

Phytic acid content and hydrochloric acid extractability of minerals in pearl millet as affected by germination time and cultivar

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Abstract

Ten pearl millet cultivars (SOSAT C-88, ZANGO, EX-BORNO, LCRI 9701, ICMV-IS 94206, ICMV-IS 94208, GWAGWA, G.I-14.9, GB 8735, G.I-297-1) were germinated for 96 h. The germinated grains were dried, polished and milled. Phytic acid contents and hydrochloric acid (HCl) extractability of minerals from the malt flours were determined at intervals of 24 h during germination. Phytic acid content decreased significantly ($P < 0.05$) with increase in germination time with concomitant increase in HCl extractable mineral contents. EX-BORNO had higher extractable Ca, while LCRI-IC 9701 had higher P, whereas Zn, Cu, Fe and I recorded high levels in ICMV-IS 94208, GB 8735, G.I-297-1 and ICMV-IS 94208, respectively. Mn did not differ ($P > 0.05$) at the various levels of germination time. There was good correlation between phytic acid reduction and increase in extractable minerals with increase in germination time.

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1. Introduction

Pearl millet is an important food crop for people living in the semi-arid region of Nigeria. The grain is comparable to some other cereal grains in term of nutrient content (Gopalan, Ramasasti, & Balasubramanian, 1989). However, it contains some antinutrient factors that affect the nutrient absorption by human body system (Sharma & Kapoor, 1997). Reduction of these antinutrient factors by malting have long been documented by other researchers (Khetarpaul & Chauhan, 1990; Obizoba & Atii, 1994) but such information is still scanty in this region. Therefore the objectives of this study were to evaluate the effect of germination time and cultivar on phytic acid content and hydrochloric acid extractability of minerals of pearl millet.

2. Materials and methods

2.1. Source of cereal grain

Ten pearl millet cultivars, SOSAT C-88, ZANGO, EX-BORNO, LCRI-IC 9701, ICMV-IS 94206, ICMV-IS 94208, GWAGWA, G.I-14.9, GB 8735 and G.I-297-1 were obtained from Lake Chad Research Institute, Maiduguri, Nigeria. These pearl millet cultivars have negligible tannin content (Badau, Nkama, & Ajalla, 2002). Chemical and reagents were obtained from recognized distributors and were of analytical grade.

2.2. Sample preparation

The cereal grains were cleaned manually to remove broken seeds, dust and other extraneous materials. Experimental samples were taken using the quartering

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procedures of Lees (1975). The cleaned grains were steeped in thrice quantity of water for 12 h with 1 h air rest after 6 h of steeping. For each air rest, the steeping water was changed. After steeping, the grains were sterilized by soaking in a solution of 1% sodium hypochlorite for 20 min before it was drained prior to germination. The steeped grains were spread on wet jute bags and covered with moist cotton cloth and left to sprout at room temperature (32 ± 3 °C) for 0, 24, 48, 72 and 96 h as described by Obizoba and Atii (1994).

After germination, the seeds were dried in Gallenkamp oven (BS model OV-160, England) at 50 °C for 24 h. Rootlets and shoots of the grains were separated from the kernels by rubbing the grain in a sieve (Endecotts Ltd, London, England) of 0.6 mm mesh size. The sieve allowed the rootlets and shoots to escape but retained the kernels (Morall, Boyd, Taylor, & Van Der Walt, 1986).

The unmalted grains and the polished malt were milled into fine flour with hammer mill (Gibbons Electric, Essex, UK) to pass through 1 mm mesh size screen for determination of phytic acid and hydrochloric acid extractability of minerals.

2.3. Total mineral determination

AOAC (1990) method was used for total mineral determination. Two grams of the oven dried unmalted and malted pearl millet cultivar flour were weighed and placed in a crucible and mineralized at 600 °C for 3 h. After cooling in a desiccator, the ashes were transferred into individual beaker and 20 ml of concentrated HNO₃ was added in each case and followed by 10 ml of H₂O₂. The mixture was heated at a temperature of 90 °C for 1 h and afterward, cooled and filtered. The filtrate was transferred into a 250 ml volumetric flask and distilled water was added to fill the flask to the mark. From this stock solution, 2 ml were pipetted into 50 ml flask and was made up to the required volume with distilled water. Mineral contents of these solutions were determined by atomic absorption spectrophotometry (Perkin–Elmer 2380, USA, 1976) for the various elements.

From stock solution of 1000 ppm, working standard solutions of the elements (BDH England) were prepared at 100 ppm by dilution. The elements included Zn, Cu, Ca, Mn, I, P, Fe, Pb and Cd. From the prepared stock solution of 100 ppm; standard solutions at 0.5, 1.0, 1.5 and 2.0 ppm were prepared for each element by dilution with distilled water. The absorbance of the standard solutions was obtained. The absorbance of the sample solutions obtained and their elemental concentration was calculated using the formula:

$$\text{Conc. (ppm) of mineral in test} = \frac{A_{\text{test}} \times \text{Conc. Std}}{A_{\text{std}}}$$

where A_{test} is the absorbance of the unknown element; A_{std} is the absorbance of standard and Conc. is the concentration.

2.4. HCl extractability of mineral

Hydrochloric acid extractability of minerals was determined by continuous shaking of 2 g of sample with 100 ml of 0.03 N HCl at 150 rpm for 3 h at 37 °C. The mixture was filtered through Whatman No.1 (ashless) filter paper and the clear supernatant was oven dried at 100 °C and the mineral estimated as described above (Chompreeda & Field, 1984; Sripriya, Anthony, & Chandra, 1997). The mineral HCl extractability was calculated as follows:

$$= \frac{\text{Mineral extracted in 0.03 N HCl} \times 100}{\text{Total mineral}}$$

2.5. Phytic acid determination

Phytic acid (phytate) content was measured by a method based on that of Davies and Reid (1979) and Griffiths (1982). About 1 g of each sample was extracted with 40 ml of 0.5 M HNO₃ for 1 h (Garcia-Esteba, Guerra-Hernandez, & Garcia-Villanora, 1999; Griffiths, 1982). It was filtered and 20 ml of 0.008 M ferric chloride solution was added to the filtrate and extracts were incubated at 100 °C for 20 min. The free Fe³⁺ remaining in the solution was then determined colorimetrically using 0.005 M ammonium thiocyanate and the iron-binding capacities of the extracts were determined by difference (Garcia-Esteba et al., 1999; Griffiths, 1982). The results were then expressed in terms of mg Fe bound per gram of sample extracted and is converted to percentage phytate by using a 3.5:6 molar ratio for Fe:P in ferric phytate (Griffiths & Thomas, 1981) and C₆P₆O₂₄H₁₈ as the empirical formula for phytic acid.

