Chemical and biological comparison of alkaline extruded and nixtamalized maize-sorghum meals[1]

Comparación química y biológica de harinas extrudidas alcalinamente y nixtamalizadas de maíz-sorgo[2]

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Recibido / *Received*: Enero / *January* 15, 2024

Aceptado / *Accepted*: Marzo / *March* 14, 2024

Publicado / Published: Junio 30, 2025 (Número 2, julio-diciembre) / *June 30, 2025* (*Number 2, July-December*)

Abstract

Maize (*Zea mays*) is the staple food of Mexico and its main form of consumption is the *tortilla*. This product is prepared using the pre-Columbian alkaline leaching technique known as nixtamalization. Sorghum (*Sorghum bicolor* L. Moench) is the second cereal cultivated in Mexico, but its use is mainly as a feed for polygastric animals because of the presence of tannins that make its protein undigestible to monogastric animals. Chemical and biological experiments using model animals have been carried out to corroborate its nutritional value compared with maize. Mixtures from zero to 100 percent maize:sorghum were prepared by nixtamalization and by alkaline extrusion. Chemical analyses of calcium and tannins concentrations, *in vitro* and *in vivo* digestibility, and PER tests were performed. Alkaline extrusion of all meals, when compared with nixtamalization improved both the *in vitro* and *in vivo* digestibility. PER values in all meals were low when compared with casein. However, nixtamalized and alkaline extruded mixtures of maize:sorghum with up to 40 per cent sorgum have comparable PER values to 100 per cent alkaline extruded and nixtamalized maize meals.

Keywords: Maize (*Zea mays*), sorghum (*Sorghum bicolor* L. Moench), biological properties, PER, digestibility

Resumen

El maíz (Zea mays) es el alimento básico de México y su principal forma de consumo es la tortilla. Este producto se prepara mediante la técnica de lixiviación alcalina precolombina conocida como nixtamalización. El sorgo (Sorghum bicolor L. Moench) es el segundo cereal cultivado en México, pero su uso es principalmente como alimento animal para poligástricos debido a la presencia de taninos que hacen que su proteína sea indigerible para los animales monogástricos. Se han realizado experimentos químicos y biológicos utilizando animales modelo para corroborar su valor nutrimental en comparación con el maíz. Se

[1] Authors' note: This paper, presented at the Third Chemical Congress of North America on Toronto, Canada, was submitted to Cereal Chemistry, an international journal, and was approved by the associate editor (Dr. Eckhoff), code 4386, along with another contribution "Rheological Properties of Extruded and Nixtamalized Corn-Sorghum Doughs and Tortillas" by Roberto Pérez-Ramírez, Javier Rodríguez-Méndez, and Carmen Durán-de-Bazúa (Code 4352) but, apparently, they were not published. For this reason, both have been submitted to this journal for its possible publication. The references are older than 5 years but they are classical studies and, thus, are still valid

[2] *Nota de los autores: Este documento, presentado en el Tercer Congreso de Química de América del Norte, en Toronto, Canadá, fue enviado a Cereal Chemistry, una revista internacional, y fue aprobado por el editor asociado (Dr. Eckhoff), clave 4386, junto con otra contribución Rheological Properties of Extruded and Nixtamalized Corn-Sorghum Doughs and Tortillas de Roberto Pérez-Ramírez, Javier Rodríguez-Méndez, and Carmen Durán-de-Bazúa (Code 4352) pero, aparentemente, no fueron publicados. Por ello,* ambos *fueron sometidos a esta revista para su posible publicación. Las referencias son anteriores a 5 años pero son clásicas y, por ello, todavía válidas*

prepararon por nixtamalización y por extrusión alcalina mezclas de cero a 100 por ciento maíz:sorgo. Se realizaron análisis químicos de calcio y taninos, digestibilidad in vitro e in vivo y pruebas REP. La extrusión alcalina de todas las harinas, en comparación con la nixtamalización, mejoró la digestibilidad tanto in vitro como in vivo. Los valores de REP para todas las harinas fueron bajos en comparación con la caseína. Sin embargo, las mezclas de maíz:sorgo nixtamalizado y alcalino extrudido con hasta un 40 por ciento de sorgo tienen valores de PER comparables a los de las harinas de maíz 100 por ciento alcalino extrudido y nixtamalizado.

***Palabras clave**: Maíz (Zea mays), sorgo (Sorghum bicolor L. Moench), propiedades biológicas, REP, digestibilidad*

Introduction

Maize (*Zea mays*) is the most important staple in Mexico. In 2023, Mexico registered the highest indicator in *per capita* human consumption of maize. According to data from the OECD[3] and FAO, consumption was seven times higher than the world average, ten times higher than the average of developed countries and six times higher than the average of developing countries (Figure 1a). These international organizations estimate that *per capita* consumption of maize in Mexico will grow to 138.5 kg in 2032. In the United States, *per capita* human consumption is very low, although it exceeds the average of developed countries (perhaps due to the Mexican influence in the diet). Its main form of consumption is the *tortilla*, made following a pre-Columbian alkaline leaching technique known as nixtamalization (Bazúa et al., 1979). Maize production in Mexico has not grown as steadily as the population resulting in increased imports of this cereal (Figure 1b, 1.43 millions/year; Figure 1c, 0.35 millones/año, giving 245 kg/persons+animals per year). Imported yellow maize does not meet the specifications necessary for good processing of maize into *masa* and results in a product of lower consumer acceptability and lower processing yields for producers.

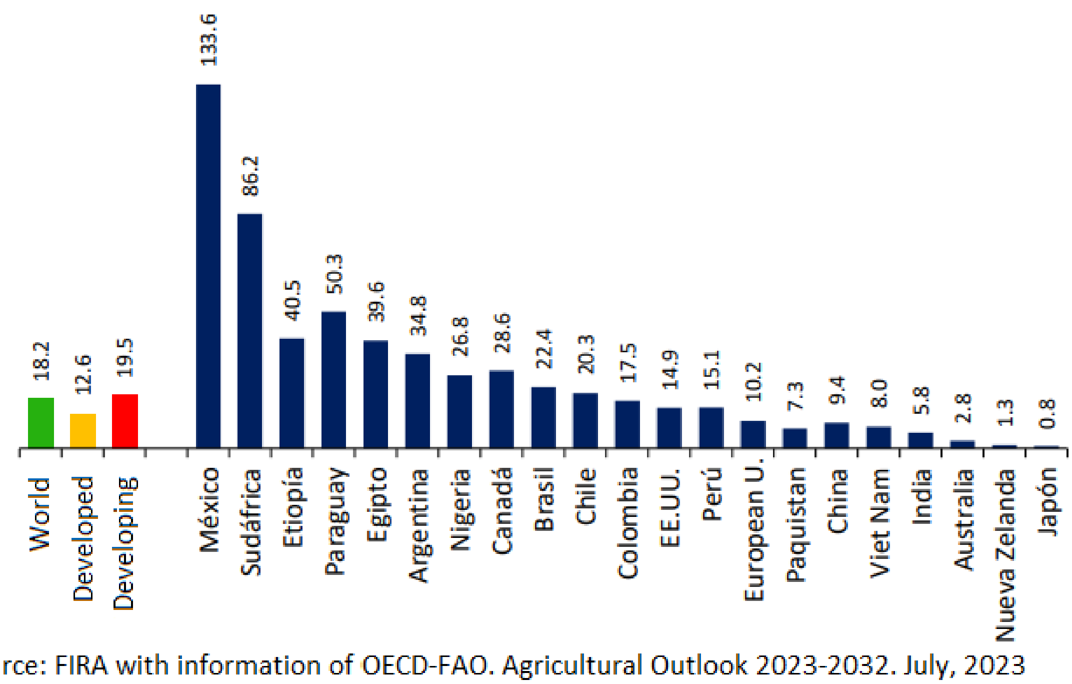


Figure 1a. Annual *per capita* human consumption of maize in kilograms per person, 2023 (Panorama Agroalimentario Maíz 2024.pdf, file:///C:/Users/Dra%20Duran/Downloads/Panorama%20Agroalimentario%20Ma%C3%ADz%202024.pdf)[4]

