proximal, sensorial, de textura y microbiológico para determinar el impacto de los ingredientes en el diseño del muffin. Se encontraron hallazgos significativos relacionados con la calidad nutricional, sensorial y textural, a pesar de no utilizar componentes importantes como la leche y la harina de trigo. Los ingredientes y la forma de su incorporación afectaron significativamente las propiedades texturales y la composición de los muffins. La incorporación de elementos con un alto contenido en lípidos (nuez pecana y chía) redujo la dureza y la gomosidad. Por el contrario, los ingredientes fibrosos con alto contenido de azúcar (jalea de manzana y chía) promovieron un bajo contenido de humedad. Además, la incorporación de jugo de mora encapsulado sugiere el aporte de funciones relacionadas con la actividad antioxidante al tratarse de un extracto rico en polifenoles. Por lo tanto, esta investigación determinó que las formulaciones 13 y 14 tenían las mejores características como productos de panadería fortificados.

Palabras clave: Sin lácteos, Sin gluten, Muffin, Textura, Superalimentos.

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1 Introduction

According to the Food and Agricultural Organization (FAO) in the year 2050, the world population will reach almost 9700 million people (FAO, 2017). Moreover, malnutrition, lack of micronutrients, being overweight, and obesity affect a high proportion of the world's population, without forgetting that the population is aging rapidly (FAO, 2017). Besides, older people are prone to malnutrition, bone and muscle loss, immunosuppression, poor life quality, mastication problems, and olfactory dysfunction (Höglund, 2017). Therefore, having a healthy diet could ensure good health and nutrition since an appropriate diet can prevent risk factors for noncommunicable diseases (Afshin *et al*., 2017).

For these reasons, the food industry is facing new challenges since it must produce innovative products to satisfy all the market demands. The development of new products using functional foods and superfoods as ingredients, with increased nutritional value, will have a positive effect on health and quality of life. Functional foods are defined as foods that improve mental and physical health by decreasing the risk of some illnesses, in addition to satisfying hunger and providing nutrients (Tacer-Caba, 2019). The functional foods categories are basic foods (food that naturally contains bioactive compounds), processed foods with added bioactive (foods processed with bioactive added), and foods enhanced to have more of a bioactive (the concentration of the bioactive is modified or concentrated) (Gul, *et al*., 2016). Superfoods are foods with high nutritional values because of the high concentration of nutrients and bioactive ingredients they contain (Devalaraja, *et al*., 2011). Some superfoods are fruits (berries, grapes, chickpeas, blueberry, raspberries, hippophagous); dried nuts (almonds, walnuts); vegetables (spinach, broccoli, avocado); pulses (red beans, lentils, cocoa); bee products (honey, waxes, royal jelly); herbs (ginger, tea, ginkgo biloba); linseed, chia, quinoa, amaranth and seaweed (spirulina, chlorella) (Proestos, 2018).

Whole oatmeal was selected for this study due to its nutrient content which includes starch, proteins, dietary fiber, and unsaturated fatty acids, in addition to its acceptability in glute-free diets (Rasane, *et al*., 2015). Honey has potential anticancer effects and has shown anti-inflammatory, antimicrobial, antioxidant, antitumoral, and antimutagenic properties (Ahmed & Othman, 2013). In the case of apples and berries, their polyphenols have shown antioxidant and anti-inflammatory activity, and anti-neurodegenerative and anticarcinogenic effects among others (Sun-Waterhouse, 2011). Chia also presents components of interest such as essential polyunsaturated fatty acids, flavanols, and phenolic acids (Mohd Ali, *et al*., 2012).

Therefore, the benefits of combining superfoods and functional foods in one product are clear. However, the product must be well accepted by consumers. The most consumed foods around the world are bakery products, being the most popular cakes, due to their sensory properties (Matsakidou, *et al*., 2010). Nevertheless, the tendency to consume healthy and natural foods, the special needs of the consumers, the world population increase, and current health problems force the bakery industry to include new ingredients and offer healthier alternatives.

Polyphenols are key components of superfoods and functional foods, unfortunately, they are unstable under some conditions such as oxidative enzymes, pH, moisture, and low and high temperature (Kuck & Noreña, 2016). There are strategies to conserve the physiological activity of these compounds, for example, microencapsulation, edible films and coatings, and vacuum impregnation (Betoret, *et al*., 2011). Microencapsulation is a strategy that protects an extensive range of molecules of biological interest including proteins, microorganisms, drugs, and bioactive molecules. In the food industry, this process is used to avoid the degradation of unstable compounds as polyphenols during food processing and storage (Betoret *et al*., 2011).

Bakery products could be considered an excellent source of nutrients since they incorporate the major nutrient groups in a sensory attractive product, however, the principal ingredients of typical bakery products are refined wheat flour, butter, milk, sugar, and egg, among others, which are common ingredients in empty calorie products (Poti, *et al*., 2014). On the other hand, replacing these ingredients with seeds and fruits can increase the nutritional content, and in turn, ingredients considered allergenic, such as milk and gluten, can be eliminated. One of the main problems in achieving a good bakery product is the technological challenge that involves replacing these ingredients and obtaining a substitute with acceptable organoleptic and sensory characteristics (Salehi & Aghajanzadeh, 2020). In this context, the present work aimed to develop novel formulations of a gluten-free and lactose-free fortified bakery product and the evaluation of their nutritional values, physical parameters, and sensory properties of a product with an increased content of lipids and protein, based on superfoods and functional foods.

2 Materials and methods

2.1 Raw materials and chemicals

Inulin, Gum Arabic, Folin-Ciocalteu reagent, absolute ethanol, sodium carbonate, oxalic acid, 2,6-

diclorofenolindofenol were purchased from Sigma-Aldrich (St. Lois. MO, USA). Fresh blackberries, apples, pecan nuts, peanuts, lemons, chia, oatmeal, honey, blueberry, and oranges were purchased in bulk from a local market. Pasteurized egg whites from a local brand were purchased in a supermarket.

