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Effect of traditional processes on phytate and mineral content of pearl millet

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Six pearl millet genotypes were used in this study: IS 91333, IS 91666, and IS 89111 for dough fermentation and IS 880004, IS 91777 and YD-X3 genotypes for Damirga flour. Investigation showed that traditional fermentation for 14 h at 37°C caused a decline in pH with time; a sharp drop was observed at the beginning, which gradually levelled off. Fermentation resulted in significant reduction of starch and phytic acid: 9.5–9.8% and 43–44%, respectively. Protein content was not affected. The Damirga process significantly elevated starch content (by 8–19%) but significantly reduced the protein and phytic acid contents (by 10.9–12.1 and 86–93%, respectively). Damirga flour was found to retain 25–84% of the major minerals (Ca, Mg, P, K and Na) and 52–65% of the minor minerals (Zn, Mn and Fe); losses occurred for all minerals except Cu. © 1998 Elsevier Science Ltd. All rights reserved

INTRODUCTION

Pearl millet [*Pennisetum glaucum* (L.) R. Br.] is one of the most important drought-tolerant crops of the tropical and subtropical regions of the world; it grows in harsh environments where other crops do not grow well. As a cereal for human food, pearl millet sustains the lives of the poorest people in Africa and Asia, and is often considered highly palatable and a good source of protein, minerals and energy. Antinutritional factors present in considerable amounts in pearl millet (Mahajan and Chauhan, 1987) limit protein and starch digestibilities (Carnovale *et al.*, 1988; Yoon *et al.*, 1983), hinder mineral bioavailability (Harland and Oberleas, 1987) and inhibit proteolytic and amylolytic enzymes (Knuckles *et al.*, 1985).

Decreasing phytic acid content of food is very advantageous, due to its influence on nutrition; therefore interest has grown to reduce its antinutritional effects (Erdman, 1979). Soaking, dehulling and fermentation are important traditional methods used to eliminate phytic acid.

In Sudan, pearl millet, locally known as Dukhn, is the third most important staple food crop after sorghum and wheat, with annual production of 800 000 metric tons, and of immensely greater nutritional significance in the diets of poor people in drier parts of the country where endemic drought causes frequent failure of other crops.

Western Sudanese people (Kordofan and Dar-fur States) process pearl millet in several types of foodstuffs such as fermented or unfermented breads (Kisra), stiff (Aseda) or thin (Nasha) porridges, alcoholic (Marisa) or non-alcoholic (Hullu-murr) beverages and Damirga, which is a fine sour and white flour obtained traditionally from pearl millet grains, which is used to make Asedat-damirga (stiff, white porridge), Nasha-beida (white nasha) or Kisra-beida (white kisra).

The objectives of the present study were to examine the biochemical changes occurring in pearl millet dough during fermentation and in manufactured Damirga flour.

MATERIALS AND METHODS

Materials

Six pearl millet genotypes obtained from the Agronomy Department, Faculty of Agriculture, University of Khartoum were used in this investigation; genotypes IS 91333, IS 91666 and IS 89111 were used for the dough fermentation; Damirga flour was obtained from IS 880004, IS 91777 and YD-X3 genotypes. Selection of genotypes was based on different levels of phytate in those samples and on the adequacy of the experimental material.

Preparation of dough

Fermented dough (Ageen) was prepared in the traditional way used in Sudanese homes as described by El Tinay *et al.* (1979). Whole milled flour was mixed with

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water (1:2 ratio) in a plastic bucket. A starter from previously fermented dough (Khumara) was then added to the mixture of flour and water (the starter of each dough was of the same genotype of millet and was about 10% of the dough volume). The dough was then incubated at 37°C. A portion of the fermented sample was withdrawn at 2-h intervals until the end of fermentation, which was terminated after 14 h (pH 3.5–3.6). The withdrawn samples were dried in an air oven at 70°C and finely ground.

