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Effect of natural fermentation on nutritive value and in vitro protein digestibility of pearl millet

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Abstract

Two pearl millet cultivars: Composite Population III, obtained from the Department of Agronomy, Faculty of Agriculture, University of Khartoum and Baladi, obtained from El Obeid Research Station, were used in this study. Investigation showed that Composite Population III is of higher tannin, total polyphenols and phytic acid and lower in in vitro protein digestibility (IVPD), which was found to be 60.5%. The two cultivars were naturally fermented for 36 h at room temperature (30±2 °C) and pH, moisture content, protein, tannin, total polyphenols, phytic acid content and IVPD were determined at 4-h intervals. Fermentation was found to cause a significant reduction in total polyphenols and phytic acid content for the two cultivars. Fermentation for 36 h at room temperature was found to cause no changes in tannin content of fermented dough for the two millet cultivars. © 2002 Published by Elsevier Science Ltd.

Keywords: Fermentation; In vitro protein digestibility; Pearl millet; Phytic acid; Tannins; Total polyphenols

1. Introduction

Bulrush millet (*Pennisetum typhoidium*), also known as pearl millet, is of the same order and height as maize and sorghum; it is the most drought-resistant millet. Pearl millet is grown annually on about 26 million ha in the arid and semi-arid tropical areas of Africa and India, principally for grain and forage. Pearl millet has a well-balanced protein, except for its lysine deficiency, with high concentration of threonine and lower (but adequate) leucine than sorghum protein. Tryptophan levels are generally higher in pearl millet than in other cereals (Chung & Pomeranz, 1985).

Pearl millet has a high nutrient content but bioavailability is low, inherently due to the presence of antinutritional factors, such as phytic acid, polyphenols and tannins. Fermentation is one of the processes known to reduce these antinutrients.

The objective of the present investigation was to study changes occurring in antinutritional factors and the in vitro protein digestibility during natural fermentation of millet.

2. Materials and methods

2.1. Materials

Two pearl millet cultivars: Composite Population III, obtained from the Department of Agronomy, Faculty of Agriculture, University of Khartoum, Sudan and Baladi (Breeder seeds, 1997), obtained from El Obeid Research Station were cleaned and ground to pass a 0.4 mm mesh.

2.2. Preparation of dough

Natural millet fermentation was carried out by mixing millet flour with distilled water (1:2 w/v). This mixture was incubated at room temperature (30±2 °C) for periods of 0, 4, 8, 12, 16, 20, 24, 28, 32, and 36 h. Samples were withdrawn and transferred to aluminium dishes and dried in a hot air oven-drier at 70 °C for 3–4 h. Dried samples were ground and stored at 4 °C for analysis. All samples were analyzed for moisture content,

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ash, crude fibre, fat, crude protein, total polyphenols, tannins, phytic acid and in vitro protein digestibility.

2.3. Proximate analysis

Moisture content, crude protein, ether extract, crude fibre and ash were determined for the oven-dried samples according to the established AOAC (1984).

2.4. Determination of total polyphenols

Total polyphenols were determined according to the Pression Blue spectrophotometric method of Price and Butler (1977).

2.5. Tannin content determination

Qualitative estimation of tannin was carried out using the modified vanillin-HCl method according to Price, Scoyoc, and Butler (1978).

2.6. Determination of phytic acid

The phytic acid content was determined by the method described by Wheeler and Ferrel (1971).

2.7. Determination of in vitro protein digestibility

This was carried out according to Saunder, Connor, Booth, Bick, and Kohler (1973).

2.8. Statistical analysis

Each determination consisted of three separate samples, which were analyzed, in triplicate and the figures were then averaged. Data were assessed by analysis of variance (ANOVA; Snedecor and Cochran, 1987) and by the Duncan's multiple range test with a probability $P \leqslant 0.05$ (Duncan, 1955).

3. Results and discussion

The chemical composition of the two pearl millet cultivars is shown in Table 1. The results are expressed on a dry basis.

Moisture content for both Composite Pop. III and Baladi, 1997 (Breeder seeds) was 6.4%. This value is lower than the range reported by Varriano Marston and Hoseney (1980) who reported that moisture content for pearl millet ranged from 7.8 to 14.2%. Also, it is lower than those reported by Khatir (1990) who gave a range from 10.6 to 11.7% for local Sudanese millet varieties.