3. Results and discussion

3.1. Calcium

The hydrochloric acid extractability of calcium in unmalted and malted pearl millet cultivars at various germination times are shown in Table 1. The Ca content of the unmalted grains varied from 53.6 to 122 mg/100 g on dry matter basis and the HCl extractability of Ca varied from 42.3 to 45.3%. HCl extractability of Ca increased progressively ($P < 0.05$) from 0 to 72 h of germination and remained almost constant ($P > 0.05$) up to 96 h of germination for each of the cultivars.

Table 1
Hydrochloric acid extractability (%) of calcium in pearl millet as affected by germination time and cultivar^f

Cultivar	Unmalted grain	Period of germination (h)				
		0 ^e	24	48	72	96
SOSAT C-88	43.1 ^d (83.5)	46.4 ^{cd} yz (81.1)	49.6 ^c yc (80.8)	54.1 ^b xyz (80.6)	58.4 ^a xyz (80.3)	60.0 ^a wxy (81.3)
ZANGO	44.3 ^c (101)	45.5 ^c yz (98.1)	51.1 ^b yz (97.7)	51.4 ^b yz (98.4)	58.2 ^a xyz (99.1)	57.7 ^a wxyz (98.8)
EX-BORNO	45.3 ^d (122)	50.8 ^c xy (119)	57.9 ^b y (119)	60.9 ^{ab} x (119)	62.1 ^a x (119)	62.4 ^a w (119)
LCRI-IC 9701	43.4 ^c (68.0)	44.4 ^c z (63.9)	46.7 ^{bc} z (57.6)	49.6 ^b z (64.1)	54.3 ^a xyz (63.7)	55.2 ^a wxyz (64.4)
ICMV-IS 94206	44.5 ^d (122)	52.8 ^c x (119)	54.7 ^{bc} yz (120)	58.4 ^{ab} xy (119)	61.2 ^a xy (117)	61.4 ^a wx (118)
ICMV-IS 94208	42.4 ^c (91.7)	46.6 ^{bc} xyz (88.5)	48.8 ^b z (88.3)	50.2 ^{ab} yz (88.3)	53.4 ^a yz (88.1)	53.7 ^a xyz (88.9)
GWAGWA	42.6 ^c (114)	46.9 ^b xyz (111)	50.1 ^{ab} yz (109)	51.3 ^{ab} yz (110)	54.0 ^a xyz (110)	53.1 ^a yz (110)
G.I-14.9	42.3 ^c (73.2)	47.4 ^b xyz (68.3)	49.5 ^{ab} z (67.0)	52.1 ^a yz (67.2)	53.7 ^a yz (67.9)	53.3 ^a yz (68.6)
GB 8735	43.5 ^b (101)	45.5 ^b zy (99.8)	47.8 ^{ab} z (97.7)	50.4 ^a yz (98.0)	57.9 ^a xyz (91.9)	50.9 ^a z (98.3)
G.I-297-1	44.1 ^b (53.6)	44.6 ^b zy (51.8)	46.4 ^b z (50.3)	51.6 ^a yz (50.4)	52.2 ^a z (50.4)	52.4 ^a yz (51.4)

^{a-d} Means within each row not followed by the same superscript are significantly different ($P < 0.05$).

^{w-z} Means within each column not followed by the same superscript are significantly different ($P < 0.05$).

The values in brackets are total calcium content (mg/100 g) of grain on dry matter basis.

^e 0 = Steeped grains.

^f Mean of triplicate determinations.

Hydrochloric acid extractability varied among cultivars at different levels of germination times. The HCl extractability varied from 44.4% to 52.8%, 46.4% to 57.9%, 49.6% to 60.9%, 52.2% to 62.1% and 50.9% to 62.4% at 0, 24, 48, 72 and 96 h of germination, respectively. The HCl extractability of calcium in unmalted grains did not differ ($P > 0.05$) among the cultivars. However there were variations as from 0 h of germination onwards. EX-BORNO had the highest ($P < 0.05$) HCl extractable Ca, followed by ICMV-IS 94206, SOSAT C-88 and ZANGO at 72 h of germination. Therefore, EX-BORNO, ICMV-IS 94206, SOSAT C-88 and ZANGO are better sources of Ca. In a similar study, Sri-priya et al. (1997) increased the HCl extractability of Ca of finger millet germinated for 24 h from 47.6% (unmalted grain) to 53.2%.

3.2. Iron (Fe)

The iron extracted from unmalted and malted grains using HCl are shown in Table 2. The Fe content of the grains varied from 16.3 to 18.3 mg/100 g on dry matter basis and the HCl extractability varied from 18.5% to 20.7%. There was no significant difference ($P > 0.05$) between the Fe extracted from unmalted and steeped grains. However, HCl extractability of iron increased rapidly ($P < 0.05$) from the beginning of the germination. There were no significant differences ($P > 0.05$) between the cultivars at various levels of germination until 72 h of germination. G.I-297-1, ZANGO, GB 8735 had higher extractable Fe contents. In a similar study, Sri-priya et al. (1997) increased the Fe extractability of finger

Table 2
Hydrochloric acid extractability (%) of iron in pearl millet as affected by germination time and cultivar^e

