

Article

Feruloylated Arabinoxylans from Nixtamalized Maize Bran By-Product as a Baking Ingredient: Physicochemical, Nutritional, and Functional Properties

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Abstract

In this study, feruloylated arabinoxylans (FAXs) extracted from nixtamalized maize bran were assessed as a functional ingredient in white bread. FAXs were added at percentages of 0.15% and 0.30% to bread, and a control sample without FAXs was prepared. Regarding texture profile analysis, hardness values in bread treated with FAXs ranged from 34.32 N (T5) to 51.03 N (T3), with all values for FAXs-added bread being lower than 64.43 N obtained for the control sample (TC). With respect to color, most of the FAX-treated samples had higher overall values than the control sample, with L^* values ranging from 50.49 (T4) to 59.40 (T6). The total color difference (ΔE) values ranged from 2.07 (T2) to 6.32 (T6), indicating differences between the control sample and the FAX-treated samples. In the analysis of proximate composition, all FAX-treated bread had higher levels of crude fiber content than the control sample, and water activity (a_w) values were lower in the control sample than in bread treated with FAXs. Regarding total phenols, FAX-treated bread ranged from 1.57 (T6) to 1.98 (T1) mgFAE/g, being higher than the 1.24 mgFAE/g found in the control sample (TC). The antioxidant capacity levels, namely, DPPH, ABTS, and FRAP, were 9.36–17.01, 8.86–17.64, and 3.05–5.07 $\mu\text{molTE/g}$, respectively. Thus, it is possible to conclude that adding FAXs to bread formulations improves the hardness, crude fiber content, and functional properties of bread.

Keywords: feruloylated arabinoxylans; maize bran by-product; white bread; physicochemical properties; proximate composition; functional properties



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1. Introduction

The nixtamalization process consists of cooking maize grain in a lime solution (calcium hydroxide) at temperatures of 85–95 °C for 30–40 min, followed by steeping for 8–16 h to produce nixtamalized dough as a final product and nixtamalized maize bran as by-products [1]. Nixtamalization partially hydrolyzes the cellulose–hemicellulose–lignin–protein structure of bran, which facilitates its detachment from maize grain [2].

Nixtamalized maize bran is composed of fibrous carbohydrate constituents, and these have promising potential for dietary fiber extraction [3]. Maize bran by-products are mainly used as raw material for animal feed and incur minimal commercial costs; apart from this, they may also provide commercial and nutritional value in food applications due to the fact that maize bran is largely composed of insoluble dietary fiber [4], with its antioxidant properties having been researched in both in vivo and in vitro studies [5]. The chemical composition of maize bran is 50% heteroxylans, 20% cellulose, 11% protein, 9% starch, 6% phenolic acids, 2% lipids, and 2% ash. Additionally, maize bran is mainly composed of feruloylated arabinoxylans (FAXs), which are associated with cellulose, a water-insoluble polysaccharide in its native form [6].

Arabinoxylans can be extracted from cereal by-products via different methods, such as water, mechanical, chemical, and enzymatic treatments, or a combination of these methods, with cereal bran being the main plant material used for their extraction [7]. In the process of arabinoxylan extraction by the methods previously mentioned, interactions with cell wall components are disrupted, and arabinoxylans are converted to water-soluble arabinoxylans [8,9]. The structure of arabinoxylans consists of a main linear backbone of xylose residues (β -xylopyranosyl form) linked by a β -(1 $\rightarrow$ 4) bond, which branch into arabinose units (α -arabynofuranosyl form), mono- and/or di-substituent at the O-3 (α -3) and/or O-2 (α -2) positions of the xylose. In addition, arabinose units can be esterified with ferulic acid at the C(O)-5 position of arabinose branches, which is a unique feature of arabinoxylans (Figure 1). Moreover, glucuronic acid and acetyl groups are also substituents of arabinoxylans [10–12].

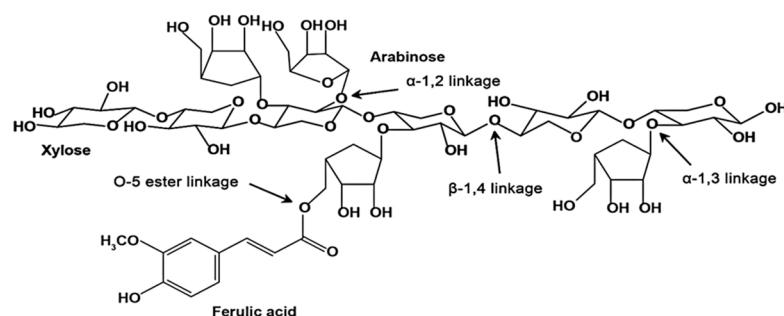


Figure 1. Chemical structure of arabinoxylans [12].