[3] Acronyms are explained at the end of this document in a Glossary

[4] Web pages are only presented in the text (not in the final list of references) [Note of the authors]

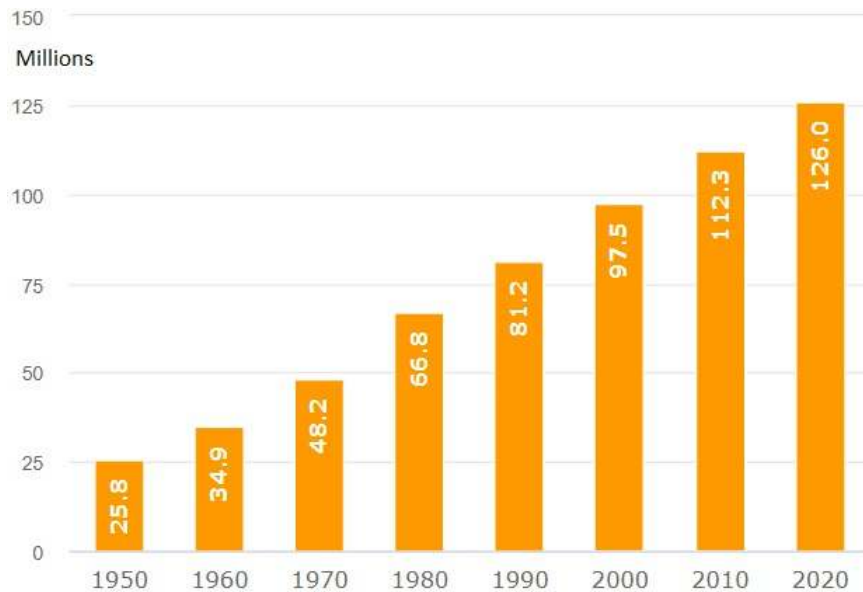


Figure 1b. Mexico's population in the last 70 years (https://cuentame.inegi.org.mx/poblacion/habitantes.aspx)

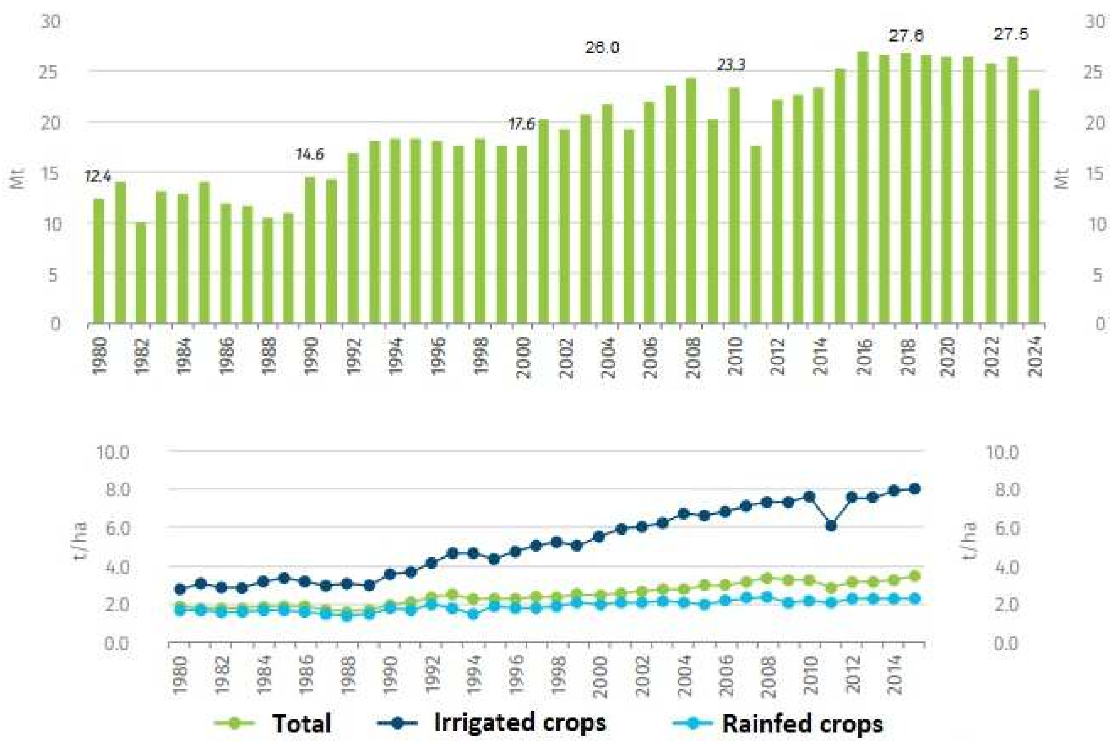


Figure 1c. Production of maize in metric tons per year and yield in tons per hectare in Mexico, 1980-2024 (Maíz para México, plan estratégico 2030, https://repository.cimmyt.org/entities/publication/7dd98fd7-5670-48bd-ae95-edcf496fad5a)

During the nixtamalization technique, maize grains are subjected to a lixiviation process in a hot alkaline solution. White maize varieties known as crystalline stand these conditions and simply lose its pericarp and become swollen, partially changing its endosperm compounds and structure. These physical and chemical changes allow the grain to be ground to a smooth dough with a perfect consistency to make tortillas. Imported yellow grain, being dried in silos with hot air and transported rather roughly becomes very brittle, and when it is being nixtamalized, grains open and release considerable amounts of endosperm and germ to the cooking and rinsing water. Thus, wastewaters from yellow versus white maize are more polluting due to the losses of yellow maize nutrients content (Durán-Domínguez-de-Bazúa, 1987). On the other side, as yellow maize comes to the factories and mills mixed with the white one, if cooking time is given to avoid these losses, the harder white maize leaves the process undercooked, giving a rather bad quality dough and rejectable *tortillas*, very brittle and without rolling characteristics. The opposite situation renders very low yields since a considerable amount of yellow maize goes to cooking and rinsing water.

Due to climate, soil, and drought conditions found in most of the maize fields in Mexico, a similar crop, sorghum (*Sorghum bicolor* L. Moench), was introduced during the seventies of the twentieth century looking for a better alternative. As a matter of fact, in Mexico, its agronomical yields are higher than those for maize at the same conditions (Betanzos, 1970) (Figure 2a). Seeds came from Africa and India, where this grain is consumed by the people in gruels (sort of Mexican atole) and chapaties (sort of tortillas).

