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Article

Effects of the addition of flaxseed and amaranth on the physicochemical and functional properties of instant-extruded cereals

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Abstract: The addition of flaxseed and amaranth on the physicochemical, functional and microstructural changes of instant-extruded cereals was evaluated. Six different mixtures were made with additions of amaranth (30%-50%) and flaxseed (10% and 15%) using maize grits and minor additives as supplementary ingredients and then extruded in a twin-screw extruder. The extrudates evaluated, had insoluble and soluble fiber contents increased with the proportion of amaranth and flaxseed. The mixture 4 (higher flaxseed content) presented highest soluble fiber percentage (1.9%). Extruded cereals had the lowest viscosity (<99.5 cp) and highest hardness values (5.2 N) whereas the dietary fiber content was highest. Fiber content increase, resulted in a higher water solubility index (WSI) (0.5) and decrease the water absorption index (WAI) (2.5). Amaranth and flaxseed incorporation increased crystallinity, resulting in a larger, and more compact laminar structure. Amaranth and flaxseed addition resulted in extruded cereals with acceptable physicochemical and functional properties.

Keywords: extruded cereals; flaxseed; amaranth; dietary fiber; extrusion-cooking

1. Introduction

Currently, an increasing trend in the demand for processed foods that include pro-health compounds such as soluble fiber is occurring due to evidence for potential health benefits to consumers, such as contributing to a reduction in various types of chronic diseases such as cancer, cardiovascular disease, type II diabetes and various gastrointestinal disorders [1]. The development of products and processes that incorporate high-fiber ingredients without altering the chemical and sensory properties of the processed foods are of interest and desirable to meet current consumer trends. A technological alternative for the incorporation of ingredients in processed products is the extrusion-cooking process, a very versatile technique widely used for the development of breakfast cereals and instant foods [2]. Some extruded products marketed as breakfast cereals have significant caloric value. One of the strategies that has allowed the food industry to reduce the energy density and produce more healthy products has been the incorporation of dietary fiber. However, the addition of dietary fiber during extrusion, especially if it is insoluble, results in products with less expansion and crispness and a higher bulk density and hardness, which are properties less preferred by consumers [3]. These characteristics can be explained by the interactions of fiber with starch that impact the mechanisms of starch gelatinization. These, in turn, are related to water absorption of the extrudates and other physicochemical transformations that occur during extrusion, such as viscoelastic properties associated with the stabilizing membranes of the bubbles formed during bubble growth in the final product [4]. Adding insoluble fiber to extruded products has been shown to decrease the proportion of starch in the food matrix, thereby reducing the water absorption capacity and, in turn, the viscosity caused by the gelatinization of the starch, which results in a reduction in the expansion of the final product [3]. A sectional reduction in the expansion of extruded products has been reported by Brennan et al. [5]. An increase in insoluble fiber results in structures with a high number of small cells and a high cell density. Several authors have reported that the bulk density is increased by adding insoluble fiber to extruded products [5,6]. Also extrusion process has been reported to cause significant effects on the dietary fiber content of extrudates [3]. These effects include the formation of resistant starch that may occur during extrusion and the formation of covalent interactions between macronutrients and insoluble components (such as insoluble fiber) that make the extrudates indigestible by amylase or protease activity [7].

The development of ready-to-eat products with high fiber contents (6 g of fiber/100 g of product) [8], it is based on the use of ingredients that meet this requirement and at the same time, provide numerous health benefits. Flaxseed and amaranth have been used specifically in extruded products due to their health benefits for consumers, but, usually, they are used individually [9–13]. Amaranth contains a good balance of amino acids, including the essential amino acid lysine, which is present in limited amounts in most cereals [14,15]. Flaxseed is low in carbohydrates (sugars and starches), high in fiber, protein and rich in polyunsaturated fatty acids, particularly alpha-linolenic acid (ALA or ALN) and linoleic acid (AL), omega-3 and omega-6 essential fatty acids, respectively [16,17]. An especially important component of these two grains is soluble fiber, considered a functional ingredient because it generates high-viscosity products by causing the gelation of chyme. Chyme acts as a network to capture glucose and cholesterol molecules in their passage through the gut, hindering their absorption and thereby decreasing blood glucose and cholesterol levels, and resulting in beneficial health effects [18].

Despite the benefits of both amaranth and flaxseed, little information has been reported on the combined effect of both ingredients on the physicochemical and functional properties of extruded products, important aspects that could be related to the functionality and acceptability of the products. Therefore, the aim of this study was to develop an extruded breakfast cereal with high-fiber ingredients and to evaluate the effects of the addition of flaxseed and amaranth on the physicochemical and functional properties of instant-extruded cereals.

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2. Materials and Methods

2.1 Materials

Amaranth (*Amaranthus hypochondriacus*), flaxseed (*Linum usitatissimum*), maize grits (grits No. 4) and minor ingredients such as sucralose, cocoa and cinnamon were obtained from a local seed distribution store (Chihuahua, Chihuahua, México). Raw materials were placed in polyethylene bags and stored at room temperature until preparation and use. All the starting materials were analysed in a proximate analysis in accordance with official methods analysis [19].