Bread making is currently a leading sector in the food industry. The annual per capita consumption of bread in Mexico is 33.5 kg, with 70–75% being white bread, including sweet bread, cookies, and cakes [13]. The Mexican Official Standard NOM-F-159-S-1979 defines white bread as “the food product made by baking fermented dough, made with wheat flour, drinking water, iodized salt, yeast and other optional ingredients and additives permitted for food production” [14].

In recent years, the incorporation of functional ingredients such as dietary fiber in food formulations, including bread, has garnered interest due to its associated health benefits. The main challenge in the use of dietary fiber polysaccharides is to achieve maximum retention of functionality in the final products to be used [15]. Due to their higher water-holding capacity, soluble fibers in bread formulations are used for water absorption in flour; in addition, their fermentation produces short-chain fatty acids that have some health-promoting properties [16].

Arabinoxylans could be a potential ingredient in baking, as they have effects on water-holding capacity and rheology [12]. The effects of arabinoxylans on bread quality can be explained by molecular interactions with water, gluten, and starch. Water-insoluble arabinoxylans generally have a negative effect on bread quality, whereas water-soluble ara-

binoxylans have a positive effect [11]. In addition, it has been proven that a small addition of water-soluble arabinoxylans has a positive effect on the physicochemical properties of bread; however, a high addition of arabinoxylans has a detrimental effect [17].

Most of the studies conducted on the effect of arabinoxylans in bread quality are based on the use of arabinoxylans primarily extracted from wheat bran, as reviewed by Saeed et al. [18] and Pietiäinen et al. [19]. Therefore, this study focuses on the influence of FAXs from maize bran by-products (obtained from the nixtamalization industry) on the physicochemical, nutritional, and functional properties of white bread.

2. Materials and Methods

2.1. Extraction and Characterization of Arabinoxylans

The extraction and characterization of the arabinoxylans used in this study followed that of Herrera-Balandrano et al.'s research [20]. In brief, nixtamalized maize bran was defatted with ethanol (1:5; w/v) and dried at 40 °C for 12 h. Then, starch was removed from bran by gelatinization in water at 90 °C (1:7; w/v) for 30 min. The maize bran was treated using NaOH 0.5 N, and arabinoxylans were extracted at different time intervals (2, 4, and 6 h); after this process, the arabinoxylans were precipitated using ethanol at 60% and ultimately freeze-dried.

For neutral sugar evaluation, the samples underwent acid hydrolysis with 2 N trifluoroacetic acid at 120 °C for 2 h. The reaction was stopped on ice, and the extracts were evaporated under air at 40 °C and rinsed twice with 200 µL of water. The evaporated extract was solubilized in 500 µL of water and quantified in a Varian 9012 HPLC with a 9040 refractive index detector (Varian, St. Helens, Australia) using a 300 × 7.8 mm Supelcogel Pb column (Supelco, Inc., Bellefonte, PA, USA) eluted with 5 mM H₂SO₄ at 0.6 mL/min at 50 °C. Protein was determined via the Bradford method, and the soluble and insoluble dietary fiber contents of FAXs were quantified by using the Megazyme® total dietary fiber kit based on the AOAC method, 991.43. The neutral sugar composition, protein, and dietary fiber content of the arabinoxylans are shown in Table 1.

Table 1. Composition and dietary fiber content in FAXs from the nixtamalized maize bran by-product used in this study.

FAXs Extraction Time	Composition (%)					Dietary Fiber (%)		
	Xyl	Ara	Gal	Glc	Pro	Ara/Xyl	Soluble	Insoluble
2 h	33.43	27.73	2.52	4.51	1.00	0.82	86.56	ND
4 h	29.69	25.89	2.50	1.98	0.86	0.87	89.95	ND
6 h	30.28	26.64	3.12	1.26	0.62	0.87	86.14	ND

FAXs = feruloylated arabinoxylans; Xyl = xylose; Ara = arabinose; Gal = galactose; Glc = glucose; Pro = protein; ND = not detected. Adapted from Herrera-Balandrano et al. [20].

