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## Effects of Malting and Lactic Fermentation on Some Chemical and Functional Properties of Maize (*Zea mays*)

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### ABSTRACT

The effects of malting and fermentation on some chemical and functional properties of maize grains were determined. White maize kernels (variety TZW, 2005 harvest) were germinated over a period of 96 h and flours of 72 h germinated grains were fermented by back-slopping. Analysis of the selected physico-chemical parameters of the resulting samples were carried out using standard analytical methods. Germination of grains resulted in significant ( $p < 0.05$ ) increase in crude protein, total soluble sugars and reducing sugars up to 72 h and ash content. Conversely, there was significant decrease ( $p < 0.05$ ) in crude fat and carbohydrates during this period. After 72 h, protein and crude fibre content reduced slightly, while carbohydrate, crude fat and ash increased though not significantly ( $p > 0.05$ ). There was also a significant decrease ( $p < 0.05$ ) in pH (6.80-5.94) as germination progressed while there was a gradual increase in TTA which was not significant ( $p > 0.05$ ). Accelerated natural fermentation of the maize flour also showed a similar trend for all the parameters. Malting and fermentation gave rise to decrease in packed bulk density ( $1.17\text{-}0.54\text{ g mL}^{-1}$ ) swelling index ( $6.47\text{-}3.46\text{ mL g}^{-1}$ ) and viscosity ( $343.33\text{-}288.26\text{ cP}$ ) while increasing the water absorption capacity ( $3.70\text{-}3.92\text{ g g}^{-1}$ ) and reconstitution index ( $4.82\text{-}6.40\text{ mL g}^{-1}$ ). There was also a significant reduction in tannin content ( $2.62\text{-}0.42\text{ g/100 g}$ ), phytates ( $2.30\text{-}0.84\text{ g/100 g}$ ), oxalates ( $2.32\text{-}0.34\text{ g/100 g}$ ), cyanide content ( $2.20\text{-}0.42\text{ mg/100 g}$ ) and fibre ( $2.12\text{-}1.11\text{ g/100 g}$ ).

**Key words:** Germination, back-slopping, accelerated natural fermentation, viscosity, nutritional quality

### INTRODUCTION

Cereal grains constitute a very large percentage of human food and animal feed worldwide (Adeyemo *et al.*, 1992). Developing countries of the world are particularly dependent on cereals for supply of both energy and proteins (El-Adawy, 1997). The storage proteins of cereals are, however, known to be deficient in some essential amino acids, notably lysine, threonine and tryptophan (Tsen *et al.*, 1996). Traditional cereal-based food products are therefore generally of poor nutritional quality for both humans and animals in terms of quality and quantity of proteins (Oluwamukomi *et al.*, 2003). Maize (*Zea mays*) which is consumed in large quantities by infants, children and adults as well as livestock in Nigeria is deficient in lysine and tryptophan (Enwere, 1998). Efforts at improving the nutritional quality of maize have been largely geared towards supplementation with legumes or animal proteins (El-Adawy, 1997) as well as genetic

engineering (Liu, 1999; ITF, 2000). Work has also been done on germination (malting) as an alternative to genetic engineering for improving the nutritive value of maize, particularly in the developed countries (Oluwamukomi *et al.*, 2003; Obasi *et al.*, 2009; Eneche, 2009).

In the developing countries, malting and fermentation have been shown to be the most effective and convenient for the improvement of nutritional value and reduction of bulk and viscosity of cereal foods (Akpapunam and Dedeh, 1995; Akpapunam *et al.*, 1996; Ariahu *et al.*, 1999). In Nigeria, maize is germinated as a traditional processing technique in the production of such beverages as sekete and pito (Adeyemo *et al.*, 1992).

There is a growing interest in the formulation of food products, using composite blends of raw, malted and fermented maize with other legumes as a way of improving nutritional quality (Sefa-Dedeh and Afaakwa, 2001; Agu and Aluya, 2004). This technology requires good knowledge of the functional properties of the flours. There is therefore, need for more information on the biochemical changes that accompany the processes of malting and fermentation of maize on which beneficial processing manipulations can be based. This study was therefore designed to investigate the effect of malting and fermentation on some chemical and functional properties of the maize grain.