Therefore, experimental research was carried out with the most promising varieties of sorghum to look for alternative processes for the use of this grain as an extender of maize for human consumption. Concerning these food technology experiments results were not so encouraging due to the nutritional value of the sorghum grain, especially for the agronomically most promising varieties that were the deep colored ones containing tannins that were not eaten by birds that are the predating animals for this grain that grows without a cover as it happens with maize that has several layers of leaves to protect the grains (Figures 2b, c).

During the first half of the seventies of the Twentieth Century, a line of research parallel to the use of sorghum, was started between the group of the maize research at UNAM and the group at INIA. This line of research was focused on the use of extrusion cooking for pre-cooking grains and reducing post-harvest losses (Crowley, 1975; Durán-Domínguez, 1974). Its application in the rural areas through the low cost units that employed power take-off units coupled to tractors, gave a new insight to the possibility of using this process for precooking brittle maize and little seeds as sorghum (Bazúa et al., 1976; Durán-Domínguez, 1978; Durán-de-Bazúa, 1977; Durán-de-Bazúa and Guerra, 1977, 1980; Guerra et al., 1983).

Meanwhile, patents have been granted in Mexico to appropriate technologies for pearling colored sorghum to remove the pericarp and testa where tannins are (Laso and Núñez, 1978). Yield of endosperm sorghum pearls was about 70 to 80 per cent, and the rest were fraction ranging 3.18 to 2.38 mm (-1/8+3/32 in). These pearled products could be ideally processed by alkaline extrusion since, for this technique granular raw materials are highly desirable to improve the equipment heat autogeneration (autogenous extruders) (Durán-de-Bazúa, coordinator, 1988).

Therefore, based on these two approaches to use sorghum or even yellow maize, the objective of this part of the research was to compare the chemical and nutritional quality of maize, sorghum, and its mixtures cooked using alkaline extrusion conditions in such a way that extruded products can be similar to traditional nixtamalized counterpart.

Products made with extruded meals were evaluated using as comparison tests material, the products obtained by traditional nixtamalization.

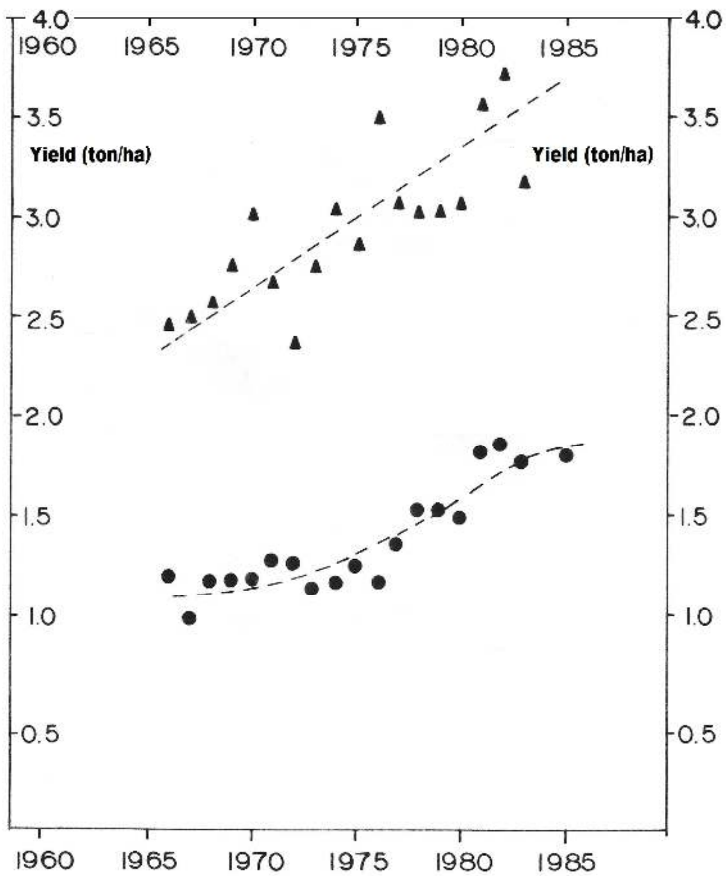


Figure 2a. Yields of maize (●) and sorghum (▲) in the Mexican fields

Figure 2b. Maize ears (*elote* with fresh leaves or *totomoxtle* in Spanish and Náhuatl) (https://pixabay.com/es/images/search/elote/)

Figure 2c. Red and white sorghum panicles (*panoja* or *panícula* in Spanish) (https://agrotendencia.tv/agropedia/agricultura/cultivos/el-cultivo-de-sorgo/; https://pixabay.com/es/photos/jowar-sorgo-karnataka-india-223591/

Materials and methods

Raw materials. To study alkaline extrusion the esperiments were carried out using white crystaline maize of the Zoapila variety of INIA (presently INIFAP, see Glossary), red-brown colored sorghum provided by *Compañía Nacional de Subsistencias Populares, Conasupo* (National Popular Subsistence Company), in Spanish. From red-brown colored sorghum, following the patented process of Kiselguhr company (Laso and Núñez, 1977) pearled sorghum was obtained (Saldaña-Morales, 1987). The following samples were studied:

1. Corn or maize 100% (M100), pearled colored sorghum 100% (SP100), and red colored sorghum 100% (SC100)
2. Mixtures of maize (M) with the two types of sorghum, 15 and 40% sorghum, either whole (SC) or pearled (SP), and 85 and 60% maize: M85-SC15, M60-SC40, and M85-SP15, M60-SP40.

Meals were labeled as follows:

M-100	Maize 100 per cent
SP-100	Pearled or decorticated sorghum 100 per cent
SC-100	Colored sorghum 100 per cent
SP15-M85	Pearled or decorticated sorghum 15 per cent- Maize 85 per cent
SP40-M60	Pearled or decorticated sorghum 40 per cent- Maize 60 per cent
SC15-M85	Colored sorghum 15 per cent- Maize 85 per cent
SC40-M60	Colored sorghum 40 per cent- Maize 60 per cent

Processes were labeled as follows:

R	Raw meals of ground grains
N	Nixtamalized meals
E	Laboratory scale Brabender extruder
E*	Bench scale low cost Mexican extruder (made by CIATECH-Conacyt)

Nixtamalization

The name of this pre-Columbian process means lime cooked maize (*nextli*=lime ashes and *tamalli*=cooked maize, maize cooked with lime ashes, Cabrera, 2002). The procedure for cooking the three grains and its mixtures using traditional nixtamalization is also described elsewhere (Durán-de-Bazúa, Coordinator, 1988; Nieto et al., 1986). Nixtamalization was carried out as follows: 1% in corn or maize mass of calcium hydroxide was added to water and grain (two parts of water per one part of grain). The grain was cooked at boiling temperature until the grains changed color from a deep orange to yellow (Valley of Mexico traditional process). A soaking period of 11 to 12 hours followed the cooking process. Cooked grains were rinsed (two parts of water per one part of grain) to eliminate excess lime and pericarp. Clean cooked grains were dried in a vacuum tray drier (70°C and 1.5 kg_f/cm^2 vacuum, for approximately 7 hours). Dried grains were ground to obtain the specific granulometry that complies with the Mexican oficial norm for nixtamalized cornmeal (DOF, 1980a). For sorghums, colored and pearled, a modified nixtamalization technique was developed to avoid any overcooking. Description of the process and conditions used are given elsewhere (Nieto et al., 1986).