Damirga flour preparation

Damirga flour was prepared as a traditional domestic art as follows. The grains were first moistened with water (approximately 20% of their weight) and then hand-pounded by wooden mortar and pestle until the required degree of dehulling was reached (about 30 min). The grains were then winnowed in the winnowing basket to remove the hulls. The bran-free kernels were soaked in water (1:2 ratio) and fermented for 72 h at ambient temperature (28–30°C). Water was then decanted and the fermented dehulled grains were sun-dried and finely ground (0.4 mm mesh).

Chemical analysis

Moisture was determined according to AACC (1980). Protein (N×6.25) was determined according to AOAC (1984). Starch was determined by the method of dispersal in $CaCl_2$, followed by iodine spectrophotometry (Kerr, 1950). Phytic acid was estimated by the method described by Wheeler and Ferrel (1971). Phosphorus was determined by the method of Chapman and Pratt (1982). Minerals were determined by atomic absorption spectrophotometry using an SP 191 Pye Unicam spectrophotometer. The results were expressed on a dry matter basis.

Statistical analysis

Each sample was analysed in triplicate and the figures were then averaged. Data were assessed by analysis of variance (ANOVA) (Snedecor and Cochran, 1987) and by Duncan's multiple range test with a probability $p \leq 0.05$ (Duncan, 1955).

RESULTS AND DISCUSSION

Fermented dough

Fermentation at 37°C for 14 h caused a gradual decline in pH of the fermented dough. The drop in pH was more pronounced during the first 2 h. Kheterpaul and Chauhan (1991) found that, after 72 h natural fermentation at 20, 25 and 30°C, the pH of pearl millet dough declined from 6.4 to 4.4, 4.0 and 3.6, respectively. The drop in pH is important in preventing the growth of food poisoning bacteria (Au and Fields, 1981). The sharp drop in pH at the beginning of fermentation agrees with El Daw (1994) who reported 16.2 and

Table 1. Changes in starch, protein avid phytate contents of IS 91333 genotype-fermented dough

Fermentation period (h)	pH	Starch (%)	Protein (%)	Phytate (mg/100 g)
0	5.0	61.0 (±0.11)^{a}	12.1 (±0.09)^{a}	795 (±0.00)^{a}
2	4.5	60.2 (±0.23)^{b}	11.7 (±0.05)^{cd}	755 (±0.10)^{b}
4	4.3	58.8 (±0.08)^{c}	11.3 (±0.05)^{e}	708 (±0.15)^{c}
6	4.2	57.2 (±0.02)^{d}	11.4 (±0.05)^{e}	660 (±0.19)^{d}
8	4.0	56.9 (±0.02)^{e}	11.6 (±0.05)^{d}	613 (±0.09)^{e}
10	3.9	56.7 (±0.07)^{e}	11.7 (±0.05)^{c}	565 (±0.22)^{f}
12	3.7	55.5 (±0.15)^{f}	11.9 (±0.05)^{b}	501 (±0.18)^{g}
14	3.5	55.2 (±0.10)^{f}	12.1 (±0.05)^{a}	453 (±0.18)^{h}

Each value is an average of three replicates expressed on dry weight basis.
Values are means (± standard deviation).
Means not sharing a common superscript letter in a column are significantly different as assessed by Duncan's multiple-range test.

Table 2. Changes in starch, Protein and phytate contents of IS 91666 genotype-fermented dough

