Comparison between food-to-food fortification of pearl millet porridge with moringa leaves and

baobab fruit and with adding ascorbic and citric acid on iron, zinc and other mineral

bioaccessibility

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Abbreviation: RNI = Recommended Nutrient Intake

ABSTRACT

Mineral deficiencies remain high in Africa, especially in rural communities subsisting on monotonous cereal-based diets. The effects of adding locally available plant foodstuffs rich in minerals, namely moringa leaf powder and promoters of mineral bioavailability, namely baobab fruit powder at 15:100 parts pearl millet porridge on mineral bioaccessibility was compared with ascorbic acid and citric acid addition. Percentage bioaccessible and total bioaccessible iron were improved only in millet+baobab and millet+citric acid, by 36%, and 52%, and by 30%, and 52%, respectively compared to millet alone. Only inclusion of citric acid improved zinc bioaccessibility. However, millet+moringa reduced bioaccessible zinc, probably because of the phenolics, very high calcium content and low organic acid content in moringa. Baobab, moringa and citric acid inclusion improved calcium and magnesium bioaccessibilities. Notwithstanding its high phenolic content, natural fortification of cereal porridge with baobab fruit enhances iron, calcium and magnesium bioaccessibility. This is probably because it is rich in both citric acid and ascorbic acid. It is nearly as effective as citric acid addition in enhancing bioaccessibility of these minerals and its inclusion in staple cereal food products could be a useful alternative or complement to conventional fortification for communities in rural semi-arid Africa.

1 Introduction

Many households in rural sub-Saharan Africa subsist on cereal-based diets for energy and micronutrients (Uusiku, Oelofse, Duodu, Bester & Faber, 2010). Such communities that are at risk of mineral deficiencies are often not reached by conventional food fortification and micronutrient supplementation strategies because of limited financial resources and inadequate health infrastructure (Dairo & Ige, 2009). Therefore, it has been advocated that a combined approach of conventional food fortification and local food-to-food fortification using locally available, micronutrient-dense foods is needed to ensure sustainable adequate micronutrient nutrition (Kruger, Mongwaketse, Faber, Van der Hoeven & Smuts, 2015).

Pearl millet is an important staple cereal crop in semi-arid Western Africa where it is widely produced and consumed (FAO, 2014). This makes pearl millet an important potential food vehicle for food-to-food micronutrient fortification in the region. However, like other cereals, it contains antinutritional factors that limit mineral bioavailability (Taylor, 2016). Thus, there is need that other locally available African plant foods, that are high in essential minerals, for example moringa leaves (Glover-Amengor, Aryeetey, Afari & Nyarko, 2016) and high in enhancers of mineral bioavailability (ascorbic acid and citric acid), for example baobab fruit (Tembo, Holmes & Marshall, 2017), be added to pearl millet-based foods to improve their mineral nutritional quality. Moringa leaves are an important cash crop in West Africa and, for example, are traditionally consumed as a sauce with rice or millet in Senegal (Fuglie, 2005). Baobab fruit is a widely consumed food across tropical Africa, including being added to millet-or sorghum-based gruels (Buchmann, Preshler, Hartl & Vogl, 2010).

Van der Merwe (2017) showed that in the presence of provitamin A-rich plant sources, baobab fruit, moringa leaves and roselle calyces can enhance the amount of bioaccessible of iron in pearl millet-based porridge. Therefore, this study evaluated the effects of inclusion of dried moringa leaf and baobab fruit powder on iron, zinc and other mineral bioaccessibilities in pearl millet porridge alone in comparison with adding ascorbic acid and citric acid, known promoters of mineral bioavailability (Lönnerdal, 2000; Iyengar, Pullakhandam, & Nair, 2010).