Alkaline extrusion

Two equipments were used for extruding the raw meals studied: A laboratory scale Brabender extruder and a bench scale low cost extruder designed and built in Mexico.

To produce the alkaline extruded meals at laboratory scale, the Brabender equipment was previously calibrated to obtained the best feeding, shear, and extrusion temperatures and the operational characteristics for each meal: The exit die had 4 mm diameter, the screw had a length:diameter ratio 3:1, and the rotational screw speed was 100 rpm. The amount of $Ca(OH)_2$ was 0.3% d.b. and the meals were rehydrated up to 20% moisture content. Average residence time in the extruder was 35±10 seconds, feeding 3 to 5 kg meal/h and temperature ranged from 150 to 170°C. After extrusion, the products were dried and ground and the meals were stored in hermetically closed containers before use (Saldaña-Morales et al., 1986).

For the extrusion process at higher scale, a low cost extruder was used (Figure 3). It was built by CIATECH (Glossary), a governmental institution that based its design in the Brady extruder (Crowley, 1975), as well as in the modifications proposed by the Colorado State University researchers (Harper, 1979). Its features and engineering characteristics as well as its limitations compared with the Brabender laboratory unit are presented elsewhere (Pérez-Ramírez and Rodríguez-Méndez, 1988). This type of extruders generates by friction the heat used for cooking the granular material. The cone displacement adjusted by closing or opening the circumference where the extruded material exits, provoques the increase or decrease of the exit temperature, according to the backpressure generated in the extrusion zone (Pérez-Ramírez and Rodríguez-Méndez, 1987). Processing conditions were obtained from preliminary experiments (Pérez-Ramírez et al., 1988): Meals granulometry (Glossary) was 20 mesh (840 µm), with a feed rate about 250 kg/h; initial moisture content was 20 per cent; initial calcium hydroxide concentration was 0.2 per cent (w.b.); extrusion temperature was around 160°C, and residence time was *ca*. 50 seconds. After extrusion, the products were dried an ground following the procedure used for nixtamalized meals, and later on used in their chemical and biological evaluations.

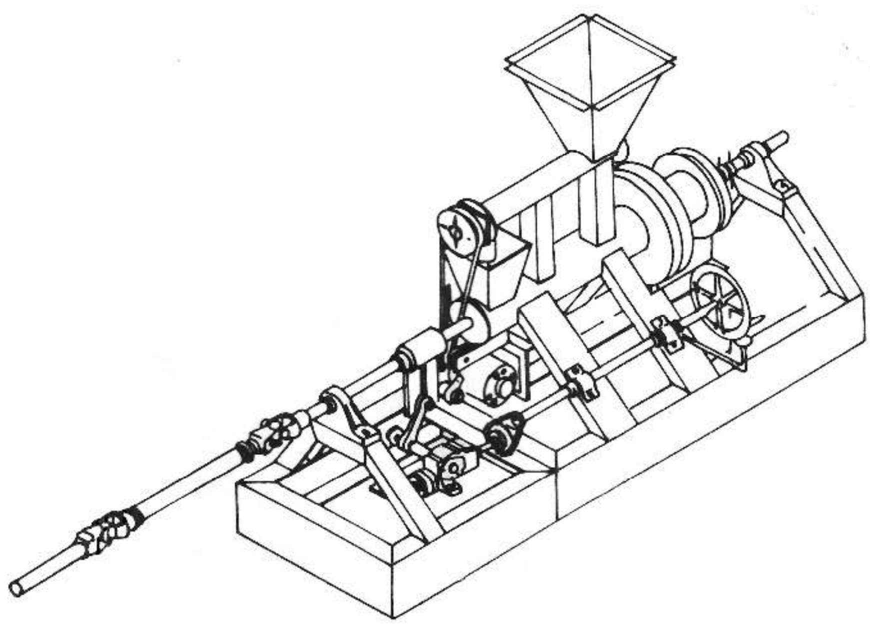


Figure 3. Low cost Mexican built extruder (CIDESI, 1981)

Tortillas preparation

The complete methodology for preparing the tortillas from the nixtamalized and alkaline extruded meals is described elsewhere (Alarcón et al., 1985).

Biological studies

Protein Efficiency Ratio, PER

One of the tests used to corroborate the effect of processing on the quality of the grains protein was the Protein Efficiency Ratio, PER, following the AOAC (1980) part 2 methodology. For this AOAC method, an additional experiment was run, using a test diet with 8 per cent casein protein. This lot of rats was added because protein content in meals studied was below the 10 per cent officially acepted value. This consideration for diets with 7 or 8% protein using a control a lot with casein at the same percentage has been previously reported (Cravioto et al., 1950; Cravioto and Cervantes, 1965; Hernández et al., 1982). The whole procedure is described by Saldaña-Morales (1987).

Aparent digestibility (*in vitro*) and digestibility (*in vivo*)
The diets prepared with the grains were marked with an indigestible substance, known as "chromium bread" (Cr_2O_3) to be determined both in the diet and in the fæces, following Schurch et al. (1950) method. For *in vitro* digestibility pepsin and HCl (10%) were used (Akbson and Stahmann, 1964). The protein content was evaluated following the Mexican official procedure NOM-F-68-S (DOF, 1980b). The procedure is also described in the previous reference (Saldaña-Morales, 1987).

Chemical analyses
Formerly known as proximate or bromatological analyses, they were performed for all samples, raw, nixtamalized, or alkaline extruded meals following the Mexican official standard analyses procedures: Ash (NOM-F-66-S) (DOF, 1978a), nitrogen contents (NOM-F-68-S) (DOF, 1980a), moisture (NOM-F-83) (DOF, 1986), fat (NOM-F-89-S) (DOF, 1978b), crude fiber (NOM-F-90-S) (DOF, 1978c). Calcium content was determined using the permanganometric method (Da-Costa-Gonçalves-Santos et al., 2024). Tannins were measured using the vanillin-HCl method (Burns, 1963) and its content compared with the literature. All analyses were made by triplicate.

Statistical analyses
Experimental data by triplicate were evaluated following variance analysis and considering probability significance levels of 5% ($p<0.05$) (Statgraphics, 2024). After the two-way ANOVA, the Tukey multiple comparison test was performed with a 95% confidence level. Both the graphs, the two-way analysis of variance and the multiple comparisons by the Tukey method were performed using the GraphPad Prism 6 software (Boston, MA, US).

Results and discussion

The overall results for these experiments are shown in Tables 1, 2, and 3. Its statistical analyses were exemplified with Graphs 1 to 7.

Chemical analyses are in Table 1.