To evaluate the ferulic acid content in the arabinoxylans, alkaline extraction (saponification) was performed using NaOH 2N for 2 h at 35 °C under argon reflux. The pH was adjusted to 2.0 with 4 N HCl. Phenolics were extracted twice with diethyl ether and evaporated at 30 °C under argon reflux. After this, quantification of ferulic acid monomers, dimers, and trimers was performed via RP-HPLC in a Waters 996 (Millipore Co., Milford, MA, USA) photodiode array detector using a 250 × 4.6 mm Alltima C18 column (Deerfield, IL, USA), and detection was carried out at a wavelength of 280 nm. The results were expressed as milligrams of every monomer, dimer, and trimer of ferulic acid per gram of the sample (mg/g). The total phenol and ferulic acid content of the arabinoxylans is shown in Table 2.

Table 2. Phenolic content in FAXs from the nixtamalized maize bran by-product used in this study.

FAXs Extraction Time	Total Phenols (mg/g)	Ferulic Acid Monomers (mg/g)	Ferulic Acid Oligomers (mg/g)
2 h	9.01	1.943	0.289
4 h	7.16	1.263	0.231
6 h	6.48	0.368	0.107

Adapted from Herrera-Balandrano et al. [20].

2.2. Treatments

A completely randomized design of seven treatments was established in this study. Each treatment involved a combination of white bread plus a specific concentration of FAXs used in a formulation (0.15 and 0.30%) at different extraction times (2 h, 4 h, and 6 h) (Table 3). A control sample was prepared that consisted of white bread without the addition of FAXs.

Table 3. Nomenclature and description of bread treatments.

Treatment Nomenclature	Treatment Description
TC (control)	Bread + 0.00% FAXs
T1	Bread + 0.15% FAXs 2 h
T2	Bread + 0.30% FAXs 2 h
T3	Bread + 0.15% FAXs 4 h
T4	Bread + 0.30% FAXs 4 h
T5	Bread + 0.15% FAXs 6 h
T6	Bread + 0.30% FAXs 6 h

FAXs = feruloylated arabinoxylans; 2 h, 4 h, 6 h = extraction time of FAXs.

2.3. Preparation of White Bread with Added FAXs

The preparation of white bread and the ingredients used in the formulations followed those by Niño-Medina et al. [21], with some modifications (Table 4). FAXs were added together with dry ingredients such as wheat flour, yeast, salt, and sugar.

Table 4. Ingredients for making bread.

Ingredient	Quantity
Flour	100 g
Water	62 mL
Salt	3 g
Sugar	4 g
Yeast	3 g
FAXs	0.15/0.30 g

After mixing the powdered ingredients for 1 min, water was added, and the mixture was kneaded for 7 min until a homogeneous dough was obtained. The dough was divided into multiple batches, placed in aluminum molds, and fermented at 40 °C for 27 min. Following fermentation, the batches were baked at 170 °C for 28 min. The samples were cooled to room temperature, and corresponding tests were performed after they were refrigerated.

2.4. Texture Profile Analysis (TPA)

TPA was conducted using a TA.XT2 Stable Micro Systems Texturometer (Surrey, England) using the Texture Exponent software. TPA was performed by evaluating parameters of hardness, elasticity, cohesiveness, gumminess, chewiness, and resilience. Measurements were taken of the bread crust in triplicate for all treatments, with the entire loaf subjected to a compression of 40% of average height in every treatment [21].

2.5. Color Evaluation

Color measurement was conducted directly on the bread, randomly taking three measurements of the crusts of each of the loaves using a Konica Minolta CR-20 colorimeter (Tokyo, Japan). Chromatic parameters were obtained using the CIELAB (L^* , a^* , b^*) and CIELCH (L^* , C^* , h^*) systems, according to Commission Internationale De L'ecclairage [22], where L^* refers to luminosity (0 = black, 100 = white), a^* indicates red (positive a) or green (negative a), b^* indicates yellow (positive b) or blue (negative b), C^* (chroma) is the saturation level of h^* , and h^* is the hue angle (0° = red, 90° = yellow, 180° = green, and 270° = blue). The online software ColorHexa [23] was used to obtain a color image using the L, a, and b values, and the equipment was calibrated before measurement.

Total color difference (ΔE) was calculated in accordance with research by Onishi et al. [24], as follows: $\left[(L_1^* - L_0^*)^2 + (a_1^* - a_0^*)^2 + (b_1^* - b_0^*)^2 \right]^{0.5}$, where L_0^* , a_0^* , b_0^* are the color values of the TC, and L_1^* , a_1^* , b_1^* are the color values of treatments, including FAXs (T1 to T6).

2.6. Proximate Composition

Proximate analysis of the bread samples was conducted following the methods specified by the Association of Official Analytical Chemists [25]. The content of moisture (method 925.09), ash (method 923.03), crude protein (method 960.52), fat (method 923.03), and crude fiber (method 920.86) were measured, and carbohydrates content was obtained by difference.