2.2 Preparation of ingredients

Before dehydration apples were washed and disinfected. Apples were sliced into discs with 2 mm thickness and impregnated in lemon juice by immersion for 3 min. Apple slices were placed over drying racks and dried in a convection oven for 30 min at 120°C. Dried apples were stored in hermetic bags at room temperature until used.

For the preparation of apple jelly, disinfected samples were chopped, blended, and cocked for 30 min with 5 mL of lemon juice and 10 g of cinnamon. The juice was recovered and cooked for another 30 min with a sweetener mixture of sugar and stevia (50:50, w/w), using a juice: sweetener ratio of 1:1. Once the jelly thickened chia was incorporated and the jelly cooled and stored until used.

Pecan nuts and peanuts were pulverized for 50 seconds at speed one using a commercial food processor. The resulting flours were bagged and stored at room temperature. Pecan nuts and peanuts were also chopped, bagged, and stored at room temperature.

Orange zests were prepared using a manual grater. Zest was bagged and stored at room temperature until use.

2.2.1 Encapsulation of blackberry juice

For the microencapsulated powder used for muffin sprinkling, blackberry juice was extracted from mature fruits using a commercial juice extractor (FPSTJE317S, Oster, China) and drinking water: fruit ratio of 1:1 (w/w). The juice was filtered and centrifuged to remove solids. The resulting supernatant was considered the initial fruit juice sample, and the pH was determined and stored at -17°C until use. The blackberry juice was mixed with Gum Arabic (10% w/v) and inulin 10% (w/v) for microencapsulation by spray-drying using a laboratory scale spray-dryer (mini spray-dryer B-290, Buchi, Switzerland). The operating conditions used were an air inlet temperature of 160°C, an outlet temperature of 60°C, a feeding flow rate of 3 mL/min, and an atomization pressure of 21 psi. The dried powder was stored in airtight bags at -17°C until used. The microencapsulation efficiency was determined from solids introduced into the fed suspension before spray drying and the total powder recovered in the cyclone dryer. PPY was calculated with Equation (1):

$$PPY = \frac{P}{Pi} \times (100) \quad (1)$$

where Pi is the weight of dry solids fed in the spray dryer, and P is the weight of the final spray-dried powder obtained.

The moisture content of the dried blackberry juice was measured using a moisture analyzer (MB23, Ohaus, USA). 1 g of samples was dried at 160°C until constant weight. All measurements were carried out by duplicate.

The total polyphenol content was determined using the Folin-Ciocalteu reagent. 1 g of sample was mixed with 3 mL of water for one hour. 0.2 mL of this aliquot was mixed with 0.8 mL of absolute ethanol and mixed for 15 min. Then the mixture was centrifuged for 15 min at 4000 rpm and 0.1 mL of the supernatant was mixed with 0.5 mL of Folin-Ciocalteu reagent. After a 5 min reaction, 0.4 mL of a saturated sodium carbonate solution was added, and the mixtures were kept in the dark at 37°C for 2 h. Absorption was measured at 765 nm using a spectrophotometer (Multiskan Go, Thermo Fisher Scientific, Waltham, MA, USA). Total phenolic compounds were expressed as milligrams of gallic acid equivalent per gram of sample. Analyses were carried out in duplicate.

Ascorbic acid content was measured by titration following the AOAC protocol (AOAC International, 2005) in triplicate. Total anthocyanins were determined following a previously described method with some modifications (Lee, *et al*., 2005). 1 g of the sample was agitated in 10 mL of distilled water for 5 min. Then the sample was centrifuged at 4000 rpm for 10 min. 0.2 mL of the supernatant was added to 1.8 mL of a potassium chloride solution (0.03M, pH 1.0) and other 0.2 mL to 1.8 mL of a sodium acetate solution (0.4 M, pH 4.5). The absorbance was measured at 520 and 700 nm respectively using a spectrophotometer (Multiskan Go, Thermo Fisher Scientific, Waltham, MA, USA). Total anthocyanins were calculated with Equation 2 and expressed as equivalents of cyanidin-3-glucoside in mg/g of sample.

$$TA(mg/L) = \frac{A \times MW \times DF \times 1000}{\varepsilon \times 1} \quad (2)$$

Where TA=total anthocyanin $A = (A_{520nm} - A_{700nm})$ pH 1.0 - $(A_{520nm} A_{700nm})$ pH 4.5; MW (molecular weight)= 449.2 g/mol for cyanidin-3-glucoside (cyd-3-glu); DF= dilution factor; 1= pathlength in cm; ε = 26 900 molar extinction coefficient, in L x mol^{-1} x cm^{-1}, for cyd-3-glu; 1000= factor for conversion from g to mg.

Color determinations of the formulations were performed using a CR-400 Chroma Meter (Konica Minolta Sensing Americas, USA). Measurement was performed using a glass transparent petri dish where the powder/juice was placed creating a 5 mm layer, standard illumination C, and illuminant D65. The determinations were repeated five times.

2.3 *Formulation and preparation of the bakery product*

The bakery product proposed is gluten and dairy-free, protein-rich and fortified. The base components of the product were oatmeal (9 g), honey (11 g), egg white (9 g), blueberry (11 g), and orange zest (1 g). Apple-chia jelly, chia, dehydrated apple, pecan nut flour, chopped pecan nuts, peanut flour, and chopped peanuts were added at different proportions. The 18 formulations (F) tested were composed of the base components and the ingredients presented in Table 1.

All based components were mixed, then the extra ingredients were added, and the batter was placed in a muffin pan and baked for 30 min at 180°C or until done in a preheated oven. After a five-minute setting period, the products were removed from the pans, sprinkled with blackberry powder, and cooled before packing and storage. All formulations were prepared in quintuplicate, and one product was prepared per replica.