Fermentation period (h)	pH	Starch (%)	Protein (%)	Phytate (mg/100 g)
0	5.1	67.3 (±0.26)^{a}	9.3 (±0.09)^{a}	530 (±0.07)^{a}
2	4.5	66.3 (±0.01)^{b}	9.0 (±0.09)^{c}	498 (±0.25)^{b}
4	4.4	64.6 (±0.04)^{c}	8.6 (±0.05)^{e}	466 (±0.17)^{c}
6	4.2	63.0 (±0.19)^{d}	8.7 (±0.05)^{d}	435 (±0.01)^{d}
8	3.9	62.6 (±0.19)^{de}	8.8 (±0.05)^{cd}	403 (±0.01)^{e}
10	3.8	62.2 (±0.25)^{e}	8.9 (±0.05)^{b}	371 (±0.01)^{f}
12	3.7	61.2 (±0.28)^{f}	9.19 (±0.05)^{b}	329 (±0.00)^{g}
14	3.5	60.7 (±0.55)^{f}	9.3 (±0.05)^{a}	297 (±0.11)^{h}

Each value is an average of three replicates expressed on dry weight basis.
Values are means (± standard deviation).
Means not sharing a common superscript letter in a column are significantly different as assessed by Duncan's multiple-range test.

Table 3. Changes in starch, protein and phytate contents of IS 89111 genotype-fermented dough

Fermentation period (h)	pH	Starch (%)	Protein (%)	Phytate (mg/100 g)
0	5.1	66.2 (±0.18)^{a}	10.1 (±0.18)^{a}	441 (±0.86)^{a}
2	4.6	65.2 (±0.09)^{b}	9.6 (±0.09)^{bc}	414 (±0.02)^{b}
4	4.4	63.7 (±0.03)^{c}	9.5 (±0.05)^{c}	388 (±0.08)^{c}
6	4.2	62.0 (±0.01)^{d}	9.5 (±0.05)^{c}	361 (±0.03)^{d}
8	4.0	61.6 (±0.10)^{e}	9.6 (±0.05)^{bc}	335 (±0.11)^{e}
10	3.9	61.3 (±0.01)^{f}	9.7 (±0.05)^{b}	309 (±0.24)^{f}
12	3.7	60.1 (±0.13)^{g}	10.0 (±0.05)^{a}	273 (±0.10)^{g}
14	3.6	59.8 (±0.05)^{g}	10.2 (±0.09)^{a}	247 (±0.23)^{h}

Each value is an average of three replicates expressed on dry weight basis.
Values are means (± standard deviation).
Means not sharing a common superscript letter in a column are significantly different as assessed by Duncan's multiple-range test.

18.5% reduction in pH of two fermented sorghum cultivars during the first 2 h of fermentation. Also, El Sharif (1993) reported that the rate of decrease in pH of Abreh dough was faster in the initial stages and slowed down as fermentation progressed.

Tables 1–3 show starch, protein and phytate contents of IS 91333, IS 91666 and IS 89111 cultivar-fermented dough. Fermentation for 14 h caused a significant decrease in the starch content from 61.0 to 55.2%, from 67.3 to 60.7% and from 66.2 to 59.8% for IS 91333, IS 91666 and IS 89111 genotypes, respectively. An appreciable decrease in starch during the first stages of fermentation may be attributed to the action of α- and β-amylases produced by microorganisms (El Tinay *et al.*, 1979), or are indigenous to the flour (Pederson, 1971). Considerable amounts of starch were hydrolysed at the beginning of the fermentation process. There were no significant differences in starch content after 12 h fermentation. El Tinay *et al.* (1979) reported that towards the end of the process, decrease of starch of fermented sorghum was very small due to the drop in pH, which inhibited the activity of α- and β-amylases.

Fermentation for 14 h caused a general significant change in protein content for the three fermented doughs investigated. Four hours fermentation reduced protein by 6.6, 5.7 and 6.05% for IS 91333, IS 91666 and IS 89111 genotypes, respectively. This drop in protein content could be attributed to the action of moulds and anaerobic bacteria, which degrade proteins and convert them to ammonia (Stainer *et al.*, 1963). Also, some strains of bacteria are known to possess deaminases, resulting in increased protein catabolism. The apparent increase in protein content of the three fermented doughs during the successive steps of fermentation could be attributed to reduction in starch content.

