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Ohmic Heating as an Emerging Technology for the Improvement of the Technno-Functional Properties of Common Bean Flour †

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Abstract: The common bean (Phaseolus vulgaris L.) is an important part of the diet in Mexico due to its high availability and low cost, which has also been associated with preventing and reducing non-communicable diseases. Due to this, the application of processing technologies has grown to improve the nutritional and bioactive profile of beans. Therefore, the processing of the raw material directly influences the techno-functional properties. The objective of this work was to demonstrate the advantage of ohmic heating over traditional cooking in bean flours, in their techno-functional characteristics and anti-nutritional factors. The results of ohmic heating did not show significant differences compared to traditional cooking; however, ohmic heating obtained higher values in foaming capacity and emulsifying. In addition, did not modify the protein solubility profile, reduced trypsin inhibitors by 25.42 to 57.44%. This suggest that ohmic heating can be an alternative to conventional treatments, in addition to reducing processing time, with greater energy efficiency, not presenting nutrient leaching.

Keywords: Ohmic heating; common bean; techno-functionality

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1. Introduction

The common bean is classified by the FAO as the most important food legume for human consumption and essential for the food security of a nation. In Mexico, it is the second in importance, only after corn (FIRA, 2016), which is distinguished by its high protein content, in addition to its contribution of carbohydrates, dietary fiber, vitamins, minerals and phytochemicals. In addition to this, the consumption of beans has been associated with the prevention and reduction of several chronic non-communicable diseases, such as cancer, obesity, diabetes and cardiovascular diseases [1], due to its content of phytochemicals, with related antioxidant activity, antimutagenic and anticancer effects [2]. However, the consumption of beans in Mexico has decreased in the last thirty years, from 16 kg to 9.9 kg of annual consumption per capita. To improve this consumption, its use as flour has been proposed, to be used as a techno-functional ingredient, to increase the nutritional value of various foods, such as tortillas, breads, cookies, fried foods, snacks, etc., because consumers prefer foods with perceived healthy properties and, at the same time, convenient for consumption, storage and handling [3].

On the other hand, the techno-functional properties are physicochemical characteristics, which in legume flours mainly depend on the presence and behavior of proteins and carbohydrates in the food matrix [2]. These properties are found by the composition and molecular structure of the individual components and the interactions that are sought between them. These characteristics are influenced by the type of processing the food is subjected to and have a direct impact on the final quality of the product.

Recently, ohmic heating has proven to be an alternative for food processing, with advantages over other technologies, due to rapid and uniform heating, which translates into reduced processing time, uniform distribution of heat in food and an energy efficiency of 90% [4]. In addition, it allows the conservation of the nutritional constituents of the raw material. This heating occurs when an electric current passes through a food causing an increase in the temperature inside it as a consequence of the resistance offered by the food to the passage of electric current (Joule effect). The amount of heat generated is directly related to the electric current and the applied [4], so it must be considered: the amount of water and ions, the shape and size of the particle, the viscosity and other characteristics of the feed [5]. However, no studies have been reported evaluating the effect of bean processing by ohmic heating. Therefore, the objective of this work was to demonstrate the advantage of ohmic heating over traditional cooking in bean flours, evaluating and comparing their techno-functional characteristics and antinutritional factors.

2. Materials and Methods

2.1. Biological Material

Seeds of common black bean (Phaseolus vulgaris L.) variety San Luis, harvest 2017 from Zacatecas, Mexico, donated by the National Institute of Agricultural and Livestock Forestry Research (INIFAP, Mexico) were used.

2.2. Ohmic Heating Cooking (OH)

The samples were conditioned with one part water and one part beans (1:1 ratio) 12 h before cooking. A batch type ohmic heater was used. The samples were ohmically heated until reaching the final heating temperature (110 °C), for 10 min and 15 min at a constant voltage of 60 V. Once the samples had been processed, they were dried in a dehydrator at 50 °C for 48 h Later they were ground (Coffee Mill) and sieved through a No. 60 mesh to obtain the flour. All ohmic flours were stored in 50 mL Corning ™ tubes at 4 °C until use.