2 Materials and Methods

2.1 Raw materials and processing

Whole pearl millet grain (variety Kuphanjala-2) was sourced from the International Crops Research Institute for the Semi-Arid Tropics, Bulawayo, Zimbabwe. Naturally dehydrated organic baobab fruit powder, obtained from the endocarp, was from Nautica Organic Trading, Durban, South Africa. Moringa leaves were obtained from the University of Pretoria experimental farm, Pretoria, South Africa. Ascorbic acid and citric acid monohydrate were from Merck-Millipore, Johannesburg, South Africa. The millet grain was milled using a laboratory hammer mill fitted with a 500 µm opening screen (Falling Number 3100, Perten Instruments, Stockholm, Sweden) to produce a wholegrain flour. Moringa leaves were sorted, rinsed with deionised water and drained on stainless steel trays for 15 min, after which they were frozen at -20°C, freeze dried and milled to fine powder using an air-cooled, knife-type laboratory mill (IKA A11, Staufen, Germany). The baobab fruit powder was vacuum packed and other flour samples were stored in zip-lock plastic bags at -20°C prior to analysis.

2.2 Preparation of pearl millet porridge

Deionised water (100 mL) was added to the whole grain millet flour (25 g). The millet slurry was heated to 100°C and maintained for 10 min with constant stirring. The cooked millet porridge was cooled, frozen at -20°C and freeze dried. The dried porridge was finely milled using the IKA A11 mill and stored at -20°C prior to further analyses.

2.3 Porridge formulations

Moringa leaf powder and baobab fruit powder were added to the cooked pearl millet porridge powder at a dry mass ratio of 15:15:100 and compared to millet porridge fortified with ascorbic acid and citric acid. The levels of ascorbic and citric acid used were equivalent to the levels in baobab powder.

2.4 Analyses

2.4.1 Moisture

Moisture contents of the plant materials were determined by an air oven, one stage, drying procedure, method 44-15A (AACC, 2000).

2.4.2 Total phenolics

The Folin-Ciocalteu method was used to quantify total phenolic content, as described by Waterman & Mole (1994) using 0.103 mol/litre HCl in methanol as the extractant at a volume to flour mass ratio of 40:1, according to the method of Price, Van Scoyoc & Butler (1978).

2.4.3 Phytate

Phytate content was determined using the extraction and assay procedure of Frühbeck, Alonso, Marzo & Santidrián (1995) based on the method of Latta & Eskin (1980). Dowex1-anion-exchange resin-AG 1 x 4 (4% Cross-linkage, chloride form, 100-200 mesh (74-149 μm) in glass

barrel Econo-columns, 7 x 5 mm was used for purification of the extracts. The standard, sodium phytate (P-8810, Sigma-Aldrich, Johannesburg, South Africa) and purified extracts were reacted with Wade reagent, after which absorbance was measured at 500 nm.

2.4.4 Condensed tannins

Condensed tannins were measured by the Vanillin-HCl method of Maxson & Rooney (1972) as modified by Price, Van Scoyoc & Butler (1978). Blanks were included to take into account the colour of the extracts. Vanillin reacts with the condensed tannins forming coloured complexes, which were measured colorimetrically at 500 nm.

2.4.5 Organic acids

The extraction and quantification of organic acids was done according to the method of Tembo et al. (2017) with slight modification. Powdered sample (2 g) was extracted in 20 ml metaphosphoric acid (0.3 g/litre and centrifuged (3500 x g, at 4°C for 10 min). The supernatants were filtered through Millipore 0.45 μm PTFE filters (Merck-Millipore, Johannesburg, South Africa) to obtain a clear filtrate. Quantification of organic acids was conducted by reversed–HPLC using acidified (pH 2.6) 0.01 mol/litre KH₂PO₄ as mobile phase, a Phenomenex-C18 column (250 x 4.6 mm, Torrance, USA), and simultaneous absorbance at 254 nm was used to detect ascorbic acid and at 210 nm to detect citric, malic and tartaric acids. The identity and quantification of the individual organic acids was confirmed by spiking with standards of these organic acid standards.

2.2.6 Minerals

Acid digestion of all the plant foods was performed using conc. nitric acid plus hydrogen peroxide according to EPA method 3051A (U.S. EPA, 2007). Iron, zinc, calcium, magnesium, phosphorus and aluminium contents of the digested flour samples were analysed by EPA method 200.7 (U.S. EPA, 1994) using inductively coupled plasma-atomic emission spectrometry (ICP-AES) (iCAP 6000 series, Thermo Fisher Scientific, Waltham, USA). Elements were analysed using wavelengths 239.5 nm for Fe, 206.7 nm for Zn, 315.8 for Ca, 285.2 nm for Mg, 214.9 nm for P and 308.2 nm for Al. To ensure accuracy, samples were analysed against National Institute for Standards and Technology traceable standards and independent quality control solutions. A calibration acceptance criteria of R²> 0.9995 was used and an internal standard technique was also used to ascertain the accuracy of the results.