Fermentation caused a significant reduction in the phytic acid content for the three genotypes. Results obtained in this study were similar to those obtained by Marfo *et al.* (1991) who reported 43.3% reduction in phytic acid content of white maize after 48 h fermentation. Many workers noticed significant reduction in phytic acid content of pearl millet flour during fermentation. Mahajan and Chauhan (1987) found that natural lactic acid fermentation for 24 h at 20, 30, 40 and 50°C reduced phytic acid by 87, 88, 90 and 91%, respectively. Kheterpaul and Chauhan (1989) reported that mixed fermentation for 72 h at 30°C reduced phytic acid by 56%. Results in this study were lower than those reported previously, which might be attributed to the relatively shorter period of fermentation.

Enzymatic hydrolysis of phytic acid by indigenous phytase of pearl millet and of fermenting microflora may account for most of the loss of phytic acid during fermentation. The low pH of the fermented dough towards the end of fermentation may provide favourable conditions for phytase activity (Dhanker and Chauhan, 1987; Mahajan and Chauhan, 1987). Thus, traditional fermentation, besides producing the desired sour taste, also eliminates considerable amounts of phytic acid.

Damirga flour

Table 4 shows the starch content of Damirga flours, which were 79, 83 and 66% for IS 880004, IS 91777 and YD-X3 genotypes, respectively. These values were significantly higher than whole unprocessed flour. The Damirga process elevated the starch content by 8–19%. The present results were in fair agreement with values obtained by Monawar (1983), who stated that starch is negatively correlated with decortication and reported an increase in starch content for pearl millet of 80% extraction rate (from 69.4 to 76.3%). Also Kenkepen (1986) found that 80% extraction rate increased starch content of pearl millet from 64.6 to 76.3%. The increased starch content of Damirga flour could be attributed to removal of the outer layers. Badi *et al.* (1976) reported that the mesocarp of pearl millet kernel contained no starch.

Damirga flour obtained from genotypes IS 880004, IS 91777 and YD-X3 contained 7.5, 8.0 and 11.4 crude protein, respectively (Table 4). These values, in comparison with the whole flour, were significantly different. The Damirga process decreased protein content of the three genotypes by 11.8, 12.1 and 10.9%, respectively. Reichert and Youg (1977) estimated the protein content of the traditionally dehulled grains of sorghum and millet to be 5–9% less than whole grain. Damirga flour in the present study retained 88–89% crude protein, which agrees with the findings of Villareal *et al.* (1991) who reported that, with 10% bran-polish removal, milled rice retained 82–90% crude protein.

The losses in protein could be attributed to the removal of hull (Bookwalter *et al.*, 1987) or to the relatively thin pericarp of pearl millet (Monawar, 1983) or to the low extraction rate during traditional dehulling estimated to be 60–65% (Badi *et al.*, 1987), which eliminate some of the protein-rich aleurone cells.

Table 5 shows the phytic acid content of the three pearl millet genotypes for the successive steps of the Damirga process (whole grain, traditionally dehulled grain and Damirga flour). Traditionally dehulled grains contained 524.1, 391.6 and 309.2 mg/100 g phytic acid for IS 880004, IS 91777 and YD-X3 genotypes, respectively. A highly significant difference in phytic acid content was noticed between the whole and dehulled grains. Compared with the whole flour, traditional dehulling resulted in a reduction range from 30 to 37%, which falls within the range of 27–53% given by Lorenz (1983) for dehulled proso millet. The range was higher than the value of 19% reported by Monawar (1983) for 80% extracted millet. Traditional dehulling eliminates phytic acid due to removal of outer layers. Tanaka *et al.* (1973) stated that phytic acid of monocotyledonous seeds is thought to be abundant in the outer layers. The higher phytic acid retention in the