Cultivar	Unmalted grain	Period of germination (h)				
		0 ^e	24	48	72	96
SOSAT C-88	19.9 ^d (17.3)	20.0 ^d (16.0)	35.5 ^c (15.3)	54.5 ^b (15.7)	64.2 ^a yz (14.3)	64.6 ^a yz (15.3)
ZANGO	20.5 ^d (18.3)	20.9 ^d (17.1)	36.2 ^c (16.5)	55.8 ^b (16.5)	66.5 ^a yz (16.5)	66.4 ^a yz (17.0)
EX-BORNO	18.5 ^d (17.8)	18.7 ^d (16.4)	34.2 ^c (15.2)	53.5 ^b (15.4)	62.7 ^a z (15.6)	62.6 ^a z (15.6)
LCRI-IC 9701	19.8 ^d (16.3)	20.0 ^d (15.8)	35.1 ^c (15.2)	54.5 ^b (15.8)	64.8 ^a yz (15.2)	65.3 ^a yz (15.1)
ICMV-IS 94206	20.7 ^d (17.2)	20.6 ^d (16.9)	35.8 ^c (15.8)	56.4 ^b (16.4)	65.5 ^a yz (16.1)	66.6 ^a yz (16.9)
ICMV-IS 94208	19.0 ^d (16.6)	19.2 ^d (15.0)	34.4 ^c (15.7)	53.5 ^b (15.7)	63.4 ^a yz (15.7)	62.5 ^a z (15.7)
GWAGWA	19.0 ^d (17.8)	19.2 ^d (17.1)	33.7 ^c (17.0)	53.1 ^b (17.6)	63.5 ^a yz (16.1)	64.7 ^a yz (16.5)
G.I-14.9	19.7 ^d (16.7)	19.8 ^d (15.8)	35.3 ^c (16.2)	55.0 ^b (16.2)	65.1 ^a yz (16.4)	64.6 ^a yz (16.0)
GB 8735	20.0 ^d (16.6)	20.0 ^d (14.9)	35.5 ^c (11.8)	55.9 ^b (15.3)	66.5 ^a yz (15.6)	65.9 ^a yz (15.5)
G.I-297-1	20.6 ^d (17.1)	20.6 ^d (15.7)	36.2 ^c (16.0)	56.4 ^b (16.2)	66.8 ^a yz (15.7)	66.9 ^a yz (15.8)

^{a-d} Means within each row not followed by the same superscript are significantly different ($P < 0.05$).

^{y-z} Means within each column not followed by the same superscript are significantly different ($P < 0.05$).

The values in brackets are total Iron content (mg/100 g) of grain on dry matter basis.

^d 0 = Steeped grains.

^e Mean of triplicate determinations.

millet germinated for 24 h from 5.90% (unmalted grain) to 8.60%.

Iron extracted from steeped grains was not significantly different ($P > 0.05$) from those of unmalted grains. Steeping did not affect the Fe extractability significantly.

3.3. Zinc

Hydrochloric acid extractability of Zinc increased when germination time increased (Table 3). The Zn extracted from unmalted and steeped grains did not differ significantly ($P > 0.05$). The total Zn content of unmalted grains varied from 2.82 to 3.24 mg/100 g on dry matter basis and the HCl extractability varied from 50.5% to 61.5%. The grains that had higher extractable Zn from the unmalted grains were

ICMV-IS 94208, GWAGWA, EX-BORNO, ZANGO and SOSAT C-88.

Similar increase in HCl extractable Zn of millet as a result of germination have been reported by Obizoba and Atii (1994). Gahlawat and Sehgel (1993) sprouted some grains for 48 h and recorded significant increase in HCl extractable Zn content. In another study by Sri-priya et al. (1997), Zn HCl extractability increased when finger millet was sprouted for 24 h.

Variations of HCl extractable Zn for different times of germination existed among the pearl millet cultivars. The values varied from 52.7% to 63.3%, 65.7% to 75.5%, 73.9% to 84.4%, 74.7% to 85.9% and 75.3% to 86.6% at 0, 24, 48, 72 and 96 h of germination, respectively. ICMV-IS 94206, ZANGO, EX-BORNO and GWAGWA had higher ($P < 0.05$) extractable Zn contents.

Table 3

Hydrochloric acid extractability (%) of zinc in pearl millet as affected by germination time and cultivar^c

Cultivar	Unmalted grain	Period of germination (h)				
		0 ^d	24	48	72	96
SOSAT C-88	59.2 ^c w (2.91)	60.4 ^c y (2.90)	73.1 ^b x (2.86)	83.0 ^a xy (2.81)	84.9 ^a w (2.87)	84.9 ^a wx (2.84)
ZANGO	60.4 ^c w (2.96)	63.0 ^c y (3.10)	75.5 ^b x (3.07)	83.9 ^a x (3.05)	84.5 ^a w (3.08)	84.7 ^a wx (3.09)
EX-BORNO	60.4 ^c w (3.01)	62.5 ^c y (2.95)	74.5 ^b x (2.93)	82.6 ^a xy (2.90)	84.3 ^a wx (2.91)	84.5 ^a wx (2.92)
LCRI-IC 9701	50.5 ^c z (2.90)	52.7 ^c z (2.71)	65.7 ^b z (2.73)	74.1 ^a z (2.70)	74.7 ^a z (2.66)	75.3 ^a z (2.74)
ICMV-IS 94206	59.5 ^c w (3.06)	61.2 ^c y (3.00)	73.2 ^b x (2.99)	82.2 ^a xy (2.81)	84.9 ^a w (3.00)	86.0 ^a w (3.01)
ICMV-IS 94208	61.5 ^c w (3.22)	62.2 ^c y (3.13)	73.8 ^b x (3.10)	84.4 ^a x (3.07)	85.9 ^a w (3.08)	86.6 ^a w (3.13)
GWAGWA	60.8 ^c w (2.82)	63.3 ^c y (2.78)	74.6 ^b x (2.77)	82.8 ^a xy (2.73)	84.2 ^a wx (2.72)	84.9 ^a wx (2.71)
G.I-14.9	53.1 ^c yz (3.27)	53.9 ^c z (3.23)	67.1 ^b z (3.21)	73.9 ^a z (3.18)	76.5 ^a z (3.16)	76.8 ^a yz (3.13)
GB 8735	58.3 ^c wx (3.19)	61.0 ^c y (3.01)	73.3 ^b x (3.12)	81.8 ^a xy (3.00)	83.1 ^a wx (3.01)	84.5 ^a wx (3.06)
G.I-297-1	57.9 ^c wx (3.24)	59.7 ^c y (3.13)	71.8 ^b xy (3.12)	79.4 ^a y (3.11)	81.5 ^a xy (3.10)	81.7 ^a xy (3.12)

^{abc} Means within each row not followed by the same superscript are significantly different ($P < 0.05$).

^{w-z} Means within each column not followed by the same superscript are significantly different ($P < 0.05$).

The values in brackets are total zinc content (mg/100 g) of grain on dry matter basis.

^d 0 = Steeped grains.

^e Mean of triplicate determinations.