2.7. Total Phenols and Antioxidant Capacity

The extraction of phenolics was performed in accordance with the procedure described by Niño-Medina et al. [26] for bound phenolics, with minor modifications. Extracts were obtained by taking 50 mg of the sample and dissolving it in 3 mL of 2-M NaOH, leaving it to shake in the dark for 2 h. Then, the pH was adjusted to 2.0 using a concentrated HCl solution, recovering the phenolic compounds with 3 mL of diethyl ether twice. The diethyl ether extracts were pooled and dried under gas nitrogen flow, and the phenolic compounds were resuspended in 2 mL of 80% methanol.

Total phenols (TPs), DPPH, ABTS, and FRAP were determined by following the method reported by López-Contreras et al. [27]. For total phenols, 200 μ L of the extract, 2600 μ L of distilled water, and 200 μ L of Folin–Ciocalteu reagent were mixed to form a solution. After 5 min, 2000 μ L of 7% Na_2CO_3 was added, the solution was stirred for 30 s, and the reaction was conducted in the dark for 90 min. The absorbance of the samples was measured at 750 nm, and their concentrations were obtained using the linear regression equation of the calibration curve established with ferulic acid at concentrations of 0–200 mg/L. The results were expressed as milligrams of ferulic acid equivalents per gram of the sample (mgFAE/g).

To determine antioxidant capacity, a DPPH (2,2-diphenyl-1-picrylhydrazyl) radical decolorization test was performed by mixing 1500 μ L of DPPH and 50 μ L of the extract. The reaction was conducted for 30 min in the dark, and the absorbance of the samples was measured at 515 nm using 80% methanol for reduction to zero and as a control for the initial absorbance measurement.

For the ABTS 2,2'-azinobis (3-ethylbenzothiazole-6-sulfonic acid) technique, a stock solution was prepared by mixing 7.4 mM ABTS reagent and 2.6 mM potassium persulfate solution (1:1); then, the mixture was left in the dark for 12 h. The solution was diluted in 80% methanol, which was adjusted to give a 0.700 nm spectrophotometer reading at 734 nm. For each sample, 50 μ L of the extract was taken and allowed to react with 1500 μ L of ABTS for 30 min in the dark. After the incubation, absorbance was read at 734 nm.

The FRAP technique was performed by mixing the following stock solutions: 300 mM sodium acetate buffer, 10 mM TPTZ (2,4,6-tripyridyltriazine complex), and 20 mM iron chloride hexahydrate at a 10:1:1 ratio at 37 °C. For each sample, 50 µL was taken and mixed with 1500 µL of the FRAP solution, with the mixture incubated for 30 min at 37 °C. Readings were taken at 593 nm using a spectrophotometer.

The levels of every antioxidant capacity assay were obtained using a linear regression equation for the calibration curves established with Trolox at concentrations of 0–500 µmol/L. The result was expressed in micromoles of Trolox equivalents per gram of the sample (µmolTE/g).

2.8. Statistical Analysis

Evaluation of the variables were performed in triplicate, and data were analyzed using a completely random design with the use of Minitab 17.0 [28]. A generalized linear model procedure was used for data analysis with the following statistical model: $y_{ij} = \mu + T_i + \epsilon_{ij}$. Here, y_{ij} is the response variables; μ represents the overall mean; T_i represents the treatment effect (TC–T6); and ϵ_{ij} represents random error with zero mean and variance σ^2 [$\epsilon_{ij} \sim N(0, \sigma^2)$]. Tukey's test was performed to compare the means between the treatments. A type I error probability of less than 0.05 ($p < 0.05$) was established to demonstrate the effect of the treatments.

3. Results and Discussion

3.1. Texture Profile Analysis (TPA)

During texture profile analysis, the food is compressed twice, and mechanical parameters were calculated from the force–deformation curves obtained [29]. The results of the TPA, obtained by adding FAXs in the production of white bread, are shown in Table 5. The control sample achieved the highest hardness score of 64.43 N, demonstrating a statistically significant difference compared to the scores of the treated samples (T1 to T6), which ranged from 34.32 N to 51.03 N. It was evident that all the samples formulated with the addition of arabinoxylans were softer than the control sample. The results obtained for springiness showed statistical differences between the treated samples, with T2 and T6 obtaining the highest levels in this parameter with a value of 0.94, whereas TC achieved the lowest value, with 0.92 N. Cohesiveness scores ranged from 0.64 N to 0.70 N, and these also showed statistically significant differences, with T4 and T6 achieving the lowest and highest values, respectively. Regarding chewiness, TC and T5 obtained the highest and lowest values, with 36.00 N and 21.43, respectively, leading to both of these samples being statistically different from all the other treated samples. Regarding resilience, values ranged from 0.20 to 0.34, with TC and T6 achieving the lowest and highest values, respectively, also being statistically different from those of all the other treated samples.