2.7 In vitro dialysability mineral bioaccessibility

The porridge formulations were subject to *in vitro* digestion to simulate human gastric and intestinal digestion. The *in vitro* dialysability method of Miller, Schricker, Rasmussen, Van Campen (1991) was used. Digestive enzymes and bile salts used were pepsin (P-7000), pancreatin (P-1750), and bile extract (B-8631) (Sigma-Aldrich, Johannesburg, South Africa). Dialysis tubing Spectra/Por 7 (\emptyset = 20.4 mm) with a molecular weight cut-off of 10 kDa was used (G.I.C. Scientific, Johannesburg, South Africa). Mineral contents of the dialysates were determined by ICP-AES as described above, but without the digestion step. Mineral bioaccessibility (%) was calculated as the percentage of the mineral in the dialysate as compared to the total mineral content in the digest.

2.8 Assessment of nutrient content in relation to nutrient requirements

from an average portion of porridge to

the recommended nutrient intakes (RNI) for women of reproductive age were calculated and expressed as a percentage of the RNI.

2.9 Statistical analyses

The compositional analyses on the plant foods were conducted in duplicate and analysed twice. Independent bioaccessibility experiments were performed two times with the intestinal step being each time performed three times. Data were analysed by one-way analysis of variance (ANOVA) using IBM SPSS Statistics 25.0, Armonk, USA. Turkey's HSD Post-hoc test was applied to determine significant differences between specific means at a confidence level of 95% (p < 0.05). Fisher's LSD Post-hoc test was also applied for pair-wise comparison between the control formulation and the treatments.

3 Results and Discussion

Except where stated otherwise, all composition data from this study are given on a dry basis.

3.1 Mineral contents

Table 1 shows that moringa leaf powder contained more than twice the iron (19.1 mg/100 g) compared to the pearl millet porridge (9.0 mg/100 g). As a result, the iron content of the composite porridge containing moringa was increased by 19%. The iron level in the moringa was within the range reported in Leone et al. (2015) and similar to the value in the USDA Food Composition Database (20.0 mg/100 g) (USDA NDL, 2018). The moringa was more than 6- and 200-fold higher in magnesium and calcium, respectively compared to the millet porridge. As a result, inclusion of moringa with millet porridge greatly increased its magnesium and calcium contents by more than 65% and 26-fold, respectively. Brilhante et al. (2017) also reports levels

as high as 1050 mg/100 g magnesium and 3650 mg/100 g calcium in moringa powder). Porridges containing moringa could substantially increase the contribution of iron, calcium and magnesium to the RNI for women of reproductive age (FAO & WHO, 2001) (Table 1).

The baobab fruit powder, although low in iron, zinc and phosphorus contained substantial levels of magnesium and calcium compared to the millet porridge, 2-fold and 24-fold higher, respectively (Table 1). However, addition of baobab to millet porridge only slightly increased the calcium and magnesium contents of the composite porridge. Regarding zinc, both moringa and baobab had much lower contents than millet porridge. Similar low levels of zinc in moringa powder (0.2–3.3 mg/100 g) (Brilhante et al., 2017) and baobab (0.4–2.4 mg/100 g) (Stadlmayr, Charrondiere, Eisenwagen, Jamnadass & Kehlenbeck, 2013) are reported. As a result, the zinc contents of the millet porridge+moringa and millet porridge+baobab formulations were slightly but not significantly lower (p < 0.05) than in the millet porridge.

Moringa was highest in aluminium (12.8 mg/100 g). High levels of aluminium in plant foods could be an indication of soil contamination (Salvo et al., 2018), and hence artificially high levels of essential minerals, particularly iron. However, the high aluminium in moringa is unlikely to be from soil contamination as other researchers have shown that it is high in aluminium (Kawo et al., 2009).

