

DYNAMICS OF FUNCTIONAL PROPERTIES OF MAIZE FLOURS FERMENTED WITH LACTIC ACID BACTERIA (LAB)-CONSORTIUM ISOLATED FROM CEREALS



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Abstract: The dynamics of functional properties of maize fermented with lactic acid (LAB) consortium from cereals were evaluated. Maize was processed into flour and fermented with LAB-consortium isolated from maize and sorghum in the following combination Lactobacillus plantarum WCFS1 + Lactobacillus rhamnosus GG, ATCC 53/03 + Lactobacillus nantensis LP33 + Lactobacillus fermentum CIP 102980 + Lactobacillus reuteri DSM 20016, and Pediococcus acidilactici DSM 20284 + Lactobacillus fermentum CIP 102980 + Lactobacillus brevis ATCC 14869 + Lactobacillus nantensis LP33 + Lactobacillus plantarum WCFS1 respectively and then naturally to determine their effect on the functional properties of maize. The result showed a gradual decrease in bulk density with increasing fermentation period from 0.82 ± 0.02 g/mL to 0.80 \pm 0.03 g/mL (natural fermentation), from 0.82 \pm 0.02 g/mL to 0.79 \pm 0.03 g/mL (LAB-consortium from maize fermentation) and from 0.82 ± 0.02 g/mL to 0.78 ± 0.03 g/mL (LAB-consortium from sorghum fermentation). The swelling capacity decreased from 0.31 \pm 0.03% (0 h) to 0.20 \pm 0.03% (48 h), from 0.31 \pm 0.03% (0 h) to 0.18 \pm 0.02% and from 0.31 \pm 0.03% to 0.19 \pm 0.01% in natural, LAB-consortium from maize and LAB-consortium from sorghum fermentation respectively. Water holding capacity decreased from 1.5 \pm 0.03 mL/g to $0.2 \pm 0.03 \text{ mL/g}$ (naturally fermentation), from $1.5 \pm 0.03 \text{ mL/g}$ to $0.4 \pm 0.02 \text{ mL/g}$ and from 1.5 \pm 0.03 mL/g to 1.0 \pm 0.03 mL/g in LAB-consortium from maize and LAB-consortium from sorghum fermentation respectively. Oil holding capacity (OHC) increased significantly (p<0.05) with increase in the fermentation periods from 8.00 \pm 0.03 mL/g to 9.50 \pm 0.02 mL/g (natural fermentation), 8.00 \pm 0.03 mL/g to 9.80 ± 0.03 mL/g (LAB-consortium from maize fermentation) and from 8.00 ± 0.03 mL/g to 9.73 ± 0.03 mL/g (LAB-consortium from sorghum fermentation). The least gelation concentration ranged from 3.0% in the unfermented sample to 6.0% in the various fermentation products. The variations differ significantly (p<0.05) with the unfermented sample. Emulsion capacity (EC) of the maize flour sample increased with increasing fermentation period from $41.03 \pm 2.48\%$ to $59.02 \pm 2.44\%$ (naturally fermentation), from $41.03 \pm$ 2.48% to 62.12 \pm 3.10% and from 41.03 \pm 2.48% to 61.34 \pm 2.10% in LAB-consortium from maize and LAB-consortium from sorghum fermentation respectively. This suggests the potentials of LAB-consortia fermentation in improving nutritional and functional properties of maize flour. Keywords: Functional properties, maize flour, fermentation, LAB-consortium.

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Introduction

Maize (*Zea mays*) belongs to the family of grasses (*Poaceae*) and is cultivated globally as one of the most important cereal crops (Ranum *et al.*, 2014). Maize contains approximately 72% starch, 10% protein, and 4% fat, supplying an energy density of 365 Kcal/100g and is grown worldwide, with the United States, China, and Brazil being the top three maize-producing countries (Ranum *et al.*, 2014; Gwirtz and Garcia, 2014). Maize is not only an important source of nutrients for human, but also a vital constituent in formulation of animal feed. It is also a raw material for manufacture of many industrial products and can be processed into a wide range of foods, snacks and beverages (CWFS, 2013; Sanni and Adesulu, 2013).

Maize (*Zea mays*) is an important cereals which serves as major source of carbohydrate, protein and calorie. However bioavailability is low due to the presence of antinutritional factors such as phytic acid, polyphenols and tannins (Maidala *et al.*, 2013). Maize contains high amount of starch and its digestibility is greatly influenced by plant type, physicochemical characteristics of the starch as well as composition, processing and storage conditions (Singh *et al.*, 2012; Olanipekun *et al.*, 2015).