The addition of FAXs to food matrices and their positive interactions with the matrix components mainly depend on their solubility, as previously reported by Herrera-Balandrano et al. [20]. The solubility of the polysaccharides added in these formulations was found to be high, with a FAXs ratio ranging from 0.82 to 0.87, similar to those reported by Morales-Ortega et al. [30].

The hardness of the bread in this study decreased from 21% (T3) to 47% (T6) after using FAXs in a formulation when preparing the bread. Our results agree with those obtained by Buksa et al. [31], who reduced hardness in bread made from rye flour up to 37% after the addition of FAXs at 1%; the explanation for this behavior is that FAXs led to an increase in the volume of bread, and hardness decreased as a result. In addition to this, our results are similar to reports by Ayala-Soto et al. [32], who evaluated the addition of FAXs in the

preparation of gluten-free bread, finding that adding 3% and 6% FAXs to the formulation decreased the hardness of the bread from 15% to 45%.

Table 5. Texture profile analysis parameters of white bread supplemented with FAXs.

Treatments	Parameter				
	Hardness (N)	Springiness	Cohesiveness	Chewiness (N)	Resilience
TC	64.43 ± 7.60 ^a	0.92 ± 0.012 ^b	0.60 ± 0.015 ^c	36.00 ± 4.63 ^a	0.20 ± 0.018 ^c
T1	40.67 ± 1.56 ^{bc}	0.93 ± 0.001 ^{ab}	0.66 ± 0.005 ^{ab}	25.30 ± 0.76 ^{bc}	0.32 ± 0.006 ^{ab}
T2	40.20 ± 2.39 ^{bc}	0.94 ± 0.001 ^a	0.67 ± 0.025 ^{ab}	25.54 ± 2.00 ^{bc}	0.30 ± 0.018 ^{ab}
T3	51.03 ± 0.07 ^b	0.93 ± 0.001 ^{ab}	0.67 ± 0.036 ^{ab}	32.33 ± 1.82 ^{ab}	0.31 ± 0.030 ^{ab}
T4	45.45 ^b ± 6.90 ^{bc}	0.93 ± 0.009 ^{ab}	0.64 ± 0.019 ^{bc}	27.24 ± 4.33 ^{bc}	0.28 ± 0.016 ^b
T5	34.32 ± 1.19 ^c	0.93 ± 0.005 ^{ab}	0.66 ± 0.010 ^{ab}	21.43 ± 1.05 ^c	0.31 ± 0.012 ^{ab}
T6	39.73 ^b ± 3.31 ^{bc}	0.94 ± 0.004 ^a	0.70 ± 0.002 ^a	26.38 ± 2.29 ^{bc}	0.34 ± 0.010 ^a

Different letters in the same column indicate statistically significant differences between treatments ($p < 0.05$). Values are reported as mean ± standard deviation ($n = 3$).

Additionally, the positive effects of arabinoxylans on the texture of bread result from their strong water-binding capacity, which prevents water loss in the initial stages of baking prior to starch gelatinization. It has been suggested that arabinoxylans interact with gluten, leading to the formation of composite, hydrated film networks and increasing the water absorption of dough during the mixing process. Apart from this, the absorbed water acts as a plasticizer in the gluten–starch matrix, decreasing the rigidity of the composite network as a result. These features ultimately aid in decreasing the hardness of bread [33,34].

3.2. Color Evaluation

The color of bread crust is the result of Maillard reaction products, which mainly form as a result of a reaction between free amino acids or proteins and carbonyl groups of reducing sugars. These reactions ultimately produce browning [35].

The results obtained for the crust color evaluation of bread made with FAXs are shown in Table 6. The luminosity values (L^*) of treated samples T5 and T6 were the highest, with values of 58.30 and 59.40, respectively, and they showed significant statistical differences from the other treated samples, whereas the lowest luminosity value was observed in T4, with 50.49.

Table 6. Color parameters in white bread supplemented with FAXs.