3.2 Inhibitors of mineral bioavailability

As stated, antinutritional factors in plant foods such as phytate, polyphenols and tannins limit bioavailability of essential minerals such as iron and zinc (Gabaza, Muchuweti, Vandamme & Raes, 2017). Table 2 shows that both moringa and baobab had high total phenolic contents, about

10 times higher than in the millet porridge. The total phenolic contents of moringa and baobab powders were in the ranges of other studies, 1390–5350 mg/100 g (Vongsak et al., 2013) and 1560–4075 mg/100 g (Kamatou, Vermaak & Viljoen, 2011; Tembo et al., 2017), respectively. The baobab also had a very high apparent tannin content (approximately 8000 mg/100 g). This was probably due to contamination of the fruit pulp with seed fragments, which were observed in this study and are unusually high in tannins (Osman, 2004). Concerning phytate, millet porridge had much higher phytate content, more than five times higher than in moringa and baobab.

3.3 Enhancers of mineral bioavailability

Table 3 shows that baobab was high in organic acids, especially ascorbic acid (140 mg/100 g) and citric acid (3345 mg/100 g) when compared to the millet porridge, which contained no detectable ascorbic acid. The citric acid content of baobab was approximately 22-fold higher compared to the millet porridge. Standlmayr et al. (2013) also reported high levels of ascorbic acid in baobab powder (126–509 mg/100 g). Also, the level is similar to those in the USDA Food Composition Database (150.0 and 272.7 mg/100 g) (USDA NDL, 2018). Likewise, the citric, tartaric and malic acid contents of baobab powder found here are similar to literature values, 3300 mg/100 g (Tembo et al., 2017), not detected and 160 mg/100 g (Magaia, Uamusse, Sjoholm & Skog, 2013), respectively. Therefore, the calculated levels of ascorbic and citric acid in the composite porridges including baobab increased substantially to 17–18 mg/100 g and more than 500 mg/100 g, respectively. Moringa was also much higher in citric acid compared to millet porridge, some 13 times higher but unlike baobab contained low level ascorbic acid (10 mg/100 g), slightly lower than the 17 mg/100 g in the literature (Gopalakrishnan et al., 2016).

Ascorbic acid and citric acid are known to be potent enhancers of mineral bioavailability in foods. They chelate minerals and keep them in a soluble and absorbable form (Lönnerdal, 2000; Iyengar, Pullakhandam, & Nair, 2010). Furthermore, ascorbic acid acts as a reducing agent for apical membrane-bound ferrireductase which reduces Fe³⁺ to Fe²⁺, the only form in which non-haem iron is transported across the intestinal membrane (Mackenzie & Garrick, 2005).

3.4 Mineral bioaccessibility

As indicated, mineral bioaccessibility from foods depends on modifiers of absorption as well as their mineral content (Krishnan, Dharmaraj & Malleshi, 2012). Table 4 shows that the percentage iron bioaccessibility was significantly increased (p < 0.05) only in the baobab- and citric acid-fortified millet porridges, in terms of percentage by 36 and 52%, respectively, and in terms of amount by 30% and 52%, respectively compared to millet porridge alone. With mineral bioaccessibility assays, the direction of the effect is a more reliable guide than the actual measured magnitude (Fairweather-Tait et al., 2005). Thus, it seems that addition of baobab and citric acid to millet porridge will have a positive effect on iron bioavailability. Assuming that this positive effect would be to improve mineral bioavailability from low to moderate, the contribution of iron from 100 g of porridge to the RNI for women of reproductive age (FAO & WHO, 2001) would increase from 29.3% to 35.1% for millet porridge+baobab and similarly from 30.6% to 36.7% for millet porridge+citric acid (Table 1).