2.3. Traditional Cooking (TC)

The beans were cooked using a "traditional" process. A 1:5 (p/v) ratio was placed and boiled for 2.5 h. The beans, as well as the cooking broth, were dehydrated at a temperature of 50 °C for 48 h. The dehydrated beans were then ground in a coffee grinder and sieved with a No. 60 mesh. Traditional Cooked Flours were stored in 50 mL Corning tubes at 4 °C until use.

2.4. Crude (C)

The raw sample was ground in a coffee grinder and sieved with a No. 60 mesh. Each flour was stored in 50 mL Corning tubes at 4 °C until use.

2.5. Water Absorption Index (WAI)

The WAI was determined with 5 g of dehydrated flour were mixed in 30 mL of distilled water in a previously weighed 50 mL centrifuge tube, vortexed for 1 min and centrifuged at 3000× *g* for 10 min at 25 °C. It was discarded the supernatant liquid and the tubes were drained, for 10 min on a paper towel and the tube was weighed. Results are expressed as grams of water retained per gram of dry solid (*g*/*g*) [6].

2.6. Oil Absorption Capacity (OAC)

The OAC was determined with 10 mL of vegetable oil were added to 1 g of each flour in a 50 mL centrifuge tube previously weighed. The suspension was stirred for 2 min, allowed to stand at 28 °C for 30 min and then centrifuged at 15,000× *g* for 20 min. The supernatant was decanted and discarded. Oil droplets adhering to the centrifuge tube were removed with cotton wool and then the tube was weighed. The result is calculated and expressed as the weight of oil absorbed per gram of flour [6].

2.7. Emulsifying Capacity (EmC)

1 g of each flour was mixed with 20 mL of water and stirred for 15 min. Then, the pH was adjusted to 7 and made up to 25 mL with distilled water. Subsequently, the 25 mL of the mixture were added with another 25 mL of corn oil in a blender and mixed for 3 min. Subsequently, the sample was centrifuged at 1300× *g* for 5 min. The emulsion is expressed in percentage terms, as the height of the emulsified layer with respect to the total liquid [6].

2.8. Foaming Capacity (FC)

This section was performed using 2 g of sample were mixed with 100 mL of water in a blender for 5 min. After this time, they were transferred to a 200 mL cylinder and the final volume was measured after 30 s pouring. FC is expressed as the percentage increase in volume compared to the original volume as shown in the formula [7].

2.9. Least Gelation Concentration (GC)

The GC was performed using suspensions at 4, 8, 12 and 14% (*p*/*v*) were prepared in distilled water, 5 mL were taken in test tubes, placed in a hot bath at 100 °C for 1 h and then in an ice bath for 1 h. Clot strength was evaluated by inverting the tube. The lowest protein concentration that formed a stable gel (remained in an inverted test tube) was considered GC [7].

2.10. Quantification of Soluble Proteins

The bean samples were defatted prior to analysis. After that, distilled water was added in a ratio of 1:10 (sample:water) and the pH was adjusted to 8.5 with 1 M NaCO. The mixture was stirred (200 rpm) for 2 h at room temperature, then centrifuged. at 10,000× *g* at 4 °C for 20 min. The supernatant was stored under refrigeration and distilled water was added to the pellet in a ratio of 1:5 (w:v). It was stirred for 2 h and centrifuged at 10,000× *g* at 4 °C for 20 min. Both supernatants were mixed and the pH was adjusted to 4.4. It was left stirring for 2 h and centrifuged at 10,000× *g* at 4 °C for 20 min. Protein isolates were stored at −80 °C [8].

Approximately 2 mg of these isolates were weighed and the protein concentration was quantified with the Pierce ™ BCA Protein Assay Kit (Thermo, Scientific).

