



## Dynamics of functional properties of sorghum flours fermented with lactic acid bacteria (LAB)-consortium isolated from cereals

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### Abstract

The functional properties of sorghum flours fermented with lactic acid (LAB) consortium isolated from fermenting maize and sorghum were evaluated. Sorghum was processed into flour and fermented with LAB-consortium previously isolated from maize (*Lactobacillus plantarum* WCFS1 + *Lactobacillus rhamnosus* GG, ATCC 53/03 + *Lactobacillus nantensis* LP33 + *Lactobacillus fermentum* CIP 102980 + *Lactobacillus reuteri* DSM 20016) and sorghum (*Pediococcus acidilactici* DSM 20284 + *Lactobacillus fermentum* CIP 102980 + *Lactobacillus brevis* ATCC 14869 + *Lactobacillus nantensis* LP33 + *Lactobacillus plantarum* WCFS1) respectively and then naturally to evaluate their effects on the functional properties of the sorghum flour at 12 h intervals. There was gradual decrease in bulk density with increasing fermentation period from  $0.86 \pm 0.03$  g/mL to  $0.81 \pm 0.03$  g/mL (natural fermentation), from  $0.86 \pm 0.03$  g/mL to  $0.80 \pm 0.03$  g/mL (LAB-consortium from maize fermentation) and from  $0.86 \pm 0.03$  g/mL to  $0.81 \pm 0.03$  g/mL (LAB-consortium from sorghum fermentation). The swelling capacity decreased from  $0.28 \pm 0.01$  % (0 h) to  $0.21 \pm 0.03$  % (48 h), from  $0.28 \pm 0.01$  % (0 h) to  $0.20 \pm 0.03$  % and from  $0.28 \pm 0.01$  % to  $0.21 \pm 0.03$  % in natural, LAB-consortium from maize and LAB-consortium from sorghum fermentation respectively. Water holding capacity decreased from  $1.6 \pm 0.01$  mL/g to  $1.2 \pm 0.02$  mL/g (naturally fermentation), from  $1.6 \pm 0.01$  mL/g to  $1.0 \pm 0.02$  mL/g and from  $1.6 \pm 0.01$  mL/g to  $1.2 \pm 0.02$  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  $8.60 \pm 0.01$  mL/g to  $9.50 \pm 0.02$  mL/g (natural fermentation),  $8.60 \pm 0.01$  mL/g to  $9.70 \pm 0.03$  mL/g (LAB-consortium from maize fermentation) and from  $8.60 \pm 0.01$  mL/g to  $9.78 \pm 0.01$  mL/g (LAB-consortium from sorghum fermentation). The least gelation concentration ranged from 3.0% in the unfermented sample to 7.0% in the various fermentation products. The variations differ significantly ( $p < 0.05$ ) with the unfermented sample. Emulsion capacity (EC) increased with increasing fermentation period from  $56.76 \pm 0.04$  % to  $63.00 \pm 0.1$  %, from  $56.76 \pm 0.04$  % to  $65.48 \pm 1.46$  % and from  $56.76 \pm 0.04$  % to  $64.66 \pm 1.98$  % in natural, LAB-consortium from maize and LAB-consortium from sorghum fermentation respectively. This suggest the potentials of LAB-consortia fermentation in improving nutritional and functional properties of sorghum flour.

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### Introduction

Sorghum (*Sorghum bicolor* (L.) Moench) is a tropical cereal grass that is thought to have first been domesticated in North Africa around 1000 BC (Atter, 2012). There are many varieties of Sorghum bicolor, ranging in colour from white through red to brown and mixed classes in the grain standards (Singh *et al.*, 2012; Attaer, 2012). Sorghum (*Sorghum bicolor*), is a source of protein, carbohydrate and calorie; however, bioavailability is low due to the presence of anti-

nutritional factors such as phytate, polyphenols and tannins (Maidala *et al.*, 2013). Sorghum contains high amount of starch and its digestibility is greatly influenced by plant type, plant microstructure, composition, processing and storage conditions (Singh *et al.*, 2012; Olanipekun *et al.*, 2015).

Fermentation is known to decrease the level of anti-nutrients in food grains and increase the starch and protein digestibility as well as nutritive value and leads to an increase in protein content, enhancement of carbohydrate accessibility and improvement in

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amino acid balance (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, aiding in food preservation (Onyango *et al.*, 2013; Ojokoh and Bello, 2014).

Fermentation using lactic acid bacteria (LAB) have been practiced 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). Lactic acid bacteria (LAB) confers preservative and detoxifying effects and LAB fermented foods boost the immune system and strengthen the body in the fight against pathogenic bacterial infections when used regularly. Thus, LAB fermentation is not only of a major economic importance, 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 sorghum flour.