2.2. Chemicals

Hydrochloric acid 37.2%, sulphuric acid 97.9%, hexane 99.8%, ethanol 99.9%, and boric acid 99.5% were all analytical grade and obtained from J.T. Baker (Mexico City, Mexico). Analytical grade sodium hydroxide (97.0%) was obtained from Sigma-Aldrich (St. Louis MO, USA). The selenium reaction mixture was obtained from Merck (Darmstadt, Germany). The kit for soluble and insoluble dietary fiber was obtained from Sigma-Aldrich (St. Louis MO, USA).

2.3. Methods

2.3.1. Mixtures preparation

Different mixtures of amaranth and flaxseed were made under a completely randomized design of two factors. Manipulate factors were flaxseed levels 10 and 15% and amaranth levels 30, 40% and 50%. The combination of levels of each factor generated a treatment structure in a complete 2X3 factorial arrangement. Nevertheless, to obtain a product with acceptable physicochemical and sensory characteristics, percentages of fat and crude fiber lower than 5.6% and 2.5%, respectively, were set, according to the literature [2,20]. These percentages were achieved by the addition of maize grits and minor ingredients (sucralose, cocoa and cinnamon). The combination of these factors and the addition of maize grits resulted in six different mixtures study (Table 1). The ingredients of each of the six treatments were mixed and evenly distributed in an industrial mixer (Bathammex, Mexico City, Mexico) for five minutes.

Table 1. Proportion of ingredients of the different mixtures

Treatment	Amaranth (%)	Flaxseed (%)	Maize grits (%)
Mixture 1	8.6	25.9	65.5
Mixture 2	7.7	30.7	61.6
Mixture 3	6.9	34.7	58.4
Mixture 4	9.8	19.6	70.6
Mixture 5	9.0	24	67
Mixture 6	8.3	27.7	64

2.3.2. Extrusion process

For the extrusion process, we used a twin-screw corotating extruder (BCTM-30, Bühler, AG,

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Uzwil, Switzerland) with a 600 mm length, a length to diameter ratio (L/D) of 20:1, a die opening of 4 mm, the screw configuration was selected specifically to create high levels of shear. The mixtures were fed to the extruder at a rate of 7.5 kg/h and were processed at a speed of 272 rpm. The moisture content of 0.22 kg water/kg dry matter was adjusted within the extruder at a fixed temperature along the extruder for all treatments of 150 °C. The extrudates of each mixture were dried at 120 °C for 15 min in a gas convection oven (Electrolux 10 GN/1, Stockholm, Sweden) with an air cross-flow velocity of 1.5 $m \cdot s^{-1}$ until the extrudates reached a moisture level range of 0.017 kg $H_2O \cdot kgss^{-1}$ to 0.031 kg $H_2O \cdot kgss^{-1}$. 6 kg of each treatment were obtained. The extrudates were packed and stored at room temperature (25 °C) until evaluation. For analysis of the extrudates, three subsamples of each treatment were used.

2.4. Analytical methods

2.4.1. Proximate analysis

The starting materials and extruded cereals were analysed for moisture, protein, fat, crude fiber, and ash content according to methods 950.02, 960.52, 920.39, 962.09, and 923.03 of AOAC [19], respectively. Carbohydrate mass was calculated as the difference between these values and the total mass. The analysis was carried out in triplicate for each treatment, and the results are expressed in g/100 g.

2.4.2. Insoluble and soluble dietary fiber

The insoluble and soluble fiber in the ingredients, the mixtures and the extruded cereals were determined with the total dietary fiber assay kit (Sigma-Aldrich, St. Louis, MO, USA) according to method 991.43 of AOAC [19]. The analysis was carried out in triplicate for each treatment, and the results were expressed in g/100 g.

2.5. Functional properties of the extruded cereals

2.5.1. Water absorption and water solubility indexes

The water absorption index (WAI) and water solubility index (WSI) were determined in triplicate following the procedures described by Anderson et al. [21]. The methods measure the quantity of water incorporated in the flour and the soluble solids that dissolve in water at 30 °C. Samples were weighed (2.5 g) into plastic tubes and mixed with 30 mL of distilled water. The samples were manually shaken, the slurries were centrifuged for 10 min at 3200 g (Thermo IEC model CL3-R, USA), and the supernatant was decanted into pre-weighed porcelain capsules. Capsules were dried for 24 h at 105 °C and weighed. The gel remaining in the tubes after decanting the supernatant was weighed. The ratio between gel-forming solids and soluble solids was measured as grams of water per gram of flour. The WAI was calculated as a percentage of remaining gel weight compared to the pre-dried weight from the extruded cereals. The WSI was calculated as a percentage of the dried supernatant weight compared to the pre-dried weight from the extruded cereals.