Table 4. Starch and protein contents of Damirga flour

Sample	IS 880004		IS 91777		YD-X3	
	Starch (%)	Protein (%)	Starch (%)	Protein (%)	Starch (%)	Protein (%)
Whole	67.0 (±0.20)^b	8.5 (±0.24)^a	70.0 (±0.47)^b	9.1 (±0.09)^a	61.1 (±0.03)^b	12.8 (±0.09)^a
Damirga flour	79.4 (±0.03)^a	7.5 (±0.05)^b	83.3 (±0.06)^a	8.0 (±0.18)^b	66.0 (±0.12)^a	11.4 (0.05)^b

Each value is an average of three replicates expressed on dry weight basis.
Values are means (± standard deviation).
Means not sharing a common superscript letter in a column are significantly different as assessed by Duncan's multiple-range test.

Table 5. Effect of Damirga process on phytic acid content

Sample	Phytate (mg/100 g)		
	IS 880004	IS 91777	YD-X3
Whole	796 (±0.88)^a	618 (±0.00)^a	443 (±0.88)^a
Dehulled	524 (±0.10)^b	392 (±5.80)^b	309 (±0.00)^b
Damirga flour	79.5 (±0.01)^c	44.2 (±0.01)^c	61.8 (±0.01)^c

Each value is an average of three replicates expressed on dry weight basis.
Values are means (± standard deviation).
Means not sharing a common superscript letter in a column are significantly different as assessed by Duncan's multiple-range test.

dehulled grains (63–70%) can be attributed to the fact that pearl millet contains considerable amounts of phytic acid in the germ (O'Dell *et al.*, 1972; Simwimba *et al.*, 1984).

Phytic acid contents of Damirga flour obtained from genotypes IS 880004, IS 91777 and YD-X3 were 79.5, 44.2 and 61.8 mg/100 g, respectively. Phytic acid reduction were 90, 93 and 86% for the three genotypes, respectively. A significant difference was observed between the value of Damirga flour and those of whole and dehulled grains.

It appears that Damirga processing is very effective in reducing phytic acid content of pear millet. This low level of phytic acid can be attributed to the cumulative effects of dehulling soaking and fermentation.

The calcium content of Damirga flour obtained from genotypes IS 880004, IS 91777 and YD-X3 was lower than that of the whole flour. However, the decrease was not significant (Table 6). The Ca content of IS 880004 genotype Damirga flour was not affected by the processing. The magnesium content of Damirga flour from IS 80004, IS 91777 and YD-X3 genotypes decreased from 0.21 to 0.05, 0.18 to 0.05 and 0.23 to 0.05%, respectively. Significant differences were found between the Mg content of whole and Damirga flour for all genotypes. The phosphorus content of Damirga flour prepared from genotypes IS 880004, IS 91777 and YD-X3 was 0.3, 0.4 and 0.3, respectively. For all genotypes, a highly significant difference was found between whole flour and Damirga flour. Monawar (1983) reported that 80% extraction rate decreased Ca, Mg and P contents of pearl millet from 0.02 to 0.01%, from 0.12 to 0.10% and from 0.24 to 0.22%, respectively. Also Sotelo *et al.* (1990) found that the removal of rice hull diminished Ca content.

The potassium of Damirga flour obtained from the three genotypes was low compared to the whole flour. However, this reduction is not significant for IS 91777 and YD-X3 genontypes, but the value for genotype IS 880003 was significantly different. Damirga flour showed a marginal and insignificant decrease in sodium contents compared to the whole flour (Table 6). Manganese contents of Damirga flour obtained from genotypes IS 880004, IS 91777 YD-X3 were 12, 13, and 13 μg/g, respectively. These values are significantly lower than the Mn values of the whole flour. Significant decrease was observed between Zn contents of whole and Damirga flour from 53 to 28, 60 to 35 and 60 to 28 μg/g for IS 880004, IS 91777 and YD-X3 genotypes, respectively. The iron content of Damirga flour obtained from genotype YD-X3 (50 μg/g) is significantly lower compared to the 100 μg/g obtained for the whole flour of the same genotype. Significant reduction in Fe content from 100 to 60 and from 70 to 50 μg/g was found for IS 880004 and IS 91777 genotypes, respectively. Bookwalter *et al.* (1987) reported a decrease from 23 to 18 and from 72 to 16 μg/g in Zn and Fe contents of pearl