The ash content was 2.1% for Composite Pop. III. and 1.2% for Baladi, 1997. Values obtained were within the range given by Hadimani and Malleshi (1995) who reported a range of 1.2–2.4%. Table 1 gives the crude fibre content of Composite Pop. III and Baladi 1997 as 2.3 and 2.0%, respectively. Table 1 shows the fat contents of Composite Pop III and Baladi, 1997 as 8.5 and 6.2%, respectively. These values are within the range of 4–9% reported by Desikachar (1975). Crude protein was 10.8% for Composite Pop. III. and 14.9% for Baladi The values obtained in this study were within the range of 9–20% reported by Desai and Zendi (1979). Sullivan et al. (1990) reported that protein content for pearl millet ranged from 8 to 19%.

3.1. Antinutritional factors in pearl millet

Tannin content, expressed as catechin equivalents (CE), is shown in Tables 2 and 3 as 0.24% and 0.12% for Composite Pop III and Baladi 1997, respectively. Differences in tannin content for the two cultivars could be due to genotype.

Total polyphenols content, expressed as catechin equivalent (CE), is shown in Tables 2 and 3 as 319 and 294 mg/100 g for Composite Pop III and Baladi 1997, respectively. This is in agreement with the fact that polyphenols content of pearl millet is fairly high, as reported by Alka-Sharma and Kapoor (1996).

Phytic acid contents were 786 and 618 mg /100 g for Composite Pop III and Baladi, respectively. Values reported here were higher than those reported by Simwemba, Hoseney, Varriano-Marston, and Zeleznak (1984). However, Khetarpaul and Chauhan (1991) reported a very high level of 990 mg /100 g. Variation in phytic acid content among different cultivars can be attributed to both genetic and environmental conditions (Simwemba, Hoseney, Varriano-Marston, & Zeleznak, 1984).

3.2. Effect of fermentation on pH of pearl millet

The pH of Composite Pop. III and Baladi 1997 fermented dough at room temperature for 36 h, is shown

Table 1
Proximate composition of two pearl millet cultivars[a]

Cultivar	Dry matter (%)	Ash (%)	Crude fibre (%)	Fat (%)	Protein (%)
Composite Population III	93.6 (0.22)	2.1 (0.15)	2.3 (0.05)	8.5 (0.38)	10.8 (0.28)
Baladi (Breeder seeds 1997)	93.6 (0.22)	1.2 (0.09)	2.0 (0.05)	6.2 (0.63)	14.9 (0.28)

[a] Values are means (±SD).

in Tables 2 and 3. Fermentation was found to cause a gradual reduction in a pH with time. The change in pH from zero to 36 h resulted in a pH drop from 5.9 to 3.6 and from 5.8 to 3.7 for Composite Pop. III and Baladi 1997, respectively. Results indicated that fermentation of pearl millet dough caused a reduction in pH and this was more pronounced after 8 h of fermentation. These results agree with those obtained by Giese (1994) who reported that, as a result of fermentation, the increased acidity and low pH enhances the keeping quality of millet foods, by inhibiting microbial growth and also contributing to the flavour of processed millet. Usha Antony, Sripriya, and Chandra (1996) reported a drop in pH from 6.4 in unfermented finger millet to 5.2 in 24 h and to 4.3 in 48 h of fermentation.

3.3. Effect of fermentation on protein content of pearl millet

The protein contents of Composite Pop. III and Baladi 1997 fermented doughs are shown in Tables 2 and 3. The protein content of the two genotypes increased initially as a result of fermentation. The increase in protein content can be attributed to microbial synthesis of proteins from metabolic intermediates during their growth cycles (Zamora & Fields, 1979).

3.4. Effect of fermentation on tannin content of pearl millet

The tannin content, expressed as catechin equivalents (CE), is shown in Tables 2 and 3 for the two millet cultivars. Fermentation for 36 h at room temperature was found to cause no changes in tannin content of fermented dough for the two millet cultivars. This result agrees with Agte, Gokhale, and Chiplonkar (1997) who reported that the levels of tannins in pearl millet were unaffected by fermentation.