2.5.2. Bulk density

The bulk density (BD) was determined according to methods published by Jin et al. [22] in which the ground extrudate (40/60 mesh) was first poured into a cylindrical container. Excess extrudate was

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then scraped off, and the net weight of the powder was then divided by the volume of the container. Bulk density was expressed in kilograms per litre (kg L^{-1}). The analysis was performed in triplicate, and mean values are reported.

2.5.3. Expansion index

The expansion index was reported as the ratio of extruded product diameter and the diameter of the die hole [23]. Values reported were means of sixty measurements.

2.6. Physical properties of the extruded cereals

2.6.1. Textural measurement: hardness and crispness

The evaluation of the hardness and crispness of the extrudates was performed according to the method described by Ding et al. [24], and carried out using a Texture Analyser TA.XT (Texture Technologies Corporation, Scarsdale, New York/Stable Micro Systems, Haslemere, Surrey, UK) configured with a 2 mm punch at a crosshead speed of 5 mm/s and a travel distance of 15 mm. Twenty-four extruded unit samples were taken randomly from each treatment and analysed. A force time curve was recorded and analysed by the Texture Exponent 32 (Surrey, UK) programme to calculate the maximum force (N) to determine the hardness and the area under the curve (N/mm) to determine the crispness.

2.6.2. Pasting properties of the extruded cereals

The amylographic viscosity profile was determined according to the method described by Sánchez-Madrigal et al. [25], with some modification, using a Rapid Visco Analyser (RVA SUPER 4 (Newport Scientific, Sydney, Australia). Flour sample suspensions were prepared by weighing 4 g of milled and dried (50 ± 2 °C, 12 h) extrudates with a 7.5 to 8.5% moisture content and a small particle size (0.25 mm) into an RVA canister and individually adjusting each sample to a total weight of 28 g using distilled water. The rotating paddles were held at 50 °C for 1 min to stabilize the temperature and ensure uniform dispersion and then heated to 92 °C at a rate of 5.6 °C/min, which was held constant for 5 min. The dispersion was then cooled to 50 °C at the same rate and was held at 50 °C for 1 min. The maximum viscosity (MaxV) at 92 °C, the minimum viscosity (MinV or lowest viscosity at the end of heating constant period at 92 °C) and the final viscosity (FinV attained during cooling to or holding at 50 °C) were recorded. The total setback viscosity or viscosity of retrogradation (final viscosity minus minimum viscosity) was calculated from these parameter values. The viscosity with RVA was obtained in RVU units (1 RVU = 10 centipoises). Each treatment was performed twice.

2.7. Scanning electron microscopy

This analysis was performed according to the method described by Sánchez-Madrigal et al. [25]. Flours of each extruded cereal with a particle size <0.15 mm and a moisture content of 1% were stuck to stubs and coated with a gold layer in a high vacuum using a Denton vacuum evaporator (Desk II), set to a pressure of 7.031 X 10^{-2} kg cm^{-2}. The samples were examined using a scanning electron microscope (JSM-5800LV, JEOL, Akishima, Japan) equipped with a secondary electron detector at an acceleration rate of 10 kV.

2.8. Statistical methods

A univariate analysis of variance was performed adjusting a model that included the main effects (flaxseed 10 and 15%; amaranth 30, 40% and 50%) and their interaction (Minitab 16). When the effect of the interaction factor or the main effects was significant (α 0.05); means comparison was performed by Tukey's test [26].

3. Results and Discussion

3.1. Raw materials characterization

Proximate analysis showed a significant difference ($p < 0.05$) between the raw materials for each of the components (Table 2) and indicated that they were of high nutritional value. Is important to highlight the protein content of amaranth is twice as high as than maize grits, whereas flaxseed has high contents of protein, fat and fiber, with a lower carbohydrate content (Table 2). These values are consistent with those reported in the literature [16,27], where it has been shown that amaranth contains a good balance of amino acids, including lysine, an essential amino acid that is not found in most cereals [14]. Flaxseed has been reported to be low in carbohydrates (sugars and starches), high in quality protein, fiber and rich in polyunsaturated fatty acids, particularly alpha-linolenic acid (ALA or ALN), an omega-3 essential fatty acid, and linoleic acid (AL), an omega-6 essential fatty oil [16]. Maize grits was the main source of carbohydrates, as shown in Table 2. In addition, it could be observed that the ingredients are high in dietary fiber and insoluble fiber (Table 3) compared with other reported, [16,27]. In terms of soluble fiber content highlights the flaxseed (9 %), followed by amaranth (1.3 %) and finally the maize grits (0.71 %).

3.2. Extrudate characterization

Proximate analysis of different extruded cereals is shown in Table 2. Chemical characteristics were significantly affected ($p < 0.05$) by the interaction of both amaranth and flaxseed additions at various proportions. The extruded cereals have a high percentage of protein compared to other commercial extruded cereals, which typically have protein content between 5 and 8% [28]. This is due to the addition of both ingredients, amaranth (17.4 %) and flaxseed (22.4 %), which have a high protein content (Table 2), increasing the protein content in each of the mixtures extruded. Additionally, the addition of amaranth could lead to a good balance of amino acids because it contains lysine is an essential amino acid, which is not found in most cereals [14], whereas flaxseed protein is rich in arginine, aspartic acid and glutamic acid, while lysine is limiting [16]. Similar findings were reported for extruded amaranth with a similar protein content [10].