Fermentation is one of the processes that decreases the level of anti-nutrients in food grains and increases the starch and protein digestibility as well as nutritive value (Singh *et al.*, 2012) and leads to an increase in protein content, enhancement of carbohydrate accessibility, improvement in amino acid balance, decrease in anti-

nutritional factors like tannin and phytic acid (Singh *et al.*, 2012). Fermented food has many beneficial products metabolized by bacteria such as biomass proteins, amino acids, vitamins, minerals, flavor and aroma compounds as well as carbohydrate. Products of respiratory and biosynthetic pathways such as lactic acid, ethanol, acetaldehyde and pyruvic acid are also produced which alters the pH of foods to levels that they control the growth of pathogenic microorganisms. This therefore enhances food safety and shelf life thus aiding in food preservation (Onyango *et al.*, 2013; Ojokoh and Bello, 2014).

Lactic acid bacteria (LAB) are a large group of closely related bacteria that have similar properties such as lactic acid production, which is an end product of the fermentation. LAB includes Lactobacillus, Lactococcus, Leuconostoc Streptococcus and species. LAB fermentation is a common way of preparing food traditionally in Africa. Some of the traditionally fermented foods in Africa include maize porridge, alcoholic beverages and dairy products. Some of the main reasons for the fermentation practice using LAB are to increase food palatability and improve the quality of food by increasing the availability of proteins and vitamins (Masood et al., 2011; Huili et al., 2011). Furthermore, LAB confers preservative and detoxifying effects on food as well. When used regularly, LAB fermented foods boost the immune system and strengthen the body in the fight against pathogenic bacterial infections. Thus, LAB fermentation is not only of a major economic importance,

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but it also promotes human health in Africa (Chelule *et al.*, 2010; Onyango *et al.*, 2013). The present study is aimed at, evaluating the effect of lactic acid bacteria (LAB)-consortium fermentation on the functional properties of maize flour.

Materials and Methods Source of materials

White variety of maize (*Zea mays*) was bought from Mushin markets of Lagos, Lagos State, Nigeria and transported to the laboratory in clean polythene bags for analysis at Federal Institute of Industrial Research Oshodi (FIIRO) where it was identified. Lactic acid bacteria were obtained from stock previously isolated from fermenting maize and sorghum. All the chemicals used were of analytical grade (AR).

Sample preparation

The raw grains of the maize were freed of foreign materials, washed with clean tap water and rinsed with distilled water. The samples were dried with hot air oven (GL, England) at 60° C for 8 h. The dried samples were milled into powder using milling machine (CNC, Germany) disinfected with 70% ethanol and stored in clean air tight containers at 4° C for further use (Singh *et al.*, 2012).

Inoculum preparation

Five (5) lactic acid bacteria previously isolated from each of fermenting maize and fermenting sorghum were combined as follows, Lactobacillus plantarum WCFS1 + Lactobacillus rhamnosus GG, ATCC 53/03 Lactobacillus nantensis LP33 + Lactobacillus fermentum CIP 102980 + Lactobacillus reuteri DSM 20016, for consortium from maize; and Pediococcus acidilactici DSM 20284 + Lactobacillus fermentum CIP 102980 + Lactobacillus brevis ATCC 14869 + Lactobacillus nantensis LP33 + Lactobacillus plantarum WCFS1, for consortium from sorghum. These were grown in a 250 ml Erlenmeyer flasks containing 210 ml MRS broth respectively, and incubated for 48 h in an orbital shaker incubator (REMI/396LAG) at 37°C for the inoculum to build-up. The inocula were harvested by centrifugation at 5000 g for 10 min and maintained in fresh MRS broth before fermentation. The washed cells were diluted using sterile distilled water to obtain 0.5 McFarland, standard (Dajanta et al., 2009).

Fermentation of maize flours

Fermentation was carried out following a modification of the method described by Sigh *et al.* (2012). The flour samples were mixed with sterile distilled water (1:2 w/v). Exactly 500 g each of the maize flours was mixed with 1000 mL of distilled water in sterile fermentation containers with the addition of 0.5 g/L potassium sorbate (to inhibit fungal growth and other contaminating organisms). The mixture was inoculated with 10 ml of 10^8 cells/mL (measured using McFarland standard) of the mixture of the lactic acid bacteria suspension and allowed to ferment. One of the set-ups was also allowed to ferment naturally without addition of potassium sorbate and starter organisms. Samples were withdrawn at 12 h intervals at periods of 0, 12, 24, 36 and 48 h for analysis.