Treatments	Parameter						Color View
	L^*	a^*	b^*	C^*	h^*	ΔE	
TC	53.22 ± 0.64 ^{bc}	12.59 ± 0.17 ^a	26.04 ± 0.05 ^a	28.97 ± 0.15 ^a	64.03 ± 0.34 ^c	----	
T1	56.16 ± 2.21 ^{ab}	12.18 ± 0.18 ^{ab}	26.61 ± 0.43 ^a	29.14 ± 0.17 ^a	65.52 ± 0.24 ^{bc}	3.04 ± 2.19 ^{bc}	
T2	53.57 ± 1.69 ^{bc}	12.45 ± 0.41 ^{ab}	27.00 ± 0.06 ^a	29.68 ± 0.11 ^a	65.29 ± 0.54 ^{bc}	2.07 ± 0.68 ^{cd}	
T3	53.99 ± 1.73 ^{bc}	9.86 ± 0.37 ^c	21.82 ± 0.03 ^c	23.60 ± 0.75 ^b	65.01 ± 0.32 ^{bc}	5.30 ± 0.41 ^{ab}	
T4	50.49 ± 0.18 ^c	12.69 ± 0.28 ^a	24.93 ± 0.36 ^b	28.21 ± 0.68 ^a	62.01 ± 1.08 ^d	2.96 ± 0.76 ^{bcd}	
T5	58.30 ± 2.21 ^a	12.16 ± 0.38 ^{ab}	26.84 ± 0.50 ^a	29.28 ± 0.77 ^a	66.47 ± 1.09 ^{ab}	5.20 ± 1.46 ^{ab}	
T6	59.40 ± 0.54 ^a	11.58 ± 0.20 ^b	26.60 ± 0.60 ^a	29.48 ± 0.61 ^a	67.44 ± 0.52 ^a	6.32 ± 0.32 ^a	

L^* = luminosity; a^* = red tendency; b^* = yellow tendency; C^* = saturation; h^* = hue, ΔE = total color difference. The letters in the columns indicate statistically significant differences between the treatments ($p < 0.05$). Values are reported as mean ± standard deviation ($n = 3$).

On the other hand, the a^* parameter also showed significant statistical differences, and T4 exhibited the highest a^* value with 12.69, whereas the lowest was found in T3, with a value of 9.86. The b^* values were lower for the T3 and T4 samples, with values of 21.82 and

24.93, respectively, and they showed a significant statistical difference from all the other samples treated, whose values were in the range of 26.04 to 27.00.

In evaluating chroma (C^*), the only sample that was statistically different was T3, which obtained the lowest value of 23.60; the values of the other samples ranged from 28.21 to 29.97. The results obtained for hue (h^*) showed significant differences between samples, with values ranging from 62.01 to 67.44, and samples T4 and T6 achieving the lowest and highest scores, respectively; however, all of the values fall under the orange zone in the color wheel.

Total color difference (also known as Delta E) (ΔE) is a standard measurement related to human visual assessment of differences in two colors. This concept was developed and introduced by the International Commission on Illumination (CIE) as a standard to measure color changes. The objective of this parameter is to identify small color differences that are larger than those that are just noticeable but smaller than the differences typically scaled in color appearance systems under optimal viewing conditions. The reference set as a guide in determining the perception of color, based on the amount of ΔE values in the range 0–100, is as follows: $\Delta E = 0.0$ – 1.0 —change not perceptible by human eyes; $\Delta E = 1.0$ to 2.0 —only an experienced observer can detect the change; $\Delta E = 2.0$ to 3.5 —an unexperienced observer can detect the change; $\Delta E = 3.5$ to 5.0 —a clear difference in color is noticed; and $\Delta E = 5.0$ or higher—two different colors noticed [36–38].

In this study, the results of the total color difference calculations showed statistically significant differences between samples, with values ranging from 2.07 to 6.32, and with T2 and T6 scoring lower and higher, respectively.

Based on the aforementioned scale, the total color difference between TC and T2, T4, and T1 was only detectable by an experienced observer. On the other hand, the total color difference between TC and T5, T3, and T6 was noticeable as two different colors. Here, we consider the color of the last-mentioned samples undesirable, as consumers will be able to notice the differences in the color of the crust.

In similar research, Zhang et al. [39] evaluated the effects of 2%, 5%, and 10% arabinoxylan-rich fractions obtained from wheat bran on bread baking performance, and the results of the total color difference measurement used were 10.86, 20.79, and 36.76, respectively, which is higher than the results obtained in our experiments.

Additionally, Han et al. [40] used arabinoxylans obtained from wheat processing wastewater, adding amounts of 2.5%, 5.0%, 7.5%, and 10% in a multigrain bread formulation, and they observed total color difference values of 8.33, 13.53, 17.66, and 23.89, respectively, which is higher than our results.