2.4 *Muffin characterization*

2.4.1 *Chemical composition*

American Official Analytical Chemistry methods were used to determine moisture content and ashes (William, 2000; AOAC International, 2005). Fat was extracted for 6 h using Soxhlet and hexane as solvent. After solvent evaporation, the percentage of fat was calculated gravimetrically. The defatted residues were dried and used for protein and ash analysis. Protein content was obtained using the Kjeldahl method and the 6.25 factor for the conversion of total nitrogen to protein content (Kirk, 1950). Carbohydrate content was obtained by subtracting moisture, ash, fat, and protein content from 100g of the product (Morillas-Ruiz & Delgado-Alarcón, 2012). Energy values were calculated using the factors 4, 9, and 4 kcal/g for protein, fat, and carbohydrate, respectively. Analysis was carried out in triplicate and mean values are presented.

Table 1. Formulation bakery product compositions (% w/w).

Formulation	Ingredients	(%)	Formulation	Ingredients	(%)
1	Apple-chia jelly	15.1	10	Chopped peanuts	14.7
				Dehydrated apple	1.3
				Chia	1.3
2	Dehydrated apple	1.6	11	Chopped pecan nuts	14.7
	Chia	1.6		Dehydrated apple	1.3
				Chia	1.3
3	Pecan nut flour	14.9	12	Pecan nut flour	14.7
	Chia	1.4		Dehydrated apple	1.3
				Chia	1.3
4	Chopped pecan nuts	14.9	13	Chopped pecan nuts	13.1
	Chia	1.4		Apple-chia jelly	13.1
5	Chopped peanuts	14.9	14	Pecan nut flour	13.1
	Chia	1.4		Apple-chia jelly	13.1
6	Peanut flour	14.9	15	Pecan nut flour	7.9
	Chia	1.4		Chopped peanuts	7.9
				Dehydrated apple	1.3
				Chia	1.3
7	Peanut flour	13.1	16	Chopped pecan nuts	6
	Apple-chia jelly	13.1		Peanut flour	7.1
				Apple-chia jelly	13.1
8	Peanut flour	14.7	17	Chopped peanuts	6
	Dehydrated apple	1.3		Chopped pecan nuts	7.1
	Chia	1.3		Apple-chia jelly	13.1
9	Chopped peanut	13.1	18	Peanut flour	6.7
	Apple-chia jelly	13.1		Pecan nut flour	8
				Dehydrated apple	1.3
				Chia	1.3

Ingredients added to 62 g of the base formulation.

2.4.2 Color analysis

Color determinations of the formulations were performed using a CR-400 Chroma Meter (Konica Minolta Sensing Americas, USA), according to the CIELAB system. Tests were performed in quintuplicate, and the L*, a*, and b values were reported. L* measured luminosity (black=0, white=100), a* colors green (-a*) and red (+a*), and b* colors blue (-b*) and yellow (+b*). The equipment was calibrated with a white ceramic plate (L* 92.49, a* 1.25, b* -1.92). For each analysis the color was measured at the crust, then the crust was carefully removed and the crumbs in the middle were exposed for color measurement finally, the color at the bottom of the sample was measured.

2.4.3 Texture analysis

Texture properties were determined using a TA.XT plus Texture Analyzer (Stable Micro Systems Ltd., Godalming, Surrey, UK). Samples were cut in cube-shaped (20 mm x 20 mm), and the bottom segment was used for mechanic measurements. A double compression test (20% of its original height) using a 40 mm diameter flat-ended cylindrical probe, at a speed of 1 mm/s was performed. Hardness, cohesiveness, compression, springiness, chewiness, and resilience were obtained from force and area measurements. Three samples from each formulation were used.

2.4.4 Microbiological analysis

The microbiological analysis was performed for the best selected formulations, following the official Mexican protocol (NOM-247-SSA1-2008) for flours and derived products. The parameters measured were colony-forming units per gram of aerobic mesophylls, total coliforms, and *Staphylococcus aureus*, in addition to the presence of *Salmonella* spp. and *Escherichia coli*.

2.4.5 Sensory evaluation

Sixty-six consumers (50% female and 50% male), participated in the study. Inclusion criteria were being over 18 years old, cookies consumers, and wishing to participate in the study. 18 formulations were evaluated in three different sessions (6 formulations per session). Formulations were presented nomadically ordered following a balanced order for each consumer (Macfie, *et al*., 1989). Each formulation was presented in a plastic container, labeled with three-digit random numbers and mineral water was available for rinsing between samples. Assessments were carried out in the laboratory with individual booths.

Subsequently, consumers responded to the CATA questions with 12 descriptors related to the sensory characteristics of the formulations. The selected terms were sweet flavor, bittersweet flavor, acid, light brown, medium brown, dark brown, sweet aroma, bittersweet aroma, bitter aroma, soft, grainy, and gritty.

2.5 Statistical analysis

All experimental data were analyzed using one-way analysis of variance (ANOVA), where $p<0.05$ was considered significant, with the software XLSTAT statistical and data analysis solution, (Addinsoft, 2020, NY, USA). The frequency with which each descriptor of the CATA questions was used to describe each formulation was determined. To identify significant differences between samples for each descriptor, the Cochran Q test was performed (Manoukian, 1986; Parente, *et al*., 2011; Ares & Jaeger, 2015). To relate the mean values of instrumental texture, color, and composition formulations to the descriptors frequencies of the tasting questions a multiple factor analysis (MFA) was used. MFA is a useful tool for analyzing different tables of variables, which can include quantitative and qualitative data (Bécue-Bertaut, *et al*., 2008; Bécue-Bertaut & Pagès, 2008). The CATA questions and MFA were carried out using FactoMineR an R package version 3.3.2.

3 Results and discussion

In this study, 18 formulations were developed using oatmeal, honey, blueberry, orange zest, egg white, peanut and pecan nut (flour and chopped), apple (jelly and dehydrated), chia seeds, and microencapsulated powder from blackberry juice sprinkled topping on the muffin (Figure 1). The encapsulated juice blackberry was added as a potential functional food ingredient with high antioxidant contents. The incorporation of ingredients recognized as superfoods can provide different effects on the nutritional and functional value and sensory, physical, and microbiological attributes of bakery products (Salehi & Aghajanzadeh, 2020).