Table 4

Hydrochloric acid extractability (%) of phosphorus in pearl millet as affected by germination time and cultivar^f

Cultivar	Unmalted grain	Germination period (h)				
		0 ^c	24	48	72	96
SOSAT C-88	19.4 ^d (60.4)	21.2 ^{cd} yz (57.3)	24.5 ^{bc} (58.8)	28.2 ^{ab} (58.4)	29.5 ^a yz (58.8)	30.4 ^a xyz (58.9)
ZANGO	19.4 ^b (51.5)	19.6 ^b yz (50.1)	23.2 ^{ab} (49.9)	26.9 ^a (50.0)	27.5 ^a z (50.1)	27.2 ^a yz (50.0)
EX-BORNO	18.6 ^d (50.5)	19.4 ^{cd} z (47.8)	23.2 ^{bc} (50.0)	27.2 ^{ab} (47.4)	29.2 ^a yz (48.1)	29.4 ^a xyz (48.0)
LCRI-IC 9701	19.7 ^d (40.0)	22.4 ^{cd} yz (37.5)	26.3 ^{bc} (38.4)	28.6 ^{ab} (38.3)	30.8 ^a yz (37.4)	31.4 ^a x (37.9)
ICMV-IS 94206	18.4 ^d (68.1)	20.2 ^{cd} yz (64.5)	24.0 ^{bc} (62.6)	25.7 ^{ab} (64.6)	28.8 ^{ab} yz (64.3)	28.7 ^a xyz (63.6)
ICMV-IS 94208	21.5 ^c (32.2)	25.1 ^{bc} y (30.0)	25.1 ^{bc} (31.3)	27.5 ^{ab} (30.5)	30.8 ^a yz (30.7)	30.3 ^a xyz (30.4)
GWAGWA	20.4 ^d (67.2)	21.5 ^{cd} yz (61.8)	25.6 ^{bc} (61.3)	28.4 ^{ab} (60.5)	31.8 ^a y (62.7)	31.1 ^a xy (64.8)
G.I-14.9	19.6 ^c (40.2)	20.6 ^{bc} yz (38.6)	24.3 ^{ab} (37.6)	25.1 ^{ab} (36.9)	27.0 ^a z (37.0)	26.5 ^a z (38.1)
GB 8735	20.2 ^c (41.0)	20.4 ^c yz (38.1)	23.8 ^{bc} (38.4)	27.1 ^{ab} (38.1)	28.0 ^{ab} yz (37.2)	28.7 ^a xyz (37.6)
G.I-297-1	20.5 ^b (30.4)	22.23 ^b yz (29.3)	24.8 ^{ab} (24.4)	27.4 ^a (23.6)	28.6 ^a yz (24.2)	28.5 ^a xyz (26.5)

^{a-d} Means within each row not followed by the same superscript are significantly different ($P < 0.05$).

^{x-z} Means within each column not followed by the same superscript are significantly different ($P < 0.05$).

The values in brackets are total phosphorus content (mg/100 g) of grain on dry matter basis.

^c 0 = Steeped grains.

^f Mean of triplicate determinations.

3.4. Phosphorus

Another important mineral considered in this study having a significant role in nutrition is phosphorus. Phosphorus hydrochloric acid extractability increased significantly ($P < 0.05$) with increase in germination time (Table 4). It increased up to 48 h of germination for most of the grains studied, after which it remained almost constant up to 96 h of germination. These findings are similar to the reports of other scientists (Khetarpaul & Chauhan, 1989; Sripriya et al., 1997).

The phosphorus content of the unmalted grains ranged from 30.4 to 68.1 mg/100 g on dry matter basis and the phosphorus extractability ranged from 18.4% to 20.5%. Variations of phosphorus extracted after various times of germination were noticed only at 0, 72 and 96 h of germination. The variations ranged from 19.4% to 25.1%, 27.0% to 31.8% and 28.5% to 31.4% at 0, 72 and 96 h of germination, respectively. The HCl extractable phosphorus contents of GWAGWA, LCRI-IC 9701, ICMV-IS 94208 and SOSAT C-88 were higher ($P < 0.05$) than in the other cultivars.

3.5. Iodine

The iodine content of unmalted grains ranged from 0.600 to 0.900 mg/100 g on dry matter basis and the HCl extractability ranged from 40.3% to 60.5%. The HCl extractability of unmalted and steeped grains, and those germinated for 24 h did not differ ($P > 0.05$) for some grains (SOSAT C-88, ZANGO, LCRI-IC 9701, ICMV-IS 94206, ICMV-IS 94208, GWAGWA). Iodine extracted from EX-BORNO and G.I-14.9 increased significantly ($P < 0.05$) from 0 to 24 h of germination. Most of the grains did not show any difference from 48 to 96 h of germination (Table 5).

There were differences in extractable iodine contents among the grains after various times of germination. The iodine extracted from the grains ranged from 42.8% to 61.6%, 45.5% to 64.9%, 48.6% to 67.9%, 52.4% to 69.0% and 52.7% to 68.6% at 0, 24, 48, 72 and 96 h of germination, respectively.

The extractable iodine content from ICMV-IS 94208, ZANGO, and ICMV-IS 94206 were higher ($P < 0.05$) than for other cultivars. For most of the grains, HCl extractability of iodine did not differ significantly ($P > 0.05$) after 24 h germination. This result agrees with the report of Obizoba and Atii (1994), where iodine was increased significantly when millet was sprouted for 36 h.

3.6. Copper

Copper was another important mineral extracted from unmalted and malted grains using HCl and the results obtained are shown in Table 6. The Cu extractability of unmalted and steeped grains did not differ significantly ($P > 0.05$). However, it increased significantly ($P < 0.05$) from 0 to 24 h of germination. The copper extracted from the germinated grains did not increase significantly ($P < 0.05$) from 48 to 96 h of germination for all the grains.