3.5. Effect of fermentation on polyphenols of pearl millet

Total polyphenols content, expressed as catechin equivalents, is shown in Tables 2 and 3. Fermentation

Table 2
Changes in pH, moisture, protein, polyphenols, tannins, phytate and in vitro protein digestibility during natural fermentation of Composite Population III[a,b]

Cultivar	Fersmentation time (h)	pH	Moisture content (%)	Protein content (%)	Polyphenols content (mg/100g)	Condensed tannins (%)	Phytate (mg/100g)	IVPD (%)
Composite Population III	0	5.9 (0.00)a	6.7 (0.40)e	10.8 (0.28)j	318.6 (0.04)a	0.24 (0.06)a	786.2 (0.00)a	60.5 (1.65)j
	4	5.7 (0.00)b	7.1 (0.51)c	11.3 (0.48)h	302.7 (0.00)b	0.24 (0.00)a	765.9 (0.00)b	62.2 (1.51)i
	8	4.7 (0.00)c	4.7 (0.21)i	11.1 (0.16)i	268.6 (0.04)c	0.24 (0.06)a	739.0 (5.10)c	67.3 (1.40)h
	12	4.4 (0.00)d	3.6 (0.25)j	13.0 (0.49)c	246.9 (0.00)e	0.24 (0.00)a	691.8 (0.00)d	73.4 (1.19)g
	16	4.0 (0.00)f	8.0 (0.42)a	12.7 (0.23)e	211.8 (0.02)h	0.24 (0.00)a	644.7 (10.20)e	78.4 (0.00)f
	20	4.1 (0.00)e	7.2 (0.25)b	13.1 (0.37)b	204.1 (0.25)i	0.24 (0.06)a	597.5 (5.10)f	80.5 (1.25)d
	24	3.9 (0.00)g	6.3 (0.21)g	12.8 (0.01)d	196.1 (0.09)j	0.24 (0.06)a	550.3 (0.00)g	86.0 (2.60)a
	28	3.7 (0.00)i	6.5 (0.10)f	12.5 (0.22)f	266.9 (0.03)d	0.24 (0.00)a	487.4 (2.50)h	79.0 (0.00)e
	32	3.8 (0.00)h	6.1 (0.20)h	13.2 (0.63)a	239.4 (0.00)f	0.24 (0.00)a	440.3 (0.00)i	80.8 (0.00)c
	36	3.6 (0.00)j	7.0 (0.49)d	12.4 (0.58)g	237.0 (0.03)g	0.24 (0.06)a	393.1 (5.10)j	82.9 (1.41)b

[a] Values are ±(SD).
[b] Means not sharing a common letter in a column are significantly different at $P \leqslant 0.05$.

Table 3
Changes in pH, moisture, protein, polyphenols, tannins, phytate and in vitro protein digestibility during natural fermentation of Baladi (1997)[a,b]

Cultivar	Fermentation time (h)	pH	Moisture content (%)	Protein content (%)	Polyphenols content (mg/100g)	Condensed tannins (%)	Phytate (mg/100g)	IVPD (%)
Baladi (Breeder seeds 1997)	0	5.8 (0.00)a	6.5 (0.10)g	14.92 (0.28)j	293.5 (0.08)a	0.12 (0.06)a	618.4 (0.00)a	61.9 (1.15)j
	4	5.6 (0.00)b	8.0 (0.56)c	16.18 (0.42)i	253.2 (0.00)b	0.12 (0.00)a	609.5 (2.05)b	63.9 (0.00)i
	8	5.2 (0.00)c	6.6 (0.27)e	19.68 (0.05)a	239.5 (0.06)c	0.12 (0.00)a	587.4 (5.10)c	69.5 (0.89)h
	12	4.4 (0.00)d	3.1 (0.58)j	17.67 (0.09)f	229.3 (0.00)d	0.12 (0.00)a	550.3 (10.20)d	74.2 (0.91)g
	16	4.2 (0.00)e	8.3 (0.56)b	17.87 (0.04)c	215.6 (0.02)f	0.12 (0.06)a	507.0 (5.10)e	76.7 (0.00)f
	20	4.1 (0.00)f	6.5 (0.10)f	18.01 (0.08)b	198.5 (0.05)I	0.12 (0.00)a	469.9 (5.10)f	78.5 (0.00)e
	24	3.9 (0.00)g	5.5 (0.15)h	17.82 (0.62)d	208.8 (0.01)h	0.12 (0.00)a	432.8 (0.00)g	79.1 (0.00)d
	28	3.9 (0.00)g	9.8 (0.21)a	17.55 (0.91)h	191.7 (0.01)j	0.24 (0.00)a	383.4 (1.50)h	86.2 (0.00)a
	32	3.8 (0.00)h	5.3 (0.21)i	17.64 (0.13)g	212.2 (0.01)g	0.12 (0.06)a	346.3 (0.00)i	79.5 (0.85)b
	36	3.7 (0.00)I	7.8 (0.50)d	17.75 (0.00)c	217.3 (0.00)e	0.12 (0.06)a	309.2 (0.00)j	79.1 (0.00)c