3.1 Encapsulation efficiency

To fortify the bakery product with antioxidant compounds, blackberry juice was microencapsulated by spray drying. The microencapsulation efficiency was 63.75 ± 3.18%; similar results to those reported in other studies regarding encapsulated blackberry juice (63.6%) (Cortes-Rodriguez *et al*., 2022). The blackberry juice powder presented a retention of 42.6% of anthocyanins (0.98 ± 0.05 mg/g) and 95.6% of total phenolic compounds (2.20 ± 0.02 mg/g)

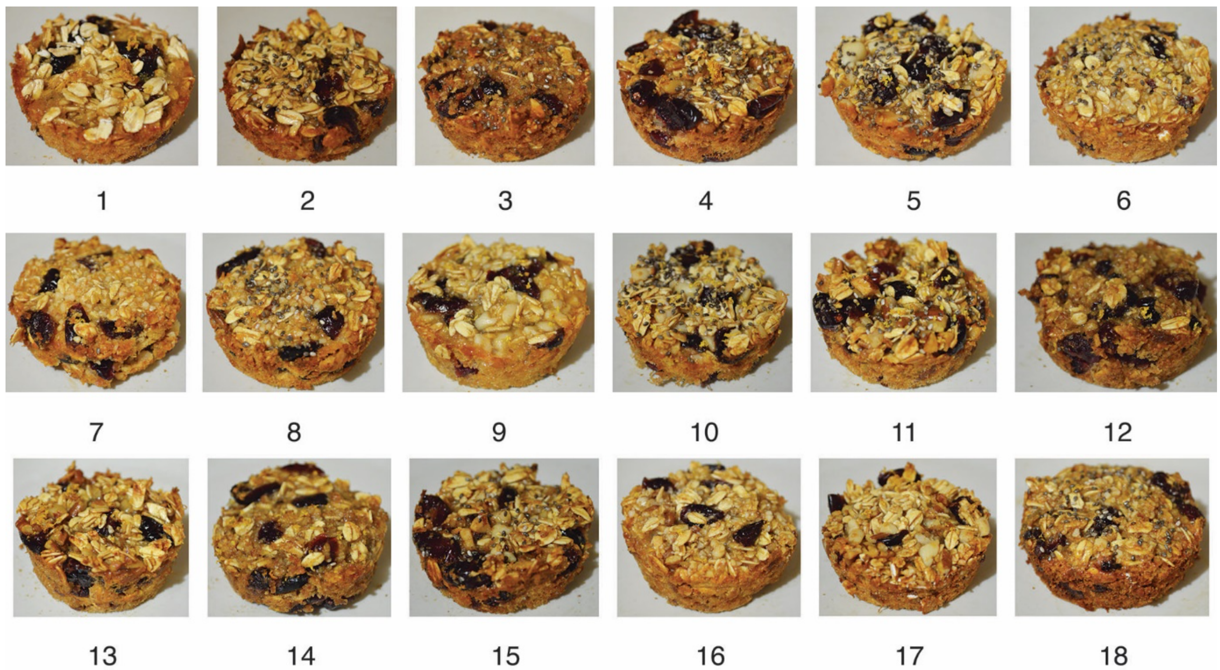

Figure 1. Bakery product formulations containing the base ingredients and extra components.

after spray drying. This trend is due to the drying temperature, which harms the content of anthocyanins since they are more sensitive to heat, as has been observed in other studies (de Carvalho *et al.*, 2017). The concentration of ascorbic was 0.45 ± 0.05 mg/g. This work presents high retention of anthocyanins and total phenolic compounds compared to a previous study, where blueberry was encapsulated by spray drying and freeze-drying and retention values close to 20% were obtained, using maltodextrin and beta-cyclodextrin like carrier agents (Wilkowska, *et al*, 2016). High retention of total phenolic compounds and good encapsulation efficiency has been previously reported for grape skin phenolic compounds using Gum Arabic (10% w/v) as a carrier (Kuck & Noreña, 2016). Good retention using Gum Arabic is related to its structure, since it is a highly branched heteropolymer sugar, and it contains a small amount of protein covalently linked to carbohydrate chains, acting as an excellent film-forming agent, that generates an effective encapsulation (Burin, *et al.*, 2011). Water activity in food powders is related to their chemical and microbial stability. The blackberry juice powder obtained showed a water activity value lower than 0.3 (0.23 ± 0.04), which indicates the stability of the compounds and prolonged shelf life (Rahman & Labuza, 2007). The color parameters observed were L 57.14 ± 2.27, a* 23.20 ± 1.62 and b* -0.06 ± 0.03. The blackberry juice powder showed in general an amethyst color. The luminosity value was high, it showed a tendency towards white, a* parameter presents a positive value with tendency to color red and b* parameter shows a negative value with tendency to blue. These results are similar to those reported for encapsulated chokeberry juice with maltodextrin-Gum Arabic like carriers (Bednarska & Janiszewska-Turak, 2019). Finally, microencapsulated powders retained high concentrations of anthocyanins and phenolic compounds, suggesting that their incorporation into muffins could provide functions related to antioxidant properties.

3.2 Nutritional analysis of muffins

Table 2 shows the compositional analysis of all formulations. The baked products contained between 11.2-21.1% of moisture, 4.9-9.5% of proteins, 1.9-17.4% of lipids, 1.5-33.7% of ash, 44.2-64.3% of carbohydrates, and calorie values between 117.8 to 240.2 kcal. The high carbohydrate percentages are related to the presence of apple jelly. In this case the high protein percentages they are attributable to the chopped peanuts and pecan nuts. Pecan nuts, as a superfood, have a high nutritional value and are rich in fat, protein and carbohydrates, micronutrients, fat-soluble bioactive, and phytochemicals. They are recognized for their positive impact on health since they can reduce cardiovascular diseases, cancer, and asthma risk, and they benefit cognitive capabilities (Alasalvar & Bolling, 2015). Peanuts are a source of energy (48-50%) and protein (25-28%), and they contain healthy nutrients such as vitamins, monounsaturated fatty acids, antioxidants, and minerals, so they are used to solve malnutrition problems (Janila, *et al.*, 2013).

