

Effects of processing and addition of a cowpea leaf relish on the iron and zinc nutritive value of a ready-to-eat sorghum-cowpea porridge aimed at young children

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Abbreviations: AR –absolute requirements; CE – catechin equivalent; db – dry basis; ESMC – extruded sorghum-micronised cowpea; HTST – high-temperature short-time; ICP-OES - inductively coupled plasma - optical emission spectrometry; RDA – recommended dietary allowance; RTE – ready-to-eat

Highlights

- HTST processing of sorghum and cowpeas produced a composite (70:30) RTE porridge.
- Adding cowpea leaves to the porridge greatly increased mineral nutritive value.
- A portion with cowpea leaves can provide up to 85% of iron RDA of young children.
- Importantly, cowpea leaves increase the bioaccessibility of zinc in the meal.

ABSTRACT

While dietary diversification of monotonous cereal-based diets using legumes and vegetables can alleviate the high prevalence of iron and zinc deficiencies in sub-Saharan African children, laborious cooking times limit the use of particularly legumes.

This study investigated the effects of high-temperature short-time (HTST) processing on sorghum (extrusion) and cowpea (micronisation), compositing sorghum-cowpea (70:30) (ESMC) in a ready-to-eat porridge and addition of cowpea leaves on iron and zinc bioaccessibilities compared to a commercial fortified maize:soy ready-to-eat porridge.

HTST processing increased iron bioaccessibility from both grains and the zinc bioaccessibility from the sorghum. One serving of ESMC porridge with cowpea leaves could contribute ≈ 85 and 18% towards the iron and zinc RDA of preschool children, compared to the commercial product at ≈ 84 and 125%, respectively. However, the higher iron and zinc bioaccessibilities from the ESMC porridge with cowpea leaves, compared to the commercial product (11.8 vs. 5.0% and 18.9 vs 2.7%, respectively) means it would provide more bioaccessible iron (2.24 vs. 0.86 mg/100 g, db) and similar levels of zinc (0.35 vs. 0.32 mg/100 g) towards the absolute/basal requirements of preschool children.

The ESMC porridge with cowpea leaves could improve the iron and zinc nutritive value of preschool sub-Saharan African children's diets.

Keywords: iron, zinc, cowpea, sorghum, ready-to-eat

1. Introduction

Globally, Africa has the highest estimated prevalence of anaemia in preschool aged (0-5 years) children at 64.6% (McLean, Cogswell, Egli, Wojdyla, & De Benoist, 2009). While

information on the prevalence of zinc deficiency is limited, it is believed that where iron deficiency persists, zinc deficiency is very likely to also occur (Ramakrishnan, 2002).

Many households in Africa, where iron and zinc deficiencies are prevalent, depend on monotonous cereal-based diets for micronutrients as well as energy (Oniango, Mutuku, & Malaba, 2003). These diets often contain high levels of antinutrients such as phytate and sometimes tannins, which reduce the already low bioavailability of the non-haem iron and zinc in the diet (Hunt, 2003). Legumes, often used to increase the protein nutritive value of the cereal based diets, have also been found to increase mineral nutritive value (Anigo, Ameh, Ibrahim, & Danbauchi, 2009).

Legume preparation, however, is time consuming and laborious, which consequently limits their use (dos Santos Siqueira, Vianello, Fernandes, & Bassinello, 2013). With the increase in urbanization and more women working outside the home, even in the low socioeconomic and/or rural areas (Tacoli, 2012), there is an increased need for affordable, culturally acceptable and convenient foods (Kennedy, Nantel, & Shetty, 2004).

The use of underutilised crops is increasing as their economic potential is realised (Gruère, Giuliani, & Smale, 2006). Underutilised crops are commercialised for various reasons including; providing culturally acceptable options, increasing commercial opportunities for local farmers, lowering production costs and/or increasing the nutritive value of the product.

Information on mineral bioavailability from processed indigenous foods, however, is still severely lacking. This is especially important in areas where mineral deficiencies are often prevalent, but interventions such as supplementation and fortification are impractical.

Sorghum and cowpeas are very important as traditional staple foods in sub-Saharan Africa, as shown by Oyarekua (2010). In a previous study with the aim of providing a high protein

quality product, we explored the potential of using sorghum and cowpeas to develop a ready-to-eat (RTE) composite porridge as a complementary food for preschool aged children (2-5 years old) (Vilakati, MacIntyre, Oelofse, & Taylor, 2015). It was found that a single serving of the RTE porridge, with a cowpea leaf relish, could meet 40% of the children's protein and lysine requirements.

In the current study the effect of high-temperature short-time (HTST) processing technologies on sorghum (extrusion cooking) and cowpea (micronisation), their compositing (70:30) and addition of a cooked cowpea leaf relish on the iron, zinc and phytate contents and iron and zinc bioaccessibilities was evaluated. The iron and zinc nutritive value of the extruded sorghum (ES)-micronised cowpea (MC) (ESMC) porridge, with or without cooked cowpea leaf relish, was also compared with a commercial fortified maize: soy RTE porridge.