## Materials and Methods

### Source of Materials

Red variety of sorghum (*Sorghum bicolor* (L.) Moench) was bought from Yaba market 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 sorghum 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 x g for 10 minutes 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 sorghum flours

Fermentation was carried out following a modification of the method described by Singh *et al.* (2012). The flour samples were mixed with sterile distilled water (1:2 w/v). Exactly 500 g of the sorghum flour 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<sup>8</sup> 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 was determined with the method described by Areemu *et al.* (2008). The emulsion activity of the various flours was determined using the method described by Suresh and Samsher (2013).

## Results

The effect of LAB-consortium fermentation on the bulk density of sorghum is presented in Figure 1. Bulk density decreased gradually with increasing fermentation period from  $0.86 \pm 0.02$  g/mL to  $0.81 \pm 0.03$  g/mL (naturally fermented), from  $0.86 \pm 0.02$  g/mL to  $0.80 \pm 0.03$  g/mL (fermented with LAB-consortium from maize) and from  $0.86 \pm 0.02$  g/mL to  $0.81 \pm 0.03$  g/mL (fermented with LAB-consortium from sorghum). The variations in the bulk density of the samples do not differ significantly ( $p>0.05$ ).

The swelling capacity (SC) of fermented sorghum flour is presented in Figure 2. The result shows that swelling capacity decreased from the initial value of  $0.28 \pm 0.01\%$  to  $0.21 \pm 0.03\%$ ,  $0.28 \pm 0.01\%$  to  $0.20 \pm 0.03\%$  and  $0.28 \pm 0.01\%$  to  $0.21 \pm 0.03\%$  in naturally, LAB-consortium from maize and LAB-consortium from sorghum fermented samples respectively.

The water holding capacity showed a decreasing trend with increasing duration of fermentation. It decreased from  $1.6 \pm 0.01$  mL/g to  $1.20 \pm 0.03$  mL/g, from  $1.6 \pm 0.01$  mL/g to  $1.00 \pm 0.02$  mL/g (fermented with LAB-consortium from maize) and from  $1.6 \pm 0.01$  mL/g to  $1.20 \pm 0.02$  mL/g (Figure 3). The variations in water holding capacity differ significantly ( $p<0.05$ ) when compared between unfermented and fermented samples.

The result of the oil holding capacity (OHC) of LAB-consortium fermented sorghum flour increased significantly ( $p<0.05$ ) with increase in the fermentation periods (Figure 4). It increased from  $8.60 \pm 0.01$  mL/g to  $9.50 \pm 0.03$  mL/g (naturally fermented), from  $8.60 \pm 0.01$  mL/g to  $9.70 \pm 0.03$  mL/g (fermented with LAB-consortium from maize) and from  $8.60 \pm 0.01$  mL/g to  $9.78 \pm 0.01$  mL/g (fermented with LAB-consortium from sorghum).

The least gelation concentration of sorghum flours ranged from 3.0% in the unfermented sample to 7.0% in the various fermentation products. The variations differ significantly ( $p<0.05$ ) with the unfermented sample.

Figure 5 presents the emulsion capacity of fermented sorghum flour. It was observed that the emulsion capacity (EC) increased with increasing fermentation period. The EC increased from the original value of  $56.76 \pm 0.04\%$  to  $63.00 \pm 0.01\%$  in naturally fermented sample, from  $56.76 \pm 0.04\%$  to  $65.48 \pm 1.46\%$  in LAB-consortium from maize fermented sample and from  $56.76 \pm 0.04\%$  to  $64.66 \pm 1.98\%$  in LAB-consortium from sorghum fermented sample. The different values obtained for the fermented product differ significantly ( $p<0.05$ ) when compared with the unfermented sample from

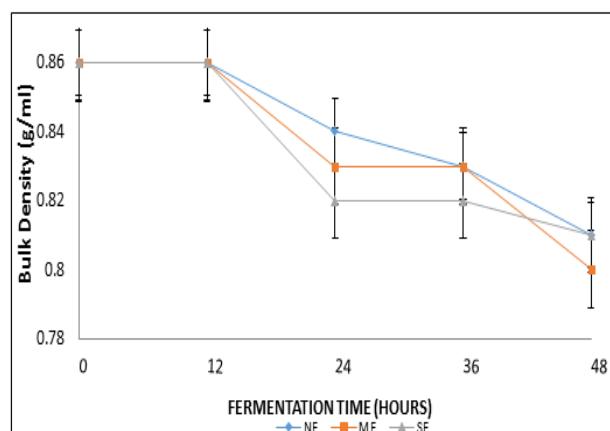


Figure 1. The Bulk Density (g/ml) of sorghum flours Fermented with LAB-consortium  
NF = Naturally fermented, MF = LAB-consortium from maize fermented; SF = LAB-consortium from sorghum fermented. Values are mean of triplicate determinations.