2.11. Lectin Quantification

An extraction of the lectins in the flour was carried out using a phosphate buffer solution (5 mM K2HPO4 and 0.15 M NaCl pH 7.4). 0.1 g of bean flour was weighed, 10 mL of phosphate buffer was added and it was left under continuous stirring for 18 h. Afterwards, the mixture was centrifuged at 12,000× *g* and the supernatant was used for the hemagglutination test. Hemagglutinizing activity is expressed in units of hemagglutinin/mg protein. Hemagglutinin units were defined as the inverse of the last dilution that showed positive agglutination [9].

2.12. Quantification of Trypsin Inhibitors

Trypsin inhibitory activity was measured as residual tryptic activity, using BAPNA (N-benzoyl-DL-arginine pnitroanilide) as substrate. Total trypsin inhibitory activity was expressed as TIU per mg of sample (TIU/mg) [9].

2.13. Statistical Analysis

All measurements were carried out as separate duplicate experiments. The data are expressed as the mean ± standard deviation (SD) and the ANOVA analysis and the comparison of means was performed using a TukeyHSD test (α = 0.05) for all treatments. All statistical analyzes were carried out in the JMP® 8.0 program.

3. Results and Discussion

3.1. Water Absorption Index (WAI)

The range OH obtained was 2.90 and 2.91 g water/g sample for OH10 and OH15 respectively (Figure 1a). This result represents a significant increase in the absorption of 29% concerning the raw product. Besides, no significant differences were found among thermal treatments (3.14 g water/g sample for traditional) (Figure 1a) and other studies [10,11]. Also, after being consistent with those reported in other studies, this finding indicated the complete cooking of beans [11]. These results may be mainly due to the albumin fraction, which, when applying electric current, can cause the ionization of the SH groups, making them more reactive, or a partial denaturation, causing higher concentration of polar groups (carboxyl, COOH, and amide, NH2 groups) [12] generated by electric current, resulting in better interaction with water.

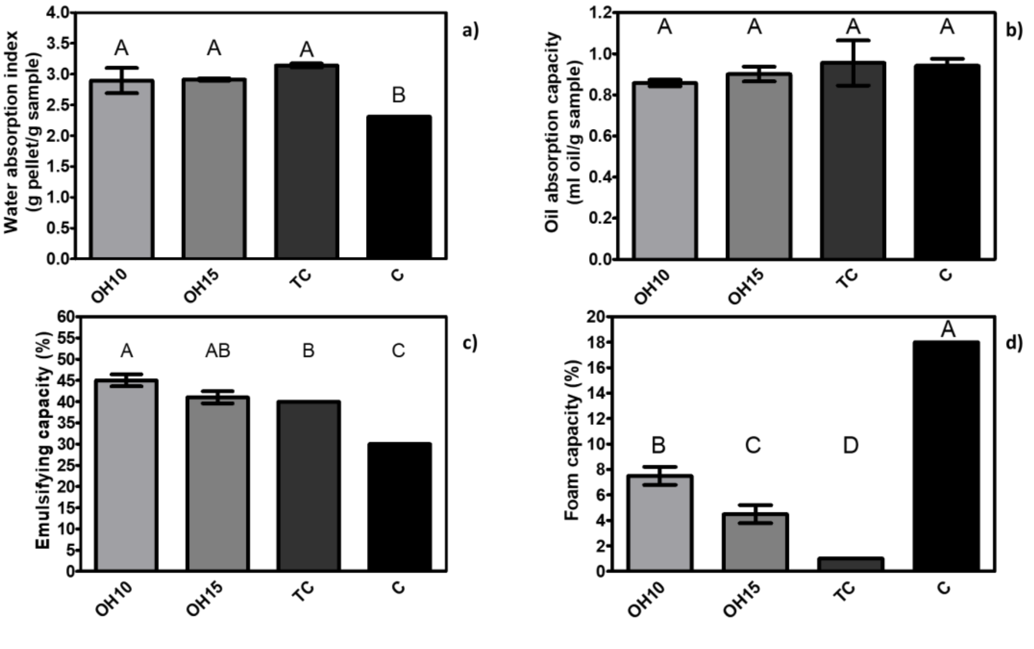

Figure 1. Techno-functional properties of bean flours processed by ohmic heating (OH10: 110 °C/10 min, OH15: 110 °C/15 min) and traditional processing (TC). Each value represents the mean of two replicates ± DS. Values with the same letter are not different from each other (Tukey α = 0.05).