2 Materials and Methods

2.1 Raw and processed materials

The sample acquisition, preparation and the formulation of the composite meals have been described in previous work (Vilakati et al., 2015). In short, red non-tannin sorghum (cultivar MR Buster) grains were decorticated, milled and extruded in a TX 32 twin-screw, co-rotating extruder (CFAM Technologies, Potchefstroom, South Africa). Cowpeas (cultivar Bechuana white) were pre-conditioned to 41% moisture, manually dehulled, micronised (Techni lamp, Johannesburg, South Africa) and then milled. Young cowpea leaves were handpicked, cleaned and boiled, as has been described for amaranth (Faber, Van Jaarsveld, Wenhold, & Van Rensburg, 2010). A commercial maize:soy RTE composite porridge (FUTURELIFE®, Durban, South Africa) formulated for children aged 1-4 years was used. The commercial product had also been fortified with multiple micronutrients, including iron, zinc and

calcium to levels of 30, 15 and 800 mg/100 g, dry basis (db), respectively (information provided on the packaging).

2.2 Analyses

2.2.1 Phytate content

Phytate content was determined using an indirect quantitative anion exchange chromatography method described by Frubeck, Alonso, Marzo, & Santidrian (1995). Glass barrel Econo-columns, 0.7 x 15 cm (BioRad, Johannesburg, South Africa), Dowex 1; anion-exchange resin-AG 1 x 4, 4% cross-linkage, chloride form, 100-200 mesh (Sigma, Johannesburg, South Africa) were used.

2.2.2 In vitro iron and zinc bioaccessibilities

Iron and zinc bioaccessibilities were determined using a dialysis method described by Luten et al. (1996) and modified by Kruger et al. (2012). In short, simulated gastric digestion was performed at pH 2 using porcine pepsin (P-700) (Sigma, Johannesburg, South Africa). Intestinal digestion was simulated at pH 7 using porcine pancreatin (P-1750), bile extract (B-8631) (Sigma, Johannesburg, South Africa) and dialysis tubing (Spectra/Por 7 (\varnothing = 20.4 mm), molecular weight cut-off of 10kDa G.I.C. Scientific, Johannesburg, South Africa). The dialysate was decanted and acidified using concentrated nitric acid to ensure that the minerals did not adsorb to the sides of the container and/or not precipitate out of solution. The iron and zinc that passed through the dialysis tubing was measured as bioaccessible. The assay is based on the theory that smaller, soluble iron and zinc compounds are better absorbed than large compounds (Fairweather-Tait et al., 2005). The gastric digestion was done in duplicate and from each gastric digestion 3 intestinal digestions were done (n=6).

Results are displayed as both the amount of bioaccessible iron and zinc (mg/ 100 g) as well as the percentage (%) of bioaccessible iron and zinc relative to respective total contents.

2.2.3 Mineral analysis

The total iron, calcium and zinc contents of the raw and processed flours, digested samples (dialysates) and blanks were determined using inductively coupled plasma optical emission spectrometry (ICP-OES), (SPECTRO ARCO, Spectro Analytical Instruments, Kleve, Germany) with a dual-view torch, spray chamber and cross-flow nebulizer. Multi-element standard solutions were prepared by dilution of stock solutions with deionized water (1000 mg/l Merck, Darmstadt, Germany) and a range of calibration standards to match expected concentration for Ca, Fe and Zn in samples were used (Operation conditions of the SPECTRO ARCO are given in Table 1). Prior to analysis of the raw undigested samples, acid assisted

Table 1: ICP-OES working conditions

Parameters	Units
RF Power	1400
Coolant flow rate (L/min)	12.0
Nebulizer flow rate (L/min)	1.00
Auxiliary flow rate (L/min)	1.00
Pump speed (rpm)	30
Rinse time (s)	30
Replicate read time (s)	15
Element	Emission line (nm)
Ca	317.9
Fe	238.2
Zn	213.9

microwave digestion was performed using ultrapure nitric acid (65%, Merck, Darmstadt, Germany) and 2 mL hydrogen peroxide (30%, Merck, Darmstadt, Germany). The iron and zinc contents of the flour samples were measured in triplicates and each dialysate (n=6) measured once.

2.2.4 Ascorbic acid determination

Ascorbic acid was determined only in the cowpea leaf relish using the method described by Nielsen (2010).

2.2.5 Statistical analysis

Data was analysed by single factor analysis of variance (ANOVA) using STATISTICA 10 (StatSoft, Johannesburg, South Africa). Fisher's LSD Post-hoc test was applied to determine significant differences between specific means at a confidence level of 95% ($p \leq 0.05$).