Table 2. Characterization of the 18 bakery product formulations.

F	Moisture (%)	Proteins (%)	Lipids (%)	Ash (%)	Carbohydrates (%)	Energy (kcal)
1	17.2 ± 0.05	4.9 ± 0.10	1.9 ± 0.08	15.9 ± 0.01	60.1 ± 0.11	153 ± 0.38
2	15.25 ± 0.09	4.9 ± 0.03	1.9 ± 0.05	33.7 ± 0.03	44.2 ± 0.07	117.8 ± 0.10
3	16.85 ± 0.06	6.9 ± 0.04	17.4 ± 0.10	11.8 ± 0.01	47 ± 0.05	205 ± 0.26
4	20.25 ± 0.16	6.9 ± 0.07	17.4 ± 0.04	8.45 ± 0.05	47 ± 0.01	205 ± 0.22
5	13.12 ± 0.05	9.5 ± 0.21	11.7 ± 0.01	17.38 ± 0.07	48.3 ± 0.22	185.8 ± 0.12
6	17.4 ± 0.04	9.5 ± 0.57	11.7 ± 0.02	13.1 ± 0.02	48.3 ± 0.05	185.8 ± 0.14
7	12.2 ± 0.05	9.5 ± 0.16	11.7 ± 0.05	2.3 ± 0.04	64.3 ± 0.06	221 ± 0.09
8	21.12 ± 0.05	9.5 ± 0.09	11.7 ± 0.08	9.38 ± 0.06	48.3 ± 0.07	185.8 ± 0.14
9	12.15 ± 0.21	9.5 ± 0.07	11.7 ± 0.01	2.4 ± 0.07	64.3 ± 0.01	221 ± 0.32
10	12.15 ± 0.08	9.5 ± 0.10	11.7 ± 0.02	18.3 ± 0.04	48.3 ± 0.03	185.8 ± 0.26
11	19.05 ± 0.11	6.9 ± 0.27	17.4 ± 0.04	9.6 ± 0.05	47 ± 0.04	205 ± 0.18
12	15.25 ± 0.12	6.9 ± 0.05	17.4 ± 0.07	13.4 ± 0.01	47 ± 0.07	205 ± 0.31
13	11.2 ± 0.10	6.9 ± 0.09	17.4 ± 0.09	1.5 ± 0.01	63 ± 0.09	240.2 ± 0.19
14	11.2 ± 0.04	6.9 ± 0.26	17.4 ± 0.05	1.5 ± 0.03	63 ± 0.05	240.2 ± 0.17
15	14.9 ± 0.07	8.2 ± 0.11	14.1 ± 0.02	15.2 ± 0.02	47.6 ± 0.04	192.9 ± 0.20
16	12.4 ± 0.04	8.2 ± 0.06	14.1 ± 0.04	1.7 ± 0.04	63.6 ± 0.03	228 ± 0.09
17	11.8 ± 0.03	8.2 ± 0.10	15 ± 0.07	1.5 ± 0.07	63.5 ± 0.04	232.8 ± 0.16
18	18 ± 0.12	8.3 ± 0.04	15 ± 0.02	11.2 ± 0.06	47.5 ± 0.07	197.6 ± 0.30

F=Formulation

Values represent means of two measurements.

The moisture content is high compared to conventional bakery products, and higher than the moisture content of rice muffins supplemented with shiitake mushroom and carrot pomace (Olawuyi & Lee, 2019). The high moisture content found in these formulations is of interest since it is associated with a fresher and softer product. The high ash content found is the result of the presence of mineral constituents (Adedeji, *et al*., 2008), which are related to formulations containing apples and peanuts as ingredients. The seven formulations with the highest carbohydrate content (60%) had apple-chia jelly as a common ingredient and increased content due to the presence of sugar. This carbohydrate content is like the content reported for rice muffins with carrot pomace (Olawuyi & Lee, 2019), muffins with grape skin flour (Bender *et al*., 2016), and 20% squash seed flour muffins (Palacio *et al*., 2018).

Formulations F5 to F10 and F15 to F18 had higher protein percentages due to the presence of peanuts since peanuts have a higher protein content than pecan nuts (Ros, 2010). These protein contents are similar to the ones obtained in rice muffins with carrot pomace (Olawuyi & Lee, 2019), and muffins with grape skin flour (Bender *et al*., 2016), higher than in corn muffins (Jauharah *et al*., 2014) and 10% squash seed flour muffins (Palacio *et al*., 2018), but lower than the content in rice muffins with shiitake mushrooms (Olawuyi & Lee, 2019), and 20% squash seed flour muffins (Palacio *et al*., 2018).

Formulations F3, F4, F11, F12, F13, and F14 showed the highest lipid content since they all had pecan nuts in their formulation. Pecan nuts have a higher lipid (Robbins, *et al*., 2011) content than peanuts (Li & Hu, 2011), which explains the differences observed. Formulations without peanuts or pecan nuts had a low lipid content (F1, F2). All the formulations had a lipid content higher than supplemented rice muffins (Olawuyi & Lee, 2019), and formulations F3-F18 had content like or higher than corn muffins (Jauharah, *et al*., 2014), muffins with grape skin flour (Bender *et al*., 2016) and squash seed flour muffins (Palacio, *et al*., 2018). One of the main benefits of the muffin designed in this research is the potential contribution of polyunsaturated fats such as oleic, linoleic, linolenic acid, and omega-3 acids, which have been reported in pecan nuts and chia (Kulczynski, *et al*., 2019; Ribeiro *et al*., 2020). Since the muffins mentioned earlier were designed with commercial fats (margarine and soybean oil), which are subjected to processes such as refining and hydrogenation, which are questionable for health. Moreover, another significant benefit of high-fat baked goods is the positive effect on texture, as they reduce hardness and elasticity and increase chewiness (Bender *et al*., 2016; Palacio, *et al*., 2018; Olawuyi & Lee, 2019).