Table 1. Chemical analyses of meals, g/100g meal (d.b.): Raw (R), nixtamalized (N), lab scale extruder (E), and bench scale extruder (E*) [Average of three values with standard deviation]

Meals		Protein$^+$	Fat	Crude fiber	Ashes	Carbohydrates
M-100	R	10.10±0.09	5.39±0.02	2.32±0.01	1.55±0.01	80.64
	N	8.95±0.07	5.07±0.01	2.11±0.01	1.44±0.01	82.43
	E	10.28±0.08	4.24±0.02	2.27±0.02	1.68±0.01	80.51
	E*	10.37±0.06	5.46±0.04	2.76±0.01	1.82±0.02	70.58
SP-100	R	9.41±0.08	1.27±0.01	0.76±0.02	1.40±0.01	87.16
	N	9.08±0.05	1.01±0.01	0.59±0.01	0.72±0.01	88.59
	E	9.45±0.10	0.32±0.01	0.62±0.01	1.01±0.01	88.54
	E*	9.70±0.09	0.97±0.02	0.66±0.02	0.98±0.01	87.69
SC-100	R	11.48±0.08	3.45±0.02	2.80±0.01	2.40±0.01	79.87
	N	8.52±0.06	2.90±0.03	2.67±0.02	2.25±0.01	83.65
	E	11.13±0.09	1.26±0.01	2.76±0.01	2.02±0.01	82.81

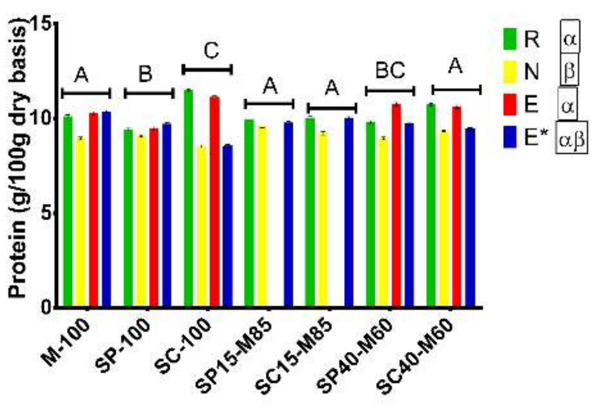
Table 1. Chemical analyses of meals, g/100g meal (d.b.): Raw (R), nixtamalized (N), lab scale extruder (E), and bench scale extruder (E*) [Average of three values with standard deviation]

Meals		Protein⁺	Fat	Crude fiber	Ashes	Carbohydrates
	E*	8.55±0.08	2.74±0.01	2.82±0.02	2.76±0.02	84.63
SP15-M85	R	9.94±0.07	4.90±0.02	2.03±0.01	1.56±0.01	81.57
	N	9.60±0.09	4.67±0.03	1.08±0.01	1.33±0.01	82.52
	E	n.d.	n.d.	n.d.	n.d.	n.d.
	E*	8.55±0.06	4.54±0.02	2.07±0.02	1.68±0.02	81.93
SP40-M60	R	9.81±0.08	3.85±0.03	1.60±0.02	1.39±0.01	83.35
	N	8.96±0.07	3.64±0.01	1.27±0.01	1.13±0.01	84.98
	E	10.75±0.09	3.08±0.02	1.35±0.01	1.42±0.01	83.93
	E*	9.73±0.08	4.09±0.01	1.58±0.02	1.50±0.02	83.10
SC15-M85	R	10.04±0.10	5.09±0.02	2.35±0.02	1.85±0.01	80.67
	N	9.22±0.09	4.65±0.01	2.04±0.01	1.36±0.01	82.63
	E	n.d.	n.d.	n.d.	n.d.	n.d.
	E*	10.02±0.08	4.29±0.02	2.27±0.01	1.76±0.01	81.66
SC40-M60	R	10.75±0.07	4.59±0.01	2.51±0.02	2.11±0.02	80.04
	N	9.35±0.04	4.27±0.03	2.15±0.01	1.39±0.01	82.83
	E	10.62±0.08	1.67±0.01	2.35±0.01	1.97±0.02	83.55
	E*	9.45±0.06	3.92±0.01	2.42±0.02	1.83±0.01	82.38

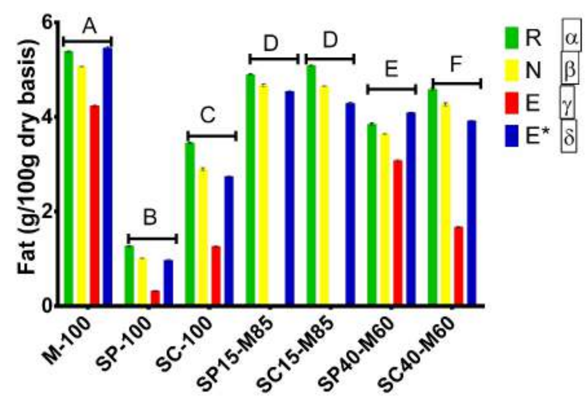
⁺Factor: Nx6.25, n.d.: not determined (because the samples were contaminated)

The parameters protein, fat, fiber, ash, and carbohydrates were analyzed by a two-way ANOVA as mentioned in Methodology. The factors were type of diet with the levels M-100, SP-100, SC-100, SP15-M85 SC15-M85, SP40-M60 and SC40-M60. The second factor was the type of processing with the following levels: Raw (R), nixtamalized (N), Extruded (E) and CIATECH extruder (E*).

In the Graphs 1 to 7, the Latin letters indicate the results of the comparisons between the first factor (diet composition). The Greek letters indicate the results of the comparisons of the second factor (process type).



Graph 1. Bars indicate mean ± SD of protein content. Protein content depends on diet type and process. Groups sharing the same letter do not differ statistically from each other. Latin letters indicate comparison between diet types and Greek letters indicate comparison between different processes. Tukey method at 95% confidence level

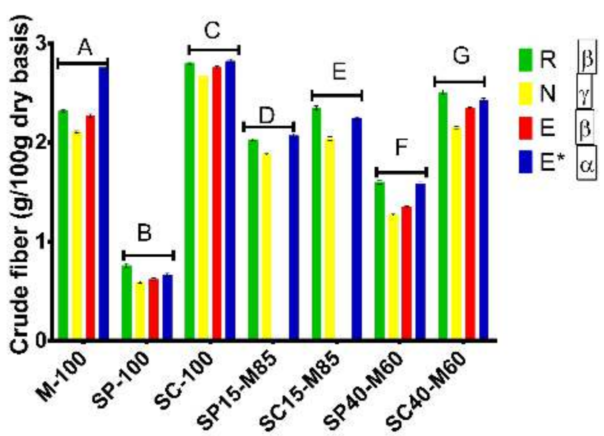


Graph 2. Bars indicate mean ± SD of fat content. Fat content depends on diet type and process. Groups sharing the same letter do not differ statistically from each other. Latin letters indicate comparison between diet types and Greek letters indicate comparison between different processes. Tukey method at 95% confidence level

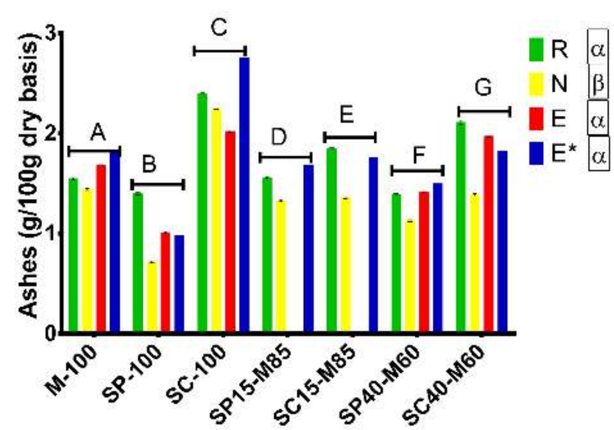
It can be seen in Graph 1 that maize diets show no statistical differences among them (letter A) indicating that protein content is not affected by the proportions of grains with the exception of the mixture containing pearled sorghum. Considering the process (Greek letters) nixtamalized diet is statistically different indicatig that this nutrient is lost in the processing wastewater (*nejayote*). For sorghum diets, pearled sorghum loses this nutrient in all diets especially compared with whole colored grain (letters B and C) due to the abrasion suffered during its pre-processing. For the process, extrusion at lab scale has the correct operating conditions. Bench scale showed statistical differences only for colored sorghum samples (100% and 40%) that perhaps were enhanced by the formation of a protein-carbohydrate complex and could not be fully determined by the Kjeldhal procedure as mentioned by Mercier et al. (1979).