3.2. Oil Absorption Capacity (OAC)

Oil pick up ranged from 0.86 and 0.90 mL of oil/g sample for OH10 and OH15 (Figure 1b). The differences found were no significant between flours obtaining for raw and traditional with 0.94 and 0.95 mL of oil/ g sample respectively, coinciding with other results [11]. The treatment, by modifying the orientation of the hydrophilic charges towards the outside, on the contrary, could cause the hydrophobic amino acids to be oriented towards the inside of the structure. According to this suggestion, it can be said that there are fewer nonpolar side chains available in the protein molecules of flour [11], which decreases their ability to bind to oil. Another option is that starch-protein interactions have been fostered, which may influence the phenomenon of reduced oil absorption. This would allow us to obtain fried products reduced in fat.

3.3. Emulsifying Capacity (EmC)

The EmC of flours processed by OH showed values ranging from 42 to 45% (Figure 1c). In comparison with TC, OH showed no differences. This results are agree with other studies with others cooking methods [13,14], and represents an approximate increase of 15% with respect to the raw product (Figure 1c). Although the compact and rigid structure of globulins, the main protein fraction of beans, does not make them proteins with good potential for emulsifying/foaming functions in the raw product [15], the results found indicate that the processed samples by ohmic heating they have the ability to develop and improve this property. Which is closely related to its water absorption capacity, since if it has a lower absorption, the amount of available water will not be enough to maintain the dispersion of the fat, which does not increase significantly in relation to the crude product.

3.4. Foam Capacity (FC)

Figure 1d shows the results obtained for the FC, where the samples of OH obtained the highest FC, a phenomenon contrary to that reported before [10], where heat treatment almost completely eliminates this capacity, due to the denaturation and/or solubilization of albumin, to which this phenomenon is attributed and is less affected by OH, as discussed in the WAI. Furthermore, as already mentioned, proteins determine important hydrodynamic functional properties such as WAI and OAC, or colloidal properties such as FC, but these are also modified by the carbohydrates and dietary fiber present in beans [10].

3.5. Least Gelation Concentration (GC)

Gelation properties observed for bean flours at different flour concentrations are shown in Table 1. It was observed that complete gelation for OH was at a concentration of 8%. These results showed gelation at concentrations lower than those reported by other studies [10,14] where the minimum gelation concentration is at a concentration of 12% for flours to which they are applied some thermal process. This could be explained by the fact that, as mentioned before, the electric current could modify the conformation of the proteins, promoting the protein-protein interaction, creating networks of intertwined polypeptides where water is trapped.

Table 1. Gelation capacity of bean flours processed with different treatments**.**

	Suspension Concentration			
Sample	**4%**	**8%**	**12%**	**14%**
OH10	-	X	X	X
OH15	-	X	X	X
TC	X	X	X	X
C	-	-	X	X

OH10: 110 °C/10 min, OH15: 110 °C/15 min. TC: Traditional cooking, C: crude. X: complete gelation, -: no gelation.

3.6. Quantification of Soluble Proteins

Figure 2a shows the protein content of bean flours. Flours processed by OH have concentration protein between 30–33 g of soluble protein/100 g of sample. These values do not present an essential statistical difference concerning the raw sample and TC. The concentrations obtained in OH flours are higher than those reported in other studies for different bean varieties [8]. Generally, a decrease in protein solubility would be expected due to the effect of thermal processing. As a consequence of protein denaturation and subsequent aggregation [16], however, a considerable reduction was not found in any of the cases. This factor has been considered the essential factor of the techno-functional properties since most of the proteins to which such effects are attributed are soluble, such as albumin and globulins [17]. We can be observed in OH10 flour, which was the best technology and functionality condition and has a high concentration of soluble proteins.