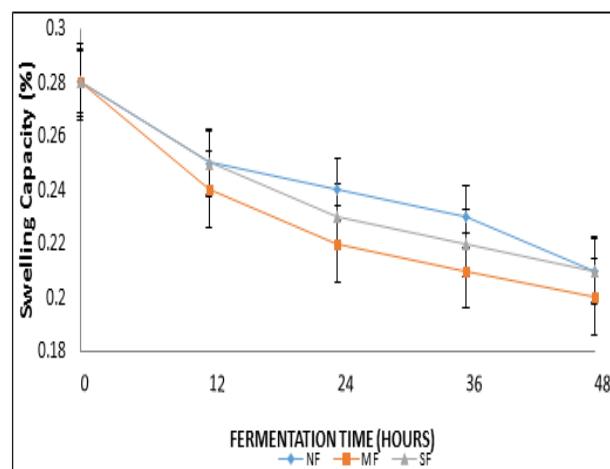


Figure 2. The Swelling capacity (%) of sorghum flours Fermented with LAB-consortium  
NF = Naturally fermented, MF = LAB-consortium from maize fermented; SF = LAB-consortium from sorghum fermented. Values are mean of triplicate determinations.

24 h to 48 h. However, the increases do not differ when compared between naturally fermented, LAB-consortium from maize and LAB-consortium from sorghum fermented samples.

## Discussion

The density of processed products determines the characteristics of its container and influences the amount and strength of packaging material (Adebawale and Maliki, 2011). Bulk density is a measure of the load the flours can carry if allowed to rest directly on one another. Decrease in bulk density is desirable in preparation of infant foods and fermentation has been reported as a traditional means of preparing low density weaning foods (Singh *et al.*, 2012). In the present study, the bulk density (BD) of

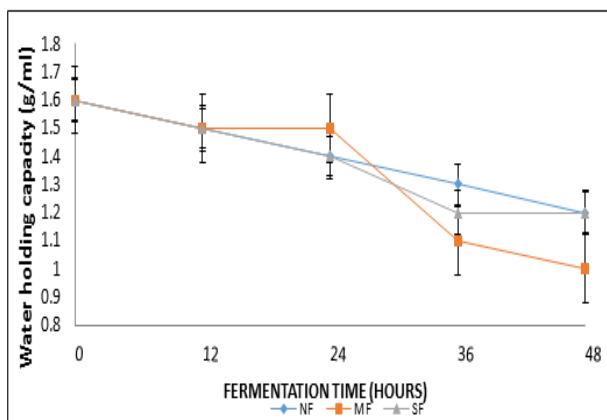


Figure 3. Water Holding Capacity (mL/g) of LAB-consortium Fermented Sorghum Flours  
NF = Naturally fermented, MF = LAB-consortium from maize fermented; SF = LAB-consortium from sorghum fermented.  
Values are mean of triplicate determinations.

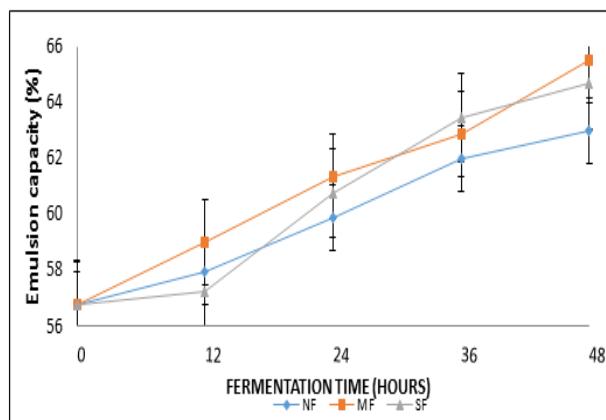


Figure 5. Emulsion Capacity (EC) (%) of LAB-consortium Fermented Sorghum Flours  
NF = Naturally fermented, MF = LAB-consortium from maize fermented; SF = LAB-consortium from sorghum fermented.  
Values are mean of triplicate determinations.