As inclusion of citric acid increased iron bioaccessibility, whereas inclusion of ascorbic acid had no significant effect (p < 0.05) (Table 4), the positive effect of baobab inclusion is likely mainly due to its high citric acid content. This is notwithstanding the fact that the molar ratios of citric

acid:iron (19:1) and ascorbic acid:iron (0.7:1) in the millet porridge+baobab were below the citric acid:iron and ascorbic acid:iron ratios of 100:1 and 2-10:1, respectively required for optimal iron absorption in cereal foods (Hunt, 2005). However, organic acids at lower ratios have been found to improve iron bioaccessibility. Porres, Etcheverry & Miller (2001) showed that addition of 600–800 mg citric acid to 100 g of a wheat-based food, similar to the 567 mg citric acid/100 g porridge in this present study, improved iron dialysability by 12-fold. The findings of this present study are in apparent contrast to those of Gabaza et al. (2018) who found % iron bioaccessibility was not improved when fermented pearl millet was enriched with baobab at a ratio 100:10. The difference is probably due to the effect of the organic acids in baobab being obscured in the study of Gabaza et al. (2018) by the effect of fermentation, which is well known to improve iron bioavailability (Proulx & Reddy, 2007).

As baobab and moringa contained high levels of total phenolics and tannins, especially in baobab, and significant levels of phytate (Table 2), the presence of these antinutritional factors likely affected iron bioaccessibility. For example, the phytate:iron molar ratios for the formulations containing baobab and moringa were high, in the range 10:1 to 12:1. Iron availability can be seriously impaired at phytate:iron molar ratios ≥1 (Hurrell, 2003). Although the porridge formulations including moringa had the highest iron contents (Table 1), there was a significant reduction in % iron bioaccessibility (by 24% compared to millet porridge only) (Table 4). This could also have been due to its high calcium content (Table 1), which was more than 10 times higher than the 50 mg/100 g reported to inhibit non-haem iron absorption in plant foods (Hunt 2003). Even though baobab contained high levels of total phenolics and tannins (Table 2), their potential negative effect on iron bioaccessibility was presumably counteracted by its high levels of organic acids.

Concerning zinc bioaccessibility, the only significant (p < 0.05) improvement compared to millet porridge alone was with citric acid inclusion and this was only small (17%) (Table 4). The high phytate:zinc molar ratios of all the porridge formulations (19.0–21:1) could explain this effect as the phytate:zinc ratios were above the critical values (≥10–14:1) at which zinc bioavailability is impaired (Hunt, 2003). Furthermore, the tannins in baobab could have also limited zinc bioaccessibility in the millet+baobab porridge. Concerning the millet+moringa porridge, this showed a significant reduction in both percentage and amount of bioaccessible zinc, by 22% and 24%, respectively, possibly due to the lowish levels of organic acids and high levels of total phenolics and calcium in moringa. Calcium becomes highly inhibitory in plant foods like millet porridge which contain high levels of phytate through forming strong insoluble complexes with phytate and zinc, consequently reducing zinc bioavailability (Lönnerdal, 2000).

Table 5 shows that only millet porridge+ascorbic acid and millet porridge+citric acid significantly increased the percentage calcium bioaccessibility, by 18% and 56% respectively. The formulations containing baobab and moringa decreased the percentage calcium bioaccessibility, presumably due to their high content of phenolics. However, there was an appreciable increase of amount of bioaccessible calcium in the composite porridges containing baobab and moringa, which is likely due to their high calcium contents (Table 1).

Percentage and amount of bioaccessible magnesium was only improved in the millet+baobab and millet+citric acid porridges, with percentage bioaccessibility increases of 12% and 25% and amounts by 22% and 25%, respectively (Table 5). This indicates that the citric acid in baobab was also promoting magnesium bioaccessibility. Moringa addition to millet porridge also appreciably increased the amount of bioaccessible magnesium, due to its high magnesium content (Table 1).

4 Conclusions

Inclusion of baobab fruit powder, like citric acid, improves the percentage and amount of bioaccessible iron in pearl millet porridge. However, inclusion of moringa leaves has a negative effect on iron bioaccessibility. Inclusion of baobab, like citric acid, also improves magnesium bioaccessibility. However, only citric acid improved zinc bioaccessibility. The positive effect of baobab is probably primarily due to its high contents of citric acid and ascorbic acid, despite its high levels of phenolics especially tannins. The negative effect of moringa is probably due to its lower levels of organic acids and its high levels of phenolics and particularly of calcium.