According to Bender et al. [41], when arabinoxylans are used in bread production, the color differences in the bread crust are mainly attributed to the coextracted impurities in arabinoxylan extracts, such as some reducing sugars (glucose and galactose) and protein, which react during the baking process and produce higher Maillard reaction products. In these cases, a higher level of browning is produced. On the other hand, Jiang et al. [42] reported that the free radical scavenging capacity of ferulic acid against intermediary compounds in the Maillard reaction also generates alterations in the reaction products during baking, which also modify the color crust.

3.3. Proximate Composition

The results of proximate composition are shown in Table 7. Here, moisture levels vary significantly among the treated samples, with samples TC and T6 having the highest moisture content of 33.45 and 33.43, in contrast to sample T3, which has the lowest moisture content of 29.31.

Table 7. Proximate composition and water activity of white bread supplemented with FAXs.

Treatments	Component (%)						aw
	Moi	Pro	Fat	Cfb	Ash	Car	
TC	33.45 ± 0.32 ^a	14.02 ± 0.68 ^a	1.00 ± 0.01 ^b	1.11 ± 0.002 ^b	4.58 ± 0.03 ^a	45.80 ± 0.40 ^b	0.92 ± 0.011 ^a
T1	32.49 ± 0.11 ^a	13.73 ± 0.44 ^a	1.11 ± 0.16 ^b	1.16 ± 0.005 ^a	4.60 ± 0.13 ^a	46.88 ± 0.26 ^b	0.89 ± 0.005 ^{ab}
T2	32.62 ± 0.10 ^a	14.21 ± 0.07 ^a	0.68 ± 0.01 ^c	1.15 ± 0.016 ^a	4.58 ± 0.15 ^a	46.73 ± 0.15 ^b	0.89 ± 0.012 ^{ab}
T3	29.31 ± 0.35 ^c	14.14 ± 0.84 ^a	1.14 ± 0.01 ^b	1.15 ± 0.006 ^{ab}	4.50 ± 0.01 ^a	49.73 ± 0.52 ^a	0.88 ± 0.028 ^b
T4	31.09 ± 0.24 ^b	14.55 ± 0.05 ^a	1.69 ± 0.01 ^a	1.15 ± 0.004 ^a	4.73 ± 0.15 ^a	46.76 ± 0.33 ^b	0.89 ± 0.010 ^{ab}
T5	30.49 ± 0.35 ^b	13.82 ± 0.27 ^a	1.44 ± 0.06 ^b	1.15 ± 0.008 ^{ab}	4.42 ± 0.10 ^a	48.66 ± 0.44 ^a	0.90 ± 0.003 ^{ab}
T6	33.43 ± 0.33 ^a	14.01 ± 0.21 ^a	0.88 ± 0.02 ^{bc}	1.16 ± 0.011 ^a	4.51 ± 0.01 ^a	45.99 ± 0.51 ^b	0.89 ± 0.006 ^{ab}

Moi = moisture; Pro = protein; Cfe = crude fiber; Car = carbohydrates; aw = water activity. Different letters in columns indicate statistically significant differences between the treatments ($p < 0.05$). Values are reported as mean ± standard deviation ($n = 3$).

There were no significant differences in protein and ash contents among the treated samples, with values ranging from 13.74 to 14.55 and 4.42 to 4.73, respectively. Differences were found in fat content, with samples T4 and T2 exhibiting the highest values (1.69%) and the lowest values (0.68%), respectively.

T2 and T6 obtained the highest crude fiber values with 1.16%, whereas the control sample had the lowest crude fiber content, with a value of 1.11. All FAX-treated samples had higher crude fiber results than the control sample (TC).

Water activity (aw) was only significantly different between TC and T3, with all the other samples having similar characteristics. The highest aw value was observed in the control sample, and these results could be attributed to the fact that FAXs (soluble dietary fiber) obtained from nixtamalized by-products held the free water in the food product, turning it into bound water, as reported previously by Herrera-Balandrano et al. [43] and by Tse and Schendel [11].

According to Irakli et al. [44], when rice bran was used to partially replace wheat flour in bread, protein, fat, and ash levels increased in the formulation. In our study, the addition of arabinoxylans (dietary fiber) only resulted in a small increase in fat compared to the control sample.

Similarly to our findings, Koegelenberg and Chimpango [45] observed a moisture level of 40.44% for bread made with pure arabinoxylan extracts and 39.69% for bread made with commercial arabinoxylans.