It is notable that the extrudates had a desirable crude fiber content (<2%; Table 2), with acceptable physicochemical and sensory characteristics for the consumer without a negative effect on the caloric and nutrient content, which could especially benefit young children [2]. Additionally, the fat content in all extruded cereals was below the minimum acceptable value of 5.6% [20], creating low-fat products also from the nutritional point of view (<5%).

Table 2. Proximate composition of the raw materials and of the extruded cereals*.

Component (%)	Amaranth	Flaxseed	Maize grits	Mixture 1	Mixture 2	Mixture 3	Mixture 4	Mixture 5	Mixture 6
Moisture	1.4^{c}	5.3^{b}	11.3^{a}	3.0^{a}	2.1^{bc}	2.4^{ab}	3.1^{a}	1.7^{c}	2.8^{ab}
Crude Fat	7.1^{b}	37.2^{a}	0.99^{c}	2.6^{b}	2.7^{b}	2.6^{b}	2.7^{b}	2.4^{b}	3.0^{a}
Crude Fiber	3.2^{b}	16.3^{a}	0.64^{c}	1.5^{b}	1.7^{ab}	1.9^{a}	1.7^{ab}	1.7^{ab}	1.9^{a}
Ash	3.0^{b}	3.3^{a}	0.55^{c}	1.6^{c}	1.7^{ab}	1.8^{a}	1.6^{c}	1.6^{c}	1.7^{b}
Crude Protein	17.4^{b}	22.4^{a}	8.7^{c}	12.2^{ab}	12.4^{a}	12.0^{bc}	12.3^{ab}	11.7^{c}	12.3^{ab}
Carbohydrate	67.9	15.5	77.8	79.1	79.4	79.3	78.6	80.9	78.3

*Means ± standard error (SE). Raw materials: SE moisture, 0.01; SE fat, 0.18; SE crude fiber, 0.05; SE ash, 0.03; SE protein, 0.2. Extruded cereals: SE moisture, 0.13; SE fat, 0.06; SE crude fiber, 0.05; SE ash, 0.02; SE protein, 0.08. Means by files with different letters show significant difference, contrast test ($p < 0.05$). Carbohydrate was calculated by difference.

Table 3. Dietary fiber content of the raw materials and of the mixtures without extruding*.

Component (%)	Amaranth	Flaxseed	Maize grits	Mixture 1	Mixture 2	Mixture 3	Mixture 4	Mixture 5	Mixture 6
SDF	1.3^{b}	9.0^{a}	0.71^{b}	0.6^{b}	1.3^{a}	1.3^{a}	1.5^{a}	1.6^{a}	1.2^{a}
IDF	11.9^{b}	50.6^{a}	7.3^{c}	9.3^{cd}	9.7^{c}	8.9^{d}	11.6^{b}	12.0^{b}	12.6^{a}
TDF	13.2^{b}	59.6^{a}	8.0^{c}	9.9^{d}	11.0^{c}	10.2^{d}	13.1^{b}	13.6^{ab}	13.8^{a}

*Means ± standard error (SE). Raw materials: SE soluble dietary fiber, 0.23; SE insoluble dietary fiber, 0.15; SE total dietary fiber, 0.32. Mixtures: SE soluble dietary fiber, 0.099; SE insoluble dietary fiber, 0.054; total dietary fiber, 0.11. Means by files with different letters show significant difference, contrast test ($p < 0.05$). SDF, soluble dietary fiber; IDF, insoluble dietary fiber; TDF, total dietary fiber.

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3.3. Dietary, insoluble and soluble fiber

Table 3 showing the soluble, insoluble fiber content of the different mixtures before being subjected to the extrusion process, in which the addition of flaxseed significant affected the soluble and total dietary fiber content. After the extrusion process of the mixtures, the results for the dietary fiber content show a significant difference between treatments ($p < 0.05$), indicating that variation in the proportions of ingredients (amaranth and flaxseed) and their interaction, significantly affects the content of both insoluble and soluble fiber in extruded cereals (Figure 1). Additionally, it could be observed that the extrusion process caused an increase in the soluble fiber content and a decrease in insoluble fiber compared to the non-extruded mixtures (Table 3, Figure 1). The extruded cereals from the mixtures 3, 4 and 5 had the highest percentage of soluble fiber but did not present a significant difference. This can be explained by the fact that extrudates 4 and 5 had the highest content of flaxseed (9.8% and 9% respectively), which is a significant source of soluble fiber, reaching up to 9 % (Table 3). In turn, extrudate 3 had the highest percentage of amaranth and flaxseed (41.7%), which resulted in high soluble fiber content. The rest of the extrudates (mixtures 1, 2 and 6) had a low percentage of soluble fiber without showing significant differences among them (Figure 1) because they had low percentages of flaxseed. Various studies have shown that dietary fiber, especially soluble fiber in extrudates, increases when they are subjected to the extrusion process [29]; during the extrusion process, shear stress caused by high screw speed occurs, which together with the high process temperatures causes chemical bond breakage, creating smaller particles that are solubilized as xylose, glucose, arabinose oligos and polymers, and, preferentially, slightly branched arabinoxylans [29,30]. Additionally, other biomolecules such as starch undergo structure changes, lead to the formation of resistant starch, another possible mechanism causing the increase in fiber during the extrusion process [7].