The Cu content of the unmalted grains ranged from 0.440 to 0.600 mg/100 g on dry matter basis and the extractable Cu ranged from 52.1% to 60.4%. There were variations of extractable Cu between the cultivars after various germination times. The extractable Cu varied from 54.5% to 66.5%, 81.2% to 86.9%, 82.4% to 90.3%, 85.0% to 91.6% and 85.3% to 92.1% at 0, 24, 48, 72 and 96 h of germination, respectively. Among the pearl millet cultivars, GB 8735, SOSAT C-88, G.I-14.9 and EX-BORNO had higher ($P < 0.05$) extractable

Table 5
Hydrochloric acid extractability (%) of iodine in pearl millet as affected by germination time cultivar^e

Cultivar	Unmalted grain	Period of germination (h)				
		0 ^e	24	48	72	96
SOSAT C-88	50.9 ^{b y} (0.840)	50.1 ^{b y} (0.800)	52.1 ^{b y} (0.810)	56.8 ^{a xy} (0.787)	58.7 ^{a xy} (0.790)	58.6 ^{a xyz} (0.810)
ZANGO	58.0 ^{c x} (0.600)	58.4 ^{c vw} (0.577)	61.7 ^{bc vwx} (0.557)	63.9 ^{ab uvw} (0.567)	67.3 ^{a uv} (0.573)	67.4 ^{a uv} (0.567)
EX-BORNO	41.5 ^{c z} (0.770)	43.5 ^{c z} (0.733)	48.3 ^{b yz} (0.727)	50.7 ^{ab yz} (0.723)	54.4 ^{a yz} (0.723)	54.3 ^{a yz} (0.730)
LCRI-IC 9701	40.3 ^{c z} (0.710)	42.8 ^{c z} (0.670)	45.5 ^{bc z} (0.663)	48.6 ^{ab z} (0.667)	52.4 ^{a z} (0.667)	52.7 ^{a z} (0.663)
ICMV-IS 94206	60.1 ^{b x} (0.800)	60.0 ^{b vw} (0.760)	63.1 ^{ab vw} (0.763)	64.7 ^{a uv} (0.733)	66.4 ^{a uvw} (0.750)	66.2 ^{a uvw} (0.767)
ICMV-IS 94208	60.5 ^{b x} (0.900)	61.6 ^{b v} (0.847)	64.9 ^{ab v} (0.857)	67.9 ^{a u} (0.850)	69.0 ^{a u} (0.847)	68.6 ^{a u} (0.847)
GWAGWA	51.2 ^{b y} (0.920)	52.2 ^{b y} (0.873)	56.4 ^{ab x} (0.863)	59.1 ^{a vwx} (0.870)	60.7 ^{a wx} (0.877)	59.7 ^{a xy} (0.883)
G.I-14.9	52.1 ^{c y} (0.740)	53.2 ^{c xy} (0.700)	57.7 ^{b wx} (0.700)	60.2 ^{ab vwx} (0.710)	62.1 ^{ab vwx} (0.717)	61.8 ^{a vwx} (0.713)
GB 8735	51.6 ^{b y} (0.810)	52.6 ^{b y} (0.767)	56.2 ^{ab x} (0.757)	57.1 ^{a x} (0.763)	59.7 ^{a yx} (0.770)	60.1 ^{a wxy} (0.777)
G.I-297-1	52.2 ^{dy} (0.890)	52.7 ^{cdy} (0.830)	56.7 ^{bc x} (0.827)	58.2 ^{ab wx} (0.833)	60.4 ^{a wxy} (0.807)	61.2 ^{a wx} (0.837)

^{a-d} Means within each row not followed by the same superscript are significantly different ($P < 0.05$).

^{u-z} Means within each column not followed by the same superscript are significantly different ($P < 0.05$).

The values in brackets are total iodine content (mg/100 g) of grain on dry matter basis.

^d 0 = Steeped grains.

^e Means of triplicate determinations.

Table 6
Hydrochloric acid extractability (%) of copper in pearl millet as affected by germination time and cultivars^c

Cultivar	Unmalted grain	Period of germination (h)				
		0 ^d	24	48	72	96
SOSAT C-88	58.6 ^c x (0.520)	61.4 ^c x (0.473)	86.6 ^b xy (0.473)	90.0 ^{ab} x (0.467)	90.8 ^{ab} y (0.477)	91.0 ^a x (0.473)
ZANGO	59.1 ^c x (0.560)	60.4 ^c xy (0.510)	84.7 ^b yz (0.500)	88.4 ^{ab} x (0.493)	89.3 ^a y (0.480)	89.9 ^a xy (0.483)
EX-BORNO	59.2 ^b x (0.470)	66.5 ^b vw (0.430)	86.2 ^a xy (0.413)	88.7 ^a x (0.403)	90.2 ^a y (0.400)	90.5 ^a x (0.410)
LCRI-IC 9701	59.3 ^c x (0.580)	60.6 ^c xy (0.520)	84.6 ^b yz (0.513)	86.6 ^{ab} xy (0.507)	88.7 ^{ab} yz (0.500)	88.9 ^a xyz (0.507)
ICMV-IS 94206	59.5 ^b x (0.580)	62.3 ^b x (0.517)	86.9 ^a xy (0.520)	88.6 ^a x (0.513)	89.7 ^a y (0.497)	89.8 ^a xy (0.497)
ICMV-IS 94208	52.1 ^c z (0.550)	54.5 ^c z (0.507)	81.2 ^b z (0.517)	82.4 ^{ab} z (0.510)	85.0 ^{ab} z (0.517)	85.9 ^a yz (0.503)
GWAGWA	53.6 ^b yz (0.600)	56.9 ^b yz (0.547)	81.8 ^a z (0.540)	83.9 ^a yz (0.520)	85.0 ^a z (0.530)	85.3 ^a z (0.540)
G.I-14.9	57.7 ^c xy (0.440)	61.3 ^c x (0.472)	86.1 ^b xy (0.393)	89.9 ^{ab} x (0.403)	90.3 ^{ab} y (0.397)	90.9 ^a x (0.400)
GB 8735	60.4 ^c x (0.550)	63.9 ^c vwx (0.517)	87.6 ^b xy (0.507)	90.3 ^{ab} x (0.503)	91.6 ^{ab} y (0.507)	92.1 ^a x (0.517)
G.I-297-1	60.4 ^b x (0.460)	62.4 ^b wx (0.440)	86.6 ^a xy (0.417)	88.5 ^a x (0.407)	88.1 ^a yz (0.410)	88.5 ^a xyz (0.417)

^{abc} Means within each row not followed by the same superscript are significantly different ($P < 0.05$).

^{v-z} Means within each column not followed by the same superscript are significantly different ($P < 0.05$).

The values in brackets are total copper content (mg/100 g) of the dry grain on dry matter basis.