3 Results and Discussion

3.1 Mineral and phytate contents

Compositing the raw and HTST treated sorghum and cowpea substantially ($p \leq 0.05$) increased the iron, zinc and calcium contents of both the raw and HTST processed composites by 47, 83, 59 and 67, 180, 410%, respectively, compared to the sorghum flours (Table 2). Adding the cooked cowpea leaf relish to the extruded sorghum-micronised cowpea (ESMC) RTE porridge greatly ($p \leq 0.05$) increased the iron and calcium contents, four and five folds, respectively, but did not affect the zinc content. Importantly, the iron content

Table 2: The effects of high-temperature short-time (HTST) processing of sorghum and cowpea and their compositing on the iron, zinc, calcium, phytate contents (mg/100 g db) and the respective phytate: mineral molar ratios

	Iron (n=3)*	[Phytate:F e]	Zinc (n=3)	[Phytate: Zn]	Calcium (n=3)	[Phytate x Ca:Zn]	Phytate (n=4)
<i>Raw and processed sorghum, cowpea and their composites</i>							
Decorticated sorghum	1.7 ^a (0.0)	[47]	0.6 ^a (0.2)	[154]	17 ^b (1)	[65]	935 ^{ab} (133)
Dehulled cowpea	4.4 ^d (0.1)	[23]	2.6 ^d (0.0)	[46]	48 ^d (0)	[56]	1220 ^c (64)
Sorghum:cowpea*	2.5 ^b (0.1)	[33]	1.1 ^b (0.1)	[87]	27 ^c (1)	[59]	971 ^{ab} (186)
Extruded sorghum (ES)	3.0 ^b (0.4)	[25]	1.0 ^b (0.0)	[89]	10 ^a (2)	[22]	901 ^a (50)
Micronised cowpea (MC)	5.0 ^e (0.3)	[21]	2.8 ^d (0.2)	[44]	51 ^d (2)	[55]	1210 ^c (106)
ESMC ready-to-eat (RTE) porridge*	3.7^{cw} (0.3)	[24]	1.5^{cw} (0.3)	[68]	22^{bw} (2)	[37]	1022^{bw}(91)
<i>Cowpea relish and other composite RTE porridges</i>							
Cowpea leaf relish	49.6^Z (3.2)	[2]	2.4^X (0.1)	[54]	374^Y (26)	[508]	1319^Y(102)
ESMC RTE porridge and cowpea leaves**	19.0^X (1.1)	[6]	1.8^W (0.2)	[67]	126^X (9)	[210]	1101^X(91)
Commercial maize: soy RTE porridge	18.7^X (1.9)	[4]	12.9^Y (0.9)	[7]	602^Z (57)	[113]	976^W(122)

Values are means with 1 standard deviation in parentheses. Mineral analyses were performed on 3 individual samples (n=3); phytate analysis was performed on two individual samples in duplicate (n = 4). ^{abc} - Mean values of raw and processed sorghum and/or cowpea flours within a column with different superscripts differ significantly (p≤0.05). ^{wxy}-Mean values of different composite dishes (**in bold**) within a column with different superscripts differ significantly (p≤0.05). *Composite ratio of cereal: legume, 70:30 (db), ** composition ratio of extruded sorghum: micronised cowpea: cooked cowpea leaves, 70:30:50 (db). ES -extruded sorghum, MC-Micronised cowpea and ESMC RTE – extruded sorghum-micronised cowpea ready-to-eat.

was increased to 19.0 mg/100 g, db, the same level as the commercial fortified maize:soy RTE porridge.

After micronisation and extrusion cooking, the cowpea and sorghum flours had significantly higher iron and zinc contents, respectively. These increases were possibly due to some iron and/or zinc contamination, occurring during the processing. The increase in iron (76%) and zinc (67%) contents after extrusion was probably due to the use of tap water, which can contain significant levels of minerals, and abrasion of the ferrous extruder screw and barrel, as was also reported in a study by Mutambuka (2013).

While the phytate contents of the raw and micronised cowpea were significantly higher than those of the raw and extruded sorghum, only compositing of HTST processed grains resulted in significantly increased ($p \leq 0.05$) phytate content (13%), compared to the extruded sorghum alone.

3.2 Mineral bioaccessibilities from the raw and processed products

The amount of bioaccessible mineral (mg /100 g food product) together with the mineral bioaccessibility (percentage of total mineral) of a specific mineral gives a good indication of the sum of the effects of the mineral bioaccessibility inhibitors and enhancers when the total mineral contents differ substantially (Kruger, Mongwaketse, Faber, van der Hoeven, & Smuts , 2015).