In relation to fiber content in food products, can be considered high in fiber if it contains 6 g of fiber per 100 g product [8]. Therefore, the extruded cereals are high-fiber products (Figure 1), due to the addition of amaranth and flaxseed, which contain a high percentage of dietary fiber; 13.1 and 59.6 % respectively (Table 3). Furthermore, with regard to the addition of high fiber ingredients, the extrusion process may be another important factor in the increase of dietary fiber in extruded cereals.

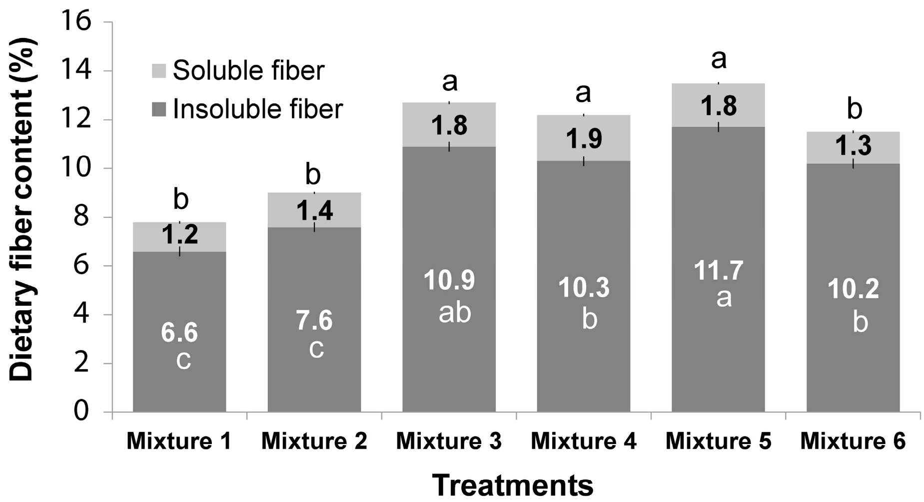

Figure 1. Insoluble and soluble dietary fiber content for columns and colour of extruded cereals. Means ± standard error (SE), SE insoluble fiber, 0.2; SE soluble fiber, 0.05. Means by columns and colours with different letters show significant differences based on contrast tests ($p < 0.05$).

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3.4. Water absorption and water solubility indexes

The WAI in the extruded cereals was significantly affected ($p < 0.05$) by the addition of the ingredients (flaxseed and amaranth) and their interaction, in the mixtures (Table 4). The extruded cereal from mixture 4 (9.8% flaxseed, 19.6% amaranth, and 70.6% maize grits) resulted in high WAI values due to its high maize grits (cornstarch) content, which, during the extrusion process, undergoes pronounced changes in gelatinization properties, favouring a higher water absorption. This is consistent with the amylographic viscosity profile, where the extrudates with the highest values of viscosity have the highest values of WAI (Figure 2). On the other hand, extrudate 3 (6.9% flaxseed, 34.7% amaranth, and 58.4% maize grits) had a low maize grits (cornstarch) content but the highest content of high fiber ingredients (41.7% amaranth and flax) and showed low WAI values, similar to extrudates 2 and 6 (Table 4). Similar effects of fiber addition on extruded products were reported by Altan et al. [31] for the extrusion of barley mixtures with tomato pomace.

WSI is an indicator of the degradation of molecular components, an example is the amount of soluble polysaccharide of the starch released after extrusion, which is a measurement of the degree of conversion of starch during extrusion [24]. Table 4 shows significant effects on the WSI of the extruded cereals by the addition of ingredients, amaranth and flaxseed, in the mixtures. Extrudates from mixtures 2, 5 and 6, with a high fiber content (Table 4), had the highest WSI values (0.5), whereas the extrudate from mixture 1, with a low fiber content, and the extrudate from mixture 4, with a high starch content, had the lowest WSI values (0.46 and 0.45, respectively). The increase in the fiber content causes an increase in the WSI values, which can be attributed to the rupture of the structural polysaccharides by the extrusion process. Similar WSI results in extruded products were reported during the extrusion of rice flour and maize blends with additions of defatted flaxseed meal (DFM). This is because DFM is high in fiber and the fiber molecules disrupt the continuous structure of the melt in the extruder, hindering elastic deformation during extrusion [13]. Similar effects on WSI values for the addition of fiber in extruded products were reported by Altan et al. [31].