## MATERIALS AND METHODS

**Source of materials:** Approximately 3.0 kg of white maize (TZW, 2005 harvest) were purchased from the Department of Agronomy, University of Agriculture, Makurdi and utilized for the research work between January-June, 2006. After manual sorting and winnowing to remove stones, debris and defective seeds, the clean seeds were packaged in a 10 L plastic bucket, which was tightly covered with a lid and stored in a refrigerator at  $8.0\pm 2^{\circ}\text{C}$  from where samples were taken for processing and analysis.

**Germination:** Germination was carried out by the method described by Ariahu *et al.* (1999). Maize grains were washed in a 5% (w/v) sodium chloride (NaCl) solution to suppress growth of moulds. They were then steeped in tap water at room temperature ( $30\pm 2^{\circ}\text{C}$ ) in a ratio of 1:3 (w/v) grain: water in a plastic bucket. The water was changed every 4 h for a total steeping time of 12 h. The seeds were then placed in a plastic basket for drainage of water after which they were spread in a single layer on a moistened jute bag and allowed to sprout in a wooden malting box for 96 h. The germinated seeds were sampled at 24, 48, 72 and 96 h and dried along with the ungerminated control to 10% moisture at  $80^{\circ}\text{C}$  in an air draft oven (Model T12H, Genlab Widnes, UK). The dried seeds were split in a disc attrition mill (Asiko All Adis, Nigeria). The testa and rootlets detached from the cotyledon during splitting were separated by winnowing. The cotyledons were then ground into flour in the disc attrition mill (Asiko All Adis, Nigeria) followed by sieving to 0.2 mm particle size. The flours were then packaged in dark coloured 500 mL plastic containers with airtight lids, stored on dry wooden shelves at room temperature ( $30\pm 2^{\circ}\text{C}$ ) and utilized within 48 h for analysis.

**Fermentation:** A portion of maize flour was subjected to accelerated natural fermentation using the method described by Ariahu *et al.* (1999). In this process, 120 g of maize flour was mixed with 80 mL of distilled water and subjected to natural fermentation in a covered 500 mL glass beaker at room temperature ( $30\pm 2^{\circ}\text{C}$ ) for 24 h. At the end of this period, 50% of the fermented mixture was used as starter for a new fermentation cycle. During the process, the pH and titratable acidity (an index of lactic bacteria activity) were monitored. The fermentation process was continued until

the pH of the medium stabilized and remained constant with further increase in fermentation cycles. The fermented concentrate was dehydrated at 60°C in a fan driven electric oven (Model T12H, Genlab Widnes, U. K.) to 10% moisture and ground in a disc attrition mill (Asiko All, Addis Nigeria) to a particle size of 0.2 mm. The flour was then packaged in dark coloured 500 mL plastic containers with airtight lids, stored on dry wooden shelves at room temperature (30±2°C) and utilized within 48 h for analysis.

### Sample analysis

**Chemical properties:** Hydrogen ion concentration (pH) was determined using the method of Egan *et al.* (1981). Titratable acidity was determined by the method of Akpapunam and Dedeh (1995). Diastatic Power (DP) was determined by the method of IOB (1982). Crude protein, moisture content, crude fibre, crude fat and ash were all determined by the methods of AOAC (2000), while carbohydrate was determined by difference (Ihekoronye and Ngoddy, 1985). Energy values were estimated by the Atwater Factor and expressed in Kilo calories/100 g (Pearson, 1976). Total available carbohydrate was determined by the Clegg-Anthrone method as described by Balami *et al.* (2004); while total soluble sugars were determined by the method of Dubois *et al.* (1956) and reducing sugars were determined by the Dinitrosalicylic acid method described by Uzomah *et al.* (2002). Tannins were determined by the Prussian blue method of Budini *et al.* (1980); phytates were determined by the method of Nkama and Gbenyi (2001), oxalates were determined by the method of Day and Underwood (1986); while hydrocyanic acid (HCN) was determined by the picrate paper kit method as described by Bradbury *et al.* (1999).