For Graph 2, fat content in diets changes according to grain type. And definitely the processing alter the fat content determination. It is known that extrusion protects fat making the products impervious to deterioration thanks to its complexation with carbohydrates (Harper, 1979; Ying et al., 2015), and due to it, its contents are much lower, especially at lab scale due to the controlled process variables.

Graphs 3 and 4 showed the statisticals results contents of crude fiber and ashes content indicating that no statistical differences were found for each diet but all were different to each other. Interestingly, processing for crude fiber had the same behavior but ashes did not change with the exception of nixtamalization where its content was reduced due to the washing of the grains after cooking.



Graph 3. Bars indicate mean ± SD of crude fiber content. Crude fiber content depends on diet type and process. Groups sharing the same letter do not differ statistically from each other. Latin letters indicate comparison between diet types and Greek letters indicate comparison between different processes. Tukey method at 95% confidence level

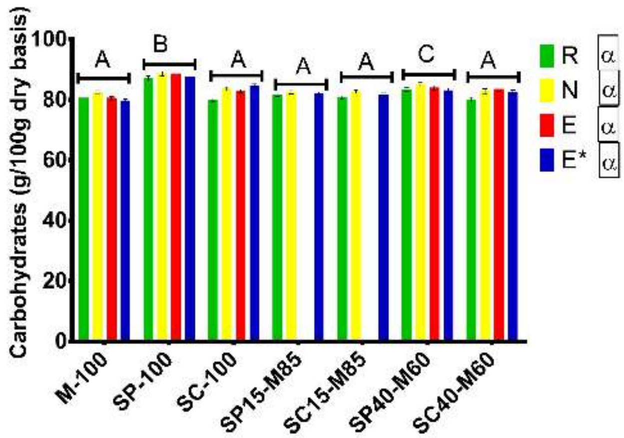


Graph 4. Bars indicate mean ± SD of ashes content. Ashes content depends on diet type and process. Groups sharing the same letter do not differ statistically from each other. Latin letters indicate comparison between diet types and Greek letters indicate comparison between different processes. Tukey method at 95% confidence level

For carbohydrates (Graph 5) neither the diet nor the process had any influence on the content. The only exception as expected was pearled sorghum and its mixture with maize.

Data on tannins and calcium, as well as *in vitro* and *in vivo* digestibilities are shown in Table 2.

With respect to tannins, there is a definite effect of extrusion on its concentration when compared with raw meals, favoring the alkaline extrusion process, especially the CIATECH extrusion coonditions. Perhaps the tannins within the high pressure hot alkaline milieu, do not react and give lower values than raw and nixtamalized meals.



Graph 5. Bars indicate mean ± SD of carbohydrates content. Carbohydrates content depends on diet type. Groups sharing the same letter do not differ statistically from each other. Latin letters indicate comparison between diet types and Greek letters indicate comparison between different processes. Tukey method at 95% confidence level

Table 2. Analyses of tannins as catechin equivalent, and calcium, g/100 g meal (d.b.), and *in vitro* digestibility and *in vivo* digestibility, in per cent: Raw (R), nixtamalized (N), lab scale extruded (E), and bench scale extruded (E*) [Average of three values]

Meals		Tannins	Calcium Before	Calcium After	*In vitro* digestibility	*In vivo* Digestibility
	Treatment					
M-100	R	0.0256	n.d.	n.d.	82.75	n.d.
	N	0.0385	0.4767	0.1290	83.86	68.80
	E	0.0206	0.1895	0.1524	89.71	n.d.
	E*	0.0085	0.1976	0.1575	88.10	81.03
SP-100	R	0.0563	n.d.	n.d.	81.56	n.d.
	N	0.0375	0.1162	0.0882	74.08	73.37
	E	0.0506	0.2052	0.1472	88.98	n.d.
	E*	0.0243	0.1976	0.1666	85.88	83.01
SC-100	R	0.1328	n.d.	n.d.	70.86	n.d.
	N	0.0758	0.8813	0.4772	60.97	40.38
	E	0.0710	0.2164	0.1804	77.72	n.d.
	E*	0.0701	0.2013	0.1806	86.29	73.30
SP15-M85	R	0.0513	n.d.	n.d.	81.86	n.d.
	N	0.0758	0.4767	0.1034	77.40	68.41
	E	n.d.	n.d.	n.d.	n.d.	n.d.
	E*	0.0329	0.1976	0.1698	87.27	82.31
SP40-M60	R	0.0639	n.d.	n.d.	83.76	n.d.
	N	0.0383	0.4725	0.0913	65.85	70.19
	E	0.0204	0.2059	0.1824	81.12	n.d.
	E*	0.0346	0.2000	0.1669	86.45	78.69
SC15-M85	R	0.0699	n.d.	n.d.	76.84	n.d.
	N	0.0698	0.4715	0.1019	77.24	74.29
	E	n.d.	n.d.	n.d.	n.d.	n.d.
	E*	0.0458	0.1976	0.1702	87.28	81.73
SC40-M60	R	0.1265	n.d.	n.d.	74.51	n.d.
	N	0.1265	0.4700	0.0773	64.42	71.50
	E	0.0546	0.2113	0.1689	84.70	n.d.
	E*	0.0612	0.2013	0.2000	88.56	78.90

n.d.: not determined because the amount of cooked meal was very low for biological experiments

Final calcium concentration was almost always lower for nixtamalized meal, since most of it goes away with the cooking and rinsing water, with the exception of colored sorghum. This meal had a very high initial calcium concentration in order to have an effect on final tannins concentration (Asropi et al., 2022). The selected initial value for extruded meal, according to the organoleptic characteristics of final *tortilla* products rendered not unduly higher values for calcium than its nixtamalized counterparts.

According to the data for *in vitro* and *in vivo* digestibility there is a definite positive effect of the extrusion process on digestibility, both *in vitro* and *in vivo*, when compared with raw and nixtamalized meals. Only for extruded and raw maize meals, there is no statistical difference ($p<0.05$) among its *in vivo* and *in vitro* digestibility. Nixtamalized sorghum products, especially, give lower values than its raw and, naturally, its extruded counterparts for *in vitro*, and even more for *in vivo* digestibility.