Despite the higher bioaccessibility of iron from cowpea flour, compositing with sorghum flour did not increase the iron bioaccessibility ($p > 0.05$) from the raw composite (sorghum 20.7% vs. composite 19.5%) or extruded sorghum (ES)-micronised cowpea (MC) ESMC RTE porridge (extruded sorghum 32.4% vs. composite 32.3%) (Table 3). This is probably because

Table 3: The effect of high-temperature short-time (HTST) processing of sorghum and cowpea and their compositing on the percentage iron and zinc bioaccessibilities (% of total iron) and amount of bioaccessible iron and zinc (mg bioaccessible iron/100 g) assessed by a dialysability assay.

	Iron bioaccessibility		Zinc bioaccessibility	
	% of total iron	mg/100 g, db	% of total zinc	mg/100 g, db
	(n=6)		(n=6)	
<i>Raw and processed sorghum, cowpea and their composites</i>				
Decorticated sorghum	20.7 ^a (3.5)	0.31 ^a (0.05)	18.2 ^c (5.7)	0.11 ^a (0.03)
Dehulled cowpea	25.2 ^{ab} (1.8)	1.28 ^c (0.09)	12.8 ^{ab} (1.7)	0.37 ^{cd} (0.05)
Sorghum:cowpea*	19.5 ^a (1.1)	0.44 ^a (0.02)	15.6 ^{bc} (3.9)	0.17 ^b (0.05)
Extruded sorghum (ES)	32.4 ^c (5.9)	0.92 ^b (0.17)	31.3 ^d (6.0)	0.30 ^c (0.06)
Micronised cowpea (MC)	40.4 ^d (4.4)	1.95 ^d (0.21)	13.5 ^{ab} (2.0)	0.36 ^d (0.05)
ESMC ready-to-eat (RTE) porridge*	32.3^{cY} (3.6)	1.05^{bW}(0.10)	11.2^X (0.8)	0.20^{bW}(0.01)
<i>Cowpea relish and other composite RTE porridges</i>				
Cowpea leaf relish	6.2^{WX} (0.6)	3.08^Y(0.28)	12.9^X (2.0)	0.30^X(0.05)
ESMC RTE porridge and cowpea leaves**	11.8^X (1.2)	2.24^X(0.34)	18.9^Y (3.4)	0.35^X(0.07)
Commercial maize: soy RTE porridge	5.0^W (0.3)	0.86^W(0.05)	2.7^W (0.3)	0.32^X(0.03)

Values are means with 1 standard deviation in parentheses, analyses were performed on two individual samples in triplicate (n = 6). ^{abc}-Mean values of raw and processed sorghum and/or cowpea flours within a column with different superscripts differ significantly (p≤0.05). ^{wxy}-Mean values of different composite dishes (**in bold**) within a column with different superscripts differ significantly (p≤0.05). *Composite ratio of cereal: legume, 70:30 (db), ** composition ratio of extruded sorghum: micronised cowpea: cooked cowpea leaves, 70:30:50 (db). ES -extruded sorghum, MC-Micronised cowpea and ESMC RTE – extruded sorghum-micronised cowpea ready-to-eat

the phytate: iron molar ratios of the composite flour at 33 and the ESMC RTE porridge at 24 (Table 2) were much higher than the critical level of 1, above which iron bioavailability is seriously impaired (Hunt, 2003).

HTST processing substantially increased the iron bioaccessibility (%) from sorghum (32.4% vs. 20.7%) and cowpea (40.4% vs. 25.2%) (Table 3). However, while the phytate: iron molar ratios were somewhat reduced (21-25) (Table 2), they were still far above the critical level of 1. The increased iron bioaccessibility (%) was probably due to the previously observed increased water absorption index (WAI) and water solubility index (WSI) of the RTE porridge (Vilakati et al., 2015). It was found that extrusion and micronisation increased the WAI and WSI by approximately 40 and 60% in sorghum and 50 and 60% in cowpea respectively, possibly resulting in more minerals solubilising from the food matrix.

The iron bioaccessibilities from the cowpea leaf relish (6.2 %) and the commercial fortified maize: soy RTE porridge (5.0%) were substantially lower than those of the sorghum and cowpea grains. This was despite the fact that the cowpea relish was found to contain 42 mg ascorbic acid/100 g (db) (data not tabulated), a known iron bioavailability enhancer (Teucher, Olivares, & Cori, 2004) and having a much lower phytate: iron molar ratio (2 vs. 21-47). The low iron bioaccessibility in the cooked cowpea leaf relish was probably due to its high tannin content (219 mg/100 g, db) (Vilakati et al., 2015). Tannins have been found to be extremely potent iron bioavailability inhibitors (Santos-Buelga, & Scalbert, 2000). However, despite the low percentage iron bioaccessibility from the cooked cowpea leaf relish (Table 3), the amount of bioaccessible iron was high (3.08 mg/100 g, db), because of the high total iron content (49.6 mg/100 g db) (Table 2). For the commercial fortified maize: soy RTE porridge it is possible that its high calcium content (602 mg/100 g db) could have decreased the iron bioaccessibility, as founded by Roughead, Zito, & Hunt (2005).