**Functional properties:** Packed bulk density and water absorption capacity were determined by the method of Okezie and Bello (1988). Viscosity was determined using the method described by Uvere *et al.* (2002) while swelling index was determined by the method of Flemming *et al.* (1974). Reconstitution index was by the method of Banigo and Akpapunam (1987).

**Statistical analysis:** Results obtained were statistically analyzed using a pre-packaged statistical software (Minitab 15) at  $p \leq 0.05$ .

## RESULTS AND DISCUSSION

Table 1 shows the change in values of pH, TTA and some carbohydrate fractions during malting, there was gradual decrease in pH and increase in acidity from 0 to 72 h (6.80-5.94) and

Table 1: Changes in some chemical properties of the maize grain during malting

Parameters	Malting time (h)					LSD
	0	24	48	72	96	
pH	6.80±0.02 <sup>a</sup>	6.71±0.01 <sup>a</sup>	6.33±0.01 <sup>ab</sup>	5.94±0.01 <sup>b</sup>	6.02±0.01 <sup>ab</sup>	0.73
TTA (%)	0.14±0.01 <sup>a</sup>	0.14±0.02 <sup>a</sup>	0.16±0.01 <sup>a</sup>	0.17±0.02 <sup>a</sup>	0.18±0.01 <sup>a</sup>	-
Total CHO (%)	56.90±0.11 <sup>b</sup>	65.71±0.30 <sup>a</sup>	66.40±0.17 <sup>a</sup>	65.85±0.10 <sup>a</sup>	34.74±0.15 <sup>c</sup>	1.60
Total sol. sugar (%)	16.86±0.31 <sup>b</sup>	12.72±0.88 <sup>d</sup>	10.04±0.45 <sup>e</sup>	20.75±1.12 <sup>a</sup>	14.39±0.76 <sup>c</sup>	1.82
Reducing sugar (%)	2.88±0.20 <sup>c</sup>	0.90±0.04 <sup>d</sup>	0.30±0.02 <sup>d</sup>	7.20±0.01 <sup>a</sup>	4.50±0.03 <sup>b</sup>	0.95
Diastatic power (°Lintner)	2.62±0.01 <sup>e</sup>	5.55±0.02 <sup>d</sup>	12.62±0.01 <sup>c</sup>	22.65±0.02 <sup>a</sup>	18.43±0.02 <sup>b</sup>	0.70

Values are Mean±SD of triplicate determinations. Means with different superscripts within the same row are significantly different ( $p < 0.05$ ). LSD: Least significant difference

(0.14 to 0.17%), respectively after which the pH increased slightly by 96 h to (6.02). This could be as a result of hydrolysis of some complex organic molecules like lipids, phytin and protein, which could be hydrolyzed to fatty acids, acid phosphates and amino acids respectively. Thus the increase in acidity accompanying malting could be an indication of the extent of hydrolysis of these complex molecules and therefore of the digestibility of malted maize. This is in agreement findings of Adeyemo *et al.* (1992). There were significant ( $p<0.05$ ) changes in values of carbohydrate fractions. Total soluble sugar and reducing sugar values decreased in the first 48 h, then increased at 72 h and dropped again by 96 h; while total carbohydrates increased steadily from 0 to 72 h (56.90-65.85%), before dropping off after 72 h to 34.74%. Akpapunam and Dedeh (1995) explained that during malting, the first 48 h are actually the period preceding sprouting, during which the growth process and metabolic activities suspended at maturation, dehydration and storage of the grain are resumed under favourable conditions of moisture and temperature. The imbibition of water during the first 24 h of soaking could have therefore paved way for breakdown of starch to simple carbohydrates *via* pre-existing hydrolytic enzymes. This breakdown does not however, show any inverse proportionality relationship as expected from hydrolytic processes only (between total available starch and total soluble sugar) because germination increases the rigour of oxidative and synthetic processes, which are energy demanding metabolic activities, thus affecting sugar content and other nutrients.

There was a significant ( $p<0.05$ ) increase in Diastatic Power (DP) from 2.62-22.65 °Lintner at 0 and 72 h, respectively. This is in agreement with Brigas *et al.* (1981), who reported that diastase (amylases and other relevant starch-degrading enzymes) increases in quantities when grains are germinated.