Determination of functional properties

Bulk density was determined according to the method given by Chau and Huang (2003). Water holding capacity

(WAC) and oil holding capacity (OHC) was determined according to the method described by Singh *et al.* (2012). Swelling capacity of the flour was determined according to the method given by Robertson *et al.* (2000). The gelation properties of the flour under study were determined with the method described by Aremu *et al.* (2008). The emulsion activity of the various flours was determined using the method of described by Suresh and Samsher (2013).

Results and Discussion

The effect of fermentation on the bulk density of maize is presented in Figure 1. Bulk density decreased gradually with increasing fermentation period. In natural fermentation, it decreased from 0.82 ± 0.02 g/mL (0 h) to 0.80 ± 0.03 g/mL (48 h), from 0.82 ± 0.02 g/mL (0 h) to 0.79 ± 0.03 g/mL (48 h) in sample fermented with LABconsortium from maize and from 0.82 ± 0.02 g/mL (0 h) to 0.78 ± 0.03 g/mL (48 h) in the sample fermented with LAB-consortium from sorghum. The variations in the bulk density of the samples do not differ significantly (p>0.05). The report of the present investigation is in agreement with the work of Singh et al. (2012) who reported a gradual decrease in bulk density of maize from 0.72 \pm 0.00 g/ml to $0.60 \pm 0.01 \text{ g/ml}$ and from 0.69 ± 0.00 to 0.61 \pm 0.01 in sorghum after 36 hours of fermentation. Adebowale and Maliki (2011) reported a gradual decrease in BD in the range of 0.80 to 0.63 g/ml with increasing fermentation period of pigeon pea flours which are comparable to the values obtained in the present investigation. Bulk density is a measure of the load the flours can carry if allowed to rest directly on one another and decrease in bulk density is desirable in preparation of infant foods; fermentation has been reported as a traditional means of preparing low density weaning foods (Desikachar, 1980; Singh et al., 2012). The density of processed products dictates the characteristics of its container or package product density influences the amount and strength of packaging material, texture or mouth feel as noted by Adebowale and Maliki (2011) and Wilhelm et al. (2004).



NF = Naturally fermented, MF = LAB-consortium from maize fermented; SF = LAB-consortium from sorghum fermented; Values are mean of triplicate determination.

The effect of fermentation on the swelling capacity (SC) of maize flours is presented in Fig 2. The result shows that swelling capacity decreased with increasing fermentation

Fig. 1: Effect of fermentation on the bulk density (g/ml) of maize flours

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period gradually. It decreased from $0.31 \pm 0.03\%$ (0 h) to $0.20 \pm 0.03\%$ (48 h), from $0.31 \pm 0.03\%$ (0 h) to $0.18 \pm 0.02\%$ and from $0.31 \pm 0.03\%$ to $0.19 \pm 0.01\%$ in natural, LAB-consortium from maize and LAB-consortium from sorghum fermentation, respectively.



NF = Naturally fermented, MF = LAB-consortium from maize fermented; SF = LAB-consortium from sorghum fermented; Values are mean of triplicate determination.

Fig. 2: Effect of fermentation on the swelling capacity (SC) (%) of maize flours

The variations in the swelling capacity of the samples differ significantly (p<0.05) when compared between unfermented and fermented samples. The decrease in the swelling capacity in this study during fermentation agreed with the reports of Adebowale and Maliki (2011) and Singh *et al.* (2012) who reported decrease in SC with increasing fermentation in sorghum, millet, sorghum and pigeon pea, respectively.

The result of the water holding capacity (WHC) of maize flours showed a decreasing trend with increasing duration of fermentation (Fig. 3). It decreased from 1.5 ± 0.03 mL/g in the raw sample to 0.2 ± 0.03 mL/g in naturally fermented sample. The decrease ranged from 1.5 ± 0.03 mL/g to 0.4 ± 0.02 mL/g and from 1.5 ± 0.03 mL/g to 1.0 ± 0.03 mL/g in samples fermented with LAB-consortium from maize and LAB-consortium from sorghum respectively. The variations in water holding capacity of the samples differ significantly (p<0.05) when compared between unfermented and fermented samples at 48 h.



 \overline{NF} = Naturally fermented, \overline{MF} = LAB-consortium from maize fermented; \overline{SF} = LAB-consortium from sorghum fermented; Values are mean of triplicate determination.