HTST processing of sorghum, but not cowpea, significantly increased the percentage and amount of bioaccessible zinc (Table 3). Alonso, Rubio, Muzquiz, & Marzo (2001) in a rat model study, observed an increase in iron absorption after extrusion of peas and kidney beans, but also did not observe any increased zinc absorption. The increase in zinc bioaccessibility from sorghum resulting from extrusion cooking may have been because of the two and three fold decrease in the phytate: zinc and phytate x calcium: zinc ratios (Table 2), respectively. HTST processing had no effect on the phytate: zinc and phytate x calcium: zinc ratios of cowpea.

Percentage zinc bioaccessibility from the commercial fortified maize: soy RTE porridge was very low, despite the low phytate: zinc molar ratio of 7, well below the critical range of 10-14, above which the bioavailability is seriously impaired (Hunt, 2003). This was probably due to the addition of soy isolate to the product, which has been found to inhibit zinc bioavailability (Lönnerdal, 2000).

3.3 Comparison of iron and zinc bioaccessibilities from the different ready-to-eat (RTE) porridge meals

The ESMC RTE porridge had substantially higher ($p \leq 0.05$) iron bioaccessibility (32.3% vs. 5.0%) compared to the commercial fortified maize: soy RTE porridge. This resulted in both porridges having similar amounts ($p > 0.05$) of bioaccessible iron (1.05 and 0.86 mg/100g db), despite the substantially higher total ($p \leq 0.05$) iron content of the commercial fortified maize: soy RTE porridge compared to the ESMC RTE porridge (18.7 vs. 3.7 mg/100 g) (Table 2). Similarly with zinc, the ESMC RTE porridge had much ($p \leq 0.05$) higher zinc bioaccessibility (11.2%) compared to the commercial fortified maize: soy RTE porridge (2.7%). The amount of bioaccessible zinc from the ESMC RTE porridge (0.20 mg/100 g db) was, however, 38%

lower than that from the commercial fortified maize: soy RTE porridge, due to the much higher zinc content of the commercial fortified maize: soy RTE composite porridge (12.9 vs. 1.5 mg/100 g db) (Table 2).

The effect of adding the cooked cowpea leaf relish to the ESMC RTE porridge on the iron and zinc bioaccessibilities varied. While the bioaccessibility of the iron decreased from 32.3 to 11.8%, the amount of bioaccessible iron doubled from 1.05 to 2.24 mg/100g, db (Table 3). Importantly, both the percentage and amount of bioaccessible iron from the ESMC RTE porridge with cooked cowpea leaves was more than double that from the commercial fortified maize: soy RTE porridge. Both the percentage and amount of bioaccessible zinc increased substantially ($p \leq 0.05$) with the addition of the cooked cowpea leaf relish. The zinc bioaccessibility (%) from the ESMC RTE porridge with cooked cowpea leaf relish was seven-fold higher than that from the commercial fortified maize: soy RTE porridge, and despite the much higher zinc content of the commercial fortified maize: soy RTE porridge, the amounts of bioaccessible zinc were similar (0.35 vs. 0.32 mg/100 g) ($p > 0.05$).

3.4 Estimated contributions of the porridges to iron and zinc requirements of preschool aged children

The contribution that a recommended portion (NICUS, 2003) of the ES, ESMC porridge, without and with cowpea leaf relish, and the commercial fortified maize: soy RTE porridges could make towards the iron and zinc recommended dietary allowances (RDA) of preschool children aged 2-3 (100 g portion) and 4-5 years (125 g portion) were calculated. Each porridge (with and without cowpea leaves) were reconstituted to 33% solid content. The RDA of children aged 2-3 and 4-5 years for iron and zinc is 7 and 10 mg/day and 3 and 5 mg/day, respectively (Institute of Medicine (IOM), 2001). Compositing the extruded