Table 2 shows changes in proximate composition during germination, there was significant ( $p<0.05$ ) increase in protein content. From 12.85% at 0 h, the protein content decreased to 10.66 before increasing to 13.68% at 48 h and 13.93% at 72 h. This could be as a result of the mobilization of storage nitrogen of the maize to produce the nutritionally high quality proteins needed by the young plant for its development. This is important to the nutrition of infants and children in developing nations who depend largely on gruels of cereals to meet their energy and protein needs. This is in agreement with Ariahu *et al.* (1999) who also reported an increase in protein content of African breadfruit. There was also a significant increase in ash content within this period ( $p>0.05$ ) from 2.28% to 3.70% while there was a significant reduction ( $p<0.05$ ) in carbohydrate, crude fat and crude fibre content as germination progressed. This could be as a result of the action of hydrolytic enzymes existing in the maize grains, coupled with the mobilization of

Table 2: Effect of malting time on the proximate composition of maize grains (% dry weight basis)

Parameters	Malting time (h)					LSD
	0	24	48	72	96	
Moisture (%)	8.10±0.16 <sup>d</sup>	10.20±0.08 <sup>b</sup>	9.70±0.01 <sup>c</sup>	10.40±0.01 <sup>b</sup>	11.20±0.03 <sup>a</sup>	0.64
Crude protein (%)	12.85±0.04 <sup>b</sup>	10.66±0.04 <sup>c</sup>	13.68±0.03 <sup>a</sup>	13.93±0.01 <sup>a</sup>	10.59±0.01 <sup>c</sup>	0.70
Crude fat (%)	4.57±0.02 <sup>a</sup>	3.84±0.04 <sup>bc</sup>	3.99±0.03 <sup>bc</sup>	3.57±0.02 <sup>c</sup>	4.11±0.05 <sup>b</sup>	0.31
Crude fibre (%)	2.21±0.15 <sup>a</sup>	1.88±0.11 <sup>a</sup>	2.24±0.04 <sup>a</sup>	2.03±0.01 <sup>a</sup>	2.26±0.03 <sup>a</sup>	-
Ash (%)	2.28±0.02 <sup>a</sup>	2.69±0.03 <sup>d</sup>	2.79±0.01 <sup>c</sup>	3.43±0.01 <sup>b</sup>	3.70±0.02 <sup>a</sup>	0.06
CHO (%)	79.82±1.16 <sup>a</sup>	78.99±1.05 <sup>b</sup>	77.41±1.62 <sup>c</sup>	77.79±1.05 <sup>c</sup>	80.29±1.01 <sup>a</sup>	1.07
Energy (K/cal)	403.05±2.35 <sup>a</sup>	401.88±1.62 <sup>b</sup>	399.93±2.12 <sup>c</sup>	399.02±1.76 <sup>c</sup>	402.56±1.86 <sup>a</sup>	1.18

Values are Mean±SD of triplicate determinations. Means with different superscripts within the same row are significantly different ( $p<0.05$ ). LSD: Least significant difference

soluble materials into roots and shoots for germination, thus reducing the level of these nutrients in the seeds. The reduction in fat content is of particular interest. Maize oil is reported to be rich in essential fatty acids (Adeyemo *et al.*, 1992), which play a very important role in the proper development of brain cells in infants and children. Thus if germination adversely affects the quantity of essential fatty acids of the grains, the need for supplementation becomes most crucial.

Table 3 shows the proximate composition of unmalted, malted and fermented maize flours, there is significant ( $p < 0.05$ ) increase in protein content from 10.64% for un-malted to 12.17% for malted maize flours. This could be as a result of mobilization of storage nitrogen of maize to aid germination. This is consistent with the findings of Malleshi *et al.* (1989) who reported significant increase in protein content, available lysine and Protein Efficiency Ratio (PER) of weaning food formulations from sprouted sorghum and cowpea seeds. The lower figure of 12.04% for protein content of fermented maize flour could be as a result of protein utilization by fermentative organisms for growth and metabolism during the process of fermentation. This is in agreement with the findings of many workers such as Ashworth and Draper (1992) who reported that fermentation decreased the concentration of total carbohydrates as a result of utilization by microorganisms. Nout (1991) as well as Lorri and Svanberg (1991) observed that fermentation decreased starch content and long chain fatty acids along with non-digestible plant components (fibre); thus improving food utilization efficiency and nutritional quality. Ash content increased significant ( $p < 0.05$ ) with germination and fermentation from 2.17-2.72%. Obasi *et al.* (2009) also observed an increase in the mineral content of pearl millet with increased soaking time.