3.4. Total Phenols and Antioxidant Capacity

The results of total phenols and the antioxidant capacity of the evaluated samples showed statistically significant differences, as presented in Table 8. The addition of FAXs positively influenced the total phenol content, as samples with the addition of FAXs ranged from 1.57 mgFAE/g (T6) to 1.98 mgFAE/g (T1) and were higher than TC, which only contained 1.24 mgFAE/g in total phenol content.

The control sample showed lower values than all the treated samples in every antioxidant capacity assay, with values of 9.36, 8.86, and 3.05 $\mu\text{molTE/g}$ in DPPH, ABTS, and FRAP, respectively. In terms of antioxidant capacity, the samples with the addition of FAXs showed results ranging from 16.03 (T5) to 17.51 (T4), 16.43 (T5) to 17.64 (T6), and 4.09 (T3) to 5 $\mu\text{molTE/g}$ in DPPH, ABTS, and FRAP, respectively.

There are few studies on the evaluation of phenolic content and antioxidant capacity in bread made with FAXs. Among these, Snelders et al. [46] evaluated the addition of FAXs in percentages from 1% to 8% in bread formulations, and the antioxidant capacity values observed via an ABTS assay ranged from 11.6 $\mu\text{molTE/g}$ to 20.7 $\mu\text{molTE/g}$, indicating that treatments with FAXs had higher antioxidant capacity potential than the control sample, which was attributable to the bound ferulic acid of FAXs. The values and behavior reported

by these authors agree with the results of this study. On the other hand, Zhang et al. [47] evaluated the use of FAXs as a supplement in bread, discovering that the antioxidant capacity of bread supplemented with FAXs was maintained after an in vitro upper tract digestion assay, indicating that ferulic acid remained attached to FAXs during digestion.

Table 8. Total phenols and antioxidant capacity of white bread supplemented with FAXs.

Treatments	Functional Property			
	Total Phenols *	DPPH **	ABTS **	FRAP **
TC	1.24 ± 0.10 ^c	9.36 ± 0.20 ^c	8.86 ± 0.20 ^b	3.05 ± 0.13 ^b
T1	1.98 ± 0.11 ^a	16.56 ± 0.20 ^{ab}	17.57 ± 0.43 ^a	4.65 ± 0.17 ^a
T2	1.93 ± 0.10 ^a	16.34 ± 0.67 ^{ab}	17.57 ± 0.94 ^a	4.24 ± 0.45 ^a
T3	1.68 ± 0.11 ^{ab}	17.01 ± 0.49 ^{ab}	17.19 ± 0.09 ^a	4.09 ± 0.71 ^{ab}
T4	1.95 ± 0.12 ^a	17.51 ± 0.60 ^a	16.68 ± 0.77 ^a	5.07 ± 0.34 ^a
T5	1.75 ± 0.17 ^{ab}	16.03 ± 0.53 ^b	16.43 ± 0.73 ^a	4.82 ± 0.16 ^a
T6	1.57 ± 0.06 ^b	16.89 ± 0.35 ^{ab}	17.64 ± 0.20 ^a	5.07 ± 0.31 ^a

* mgFAE/g, ** $\mu\text{molTE/g}$, Different letters in columns indicate statistically significant differences between the treatments ($p < 0.05$). Values are reported as mean $\pm$ standard deviation ($n = 3$).

The ferulic acid units that are bound to arabinoxylans are responsible for all FAX-treated samples having higher total phenol content and higher antioxidant capacity levels than the control sample (CT). This could be explained by the structure of ferulic acid, which consists of a phenolic nucleus with an unsaturated side chain; in addition, an OH group in the ortho position is present, together with a methoxyl group that acts as an electron donor. This configuration produces a resonance-stabilized phenoxy radical that has high antioxidant potential [48,49]. Furthermore, ferulic acid oligomers (especially dimers) have been shown to have higher antioxidant levels than ferulic acid in monomer form [50].

4. Conclusions

The results obtained in this study showed that the use of FAXs from nixtamalized maize bran by-products positively influenced the physicochemical and functional properties of bread when used as a baking ingredient. In the texture profile analysis performed, the hardness and chewiness decreased with the use of FAXs. In the color evaluation performed, most of the treated samples had higher luminosity values (L^*), and no changes were observed in chromaticity (C^*). Regarding proximate composition, the content of protein was not affected, the content of crude fiber was higher, and water activity decreased in samples treated with FAXs. With regard to functional properties, the total phenol content and antioxidant capacity of the bread supplemented with FAXs were higher than those of the control sample. We conclude that the addition of FAXs improves the quality of bread, and this information could be useful for both the baking industry and consumers. However, further investigation, specifically sensory analysis and the evaluation of FAX's health-improving effects, is necessary.

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