3.5. Bulk density

The bulk density of extruded cereals shows significant changes ($p < 0.05$) due to the addition of the ingredients in the mixtures (Table 4). The extrudate from mixture 6, with a high fat content, presented the lowest density value due to the fat content. This is because fat has low density values and also because the oils contained in the cereals, when extruded, undergo an emulsion process due to the strong pressure to which they are subjected. The fine drops of fat are coated by starches and proteins, leaving the fat encapsulated and causing a decrease in the density [32]. The extruded cereals from the mixtures 2 and 4 were high in protein and presented the highest density values. The rigid tertiary structure, high cohesiveness, high molecular weights and structural functions in cereal proteins such as corn, can increase the density of food products [33]. Ryu et al. [23], reported that the density of an extruded product is strongly affected by water, the amount of fiber, and the fat and starch content. The extruded cereals from the mixtures 6 and 3, with similar amaranth contents (27.6 and 34.7 g/100 g respectively), resulted in the lowest density values (4.6 and 4.7 kg L^{-1}, respectively; Table 4). These results are consistent with those shown by Ilo et al. [9], who evaluated the effect of extrusion-cooking process on the properties of extruded rice flour and amaranth blends and observed that amaranth had an important influence on the product density, resulting in a minimum value at an amaranth content of 30 g/100 g. Another report showed that the extrusion of rice flour and corn

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fortified with linseed leads to increases in the bulk density values [12].

3.6. Expansion index

The addition of the different ingredients in their different proportions, and their interaction, significantly affected the expansion of extruded cereals ($p < 0.05$) (Table 4). The expansion characteristic of instant extruded cereals is very important since it is directly related to consumer acceptability, because extruded cereals typically have an inflated, lightweight and crunchy structure [34]. The presence of starch is an important factor for adequate expansion in extruded products [35]. For example, the extrudate of the mixture 4 containing a higher percentage of maize grits (starch) showed a higher expansion index (3.33), followed by the extruded mixture 5 (3.17) and finally the extruded cereals containing less amount of maize grits and that did not have significant difference between them presented the lower values of expansion index, these were the extruded ones of mixtures 1, 2 and 6 (Table 4). It should be noted that, as they contain a higher starch content, mixtures with higher expansion values also contain a lower percentage of the components with high dietary fiber content: amaranth and flaxseed; while mixtures 1, 2 and 6 with higher content of these ingredients presented lower expansion rates, because a high content of dietary fiber can significantly affect the expansion of extruded products. The effect of the fiber on the expansion of the extruded products depends mainly on its interactions with the starch and, therefore, on the type and amount of fiber. Insoluble fiber significantly reduces expansion volumes and increases the density of extruded products; conversely, soluble fiber leads to better expansion volumes, while less affecting the bulk density of the extruded products compared to the insoluble fiber components. The difference in expansion behavior between soluble and insoluble fiber can be explained by their interactions with starch, differences in water absorption and plasticizing behavior, but also by the physicochemical transformations they undergo during extrusion [3]. This is consistent with the results reported by Altan et al. [31], who made extruded barley and tomato pomace, noting that by increasing the level of tomato pomace, the expansion of the final product decreased, because the increase in the rate of expansion depends on the gelatinization of the starch that decreased when adding the tomato pomace (fiber) to the different mixtures. Several authors have reported that the expansion index decreased by adding insoluble fiber to extruded products [35–37].

3.7. Textural measurement (hardness and crispness)

An important quality parameter of ready-to-eat cereals is texture. Table 4 shows the values of hardness and crispness of extruded cereals made from the different mixtures of ingredients. The crispness of the extruded cereals was not significantly different ($p > 0.05$) among the various mixtures. Similar findings have been reported for corn extruded with amaranth, where it was found that an amaranth content from 20 to 35% had no substantial effect on crispiness [11], in ranges similar to those used in this study (19.6-34.7%).

Table 4. Functional and physical properties of the extruded cereals*.

Treatments	BD (Kg L^{-1})	EI	WSI	WAI	Hardness (N)	Crispness (N/mm)	MaxV (cp)	MinV (cp)	FinV (cp)	Setback viscosity (cp)
Mixture 1	0.5^{a}	3.11^{c}	0.46^{bc}	2.9^{b}	5.0^{ab}	26.9^{a}	86.2^{b}	69.9^{abc}	151.0^{a}	81.1^{a}
Mixture 2	0.49^{abc}	3.10^{c}	0.5^{a}	2.5^{c}	4.9^{bc}	25.4^{a}	75.3^{bc}	65.7^{bc}	124.5^{bc}	58.9^{c}
Mixture 3	0.47^{cd}	3.06^{d}	0.49^{ab}	2.5^{c}	4.7^{c}	24.9^{a}	66.7^{c}	63.2^{c}	111.1^{c}	48.0^{c}
Mixture 4	0.49^{ab}	3.33^{a}	0.45^{c}	3.4^{a}	4.7^{c}	25.7^{a}	99.3^{a}	79.8^{a}	162.5^{a}	82.7^{a}
Mixture 5	0.47^{bcd}	3.17^{b}	0.5^{a}	2.9^{b}	5.2^{a}	26.7^{a}	85.6^{b}	75.2^{ab}	154.8^{a}	75.1^{ab}
Mixture 6	0.46^{d}	3.09^{c}	0.5^{a}	2.5^{c}	4.8^{bc}	26.1^{a}	76.1^{bc}	70.2^{abc}	130.5^{b}	60.5^{bc}