Food-to-food fortification of cereal porridge with baobab fruit seems to be nearly as effective as citric acid addition in enhancing bioaccessibility of iron and magnesium. As baobab fruit is a very popular food in semi-arid tropical Africa, its inclusion in staple cereal food products could be a useful alternative or complement to conventional fortification in rural communities.

Competing interests

The authors declare no competing interests.

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Mineral contents (mg/100 g dry basis) of pearl millet porridge, baobab fruit powder, moringa leaf powder and their composites, and their estimated percentage nutrient contribution of an average porridge portion size (100 g dry weight basis) to the daily Recommended Nutrient Intake (RNI) for women of reproductive age.

| Plant foods and porridge formulations (dry mass ratios) | Fe | Zn | Ca | Mg | P | Al | Percentage contribution of Fe to RNI (29.4 mg/day) ³ | Percentage contribution of Zn to RNI (9.8 mg/day) ⁵ | Percentage contribution of Ca to RNI (1000 mg/day) | Percentage contribution of Mg to RNI (220 mg/day) |
|---|-----------------------------|-------------------------|-----------------------|----------------------|---------------------------|--------------------|---|--|--|---|
| Millet porridge | $9.0^{\circ} \pm 0.1^{1,2}$ | $6.6^{d} \pm 0.1$ | $20^a \pm 0$ | $137^a \pm 2$ | 306 ^{bcd} ± 4 | $3.1^{a} \pm 0.3$ | 30.6 | 67.3 | 2 | 62.3 |
| Baobab | $3.7^{a} \pm 0.2$ | $0.9^{a} \pm 0.0$ | 470 ^b ± 11 | 259 ^b ± | $50^a \pm 1$ | $3.1^{a} \pm 0.3$ | Not applicable | Not applicable | Not applicable | Not applicable |
| Moringa | 19.1 ^f ± 0.1 | $2.0^{b} \pm 0.0$ | 4252° ± 72 | 896° ± 28 | $330^{cd} \pm 3$ | $12.8^{b} \pm 0.1$ | Not applicable | Not applicable | Not applicable | Not applicable |
| Millet porridge+Baobab (100:15) | $8.6^{b} \pm 0.0$ | $6.3^{cd} \pm 0.2$ | $72^a \pm 1$ | 150 ^a ± | 282 ^b ± | $3.6^{a} \pm 0.3$ | 29.3 (35.1 ⁴) | 64.3 | 7.2 | 68.2 |
| Millet porridge+Moringa (100:15) | 10.7° ± 0.6 | 6.4 ^{cd} ± 0.4 | 547 ^b ± 14 | 227 ^b ± 5 | 318 ^{cd} ± 11 | $4.2^{a} \pm 0.3$ | 36.4 | 65.3 | 54.7 | 103.2 |
| Millet porridge+Baobab+Moringa | $9.9^{d} \pm 0.0$ | 5.9° ± 0.1 | 547 ^b ± | 231 ^b ± | 293 ^{bc} ± | 4.5° ± 0.8 | 33.7 | 60.2 | 54.7 | 105 |

(100:15:15)

| Millet porridge+Ascorbic acid ⁶ | 9.0 | 6.6 | 20 | 137 | 306 | 3.1 | 30.6 (36.74) | 67.3 | 2 | 62.3 |
|--|-----|-----|----|-----|-----|-----|--------------|------|---|------|
| Millet porridge+Citric acid ⁶ | 9.0 | 6.6 | 20 | 137 | 306 | 3.1 | 30.6 (36.74) | 67.3 | 2 | 62.3 |

 $^{^{1}}$ Values are the means \pm 1 SD of at least two samples of each plant food analysed independently in duplicate (n = 4),

 $^{^{2}}$ Means with different superscripts letters in a column differ significantly (p < 0.05).