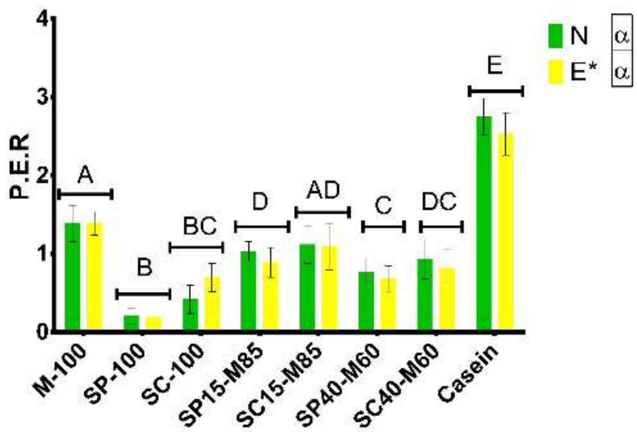
However these results are not comparable with the PER data obtained (Table 3 and Graphs 6 and 7), since extrusion apparently does not give any statistical difference with nixtamalization to the protein efficiency ratio of the protein content in the meals.

Table 3. Protein content in diets, percent nitrogen (x 6.25) and protein efficiency ratio (PER) with standard error for nixtamalized (N) and CIATECH extruder processed meals (E*)

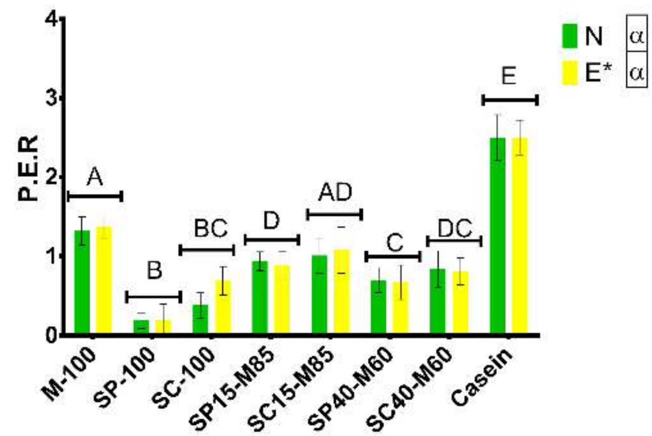
			P E R	
Meals		Protein in diet	Experimental value	Adjusted value
M-100	N	7.25	1.39±0.23	1.32±0.18
	E*	8.81	1.39±0.15	1.37±0.14
SP-100	N	7.63	0.21±0.10	0.19±0.10
	E*	8.19	0.19±0.22	0.19±0.21
SC-100	N	7.00	0.42±0.18	0.38±0.16
	E*	7.38	0.70±0.18	0.69±0.18
SP15-M85	N	8.13	1.03±0.13	0.94±0.12
	E*	8.69	0.89±0.19	0.88±0.18
SP40-M60	N	7.88	0.77±0.18	0.70±0.16
	E*	8.44	0.68±0.17	0.67±0.22
SC15-M85	N	8.13	1.12±0.24	1.01±0.22
	E*	8.31	1.09±0.29	1.08±0.29
SC40-M60	N	8.62	0.93±0.25	0.84±0.23
	E*	8.44	0.82±0.23	0.81±0.17
Casein	N	10.00	2.75±0.23	2.50
	E*	10.00	2.53±0.27	2.50
Casein	N	8.00	2.44±0.29	n.d.
	E*	8.00	2.45±0.22	n.d.

Analyzing the results obtained, as cereals are not a good source of protein, and maize and sorghum are not the exception, PER values are considerable lower than those obtained for casein fed rats, both with 10 and 8 per cent protein.

With these experiments what was looked for was the posible difference between the new technology, alkaline extrusion, and the traditional lime-cooking technique, nixtamalization. According to them, no statistical difference ($p<0.05$) in PER data was found among the two cooking processes indicating that alkaline extrusion may substitute traditional lime cooking (nixtamalization) with the advantages of much less processing time, energy saving, and finally, the most important item besides the energy savings, the high reduction of water needed for its processing with no wastewater produced (Hernández-Morales et al., 2019; Durán-Domínguez-de-Bazúa et al., 2021).



Graph 6. Bars indicate mean ± SD of PER (experimental value). Protein efficiency ratio (PER) depends on diet type. Groups sharing the same letter do not differ statistically from each other. Latin letters indicate comparison between diet types and Greek letters indicate comparison between different processes. Tukey method at 95% confidence level



Graph 7. Bars indicate mean ± SD of PER (adjusted value). Protein efficiency ratio (PER) depends on diet type. Groups sharing the same letter do not differ statistically from each other. Latin letters indicate comparison between diet types and Greek letters indicate comparison between different processes. Tukey method at 95% confidence level

It is important to point out that pearling has a definite effect on protein quality, since aleurone is lost during the abrasion process. In contrast, colored sorghum, although it has not such low PER value as pearled sorghum, when it is nixtamalized, a very low *in vivo* digestibility was found. This might be perhaps due to the high calcium concentration since for its alkaline extruded counterparts this effect is not noticed, and neither for its mixture with maize at 40:60 per cent ratio with comparable calcium concentration final content.

It has been already reported that there were no effects of nixtamalization on protein value for each grain or grain mixture studied (Table 4).

Table 4. Effect of nixtamalization on PER of meals (Nieto and Durán-de-Bazúa, 1988)

(A) Nixtamalized *versus* raw meals			
Samples	T	d.f.	scores*
Nixtamalized *versus* raw maize	3.166	14	A
Nixtamalized *versus* raw colored sorghum	-2.700	13	A
Nixtamalized *versus* raw pearled sorghum	4.667	14	B
Nixtamalized *versus* raw mixture maize:pearled sorghum (60:40)	1.602	13	A

Table 4. Effect of nixtamalization on PER of meals (Nieto and Durán-de-Bazúa, 1988)

(B) Differences among nixtamalized meals			
Samples	T	d.f.	scores*
Nixtamalized maize *versus* nixtamalized colored sorgum	6.803	14	B
Nixtamalized maize *versus* nixtamalized pearled sorgum	6.915	14	B
Nixtamalized maize *versus* nixtamalized mixture maize:pearled sorghum (60:40)	4.309	14	B
Nixtamalized colored sorghum *versus* nixtamalized mixture maize:pearled sorghum (60:40)	1.132	13	A
Nixtamalized colored sorghum *versus* nixtamalized mixture maize:colored sorghum (60:40)	-3.418	14	A
Nixtamalized pearled sorghum *versus* nixtamalized mixture maize:colored sorghum (60:40)	-2.719	13	A

*t scores for 99.9 per cent level of certainty for both upper and lower limits: 13 d.f. (degrees of freedom), 4.221; 14 d.f., 4.140
a = No significant difference among meals, b = Significant difference at 99.9 per cent level of certainty

The exception is pearled sorghum meal using protein efficiency ratio t scores or t values from the statistical analysis results at 99.9 per cent certainty level. For this meal, nixtamalization has an apparent improving effect.

Conclusions

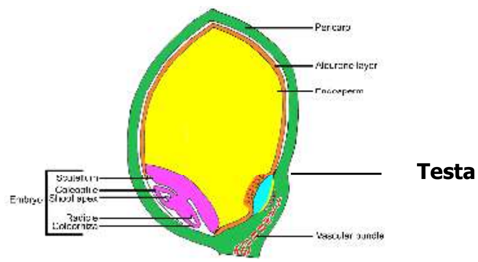
According to the results obtained, it may be said that alkaline extrusion do not reduce the nutritional value of maize but, on the contrary, maintains the quality of the protein in the extruded cereal meals, and improves its digestibility, both *in vitro* and *in vivo*. The use of whole colored and pearled sorghum in mixtures with maize reduces the quality of the protein of nixtamalized and alkaline extruded meals but, for the alkaline extruded meals the digestibility both *in vitro* and *in vivo*, is not significantly reduced. Therefore, if there is a necessity to use sorghum to extend maize products for human consumption, the mixture will have a similar digestibility and will, energetically, be as good as the one hundred per cent maize products. Definitely, the cereals are not a good protein source but an energy soource, and thus, these grains represent an adequate supply to improve the caloric intake of the Mexican and the rest of the world population.