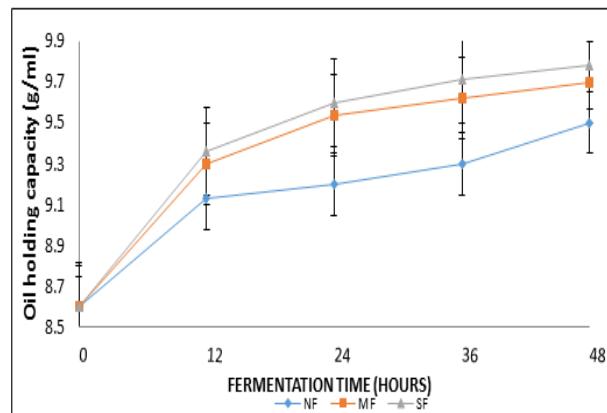


Figure 4. Oil Holding Capacity (mL/g) of LAB-consortium Fermented Sorghum Flours  
NF = Naturally fermented, MF = LAB-consortium from maize fermented; SF = LAB-consortium from sorghum fermented.  
Values are mean of triplicate determinations

sorghum decreased  $0.86 \pm 0.02$  g/mL to  $0.81 \pm 0.02$  g/mL. This report is in agreement with the work of Singh *et al.* (2012) who reported a gradual decrease in bulk density of sorghum from  $0.69 \pm 0.00$  to  $0.61 \pm 0.01$  after 36 hours of natural fermentation. Also, Ocheme *et al.* (2015) noted decrease in BD of sorghum from  $0.77 - 0.70$  g/ml after 72 h germination which was attributed to breakdown of complex compounds such as protein as a result of modifications that occurred during germination.

The effect of fermentation on the swelling capacity (SC) of sorghum flour decreased with increasing fermentation period from 0.28% to 0.20. The variations in the swelling capacity differ significantly ( $p < 0.05$ ) when compared between unfermented and fermented samples. The decrease agreed with the reports of Adebawale and Maliki (2011) and Singh *et al.* (2012) who reported decrease in SC with increasing

fermentation in sorghum, millet, sorghum and pigeon pea respectively.

In the present study, the water holding capacity (WHC) showed a decreasing trend with increasing duration of fermentation. The report of decrease in WHC of sorghum flour from 1.26-1.03 ml/g by Singh *et al.* (2012) agreed 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 Adebawale 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. The sample fermented with the LAB consortia showed low absorption capacity than the unfermented sample and naturally fermented sample. Water binding capacity is a useful indication for the incorporation of flour into aqueous food formulation especially those involving dough. WHC gives an indication of the amount of water available for gelatinization 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).

In the present study, the oil holding capacity increased significantly with increasing fermentation periods. The highest increase was observed in samples fermented with LAB-consortia. OHC of sorghum increased from  $8.60 \pm 0.01$  mL/g to  $9.78 \pm 0.02$  mL/g. The variations in Oil holding capacity (OHC) of the sample differ significantly ( $p < 0.05$ )

when compared between unfermented and fermented samples. The work of Singh *et al.* (2012) who reported that fermentation increased the OHC in the range of 8.0 to 9.7 for sorghum, millet and maize is in agreement with the present study. The increase in the OHC suggests that the flours could be useful in food formulation where an oil holding capacity is a factor (Singh *et al.*, 2012). The water and oil binding capacity of food is dependent on amino acid composition, protein conformation and surface polarity or hydrophobicity (Suresh and Samsher, 2013). The ability of the proteins of these flours to bind with oil makes it useful in food system where optimum oil absorption is desired such as in sausages (Suresh and Samsher, 2013).

In the present study the least gelation concentration decreased with increasing fermentation period. It ranged from 4.0% in the unfermented sample to 7.0% in 36 h and 48 h naturally fermented sample. Gelation power is an index of gelling tendency of sample and it is an important factor in food preparations (Adebawale and Maliki, 2011). Decrease in gelation power with increasing fermentation time in pigeon pea has been reported by Adebawale and Maliki (2011) which agreed with the present investigation. The variations in the gelation capacities of the present investigation could be attributed to the relative ratios of proteins, carbohydrates and lipids that make up the flours which suggests that interactions between the various components may have a significant impact on the functional properties of the products (Adebawale and Maliki, 2011).

The result of the emulsion capacity was observed to increase with increasing fermentation period from  $6.76 \pm 0.04\%$  in the unfermented sample to  $65.48 \pm 1.46\%$  in LAB-consortium from maize fermented sample. The values obtained in the present study is 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 of proteins (Suresh and Samsher, 2013). Moreover, hydrophobicity of protein has been attributed to influence their emulsifying properties (Kaushal *et al.*, 2012).

## Conclusion

The present study has shown that the functional properties of sorghum flours improved after natural fermentation, LAB-consortium from maize and LAB-consortium from sorghum fermentation. The highest improvement were observed more in the consortia fermented samples than the naturally fermented

sample. This is an indication that LAB-consortium isolated from cereals as starter cultures are ideal in improving the nutritional qualities of staple cereal products.

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