³RNI at 10% iron bioavailability; ⁴Percentage contribution of iron to the RNI at 12% bioavailability (numbers in brackets); ⁵RNI at low (15%) zinc bioavailability (FAO & WHO, 2001)

⁶Quantities of ascorbic acid and citric acid used were equivalent to the levels in the baobab fruit powder

Table 2

Total phenolic, tannin and phytate contents of pearl millet porridge, baobab fruit powder and moringa leaf powder (mg/100 g dry basis), and their phytate:iron molar ratios.

| Plant foods and porridge formulations (dry mass ratios) | Total phenolics | Tannins ² | Phytate | Phytate:iron molar ratio |
|---|----------------------|----------------------|------------------------|--------------------------|
| Millet porridge | $278^a \pm 42^{1,3}$ | Not detected | 1417 ^b ± 65 | 13:1 |
| Baobab | $2560^{b} \pm 156$ | 8353 ± 201 | $254^a \pm 1$ | 5.8 |
| Moringa | $2650^{b} \pm 42$ | 37 ± 1 | 268 ^a ± 11 | 1.2 |
| Millet porridge+Baobab (100:15) ⁴ | 573 | 1090 | 1265 | 12:1 |
| Millet porridge+Moringa (100:15) ⁴ | 587 | 5 | 1267 | 10:1 |
| Millet porridge+Baobab+Moringa (100:15:15) ⁴ | 813 | 968 | 1150 | 10:1 |

 $^{^{1}}$ Values are the means \pm 1 SD of at least two samples of each plant food analysed independently in duplicate (n = 4),

 $^{^2}$ Tannin content values are the mean \pm 1 SD of four samples of each food analysed independently in duplicate (n = 8)

 $^{^{3}}$ Means with different superscripts letters in a column differ significantly (p < 0.05).

⁴Total phenolics, tannins and phytate values of porridge formulations were calculated from the individual plant food values

Organic acid contents (mg/100 g dry basis) of pearl millet porridge, baobab fruit powder and moringa leaf powder and their composites and the organic acid:iron molar ratios.

| Plant food and porridge formulations (dry mass ratios) | Ascorbic acid | Citric acid | Tartaric acid | Malic acid | Ascorbic acid:iron molar ratio | Citric acid:iron molar ratio |
|---|----------------------|-------------------------|-----------------------|----------------------|--------------------------------------|------------------------------------|
| Millet porridge | Not detected | $150^{a} \pm 9^{1,2}$ | 713 ^b ± 66 | 194 ^a ± 9 | 0.0:1 | 5:1 |
| Baobab | 140 ^b ± 7 | $3345^{c} \pm 63$ | Not detected | 210 ^b ± 9 | 12.0:1 | 263:1 |
| Moringa | $10^{a} \pm 1$ | 2017 ^b ± 113 | $228^a \pm 26$ | Not detected | 0.2:1 | 31:1 |
| Millet porridge+Baobab (100:15) ³ | 18 | 567 | 620 | 196 | 0.7:1 | 19:1 |
| Millet porridge+Moringa (100:15) ³ | 1 | 393 | 650 | 169 | 0.0:1 | 11:1 |
| Millet porridge+Baobab+Moringa (100:15:15) ³ | 17 | 734 | 575 | 173 | 0.6:1 | 22:1 |

 $^{^{1}}$ Values are the means \pm 1 SD of at least two samples of each plant food analysed independently in duplicate (n = 4),

 $^{^{2}}$ Means with different superscripts letters in a column differ significantly (p < 0.05).

³Organic acid values of porridge formulations were calculated from the individual plant food values

Effects of adding baobab fruit powder and moringa leaf powder alone, and in combination, to pearl millet porridge on percentage and total iron and zinc bioaccessibilities of the porridges.