3.3 Color analysis

Color data of the different formulations are presented in Table 3. The bottom part of the samples had the same L, a*, and b* values as the crumbs in the middle. A significant difference was observed regarding the luminosity of the products. The luminosity values of the crust and crumb decreased as the formulations became more complex. The luminosity tended to darkness. The highest luminosity values

Table 3. Color and texture profile analysis of bakery products.

F	Crust L*	Crust a*	Crust b*	Crumb L*	Crumb a*	Crumb b*	Hardness (N)	Adhesiveness (N)	Springiness (cm)	Cohesiveness	Gumminess (N)	Chewiness (Kg cm)
1	54.05^{a}	7.29^{abcd}	24.38^{a}	46.81^{abcd}	11.23^{a}	26.41^{a}	3.15^{ab}	-0.63^{ab}	0.57^{a}	0.17^{a}	5.60^{abc}	3.17^{abc}
2	48.53^{abc}	7.21^{abcd}	22.45^{ab}	43.23^{cdefg}	13.72^{a}	25.52^{a}	—	—	—	—	—	—
3	40.82^{cd}	7.96^{abc}	17.27^{ab}	38.76^{g}	10.09^{a}	17.11^{cd}	2.85^{bc}	-0.40^{a}	0.41^{a}	0.14^{a}	4.05^{bc}	1.67^{c}
4	45.35^{abcd}	6.48^{bcd}	18.76^{ab}	41.98^{defg}	10.09^{a}	19.87^{abcd}	—	—	—	—	—	—
5	45^{abcd}	4.68^{d}	15.43^{b}	$49.70 \pm 2.4\ ^{ab}$	11.81^{a}	26.95^{a}	—	—	—	—	—	—
6	46.96^{abcd}	6.60^{bcd}	18.56^{ab}	47.59^{abcd}	12.18^{a}	25.35^{ab}	—	—	—	—	—	—
7	48.15^{abcd}	9.46^{a}	22.68^{a}	46.12^{abcdef}	10.77^{a}	22.02^{abcd}	3.36^{ab}	-0.76^{b}	0.76^{a}	0.14^{a}	5.57^{abc}	4.31^{ab}
8	45.60^{a}	8.12^{a}	19.49^{ab}	44.30^{a}	13.62^{a}	25.13^{a}	—	—	—	—	—	—
9	50.99^{ab}	8.54^{ab}	23.68^{a}	52.12^{a}	9.77^{a}	24.6^{abcd}	2.15^{c}	-0.52^{bc}	0.58^{a}	0.14^{a}	3.12^{c}	1.84^{c}
10	48.14^{abcd}	5.77^{cd}	17.22^{ab}	48.96^{abc}	10.44^{a}	23.72^{abcd}	—	—	—	—	—	—
11	43.81^{bcd}	7.28^{abcd}	20.43^{ab}	41.64^{defg}	11.7^{a}	23.49^{abcd}	—	—	—	—	—	—
12	38.45^{d}	8.61^{ab}	17.81^{ab}	41.11^{efg}	10.58^{a}	21.16^{abcd}	3.82^{a}	-0.53^{ab}	0.61^{a}	0.17^{a}	6.36^{ab}	3.85^{ab}
13	46.28^{abcd}	7.43^{abc}	21.20^{ab}	37.99^{g}	10.38^{a}	16.91^{d}	2.9^{b}	-0.51^{ab}	0.66^{a}	0.13^{a}	3.81^{c}	2.60^{bc}
14	43.14^{bcd}	8.58^{ab}	20.14^{ab}	39.90^{fg}	9.83^{a}	17.19^{bcd}	2.85^{bc}	-0.47^{a}	0.72^{a}	0.12^{a}	3.40^{c}	2.42^{bc}
15	48.06^{abcd}	7.20^{abcd}	21.3^{ab}	44.14^{bcdefg}	11.09^{a}	21.48^{abcd}	3.8^{a}	-0.4^{a}	0.66^{a}	0.18^{a}	7.27^{a}	5.07^{a}
16	45.45^{abcd}	7.48^{abc}	19.18^{ab}	42.46^{defg}	10.94^{a}	19.72^{abcd}	2.91^{b}	-0.60^{ab}	0.67^{a}	0.15^{a}	4.35^{bc}	2.92^{bc}
17	49.52^{abc}	7.66^{abc}	22.04^{ab}	45.63^{bcdef}	11.9^{a}	23.76^{abcd}	—	—	—	—	—	—
18	42.64^{bcd}	7.57^{abc}	17.08^{ab}	41.94^{defg}	13.02^{a}	22.22^{abcd}	—	—	—	—	—	—
p-values	0.001	0	0.008	< 0.0001	0.121	0.001	0	0.019	0.134	0.07	0.001	0.001

Values represent means with standard deviations. Different superscript letters indicate statistical differences (p<0.05) according to Turkey's test. p-values < 0.05 indicate statistically significant differences. Abbreviations: F (formulation); N (Newton, Kg·m/s^2).

were obtained in those formulations where apples, in both presentations, and peanuts were present. On the other hand, formulations with pecan nuts generated the lowest luminosity values. Darker colors have been previously related to Maillard reactions and the presence of condensation reactions between the different components, such as sugar and proteins that can occur at high baking temperatures (Mrabet *et al*., 2016).

The a* parameter presented a tendency towards red, with a significant difference in the crust and a nonsignificant difference in the crumbs. The increase in the redness of the formulations is related to the presence of carotenoids (Nath, *et al*., 2018), in this the blueberries. Finally, the b* parameter tended yellow. This coloration is related to the presence of oatmeal as part of the base ingredients and like in luminosity, the intensity of the color diminishes as the formulations became more complex.