^d 0 = Steeped grains.

^e Mean of triplicate determinations.

Cu contents. The copper extracted from the malted grains increased significantly ($P < 0.05$) until 24 h of germination and beyond this, there was no significant ($P > 0.05$) change. Nevertheless, very small amount is required for normal body function (Davidson, Passmore, Brock, & Truswell, 1975). Khetarpaul and Chauhan (1989) have reported similar results. They found out that HCl extractability of Cu could be increased from 35.2% to 56.5% by germinating the grain for 24 h at 30 °C. Identical results were obtained by Sripriya et al. (1997); Obizoba and Atii (1994) and Gahlawat and Sehgel (1993).

3.7. Manganese

Table 7 shows the HCl extractability of manganese in unmalted and malted grains. The extractability of Mn in unmalted and steeped grains did not differ significantly ($P > 0.05$). The HCl extractability of Mn for SOSAT C-88, ZANGO and G.I-297-1 increased rapidly ($P < 0.05$) from 0 to 48 h of germination and then became steady up to 96 h. Mn extracted from EX-BORNO, LCRI-IC 9701, ICMV-IS 94208 and GWAGWA increased from 0 to 24 h and then became steady up to 96 h of germination.

The Mn content and hydrochloric acid extractability did not differ significantly ($P > 0.05$) among the grains. However, there was significant ($P < 0.05$) increase of HCl extractable Mn contents during germination for each of the grains. The maximum value was reached at 48 h of germination for most of the cultivars studied. Similar reports have been published previously. When finger millet was germinated for 24 h, the Mn extractability increase from 4.27 to 4.69 mg/100 g (Sripriya et al., 1997) and when pearl millet was germinated for 24 h, it increase from 0.790 to 0.900 mg/100 g (Khetarpaul & Chauhan, 1989).

3.8. Lead and cadmium

There were traces of lead and cadmium in some of the pearl millet cultivars. LCRI-I 9701, G.I-14.9 and G.I-297-1, had traces of Pb in them. Traces of Cd were detected in ICMV-IS 94208 and G.I-297-1.

3.9. Recommended daily intake of minerals

The recommended daily intake (RDI) of major minerals and their main sources among the pearl millet cultivars are shown in Table 8. Ca was extracted more in EX-BORNO, while that of Fe, Zn, P and I were ZANGO, ICMV-IS 94208, GWAGWA and ICMV-IS 94208, respectively. The grains are better sources of Fe and Zn, followed by Ca. Pearl millet is not a good source of P and I. Malting has improved the quality of the minerals in pearl millet cultivars. However, it is advisable that people inhabiting the semi-arid tropics whose staple food is pearl millet need to supplement I, P and Ca from other sources.

It has been observed in this study, that malting has significantly improved the bioavailability of minerals of these pearl millet cultivars. SOSAT C-88, ZANGO, EX-BORNO, and ICMV-IS 94206 have been identified as better sources of dietary Ca. GWAGWA had higher P and Zn extracted from LCRI-9701 and G.I-14.9 were lower than the other cultivars. There was no variation of Cu extractability among the cultivars. Similarly, manganese did not show significant difference ($P > 0.05$) among cultivars. The Fe extractability did not differ among cultivars. ZANGO, ICMV-IS 94206 and ICMV-IS94208 were, identified as better sources of I. Traces of Pb and Cd were identified in few of the pearl millet cultivars.

These minerals extracted from the various pearl millet cultivars perform specific function in the human body.

Table 7
Hydrochloric acid extractability (%) of manganese in pearl millet as affected by germination time and cultivar^c

Cultivar	Unmalted grain	Period of germination (h)				
		0 ^d	24	48	72	96
SOSAT C-88	80.5 ^c (2.91)	80.6 ^c (2.88)	89.5 ^b (2.77)	95.2 ^a (2.88)	97.7 ^a (2.89)	98.1 ^a (2.85)
ZANGO	77.7 ^c (3.01)	78.6 ^c (2.88)	88.4 ^b (2.85)	93.3 ^a (2.74)	94.2 ^a (2.97)	94.1 ^a (2.95)
EX-BORNO	80.7 ^c (3.26)	82.6 ^c (3.04)	90.3 ^b (2.97)	93.2 ^{ab} (2.97)	96.5 ^a (2.89)	96.5 ^a (2.95)
LCRI-IC 9701	83.2 ^c (2.82)	83.5 ^c (2.71)	93.3 ^b (2.65)	97.1 ^{ab} (2.71)	98.4 ^a (2.67)	99.0 ^a (2.71)
ICMV-IS 94206	82.5 ^b (2.96)	82.5 ^b (2.95)	93.5 ^a (2.70)	96.6 ^a (2.85)	97.5 ^a (2.79)	97.7 ^a (2.85)
ICMV-IS 94208	80.3 ^c (2.76)	81.5 ^c (2.71)	90.4 ^b (2.65)	93.6 ^{ab} (2.66)	95.5 ^a (2.77)	96.3 ^a (2.70)
GWAGWA	78.4 ^c (2.67)	79.4 ^c (2.60)	90.5 ^b (2.50)	91.3 ^{ab} (2.46)	95.7 ^a (2.54)	96.2 ^a (2.58)
G.I-14.9	80.2 ^b (2.83)	81.3 ^b (2.75)	93.4 ^a (2.70)	95.4 ^a (2.68)	97.2 ^a (2.69)	97.6 ^a (2.77)
GB 8735	80.6 ^b (3.55)	82.3 ^b (3.37)	94.4 ^a (3.31)	96.6 ^a (3.35)	98.0 ^a (3.35)	98.4 ^a (3.35)
G.I-297-1	83.5 ^c (2.78)	82.5 ^c (2.68)	90.3 ^b (2.69)	97.1 ^a (2.70)	98.6 ^a (2.73)	99.0 ^a (2.75)

^{abc} Means within each row not followed by the same superscript are significantly different ($P < 0.05$).

The values in bracket are total manganese content (mg/100 g) of the dry grain on dry matter basis.

^d 0 = Steeped grains.

^c Mean of triplicate determinations.