*Means ± standard error (SE). SE expansion index, 0.007; SE bulk density, 0.006; SE WSI, 0.007; SE WAI, 0.05; SE Hardness, 0.16; SE Crispness, 1.3; SE MaxV, 2.4; SE MinV, 2.3; SE FinV, 2.8; SE setback viscosity, 2.9. Means by columns with different letters show significant difference, contrast test ($p < 0.05$). BD, bulk density; EI, expansion index; MaxV, maximum viscosity; MinV, minimum viscosity; FinV, final viscosity.

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However, the hardness of the cereals was significantly affected ($p < 0.05$) by the added ingredients (flaxseed and amaranth) and their interaction. Extruded cereals from mixture 5, which had a high dietary fiber content showed higher hardness, characteristics typical of what these ingredients impat to food products. Indeed, the addition of dietary fiber most often leads to reduce expansion volumes and increases the density of the extruded cereals, inducing harder textures and less crispiness [3]. This result is consistent with the results reported by Ganorkar and Jain [12], who showed that an increase in added flaxseed causes an increase in the hardness of the extruded products. In other studies, such as that by Brennan et al. [5], have reported an increased breaking force with increasing wheat bran content up to 15% in extruded breakfast cereals. In contrast, it has also been reported that soluble fibers, such as inulin, deliver a more favourable texture compared to insoluble fibers, such as cereal bran fiber [6]. This was corroborated by Brennan et al. [5], who observed a slight change when adding either inulin or guar gum to extruded corn flour. This can be corroborated in our results, where the extruded cereals from mixture 3 and 4 (Figure 1) contained the highest percentage of soluble fiber and the lowest hardness values (4.7 N; Table 4).

3.8. Pasting properties of the extruded cereals

The addition of amaranth and flaxseed to the mixtures significantly affected ($p < 0.05$) the amylographic viscosity profile (RVA) of the extruded cereals (Table 4). The MaxV values obtained for each of the extruded cereals were low (Table 4, Figure 2). Mixture 4, with a high starch content (70.6% maize grits), had the highest MaxV value, whereas mixture 3, with a lower percentage of starch (58.4% maize grits), had the lowest value (Figure 2). A similar trend on other viscosity parameters was observed (e.g., MinV, FinV, and setback viscosity; Table 4), where the starch content strongly influences the pasting properties of the extruded cereal. The structure of the starch granules can be damaged by the extrusion process due to the combination of shear force and temperature inside the barrel [38]. On the other hand, it is possible that the formation of complex structures during extrusion cooking through interactions between starch-lipid complexes and/or starch-protein walls prevents adequate gelatinization of the starch [39,40]. Another factor influencing the pasting properties of extruded cereals is the presence of dietary fiber, which leads to a decrease in the fraction of water-swelling starch, due to its replacement by the fiber [37]. All these factors impede complete starch gelatinization, causing a decrease in viscosity. Similar results were observed in a study where increasing the amaranth level in rice-amaranth blends was shown to generally decrease the viscosity of extruded products [9]. In extruded blends of cornstarch, whey protein concentrate and Agave tequilana fiber, extruded samples with agave fiber showed significantly lower peak, minimum, and final viscosity compared to extruded samples without agave fiber. These results were attributed to the interference of fiber with the gelatinization or water absorption of starch granules [37].

Additionally, low setback values were found for all extruded cereals, indicating low rates of starch retrogradation and syneresis. During cooling, reassociation of starch molecules, especially amylose, will result in the formation of a gel structure, and viscosity will therefore increase to reach the final viscosity. This phase is commonly described as the setback region during which retrogradation and reordering of starch molecules occurs [41].