These results provide a caramel color in all the samples, which are a result of the ingredients and their interactions (Alasalvar & Bolling, 2015). The incorporation of ingredients like apple (jelly and/or dehydrated), pecan nut, and peanuts (chopped or flour), are responsible for the caramel color observed.

3.4 Texture profile analysis

Cohesiveness is the maximum resistance that a material supports and is related to the force necessary to break a product into crumbs or pieces, it includes fracturability, chewiness, and gumminess properties. Food with strong cohesion will be more tolerant during handling, packaging, and transport ensuring it will arrive to the consumer in the desired presentation (Barbosa-Cánovas & Juliano, 2012). In this context, 9 of the 18 formulations (F2, F4, F5, F6, F8, F10, F11, F17, and F18) presented out-of-range and non-quantifiable values, showing low cohesiveness, and therefore were discarded.

Dough viscosity together with air incorporation plays an important role in the texture and appearance of the final product. In this study, we exanimated the effect of the different ingredients on the baked product texture. Results displayed that hardness, adhesiveness, gumminess, chewiness, and resilience were statistically significant (Table 3). For formulations F1, F7, F12, and F15 an increase in hardness translated as an increment in adhesiveness, gumminess, chewiness, and resilience. In addition, these formulations showed less water, indicating a higher content of other constituents such as proteins, lipids, or carbohydrates. Thus, results suggest that ingredients influenced the product texture. Previous studies have demonstrated that the baked-product quality is widely led by the nature and abundance of the ingredients, which influence mainly in their physical properties (Maache-Rezzoug, *et al*., 1998).

Hardness was mainly linked with dehydrated apples (F12 and F15) and apple-chia jelly (F1 and F7). However, the mixture of apple-chia jelly with other ingredients (F9, F13, F14, and F16) displayed a decrease in hardness. The dehydrated apple has fiber and sugars that promote hardness. This behavior has been observed in muffins made with high fruit fiber (Vasantha Rupasinghe, *et al*., 2009; Qureshi *et al*., 2017). Mildner-Szkudlarz *et al*. (2016) reported an increase in the hardness of fiber-enriched muffins related to the dilution of starch. They also observed that hardness is affected by the amount of water absorbed by the fiber fraction of the ingredients. Moreover, the high content of sugars acts as a hardener due to their crystallization (Maache-Rezzoug *et al*., 1998), which makes a crunchier product, modifying all texture parameters. Another study observed a correlation between hardness and cane bagasse incorporation, indicating that the fibrous components directly affect bakery products' texture properties.

Table 4. Descriptors frequency during the evaluation of each formulation.

Formulation	Flavor			Color			Aroma			Texture		
	Sweet *	Bittersweet*	Acid*	Light brown*	Medium brown*	Dark brown*	Sweet*	Bittersweet*	Bitter*	Soft*	Grainy*	Gritty*
1	32	24	6	24	30	8	13	42	7	23	33	6
2	24	30	8	16	35	11	10	37	15	17	39	6
3	27	28	7	8	30	24	14	41	7	16	39	7
4	28	28	6	12	32	18	13	37	12	16	40	6
5	29	26	7	28	29	5	10	39	13	16	40	6
6	20	34	8	22	24	16	13	36	13	12	40	10
7	37	16	9	35	19	8	8	30	24	25	26	11
8	20	27	15	26	34	2	7	40	15	16	35	11
9	43	17	2	43	17	2	7	36	19	30	30	2
10	22	26	14	39	20	3	9	35	18	15	41	6
11	31	24	7	24	28	10	17	38	7	19	38	5
12	27	24	11	12	29	21	16	37	9	15	32	15
13	44	16	2	15	33	14	9	42	11	27	30	5
14	38	24	6	9	22	31	23	30	9	29	19	14
15	16	33	7	17	36	9	11	40	11	9	43	10
16	42	17	3	9	29	24	15	38	9	19	34	9
17	40	18	4	14	32	16	17	35	10	11	47	4
18	21	29	12	9	35	18	10	42	10	10	37	15

*Represent p<0.0001.

Another study observed a correlation between hardness and cane bagasse incorporation (Morales-Tapia *et al*., 2023). Therefore, this information suggests that the fibrous components directly affect bakery products' texture properties. On the other hand, lipids and proteins from ingredients seem to not influence the texture properties of the formulations.

From the results of the texture profile analysis, the formulations with greater hardness, gumminess, chewiness, and resilience were discarded (F12 and F15). The remaining 7 formulations have acceptable organoleptic and sensory properties for a lactose-free bakery.

3.5 CATA question

Frequency of the terms used in the CATA questions to describe the different formulations are shown in Table 4. According to the Cochran Q test, all the descriptors presented significant differences in the frequency of use for the description of the 18 formulations. The descriptors that were most frequently mentioned were bittersweet, aroma, and grainy. On the other hand, those that were less frequently mentioned were: light brown and gritty. These results suggest that the CATA questions were adequate for detecting differences in consumer perception of the formulations. These results coincide with other studies where that have evaluated different types of products (Lado, *et al*., 2010; Lazo, *et al*., 2016; Schouteten, *et al*., 2017; Alencar *et al*., 2019). Furthermore, it is confirmed that the descriptors used were adequate for the sensory characterization of the formulations.

3.6 Correlation of sensory and instrumental variables

Figures 2A and 2B, show the graphical representation obtained through the MFA of the formulations and the sensory and instrumental variables. The first two factors of MFA explain 54% of the data variability. The map shows (Figure 2A) that the formulations are spatially distributed, which suggests that the formulations were perceived as different at the sensory level, and these differences are reflected in the significant differences observed in the levels of the instrumental measurements. Figure 2B, shows the MFA performed on the sensory and instrumental variables, as well as the composition of the formulations. The first factor is positively correlated with sweet flavor, percentage of carbohydrates, soft texture, and energy (kcal). However, it is negatively correlated by the variables of flavor (bittersweet), composition (ash and humidity percentages), sensory texture (grainy), instrumental (cohesiveness, gumminess, resilience, hardness), and color (a * -crumb and b * crumb). On the other hand, the second factor is positively correlated with light brown, L-crumb, L-crust, and mainly a bitter aroma. In contrast, the second factor negatively correlates with dark brown, sweet aroma, lipids, gritty, and adhesiveness. It is important to mention that the CATA question descriptors are related to some of the instrumental variables such as the soft and sweet flavor attributes, with the percentage of carbohydrates and energy.