Fig. 3: Effect of fermentation on the water holding capacity (WHC) (mL/g) of maize flours

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Water holding capacity was found to be highest in unfermented sample $(1.5 \pm 0.03 \text{ mL/g})$ followed by LAB-consortium from sorghum fermented sample $(1.0 \pm 0.03 \text{ mL/g})$

mL/g) and LAB-consortium from maize fermented sample $(0.4 \pm 0.02 \text{ mL/g})$ while the naturally fermented sample was the least $(0.2 \pm 0.03 \text{ mL/g})$ after 48 h. The variations in water holding capacity of the samples differ significantly (p<0.05) when compared between unfermented and fermented samples. Beugre et al. (2014) reported increase in water absorption capacity from 1.2 -1.8 ml/g in maize which disagrees with the present study. The report of decrease in WHC of maize and sorghum flours from 0.92-0.77 ml/g and 1.26-1.03 ml/g respectively reported by Singh et al. (2012) is consistent with the present study. Also, Elkhalifa et al. (2005) reported decrease in WHC after fermentation of sorghum for 8-24 h. Gernah et al. (2011) and Ocheme et al. (2015) noted increase in water absorption capacity of maize and sorghum after malting and germination respectively. The result of this study is also comparable to the work of Adebowale and Maliki (2014) who reported decrease in WHC of pigeon pea from 142.0 g/100g to 113.0 g/100g after a 5-day fermentation. Water binding capacity is a useful indication for the incorporation of flours into aqueous food formulation especially those involving dough. WHC gives an indication of the amount of water available for gelatilization and low absorption capacity is desirable for making thinner gruels as reported by (Singh et al., 2012). The result of this study suggests that the fermented flours may find application in preparation of weaning foods and in the production of some baked products (Singh et al., 2012). Also, LAB-consortium fermentation of achieving low water absorption for maize flours.

The result of oil holding capacity (OHC) of maize flour under study increased significantly (p<0.05) with increase in the fermentation periods. It increased from the initial value of 8.00 \pm 0.03 mL/g in the raw sample to 9.50 \pm 0.02 mL/g in natural fermentation, from 8.00 \pm 0.03 mL/g to 9.80 \pm 0.03 mL/g in LAB-consortium from maize fermented sample and from 8.00 \pm 0.03 mL/g to 9. 73 \pm 0.03 mL/g in LAB-consortium from sorghum fermented sample (Figure 4). The variations in Oil holding capacity (OHC) of the samples differ significantly (p<0.05) when compared between unfermented and fermented samples. The increase in OHC was found to be highest in maize fermented with LAB-consortium from maize (9.80 ± 0.03) mL/g), followed by LAB-consortium from sorghum fermented sample (9.73 \pm 0.02 mL/g) and the naturally fermented sample $(9.50 \pm 0.03 \text{ mL/g})$ while the unfermented sample was the least (8.00 \pm 0.03 mL/g). This suggests the effectiveness of the LAB-consortia in improving the OHC more than the natural fermentation. The variations in Oil holding capacity (OHC) of the samples differ significantly (p<0.05) when compared between unfermented and fermented samples. Singh et al. (2012) reported that fermentation increased the OHC in the range of 8.0 to 9.7 for sorghum, millet and maize which is in agreement with the present investigation. The result of this study is also comparable to the report of Acuña et al. (2012) for soybean. Elkahlifa et al. (2005) reported about 7% increase in the oil absorption capacity of sorghum fermented for 8 h. The increase in the OHC suggests that the flours could be useful in food formulation and fortification where an oil holding capacity is a factor (Singh et al., 2012). The water and oil binding capacity of food protein is dependent on the intrinsic factors like amino acid composition, protein conformation and surface polarity or hydrophobicity. The ability of the proteins of these flours to bind with oil makes it useful in

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food system where optimum oil absorption is desired. This makes flour to have potential functional uses in foods such as sausage production (Suresh and Samsher, 2013).



 \overline{NF} = Naturally fermented, \overline{MF} = LAB-consortium from maize fermented; \overline{SF} = LAB-consortium from sorghum fermented; Values are mean of triplicate determination.