Table 6. Mineral content of Damirga flour

Genotype	Sample	Ca (%)	Mg (%)	P (%)	K (μg/g)	Na (μg/g)	Zn (μg/g)	Mn (μg/g)	Cu (μg/g)	Fe (μg/g)
IS 880004	Whole	0.05 (±0.00)	0.21 (±0.00)	0.99 (±0.03)	100 (±0.00)	5 (±0.00)	53 (±0.230)	23 (±0.10)	13 (±0.07)	110 (±0.0)
	Damirga flour	0.05 (±0.00)	0.05 (±0.01)	0.33 (±0.00)	50 (±0.00)	3 (±0.00)	28 (±0.21)	13 (±0.71)	13 (±0.99)	60 (±0.01)
IS 91777	Whole	0.08 (±0.03)	0.18 (±0.00)	0.88 (±0.03)	100 (±0.00)	8 (±0.00)	60 (±0.62)	20 (±0.15)	20 (±0.15)	70 (±0.00)
	Damirga flour	0.05 (±0.01)	0.05 (±0.00)	0.36 (±0.03)	60 (±0.00)	7 (±0.00)	35 (±0.63)	13 (0.70)	13 (±0.70)	50 (±0.00)
YD-X3	Whole	0.05 (±0.00)	0.25 (±0.00)	0.55 (±0.04)	70 (±0.00)	4 (±0.00)	60 (±0.35)	18 (±0.01)	18 (±0.01)	100 (±0.01)
	Damirga flour	0.04 (±0.00)	0.05 (±0.01)	0.25 (±0.00)	60 (±0.00)	3 (±0.00)	28 (±0.42)	13 (0.71)	13 (±0.71)	50 (±0.00)

Table 7. Mineral retention percentage in Damirga flour

Genotype	Major minerals retention (%)				Minor mineral retention (%)				
	Ca	Mg	P	K	Na	Zn	Mn	Cu	Fe
IS 880004	100	24	33	52	65	52	55	100	57
IS 91777	62	29	41	60	87	58	65	100	71
YD-X3	89	22	45	88	87	46	74	100	38
Mean	84	25	39	67	80	52	65	100	55

millet as a result of 50% extraction. Sotelo *et al.* (1990) stated that the dehulling of rice significantly diminished the amount of Fe. The copper contents of the three genotypes investigated showed no change as a result of Damirga preparation (Table 6). This is in agreement with the findings of Villareal *et al.* (1991), who stated that losses were noted in nearly all minerals except Cu during the bran-polish of rice.

The mean retention percentages of major minerals (Ca, Mg, P and Na) and minor minerals (Zn, Mn and Fe) were found to range between 25 and 84 and 52 and 65%, respectively (Table 7). Values for Fe, Zn and P were in line with the findings of Villareal *et al.* (1991), who reported that with 10% bran-polish removal, milled rice retained 33–55% of P, K, Mg and Mn and 54–66% of Ca, Fe and Zn. In pearl millet a greater concentration of minerals is located in the covering layers and germ than the endosperm portion of the kernel (Varriano-Mariston and Hoseney, 1980). Therefore, removal of the outer layers may account for mineral losses in Damirga flour. Carr (1960) stated that small losses of nutrients were found to occur for pearl millet as a result of traditional grinding. Also, Goswami *et al.* (1969) reported that removal of pericarp affects the mineral content in sorghum and millet.

In general, Damirga flour, which is highly acceptable to consumers. is characterized by an improved visual appearance, low phytic acid content and improved nutritive value.

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