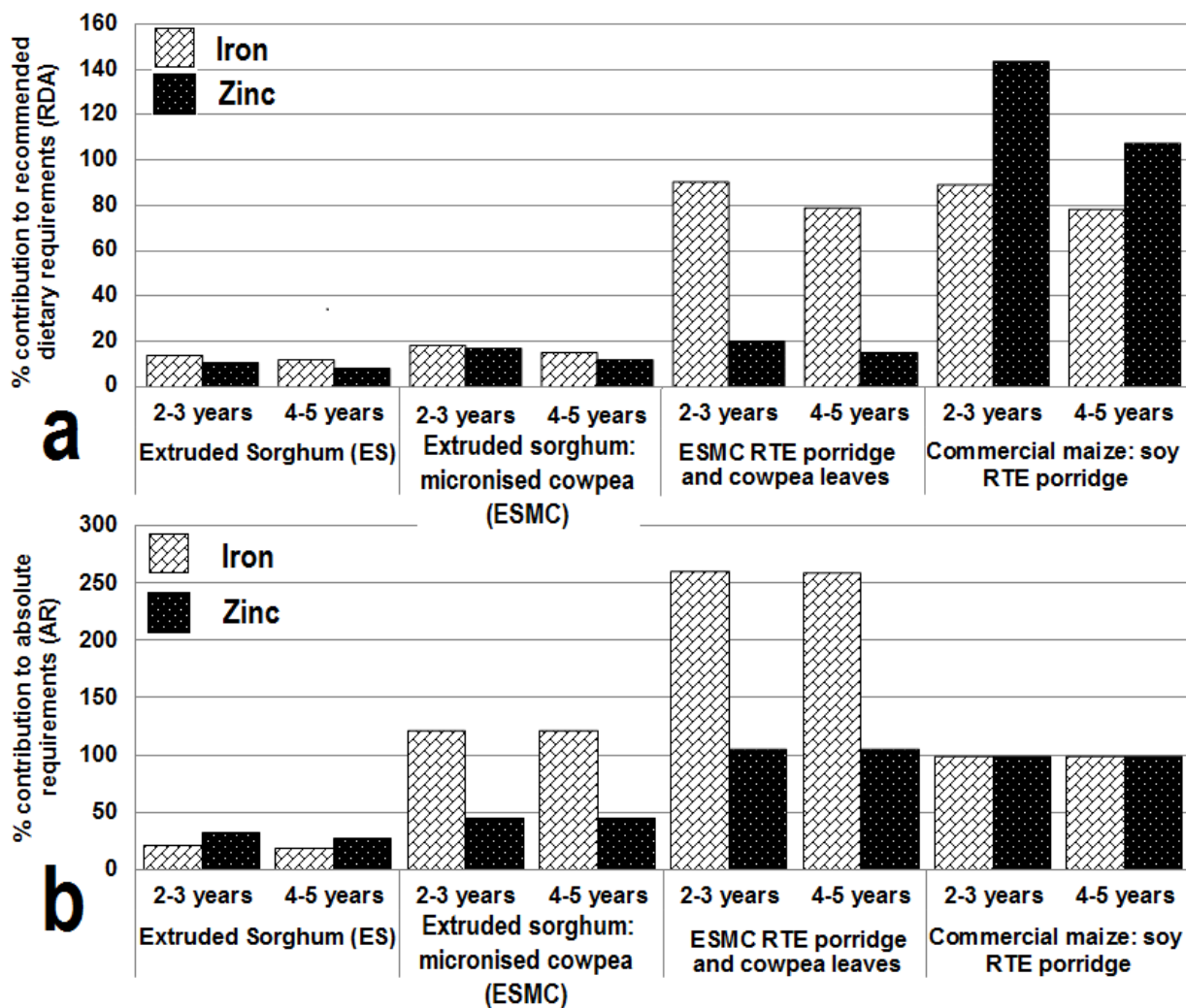


Figure 1: The contribution that 100 and 125 g* (as consumed) ready-to-eat (RTE) porridge (reconstituted to 33% solid content) can make towards (a) the recommended dietary allowance (RDA)** and (b) absolute requirements (AR)*** of pre-school children aged 2-3 and 4-5 year old, respectively.

*Recommended portion size per serving as consumed is according to the Nutrition Information Centre of the University of Stellenbosch (NICUS) (2003). **RDA for iron and zinc: 7 and 3 mg/day for children aged 2-3 years and 10 and 5 mg/day for those aged 4-5 years, respectively (Institute of medicine (IOM), 2001). ***AR absolute requirements is the amount of bioavailable nutrient that is required to be absorbed and utilized and then the contribution that the amount of bioaccessible iron and zinc (Table 3) in each meal can make towards the that for iron (World Health Organization (WHO), 2008) and zinc (Food and Agriculture Organization/World Health Organisation (FAO/WHO), 2001): 0.46 and 0.92 mg/day for children aged 2-3 years, respectively and 0.50 and 1.09 mg/day for those aged 4-5 years, respectively. The percentage contributions are expressed relative to the contribution of the commercial product which is displayed as 100.

sorghum with the micronised cowpea increased the contribution the RTE porridge could make towards the RDAs of children aged 2-3 and 4-5 years (averaged across age groups) from 13 to 17% for iron and 10 to 15% for zinc, compared to extruded sorghum alone (Figure 1a). Further, inclusion of the cooked cowpea leaf relish with the ESMC RTE porridge increased the contribution to the iron RDA of preschool children to an average of 85%, the same as the commercial fortified maize: soy RTE porridge (84%). The contribution of the commercial fortified maize: soy RTE porridge to the zinc RDA was much higher (125%) compared to the ESMC RTE porridge without or with the cowpea leaf relish, which could only provide on average 15 and 18%, respectively.