Glossary

Acronyms or terms	Meaning
Atole	A drink made of nixtamalized maize. The word “atole” comes from the Nahuatl language ,“atolli”, which means “maize drink”, due to its root of “atl” water and “tlaolli”, maize (Cabrera, 2002)
b. C.	Before Christ
CIATECH	*Centro de Investigaciones y Asistencia Tecnológica del Estado de Chihuahua, A.C., Conacyt*, in Spanish. Research and Technological Assistance Center of the State of Chihuahua, A.C., Conacyt
CIDESI	*Centro de Ingeniería y Desarrollo Industrial del Instituto Politécnico Nacional* (Center for Engineering and Industrial Development of the National Polytechnical Institute, in Spanish)
Conacyt	*Consejo Nacional de Ciencia y Tecnología*, in Spanish. Mexico’s National Council for Science and Technology
d.b.	Dry basis

Acronyms or terms	Meaning
Digestibility, apparent (*in vitro*) and true (*in vivo*)	The 'apparent digestibility' (AD) of a protein is the ratio of the difference of the ingested and fæcal nitrogen to the ingested nitrogen, expressed as a percentage, while the 'true digestibility' (TD) makes allowance for the nitrogen in the fæces of non-dietary origin (the so-called metabolic nitrogen, F_0)
Granulometry	Measurement of the size distribution in a collection of grains. It is also called particle size distribution test. It is often characterized by the percentage of particles with certain diameter ranges (in micron meters) (Wikipedia, 2024). Mesh size is straightforward, it measures the number of openings in a mesh that make up one linear inch (2.54 cm)
ICRISAT	International Crop Research Institute for Semi-Arid Tropics, Hyderabad, India. A Mexico's fraternal center is located at El Batán, State of Mexico, Mexico
INIA-SARH	*Instituto Nacional de Investigaciones Agrícolas de la Secretaría de Agricultura y Recursos Hidráulicos*, in Spanish. Mexico's National Institute of Agricultural Research of the Secretary of Agriculture and Water Resources
INIFAP	*Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias*, in Spanish. The fusion of three institutes into one, *INIA*, *INIF*, and *INIP*, during the government of Salinas-de-Gortari, reducing their capabilities and personnel
Maize	Word coming from the Taino language, since the Spaniards with Columbus arrived in 1492 to the Caribbean islands believing it was India, and that was the language spoken there (a now-extinct Arawakan language, once predominated in the Antilles and was the first language to be encountered by Europeans). Its scientific name is *Zea mays*. This grain originated in Mexico where it was called *tlaolli* in the Nahuatl language (Cabrera, 2022). Domesticated grains, dated 4000 years b. C., were found in a cave in Puebla, Mexico. Its cultivation expanded to the whole American continent and later on, after the Spanish conquest in 1521, to the rest of the world. The maize grain is covered by a coat called testa. The testa is fused with the pericarp (fruit wall) and both form a single outermost layer. Internally, the grain is unequally divided into two parts by epithelium. The upper big part is the endosperm and the lower small part is the embryo
M100	Maize, 100%
M85-SP15, M85-SC15, M60-SP40, M60-SC40	Mixtures of maize with the two types of sorghum, White (SB) and 'pearled' or decorticated (SP) at different proportions
Masa	Dough in Spanish
Panicle	A much-branched inflorescence. Some authors distinguish it from a compound spike inflorescence, by requiring that the flowers (and fruit) be pedicellate (having a single stem per flower). The branches of a panicle are often racemes (*racimo* in Spanish, i.e. grapes). A panicle may have determinate or indeterminate growth (Wikipedia, 2024)
SC100	Red colored sorghum, 100%
Sorghum	*Sorghum bicolor* L. Moench is a grain originally from Africa where it was domesticated around 8000 b. C. in Ethiopia and Sudan. It later spread to East and South Africa. Wall paintings and archaeological excavations have provided evidence of the cultivation of sorghum in Egypt in the 7th century b. C. (https://www.alimentarium.org/en/fact-

Acronyms or terms	Meaning
	sheet/sorghum#:~:text=Sorghum%20originated%20in%20the%20heart, the%207th%20century%20BCE). It has the same parts as maize grains: Testa, pericarp, endosperm, and embryo:



(Courtesy of Ravi-Shankar and Dayanandan, 2020)

It was introduced to Mexico in the second half of the Twentieth Century because at that time maize was the staple food of people and it was not allowed to use it for feeding cattle. For that reason people in Mexico associates sorghum as an animal feedstuff and not to be edible by persons. Mexico has become the highest importer of sorghum in the world in spite of being the fourth producing country with a volume equivalent to 6,202,920 Tons/year. It presents an internal deficit for animal feed and beer production importing 1,878,474 tons/year (FAO, 2017). In Africa is just the opposite: Sorghum is for people and not for animals

SP100	'Pearled' or decorticated sorghum, 100%
Tamales	The word tamal comes from the Nahuatl "tamalli" (maize dough cooked with lime, Cabrera, 2002). Mexico is the country with the greatest variety of tamales in the world. There are more than 500 types in Mexico
Tortilla	Spanish name for maize made flat unleavened bread cooked on a hot surface. The full grains are cooked with lime or $Ca(OH)_2$
Totomochtle, totomoxtle	Leaves that wrap around the cob and are used in Mexico for forage, for cigarette formation, or to prepare autochthonous foods such as tamales. It come from the Nahuatl "totomochtli" (Cabrera, 2002)
w.b.	Wet basis

Acknowledgements

Brabender laboratory scale extruder and scientific and technical support were provided by Mrs. Yoja Gallardo, M.S. (*Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, ENCB-IPN*, Mexico City Mexico) and Mr. René Manzur (*Instituto de Investigaciones para la Industria Alimenticia*, La Habana, Cuba). Authors acknowledge Mr. F. Laso, P.E. and Víctor Núñez, P.E. for their helpful advice during the course of this research and the provision of whole and pearled sorghum supplied by Kieselguhr Co. Technical assistance of Mr. Héctor Cejudo, MS., and Mrs. Ofelia Ramírez of Quality Laboratories, INIA, for tannins analyses is acknowledged. Authors also thank to *INIA* for providing maize white maize grain. Finantial support was granted by *Conacyt*, Mexico, through the Project PCALBNA-022816 -including the construction of the low cost extruder and other materials- and by Organization of American States, O.A.E., in Mexico, *OEA* in Spanish, through the Project "*Industrialización del Maíz para Consumo Humano*" (Industrialization of maize for human consumption, in Spanish).

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[5] Nahuatlism is the term used to refer to words in the Spanish language that originate from Nahuatl. It is also known in Spanish by the name "aztequismo" (or "Aztec-ism") Many nahuatlisms are only known in Mexican Spanish, since the majority of Nahuatl speakers are concentrated in that country

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[6] Polyglucids would be the right term instead of polysaccharides since the monomer is glucose not sucrose (note of last author)