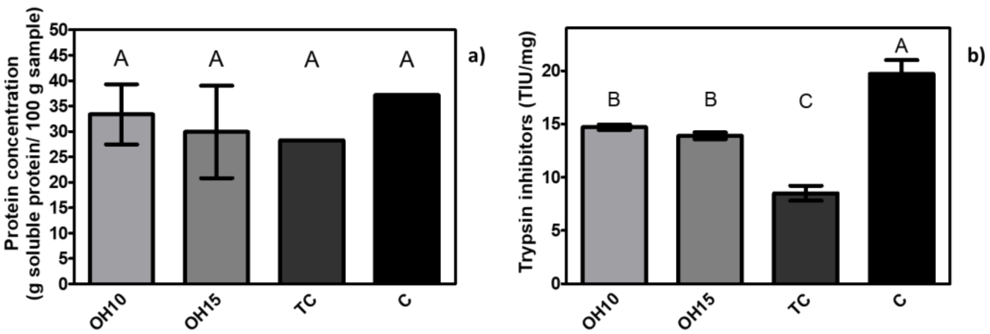

Figure 2. (**a**) Protein concentration and (**b**) trypsin inhibition for each flour sample. OH10: 110 °C/10 min, OH15: 110 °C/15 min, TC: Traditional cooking, C: crude. Each value represents the mean of two replicates ± DS. Values with the same letter are not different from each other (Tukey α = 0.05).

3.7. Quantification of Trypsin Inhibitors

Figure 2a shows the trypsin inhibitors content for all flours analyzed, obtaining a concentration of 19.72 ± 1.29 UIT/mg for the raw product. Finding the OH process showed a significant reduction about 25% regarding raw flour. It has been reported that by nature of the matrix, reaching a temperature of 110 °C, the union of ions with the proteins can occur or that cross-links may occur during denaturation, due to the effect of temperature, which can significantly alter the thermal stability of proteins, resulting in more excellent resistance to heat [18], which could result in a lower reduction of inhibitors in comparison with TC. Despite this, it has been reported that at least a concentration of 18.1 UIT/mg of trypsin inhibitors is needed to form complexes [19]. Based on this concentration, it can be seen that each of the OH treatments has a lower concentration.

3.8. Quantification of lectins

It was observed that the raw bean flour presented a higher concentration of hemagglutination (688.17 hemagglutination/mg protein), which agrees with that reported in the literature [8,20], compared with the samples subjected to heat treatment (TC and OH), in which protein was not detectable. These results are where after the thermal process, hemagglutination is not observed [20]. OH process ensures the inactivation of these antinutritional compounds, that could be a change in conformation caused by the reorientation of charges by the electric current described above, causing the protein to no longer be structurally available.

4. Conclusions

Processed common bean flours by OH showed the highest values for EmC and FC with respect to the conventional method and there were no statistical differences for the rest of the techno-functional properties. On the other hand, for the antinutritional compounds, no lectins were detected in any of the treatments, there was a reduction in the trypsin inhibitors and there were no changes in the solubility profile of the proteins. These results suggest that the processing of beans by OH can be an innovative alternative to TC for obtaining bean flours, with the advantages of having greater energy efficiency (90%), reduction of processing time (10 min vs. 2 h), which in the long run would also represent significant financial savings. In addition, it does not present leaching of the nutritional compounds of the beans, since it is not lost in the cooking broth, with the plus of having techno-functional characteristics similar to those obtained by conventional treatment, and can be a potential ingredient in the formulation of new functional products, which could translate into increased consumption and production of this legume.

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Conflicts of Interest:

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