When evaluating the contribution that bioaccessible minerals can make towards the absolute/basal requirements (AR), expressed as mg bioavailable iron or zinc/day (Figure 1b), the direction of the effect is more reliable than the magnitude (Fairweather-Tait et al., 2005). For this reason, the percentage contribution that each porridge could make towards the absolute iron and zinc requirements is expressed relative to the commercial fortified maize: soy RTE porridge's contribution ($\% \text{ contribution of RTE porridge} / \% \text{ contribution of commercial fortified maize: soy RTE porridge} \times 100$).

While the extruded sorghum could contribute approximately 20 and 30%, respectively of what the commercial fortified maize: soy RTE porridge could towards the iron and zinc AR, the ESMC RTE porridge could contribute the same as the commercial product towards the iron AR and almost 50% towards the zinc AR. Addition of the cooked cowpea leaf relish doubled the contribution that the ESMC RTE porridge could make towards the absolute iron requirements compared to the commercial fortified maize: soy RTE porridge. Importantly, despite the small contribution the ESMC RTE porridge with cooked cowpea leaf relish could

make towards the zinc RDA ($\approx 18\%$) compared to the commercial fortified maize: soy RTE porridge ($\approx 125\%$), the contribution towards the zinc AR is similar.

4. Conclusion

Despite the lower total iron and zinc contents of the extruded sorghum-micronised cowpea RTE porridge and cooked cowpea leaf relish meal, the bioaccessibility of the intrinsic iron and zinc is much higher compared to a commercial fortified maize: soy RTE product. If the intrinsic iron and zinc contents of a sorghum: cowpea RTE porridge can be increased further, for example through high iron and zinc cultivar selection, it has the potential to have an iron and zinc nutritive value far superior to the commercial fortified maize: soy RTE product.

The ESMC RTE porridge on its own and even more so when consumed as a meal with added cooked cowpea leaf relish, shows it could improve the iron and zinc nutritive value of diets of sub-Saharan African children. Hence, this composite RTE porridge produced from sorghum and cowpea, grains that are staples in Africa, could be a sustainable locally produced or commercial alternative to imported commercial fortified “maize: soy blend” type products for complementary feeding of young children.

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5 References

Alonso, R., Rubio, L. A., Muzquiz, M., & Marzo, F. (2001). The effect of extrusion cooking on mineral bioavailability in pea and kidney bean seed meals. *Animal Feed Science and Technology*, *94*, 1-13.

Anigo, K. M., Ameh, D. A., Ibrahim, S., & Danbauchi, S. S. (2009). Nutrient composition of commonly used complementary foods in North western Nigeria. *African Journal of Biotechnology*, *8*, 4211-4216.

dos Santos Siqueira, B., Vianello, R. P., Fernandes, K. F., & Bassinello, P. Z. (2013). Hardness of carioca beans (*Phaseolus vulgaris* L.) as affected by cooking methods. *LWT-Food Science and Technology*, *54*, 13-17.

Faber, M., Van Jaarsveld, P., Wenhold, F., & Van Rensburg, J. (2010). African leafy vegetables consumed by households in the Limpopo and KwaZulu-Natal provinces in South Africa. *South African Journal of Clinical Nutrition*, *23*, 30-38.

Fairweather-Tait, S., Lynch, S., Hotz, C., Hurrell, R., Abrahamse, L., Beebe, S., Bering, S., Bukhave, K., Glahn, R., Hambidge, M., Hunt, J., Lonnerdal, B., Miller, D., Mohktar, N., Nestel, P., Reddy, M., Sandberg, A-S., Sharp, P., Teucher, B., & Trinidad, P. T. (2005). The usefulness of in vitro models to predict the bioavailability of iron and zinc: A consensus statement from the HarvestPlus expert consultation. *International Journal of Vitamin and Nutritional Research*, *75*, 371-374.

Food and Agriculture Organization/World Health Organisation (FAO/WHO). (2001). Human Vitamin and Mineral Requirements. Rome: FAO <http://www.fao.org/docrep/004/Y2809E/Y2809E00.HTM>.

Accessed 20 August 2015

Frubeck, G., Alonso, R., Marzo, F., & Santidrian, S. (1995). A modified method for the indirect quantitative analysis of phytate in foodstuffs. *Analytical Biochemistry*, 225, 206-212.

Gruère, G. P., Giuliani, A., & Smale, M. (2006). Marketing underutilized plant species for the benefit of the poor: a conceptual framework. *IFPRI Environmental and Protection Technology (EPT) Discussion Paper*, (154). <http://ssrn.com/abstract=916572> or <http://dx.doi.org/10.2139/ssrn.916572>. Accessed 13 August 2015.

Hunt, J. R. (2003). Bioavailability of iron, zinc, and other trace minerals from vegetarian diets. *American Journal of Clinical Nutrition*, 78, 633S-639S.