| Porridge formulation (dry mass ratios) | Percentage bioaccessible iron | Amount of bioaccessible iron (mg/100 g, dry basis) | Percentage bioaccessible zinc | Amount of bioaccessible zinc (mg/100 g, dry basis) |
|--|-------------------------------|--|---------------------------------|--|
| Millet Porridge | $6.66^{ab} \pm 0.47^{1,2}$ | $0.60^{a} \pm 0.04$ | $30.5^{bc} \pm 3.9$ | $2.01^{bc} \pm 0.26$ |
| Millet porridge+Baobab (100:15) | $9.05^{cd} \pm 1.94 (36\%)^3$ | $0.77^{bc} \pm 0.17 (30\%)$ | $33.1^{\circ} \pm 3.9$ | $2.07^{bc} \pm 0.24$ |
| Millet porridge+Moringa (100:15) | $5.00^{a} \pm 0.28 \ (-24\%)$ | $0.53^{a} \pm 0.03$ | $23.8^{a} \pm 3.8 \ (-22\%)$ | 1.52 ^a ±0.24 (-24%) |
| Millet porridge+Baobab+Moringa (100:15:15) | $6.07^{ab} \pm 0.44$ | $0.60^{a} \pm 0.04$ | $31.7^{bc} \pm 2.2$ | $1.87^{ab} \pm 0.13$ |
| Millet porridge+Ascorbic acid ⁴ | $7.53^{bc} \pm 0.35$ | $0.68^{ab}\pm0.03$ | $27.5^{ab} \pm 2.7$ | $1.81^{ab} \pm 0.18$ |
| Millet porridge+Citric acid ⁴ | $10.11^{d} \pm 1.40 (52\%)$ | $0.91^{\circ} \pm 0.13 (52\%)$ | $35.8^{\circ} \pm 1.8 \ (17\%)$ | $2.36^{\circ} \pm 0.12 (17\%)$ |

 $^{^{1}}$ Values are the means \pm 1 SD of two completely independent dialysability experiments, with the intestinal step being each time performed in triplicate (n = 6).

 $^{^{2}}$ Means with different superscripts letters in a column differ significantly (p < 0.05)

 $^{^{3}}$ Percentage difference in brackets is significantly different when compared to millet porridge alone using Fisher's LSD test (p < 0.05)

⁴Quantities of ascorbic acid and citric acid used were equivalent to the levels in the baobab fruit powder

Effects of adding baobab fruit powder and moringa leaf powder alone, and in combination, to pearl millet porridge on percentage and total calcium and magnesium bioaccessibilities.

| Porridge formulations (dry mass ratios) | Percentage bioaccessible calcium | Amount of bioaccessible calcium (mg/100 g, dry basis) | Percentage bioaccessible magnesium | Amount of bioaccessible magnesium (mg/100 g, dry basis) |
|--|----------------------------------|---|------------------------------------|---|
| Millet porridge | $23.3^{\circ} \pm 1.6^{1,2}$ | $4.6^{a} \pm 0.3$ | $35.5^{a} \pm 1.0$ | $48.7^{a} \pm 1.4$ |
| Millet porridge+Baobab (100:15) | $20.1^{bc} \pm 3.3 (-14\%)^3$ | $14.6^{b} \pm 2.4 \ (220\%)$ | $39.6^{b} \pm 2.6 (12\%)$ | $59.4^{b} \pm 3.8 \ (22\%)$ |
| Millet porridge+Moringa (100:15) | $14.5^{a} \pm 0.5 \ (-37\%)$ | $79.3^{\circ} \pm 3.0 \ (1641\%)$ | $36.1^a \pm 1.1$ | $82.3^{\circ} \pm 2.5 \ (69\%)$ |
| Millet porridge+Baobab+Moringa (100:15:15) | 17.2 ^{ab} ± 1.7 (-26%) | $94.0^{d} \pm 9.0 \ (1967\%)$ | $37.2^{ab} \pm 3.1$ | $86.1^{\circ} \pm 7.1 \ (77\%)$ |
| Millet porridge+Ascorbic acid ⁴ | $27.4^{\rm d} \pm 1.9 (18\%)$ | $5.4^{a} \pm 0.4$ | $37.5^{ab} \pm 1.1$ | $51.5^{a} \pm 1.5$ |
| Millet porridge+Citric acid ⁴ | $36.4^{\rm e} \pm 2.2 \ (56\%)$ | $7.1^{a} \pm 0.4$ | 44.4°± 1.6 (25%) | $60.9^{b} \pm 2.3 \ (25\%)$ |

 $^{^{1}}$ Values are the means \pm 1 SD of two completely independent dialysability experiments, with the intestinal step being each time performed in triplicate (n = 6).

 $^{^{2}}$ Means with different superscripts letters in a column differ significantly (p < 0.05)

 $^{^{3}}$ Percentage difference in brackets is significantly different when compared to millet porridge alone using Fisher's LSD test (p < 0.05)

⁴Quantities of ascorbic acid and citric acid used were equivalent to the levels in the baobab fruit powder