[a] Values are means ±(SD).
[b] Means not sharing a common letter in a column are significantly different at $P \leqslant 0.05$.

was found to cause a significant ($P \leqslant 0.05$) decrease in polyphenols from 319 to 196 mg/100 g after 24 h and from 294 to 199 mg /100 g after 20 h for Composite Pop. III and Baladi, respectively. This result agrees with Dhankher and Chauhan (1987) who reported a decrease in polyphenols of pearl millet with increasing fermentation time. Reduction in polyphenols may be due to activation of polyphenol oxidase (Dhankher & Chauhan, 1987). The increase in total polyphenols towards the end of fermentation could be attributed to higher losses in carbohydrate components.

3.6. Effect of fermentation on phytic acid content of pearl millet

Tables 2 and 3 show changes in phytic acid contents of fermented Composite Pop. III and Baladi 1997 doughs at room temperature for 36 h. Significant ($P \leqslant 0.05$) reduction was observed as a result of fermentation. Phytic acid contents decreased from 786 to 393 mg/100 g and from 618 to 309 mg/100 g for Composite Pop. III and Baladi, respectively. The percent decrease in phytic acid after 36 h was 50% for both cultivars. Khetarpaul and Chauhan (1990) reported that phytic acid is almost eliminated in pearl millet flour fermented at 30 °C for 72 h. Mahajan and Chauhan (1987) reported that endogenous phytase of pearl millet contributed significantly to the reduction of the phytate content of fermented pearl millet flour which was dependent on pH and temperature of fermentation. Microbial phytase, was reported to be present in several microorganisms; it hydrolyses phytic acid during fermentation of autoclaved flour, accounting for a reduction in phytic acid content in the autoclaved fermented product (Daniels & Fisher, 1981; Lopez, Gordon, & Fields, 1983).

3.7. Effect of fermentation on in vitro protein digestibility (IVPD) of pearl millet

The IVPD of Composite Pop. III and Baladi fermented dough is shown in Tables 2 and 3. Fermentation was found to cause a significant ($P \leqslant 0.05$) improvement in IVPD for the two pearl millet cultivars. The increase was from 60.5 to 86.0% (24 h) and from 61.9 to 86.2% (28 h) for Composite Pop. III and Baladi, respectively. Microflora may produce proteolytic enzymes during fermentation which may be responsible for the increased protein digestibility (Steinkraus, Lee, & Buck, 1965; Hasseltine, 1983). In addition, the elimination of phytic acid contributes to the improvement in protein digestibility in fermented millet (Khetarpaul, 1988). Thus, fermentation offers unique nutritional advantages for making protein of coarse-grained pearl millet more digestible, possibly by significantly reducing its phytate content and decreasing the level of polyphenols.

4. Conclusion

The proximate composition of the two cultivars of pearl millet showed that Baladi cultivar had a higher protein content but lower fat, ash, polyphenols, tannins and phytate contents compared to Composite Pop. III. In line with the lower value for polyphenols, tannins and phytate, Baladi had the higher protein digestibility. Results showed that natural fermentation of pearl millet decreases polyphenols and phytic acid and causes no changes in tannin contents of the two millet cultivars. Also, pearl millet protein digestibility is elevated, indicating that natural fermentation is associated with improvement in the nutritional quality of the grain.

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