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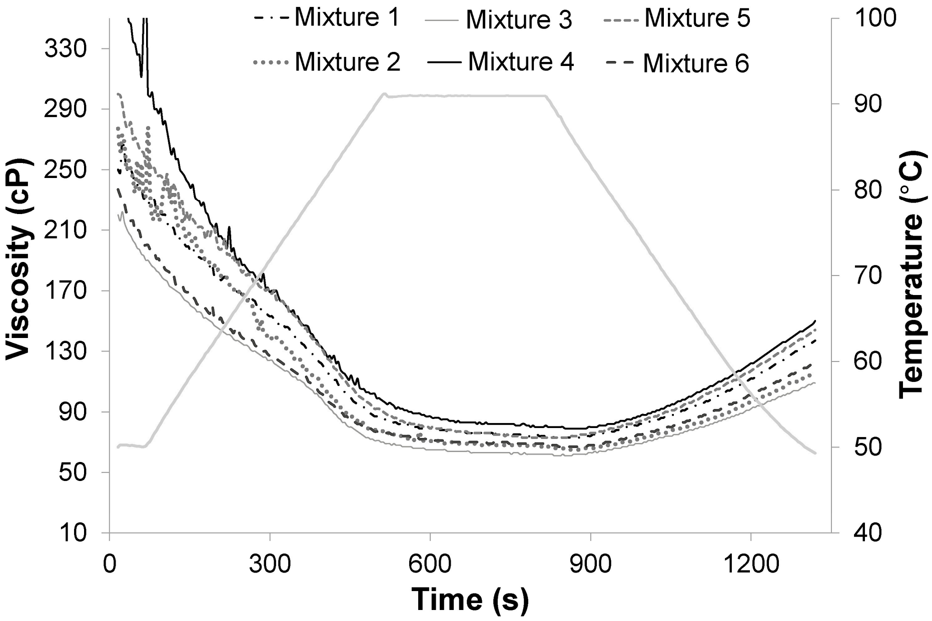

Figure 2. Amylographic viscosity profile of extruded cereals obtained with Rapid Visco Analyser (RVA).

3.9. Scanning electron microscopy

The scanning electron micrographs revealed the impact of the different ingredients (Figure 3). After extrusion, can be observed that combination of shear force and temperature inside the barrel caused microstructural changes in the extrudates of the different treatments [42]. The microsestructural analysis shows, that the addition of amaranth and flaxseed, increased the fiber content in the mixtures, resulting in compact agglomerates, increased crystallinity and larger, more compact laminar structures (Figure 3c-f). This can been attributed to high protein (12%) and fiber (9-13%) content, the later has a tendency to rupture cell walls and promotes breakage of air cells during extrusion, which prevents matrices from expanding [43], resulting in harder textures, higher densities and more compact structures as shown in the micrographs (Figure 3c-f). Similar results were found by Zhang et al. [29] and Cueto et al. [42].

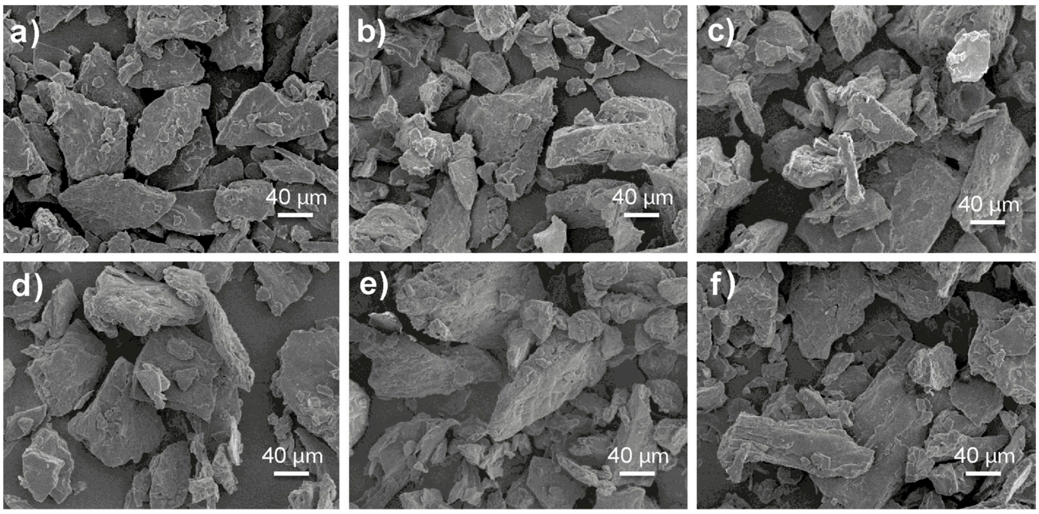

Figure 3. Micrographs of the extruded cereals. (a) Extruded cereal from mixture 1, (b) Extruded cereal from mixture 2, (c) Extruded cereal from mixture 3, (d) Extruded cereal from mixture 4, (e) Extruded cereal from mixture 5, (f) Extruded cereal from mixture 6.

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4. Conclusions

In this study, we show that different levels of bioactive ingredients such as amaranth and flaxseed in the development of extruded breakfast cereal have a significant impact on the functional and physicochemical properties of the cereal. Extruded cereals were obtained with high protein content (>12%), which is more than other commercial breakfast cereals, with a healthy fat content (<5%) and a high content of soluble and insoluble dietary fiber.

Another important ingredient that serves as a basis for the production of extruded breakfast cereal expansion is starch (maize grits). However, with low levels of starch (maize grits) and high levels of fiber in the mixtures, the viscosity development was minimized, the WAI decreased and the WSI increased, resulting in compact agglomerates, increased crystallinity and the formation of a larger, more compact laminar structure. These results suggest that the addition of high-fiber grains such as flaxseed (6.9-9.8%) and amaranth (19.6-34.7%) in breakfast cereals processed by extrusion-cooking results in a product of good nutritional quality, with functional properties and acceptable physicochemical characteristics.

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