Table 8
RDI of some important minerals obtained in some unmalted and malted pearl millet cultivars

Cultivar	Total mineral content (mg/100 g)		Quantity (g) of grain that meets RDI	
	Unmalted	Malted ^b	Unmalted	Malted
			Ca (1000 mg) ^a	
EX-BORNO	122	119	1808	1353
ICMV-IS 94206	122	117	1842	1397
SOSAT C-88	83.5	80.3	2584	2132
ZANGO	101	99.1	2174	1733
			Fe (10 mg)	
G.I-297-1	17.1	15.7	284	95.2
ZANGO	18.3	16.5	267	90.9
GB 8735	16.6	15.6	301	96.2
ICMV-IS 94206	17.2	16.1	281	95.2
			Zn (15 mg)	
ICMV-IS 94208	3.22	3.08	758	566
SOSAT C-88	2.91	2.87	872	615
ICMV-IS 94206	3.06	3.00	824	588
ZANGO	3.26	3.08	761	577
			P (700 mg)	
GWAGWA	67.2	62.7	5109	3518
LCRI-IC 9701	40.0	37.4	8883	6087
ICMV-IS 94208	32.2	30.7	10116	7400
SOSAT C-88	60.4	58.8	5983	4046
			I (150 mg)	
ICMV-IS 94208	0.900	0.847	27523	25685
ZANGO	0.600	0.573	43103	38860
ICMV-IS 94206	0.800	0.750	31185	30120
G.I-14.9	0.740	0.717	38860	33708

Values in brackets are the recommended daily intake (RDI) for adult males (31–50 years).

^a RDI Chart retrieved from <http://www.daily-vitamins.com/rda.html> on 13th June, 2004.

^b Germinated for 72 h.

Calcium helps in bone development and its deficiency can lead to improper development of bone in growing children leading to various deformities of the skeletal system (Gahlawat & Sehgel, 1993). Calcium contributes towards bone and teeth formation, muscle contraction and

blood clotting (Igoe, 1989). Iron is necessary for the prevention of anemia, (Igoe, 1989). Zinc functions as a nutrient and dietary supplement (Igoe, 1989).

Zinc functions as a nutrient and dietary supplement. It is believed to be necessary for nucleic acid

metabolism, protein synthesis and cell growth (Igoe, 1989). Deficiency of calcium, iron and Zinc are capable of producing severe impairment of health (Reinhold, 1988).

Phosphorus is an important mineral associated with calcium in bone. The calcium/phosphorus ratio in bone is 2:1. The imbalance of these minerals may lead to bone disorder known as Osteoporosis which is a major public health problem, as it may cause serious complication leading to incapacitation and requiring costly medical care (Spencer & Karmar, 1988).

Manganese functions as a nutrient and dietary supplement. It is needed in a very small amount (Robinson, 1973).

Lead and cadmium are harmful elements and their presence in food is not a healthy development (Bryce-Smith, 1980; Robinson, 1973).

3.10. Phytic acid

The phytic acid content of pearl millet cultivars are presented in Table 9. The phytic acid content of the unmalted grain ranged from 2.91% to 3.30%. The phytic acid decreased significantly ($P < 0.05$) during steeping. LCRI-IC 9701 was the only pearl millet cultivar whose phytic acid content decreased significantly ($P < 0.05$) within the first 24 h of germination. The phytic acid content did not show any appreciable decrease from 72 up to 96 h of germination. The phytic acid content of the various grains did not differ ($P > 0.05$) among cultivars at different levels of germination times. Germination had significantly ($P < 0.05$) reduced the phytic acid content of the grains.

Generally, cereal has been regarded as the major source of dietary phytate (Harland, 1989; Reddy, Pierson, Sathe, & Salunke, 1989). The majority of ingested phytate is undergraded during transit through gastroin-

testinal tract (Graf & Easton, 1990). Metal phytate complexes are highly insoluble over a wide pH range (Graf, Mahoney, Bryant, & Easton, 1984). One phytate molecule can bind up to six divalent cations, and the metal could possibly bridge at least two phytate molecules, depending on the redox state (Graf & Easton, 1990). Phytic acid is a powerful inhibitor of iron-driven hydroxyl radical formation because it forms a catalytically inactive iron chelate (Graf et al., 1984). These actions of phytic acid may contribute to its antioxidant activity on metal-catalyzed lipid peroxidation (Lee & Hendricks, 1997).

Steeping and germination decreased the phytic acid content of pearl millet cultivars. Gupta and Sehgal (1991) have observed decrease in phytic acid contents of cereal grains used for preparing weaning foods as a result of soaking and germination. The decrease in the level of phytic acid during soaking may be attributed to leaching out into soaking water under the concentration gradient (Abdullah, Baldwin, & Minor, 1984). Other researchers have reported decrease in the level of phytic acid during soaking (Khokhar & Chauhan, 1986; Ologhobo & Fetuga, 1984) and germination (Borade, Kadam, & Salunke, 1984; Mandal, Burman, & Biswas, 1972) due to phytase activity in the germinating grains (Rao & Deosthale, 1982). Agte and Sandhana (1997) reported that soaking of wheat batter is beneficial because it reduces phytic acid by 40%.

Phytase activity was found during germination of wheat, barley, rye and oats which hydrolyse phytate to phosphate and myoinositol phosphates (Larsson & Sandberg, 1992). Indumadhari and Agte (1992) reported that the increase in HCl extractable minerals in cereals could be attributed to reduction in phytate.

Germination of pearl millet grains at 30 °C for 24 h reduced the phytic acid by more than 50% (Khetarpaul & Chauhan, 1990). The inherent phytase activity in pearl millet grains (Mahajan & Chauhan, 1987) hydro-

Table 9
The phytic acid content (%) of pearl millet as affected by germination time and cultivar^f