Overall, the results show that F13 and F14 were perceived as sweet and soft-textured, with a high percentage of carbohydrates and energy. Meanwhile, F15 and F6 were perceived with bittersweet flavor, percentages of ash and humidity, and grainy, cohesiveness, gumminess, resilience, hardness texture, and color (a* and b* crumb). The F9 was distinguished by its light brown, L-crumb, and L-crust and by its bitter aroma. Finally, the F3 and F12 were perceived as dark brown, with a sweet aroma, gritty, adhesiveness, and high content of lipids.

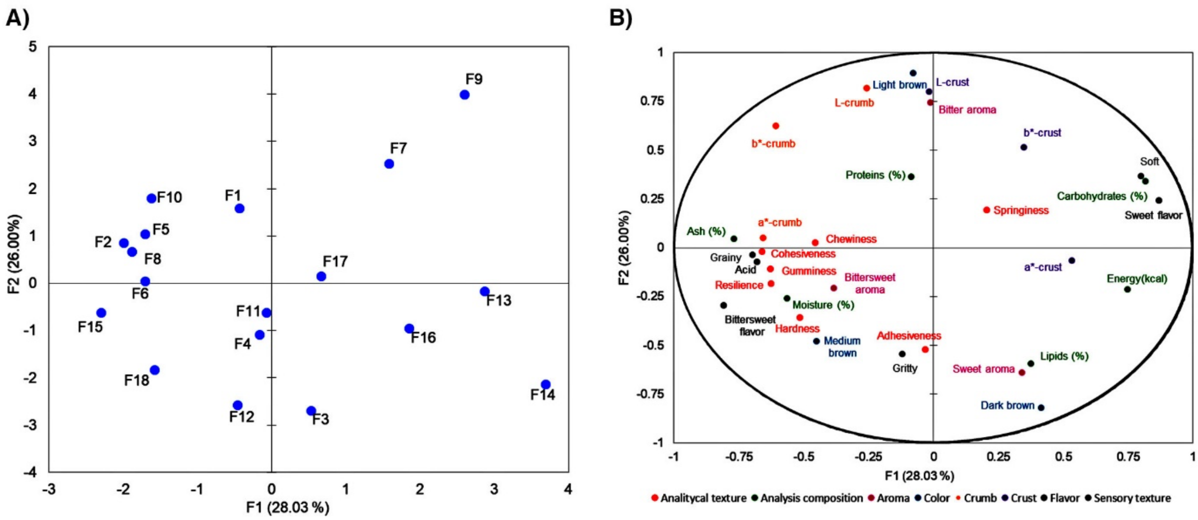

Figure 2. Multiple factorial analyses were carried out on the descriptors of the CATA questions, analytical texture and the composition of the formulations: (A) formulations, B) variables.z

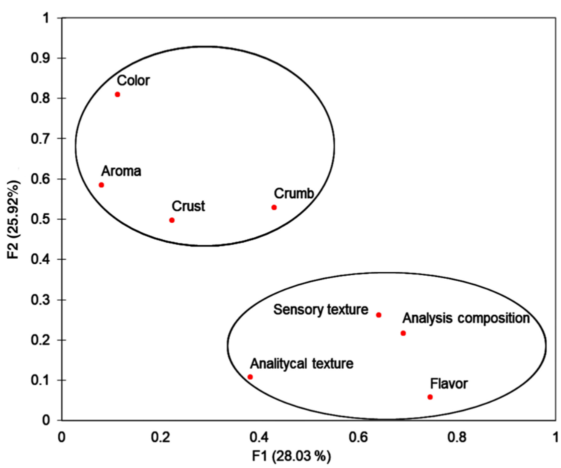

Figure 3. Representation of sensory, instrumental and composition variables in the first two factors, obtained through the MFA.

Figure 3 shows the spatial representation of the variables analyzed using the MFA. It is observed that the analytical texture and composition measurements are correlated with taste and sensory texture, while the color and aroma are related to crust and crumb. Through this analysis, the instrumental variables that influence the sensory characteristics of the formulations are confirmed.

3.7 Microbiological analysis

From the nutritional values, sensorial and texture analysis two formulations were selected F13 and F14. Microbiological analysis was carried out for both formulations 12 h after baking. The presence of Staphylococcus aureus, Salmonella spp., Escherichia coli, aerobic mesophiles, and total coliforms was evaluated. Results showed values under the permissible limits of the official Mexican protocol (NOM-247-SSA1-2008). Therefore, the baking product generated is microbiologically stable. Ingredients such as nuts, sugar, and spices are considered sources of bacterial spores, however, the baking process allows the inactivation of this type of microorganisms (Cook & Johnson, 2012).

Conclusions

This study evaluated the incorporation of ingredients recognized as superfoods in the design of a gluten- and lactose-free bakery product. Although critical ingredients were not considered in the muffin-making, such as milk and wheat flour, significant findings regarding nutritional, sensory, and textural quality were found. The effect of incorporating various ingredients with high lipids content (pecan nut and chia) reduced hardness and gumminess. In this sense, fibrous ingredients with high sugar (Apple-chia jelly) promoted balanced moisture content, which could impact the sanitary quality. Moreover, incorporating encapsulated blackberry juice could provide functions related to antioxidant activity since it is an extract rich in polyphenols. Therefore, the mixture of these ingredients makes the muffins more appetizing and nutritionally more attractive. Nonetheless, further research is recommended to study the concentration of flavonoids, phenolic compounds, and their antioxidant capacity in the final product. It would also be interesting to incorporate the blackberry juice powders before baking to determine the encapsulation process's effectiveness on the bioactive compounds.

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