Fig. 4: Effect of fermentation on the oil holding capacity (OHC) (mL/g) of maize flours

The least gelation concentration of maize flours ranged from 3.0% in the unfermented sample to 6.0% in the various fermentation products (Table 1). The variations differ significantly (p < 0.05) with the unfermented sample. Gelation power is an index of gelling tendency of sample and it is an important factor in food preparations (Adebowale and Maliki, 2011). In the present study the least gelation concentration decreased with increasing fermentation period. In the maize flours, it ranged from 3.0% in the unfermented sample to 6.0% in the various fermentation products. The variations differ significantly (p<0.05) with the unfermented sample. Adebowale and Maliki (2011) also reported decrease in gelation power with increasing fermentation time in pigeon pea which agreed with the present investigation. The variations in the gelation capacities of the present investigation could be attributed to the relative ratios of different constituents such as proteins, carbohydrates and lipids that make up the flours which suggest that interactions between the various components may have a significant impact on the functional properties of the products (Adebowale and Maliki, 2011).

Table 1: Effect of fermentation on the least gelation concentration of maize flour under study

Flour Conc. (%)	0 h		12 h			24 h			36 h			48 h	
	F*	NF	MF	SF	NF	MF	SF	NF	MF	SF	NF	MF	SF
1.0	V	V	V	V	V	V	V	V	V	V	V	V	V
2.0	V	V	V	V	V	V	V	V	V	V	V	V	V
3.0	G	V	V	V	V	V	V	V	V	V	V	V	V
4.0	G	V	G	G	V	V	V	V	V	V	V	V	V
5.0	G	G	G	G	V	G	G	V	G	G	V	G	G
6.0	G	G	G	G	G	G	G	G	G	G	G	G	G
7.0	G	G	G	G	G	G	G	G	G	G	G	G	G
8.0	G	G	G	G	G	G	G	G	G	G	G	G	G
9.0	G	G	G	G	G	G	G	G	G	G	G	G	G
10.0	G	G	G	G	G	G	G	G	G	G	G	G	G
LGC	3.0 ^a	$5.0^{b,d}$	$4.0^{a,b}$	$4.0^{a,b}$	6.0°	$5.0^{b,d}$	$5.0^{b,d}$	$6.0^{c,d}$	$5.0^{b,d}$	$5.0^{b,d}$	$6.0^{c,d}$	$5.0^{b,d}$	$5.0^{b,d}$

 F^* = Unfermented sample; NF = naturally fermented; MF = fermented with LAB consortium from maize; SF = fermented with LAB consortium from sorghum; V = viscous; G = gel; LGC = Least gelation concentration; Values with the same superscript are not significantly difference (P>0.05)

The result of the emulsion capacity (EC) of the maize flour sample increased with increasing fermentation period. The EC of maize increased from $41.03 \pm 2.48\%$ (0 h) to $59.02 \pm 2.44\%$ (48 h) in naturally fermented sample, from $41.03 \pm 2.48\%$ (0 h) to $62.12 \pm 3.10\%$ (48 h) in LAB-consortium from maize fermented sample and from $41.03 \pm 2.48\%$ (0 h) to $61.34 \pm 2.10\%$ (48 h) in LABconsortium from sorghum fermented sample. The variations in the emulsion capacity of maize compared favourably between the naturally fermented, LABconsortium from maize and LAB-consortium from sorghum fermented samples but do not show significant different statistically (p>0.05) except 48 h LABconsortium from maize fermented sample which differ significantly with the 48 h naturally fermented sample. However, the values obtained for EC in fermented maize differ significantly (p<0.05) when compared with the unfermented maize sample from 12 h to 48 h (Fig. 5). The different values obtained for the fermented product differ significantly (p<0.05) when compared with the unfermented samples in each substrate from 24 h to 48 h. However, the increases do not differ significantly (p>0.05) when compared between naturally fermented, LABconsortium from maize and LAB-consortium from sorghum fermented samples. The values obtained in the present study are comparable to the work of Suresh and Samsher (2013) who reported 43.88% and 41.48% in wheat and rice flours respectively. Difference in the EC of the various samples may be related to solubility proteins as noted by Suresh and Samsher (2013). Hydrophobicity of protein has been attributed to influence their emulsifying properties (Kaushal *et al.*, 2012).



NF = Naturally fermented, MF = LAB-consortium from maize fermented; SF = LAB-consortium from sorghum fermented; Values are mean of triplicate determination.

Fig. 5: Effect of fermentation on the emulsion capacity (EC) (%) of maize flours

Conclusion

The functional properties of maize flours improved after natural fermentation, LAB-consortium from maize and LAB-consortium from sorghum fermentation. The highest improvements were observed more in the consortia fermented samples than the naturally fermented samples. These suggest the possible use LAB-consortium fermentation isolated from cereals as starter organisms in improving the nutritional qualities of local staple cereal products.

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Conflict of Interest

The authors have declared no conflict of interest.

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