Table 4 presents some selected toxins and anti-nutritional factors of unmalted, malted and fermented maize flours, there was a significant ( $p < 0.05$ ) reduction in all toxins and anti-nutritional factors. This could be due to the leaching effect of soaking as well as degradation of toxic components. It has been reported that soaking reduced phytic acid levels in some legumes by up to 45-50%; as well as oligosaccharides, trypsin inhibitors, hemagglutinins and saponin levels in

Table 3: Proximate composition of unmalted, malted and fermented maize grains (% dry weight basis)

Parameters	Unmalted	Malted	Fermented	LSD
Moisture (%)	9.39±0.03 <sup>b</sup>	9.23±0.03 <sup>c</sup>	10.08±0.02 <sup>a</sup>	0.03
Crude protein (%)	10.64±0.03 <sup>c</sup>	12.17±0.03 <sup>a</sup>	12.04±0.03 <sup>b</sup>	0.07
Crude fat (%)	4.02±0.03 <sup>a</sup>	3.81±0.02 <sup>b</sup>	3.76±0.02 <sup>c</sup>	0.04
Crude fibre (%)	2.12±0.04 <sup>a</sup>	1.82±0.03 <sup>b</sup>	1.78±0.02 <sup>b</sup>	0.09
Ash (%)	2.17±0.06 <sup>c</sup>	2.25±0.03 <sup>b</sup>	2.72±0.03 <sup>a</sup>	0.07
Carbohydrate (%)	80.87±0.03 <sup>a</sup>	79.90±0.05 <sup>b</sup>	79.66±0.02 <sup>c</sup>	0.12
Energy (K/cal)	402.45±0.02 <sup>b</sup>	402.96±0.02 <sup>a</sup>	400.77±0.06 <sup>c</sup>	0.12

Values are Mean±SD of triplicate determinations. Means with different superscripts within the same row are significantly different ( $p < 0.05$ ). LSD: Least significant difference

Table 4: Some toxins and anti-nutritional factors of unmalted, malted and fermented maize grains (% dry wt. Basis)

Parameters	Unmalted	Malted	Fermented	LSD
Tannins (g/100 g)	2.62±0.01 <sup>a</sup>	1.36±0.03 <sup>b</sup>	0.42±0.04 <sup>f</sup>	0.48
Phytates (g/100 g)	2.30±0.07 <sup>a</sup>	1.62±0.02 <sup>b</sup>	0.84±0.08 <sup>c</sup>	0.75
Oxalates (g/100 g)	2.32±0.01 <sup>a</sup>	1.22±0.06 <sup>b</sup>	0.34±0.07 <sup>e</sup>	0.63
Cyanide (mg/100 g)	2.20±0.01 <sup>a</sup>	1.08±0.03 <sup>b</sup>	0.42±0.05 <sup>c</sup>	0.06
Crude fibre (g/100 g)	2.12±0.04 <sup>a</sup>	1.62±0.03 <sup>b</sup>	1.11±0.02 <sup>c</sup>	0.07

Values are Mean±SD of triplicate determinations. Means with different superscripts within the same row are significantly different ( $p < 0.05$ ). LSD: Least significant difference

Table 5: Some functional properties of unmalted, malted and fermented maize flours