Institute of Medicine (IOM). (2001). Dietary Reference Intakes for Vitamin A, Vitamin K, Arsenic, Boron, Chromium, Copper, Iodine, Iron, Manganese, Molybdenum, Nickel, Silicon, Vanadium, and Zinc. Washington DC: National Academies Press. <http://www.nap.edu/catalog/10026.html>. Accessed 12 May 2013.

Kennedy, G., Nantel, G., & Shetty, P. (2004). Globalization of food systems in developing countries: a synthesis of country case studies. *Food and Agriculture Organization of the United Nations*, 83, 1-26.

Kruger, J., Mongwaketse, T., Faber, M., van der Hoeven, M., & Smuts, C. M. (2015). Potential contribution of African green leafy vegetables and maize porridge composite meals to iron and zinc nutrition. *Nutrition*, 31, 1117-1123.

Kruger, J., Taylor, J. R. N., & Oelofse, A. (2012). Effects of reducing phytate content in sorghum through genetic modification and fermentation on in vitro iron availability in whole grain porridges. *Food Chemistry*, 131, 220-224.

Lönnerdal, B. (2000). Dietary factors influencing zinc absorption. *Journal of Nutrition*, 130, 1378S-1383S.

Luten, J., Crews, H., Flynn, A., Van Dael, P., Kastenmayer, P., Hurrell, R., Deelstra, H., Shen, L-H., Fairweather-Tait, S., Hickson, K., Farré, R., Schlemmer, U., & Frøhlich, W. (1996). Interlaboratory Trial on the Determination of the In Vitro Iron Dialysability from Food. *Journal of the Science of Food and Agriculture*, 72, 415-424.

McLean, E., Cogswell, M., Egli, I., Wojdyla, D., & De Benoist, B. (2009). Worldwide prevalence of anaemia, WHO vitamin and mineral nutrition information system, 1993–2005. *Public Health Nutrition*, 12, 444-454.

Mutambuka, M. (2013). Iron bioavailability and consumer acceptability of extruded common bean (*Phaseolus vulgaris*) flour. *PhD Thesis*, Iowa State University, Iowa.

Nielsen, S. S. (2010). *Food analysis laboratory manual. Food science texts series*. (16th ed.). New York: Springer, (Chapter 7).

Nutrition Information Centre of the University of Stellenbosch (NICUS). (2003). Dietary reference intakes (DRI's). Compiled by the Nutrition Information Centre of the University of Stellenbosch (NICUS), Cape Town. <http://www.sun.ac.za/english/faculty/healthsciences/nicus/how-to-eat-correctly/nutrients/dri>. Accessed 15 July 2014.

Oniango, R., Mutuku, J., & Malaba, S. J. (2003). Contemporary African food habits and their nutritional and health implications. *Asia Pacific Journal of Clinical Nutrition*, 12, 331-336.

Oyarekua, M. A. (2010). Sensory evaluation, nutritional quality and antinutritional factors of traditionally co-fermented cereals/cowpea mixtures as infant complementary food. *Agriculture and Biology Journal of North America*, 1, 950-956.

Ramakrishnan, U. (2002). Prevalence of micronutrient malnutrition worldwide. *Nutrition Reviews*, 60, S46-S52.

Roughead, Z. K. F., Zito, C. A., & Hunt, J. R. (2005). Inhibitory effects of dietary calcium on the initial uptake and subsequent retention of heme and nonheme iron in humans: comparisons using an intestinal lavage method. *The American Journal of Clinical Nutrition*, *82*, 589-597.

Santos-Buelga, C., & Scalbert, A. (2000). Proanthocyanidins and tannin-like compounds – nature, occurrence, dietary intake and effects on nutrition and health. *Journal of the Science of Food and Agriculture*, *80*, 1094-1117.

Tacoli, C. (2012). Urbanization, gender and urban poverty: paid work and unpaid carework in the city. Human Settlements Group, International Institute for Environment and Development. *International Institute for Environment and Development (IIED) and United Nations Population Fund (UNFPA)*. <http://www.unfpa.org/sites/default/files/resource-pdf/UEPI%207%20Tacoli%20Mar%202012.pdf>. Accessed: 25 May 2015.

Teucher, B., Olivares, M., & Cori, H. (2004). Enhancers of iron absorption: ascorbic acid and other organic acids. *International Journal for Vitamin and Nutrition Research*, *74*, 403-419.

Vilakati, N., MacIntyre, U., Oelofse, A., & Taylor, J. R. N. (2015). Influence of micronisation (infrared treatment) on the protein and functional quality of a ready-to-eat sorghum-cowpea African porridge for young child-feeding. *LWT-Food Science and Technology*, *63*, 1191-1198.

World Health Organization (WHO). (2008). Iron deficiency anaemia; assessment, prevention and control. Geneva: WHO Press. http://apps.who.int/iris/bitstream/10665/66914/1/WHO_NHD_01.3.pdf?ua=1. Accessed 19 August 2015.