S/No	Material	Unmalted Grain	Germination time (h)				
			0 ^c	24	48	72	96
<i>Pearl millet cultivars</i>							
1	SOSAT C-88	2.91 ± 0.37 ^a	1.58 ± 0.37 ^b	0.826 ± 0.344 ^{bc}	0.409 ± 0.223 ^c	0.282 ± 0.039 ^c	0.263 ± 0.065 ^c
2	ZANGO	2.96 ± 0.38 ^a	1.63 ± 0.31 ^b	0.816 ± 0.364 ^b	0.414 ± 0.196 ^c	0.289 ± 0.043 ^c	0.280 ± 0.025 ^c
3	EX-BORNO	3.16 ± 0.27 ^a	1.97 ± 0.37 ^b	1.20 ± 0.41 ^{bc}	0.501 ± 0.301 ^c	0.370 ± 0.161 ^d	0.378 ± 0.174 ^d
4	LCRI-IC 9701	3.26 ± 0.32 ^a	1.93 ± 0.40 ^b	1.15 ± 0.42 ^b	0.530 ± 0.336 ^{cd}	0.402 ± 0.177 ^d	0.416 ± 0.196 ^d
5	ICMV-IS 94206	3.19 ± 0.36 ^a	2.00 ± 0.35 ^b	1.14 ± 0.46 ^b	0.513 ± 0.337 ^{cd}	0.376 ± 0.136 ^{bc}	0.380 ± 0.143 ^c
6	ICMV-IS 94208	2.98 ± 0.37 ^a	1.61 ± 0.33 ^b	0.89 ± 0.37 ^{bc}	0.410 ± 0.203 ^{bc}	0.271 ± 0.054 ^c	0.262 ± 0.036 ^c
7	GWAGWA	3.30 ± 0.28 ^a	1.92 ± 0.39 ^b	1.42 ± 0.20 ^b	0.524 ± 0.322 ^c	0.391 ± 0.188 ^c	0.393 ± 0.192 ^c
8	G.I-14.9	3.25 ± 0.34 ^a	2.00 ± 0.34 ^b	1.19 ± 0.33 ^{bc}	0.508 ± 0.306 ^c	0.411 ± 0.203 ^c	0.414 ± 0.207 ^c
9	GB 8735	3.21 ± 0.27 ^a	1.97 ± 0.36 ^b	1.12 ± 0.43 ^{bc}	0.523 ± 0.328 ^c	0.413 ± 0.212 ^d	0.403 ± 0.194 ^d
10	G.I-297-1	3.18 ± 0.27 ^a	1.97 ± 0.39 ^b	1.18 ± 0.39 ^b	0.495 ± 0.326 ^c	0.364 ± 0.107 ^c	0.360 ± 0.101 ^c

^{a-d} Means within each row not followed by the same superscript are significantly different ($P < 0.05$).

^c 0 = steeped grains.

^f Mean ± Std of triplicate determinations.

Table 10
Correlation analysis between phytic acid and hydrochloric acid extractability of minerals in pearl millet cultivars

	Phytic acid	Ca	Fe	Zn	P	Cu	Mn
Ca	-0.89 ^a						
Fe	-0.87 ^a	0.78 ^a					
Zn	-0.65	0.63	0.68				
P	-0.88 ^a	0.56	0.47	0.44			
Cu	-0.55	0.45	0.46	0.81 ^a	0.47		
Mn	-0.51	0.41	0.43	0.76 ^a	0.43	0.87 ^a	
I	-0.48	0.68	0.78 ^a	0.65	0.66	0.43	0.40

^a significant at 5 % level ($P < 0.05$).

lyze phytic acid during germination and account for reduction in phytic acid in sprouts. Similar reduction in phytate during germination (Kumar, 1989) have been reported.

Germination has been reported to reduce phytate in various food grains (Hsu, Leung, Finney, & Morad, 1980; Khokhar & Chauhan, 1986) and to increase inorganic and HCl-extractable divalent minerals (Beal, Finnney, & Mehta, 1984; Rao & Deosthale, 1983).

A lot of reports on the detrimental effects of phytic acid have been published (Clydesdale, 1988; Reinhold, 1988; and Spencer & Karmer, 1988) but recent findings have shown that it has some beneficial effects. Recent information showed that phytates possess potential ability to lower blood glucose, reduce cholesterol and triacylglycerols, and reduce the risks of cancer and heart disease (Burgess & Gao, 2002; Cornforth, 2002; Jenab & Thompson, 2002). Generally, increase in the bioavailability of minerals in pearl millet cultivars experienced in this study could be a result of the decrease in the phytic acid level of these grains during germination.

3.11. Correlation analysis

Correlation between phytic acid content and HCl extractability of minerals of the grains are presented in Table 10. There were significant correlation between phytic acid content and most of the minerals extracted from the grains. Phytic acid content was negatively correlated with Ca ($r = -0.89$; $P < 0.05$), Fe ($r = -0.87$; $P < 0.05$), and P ($r = -0.88$; $P < 0.05$). Ca was positively correlated with Fe ($r = 0.78$; $P < 0.05$). Fe was positively correlated with I ($r = 0.78$; $P < 0.05$). Zn was positively correlated with Cu ($r = 0.81$; $P < 0.05$), and Mn ($r = 0.76$; $P < 0.05$), whereas Cu was positively correlated with Mn ($r = 0.87$; $P < 0.05$).

In a similar study, Khetarpaul and Chauhan (1989) reported significant high negative correlation between phytic acid levels and extractable iron content ($r = -0.9999$; $P < 0.01$), Zinc ($r = -0.893$; $P < 0.01$) and Calcium ($r = -0.9995$; $P < 0.01$) and a significant but low correlation with copper ($r = -0.662$; $P < 0.05$), but not with manganese ($r = -0.289$; $P > 0.05$).

The relationship between the phytic acid content and most of the mineral contents studied was an inverse one, as the phytic acid content decreased, the extractability of the minerals increased. This confirms earlier reports that the increase in the minerals bioavailability could be increased by germination (Khetarpaul & Chauhan, 1989). Since during germination, phytase is produced which breakdown the phytic acid substance that bind minerals making them available for the body system (Geetha, Virupaksha, & Shadaksharaswamy, 1997; Raindran, 1991).

4. Conclusion

The germination of various pearl millet cultivars increased significantly the HCl extractable parts of Ca, Fe, Zn, P, I, Cu and Mn, and also reduced significantly ($P < 0.05$) the phytic acid content of the pearl millet cultivars. Calcium was extracted more in EX-BORNO, ICMV-IS 94206 and SOSAT C-88, iron was more in G.I-297-1, ZANGO, GB 8735, zinc was extracted more in ICMV-IS 94206, ZANGO, EX-BORNO and GWAGWA, phosphorus more in GWAGWA, LCRI-IC 9701, ICMV-IS 94208 and SOSAT C-88, Copper more in GB 8735, SOSAT C-88, G.I-14.9 and EX-BORNO, and the extractable part of Mn was almost the same in the various cultivars. Iodine was extracted more in ICMV-IS 94208, ZANGO and ICMV-IS 94206.

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