Parameters	Unmalted	Malted	Fermented	LSD
Bulk density (g mL <sup>-1</sup> )	1.17±0.02 <sup>a</sup>	0.86±0.01 <sup>b</sup>	0.54±0.01 <sup>b</sup>	0.06
Swelling Index (mL g <sup>-1</sup> )	6.47±0.02 <sup>a</sup>	3.93±0.01 <sup>b</sup>	3.46±0.01 <sup>c</sup>	0.08
Water Absorption Capacity (g sec <sup>-1</sup> )	3.70±0.03 <sup>a</sup>	3.83±0.01 <sup>a</sup>	3.92±0.05	-0.35
Viscosity (cps)	343.33±0.97 <sup>a</sup>	324.46±1.06 <sup>b</sup>	288.26±0.58 <sup>c</sup>	2.02
Reconstitution index (mL g <sup>-1</sup> )	4.82±0.40 <sup>c</sup>	5.48±0.42 <sup>b</sup>	6.40±0.20 <sup>a</sup>	0.90

Values are Mean±SD of triplicate determinations. Means with different superscripts within the same row are significantly different (p<0.05). LSD: Least significant difference

different seeds. Khetarpaul and Chauhan (1989) also reported a decrease in tannins and lecithins (which inhibit trypsin activity and consequently reduce protein utilization) with acid fermentation. This has been corroborated by Ahrens *et al.* (1989), who showed that lactic acid fermentation of sorghum increased the protein efficiency ratio from 2.06 to 2.34 in rats. Generally, the values of all the toxins and anti-nutritional factors were found to be well below safe limits.

Table 5 shows the effect of malting and fermentation on some functional properties of maize flours, the unmalted flour had a higher bulk density (1.17 g mL<sup>-1</sup>) which was significantly (p<0.05) higher than that of malted flour (0.86 g mL<sup>-1</sup>) and fermented flour (0.54 g mL<sup>-1</sup>). This could be because malting and fermentation tend to soften the seeds, thus making milling easier, with smaller particle sizes than unmalted grain, hence the reduction in bulk density. The significance of this is that the less bulky flours will have higher nutrient density, since more flour can be packaged in the same given volume. The unmalted flour also had a higher swelling index (6.47 mL g<sup>-1</sup>), which was significantly (p<0.05) higher than that of malted (3.93 mL g<sup>-1</sup>) and fermented (3.46 mL g<sup>-1</sup>) flours. This could be as a result of the swelling of the starch granules, which leads to disruption of some of the intermolecular hydrogen bonds, thus allowing more water to enter and enlarge the granules (Ihekoronye and Ngoddy, 1985). The malted and fermented flours, whose starches had already been dextrinized, could not swell as much. Swelling capacity can be an index of stickiness of the resultant product.

There was an increase in water absorption capacity, with malted and fermented flours taking up more water, though the difference was not significant (p>0.05). This increased solubility could be as a result of the increase in amount of soluble sugars present in the malted and fermented flours. The reconstitution index of fermented maize flours was significantly (p<0.05) higher than that of unfermented flours. This means that fermented flours, which have better water absorption capacity, are easier to reconstitute in water when needed. Eneche (2009) also reported an increase in water absorption with soaking of maize grains.

There was a significant (p<0.05) decrease in viscosity from unmalted flour (343.33 cP) to malted flour (324.46 cP) and fermented flour (288.26 cP). This reduction in viscosity could be due to starch degradation caused by the action of hydrolytic enzymes ( $\alpha$  and  $\beta$  amylases) that developed during the germination process, thus hydrolyzing some of the starch into limit dextrins and maltose, which do not swell when cooked. Fermentation further decreases the total amount of carbohydrates and other nutrients, since microbial activity requires energy and nutrients (Gaman and Sherrington, 1990). Flour prepared from sprouted and fermented grains can therefore be used in greater amount to give the same viscosity as flour from unmalted grains, thereby giving higher nutrient and energy density. The lowered starch complexity, resulting from the dextrinogenic (viscosity-reducing) effect of enzymic modification of starch, as well as the partial digestion by enzymes during germination therefore help in its utilization.

## CONCLUSION

This study indicates that germination and fermentation of white maize results in the enhancement of its nutritional quality, as observed by the significant increase in quantity of protein, reduced bulk/viscosity and increased nutrient density as well as reduction in toxins and anti-nutritional factors. These observations could be advantageously utilized to improve nutrition of infants and children, particularly in the developing countries, where maize is consumed in large quantities. The associated reduction in carbohydrate and fat can be compensated for by blending with flours of other legumes/oilseeds.

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