

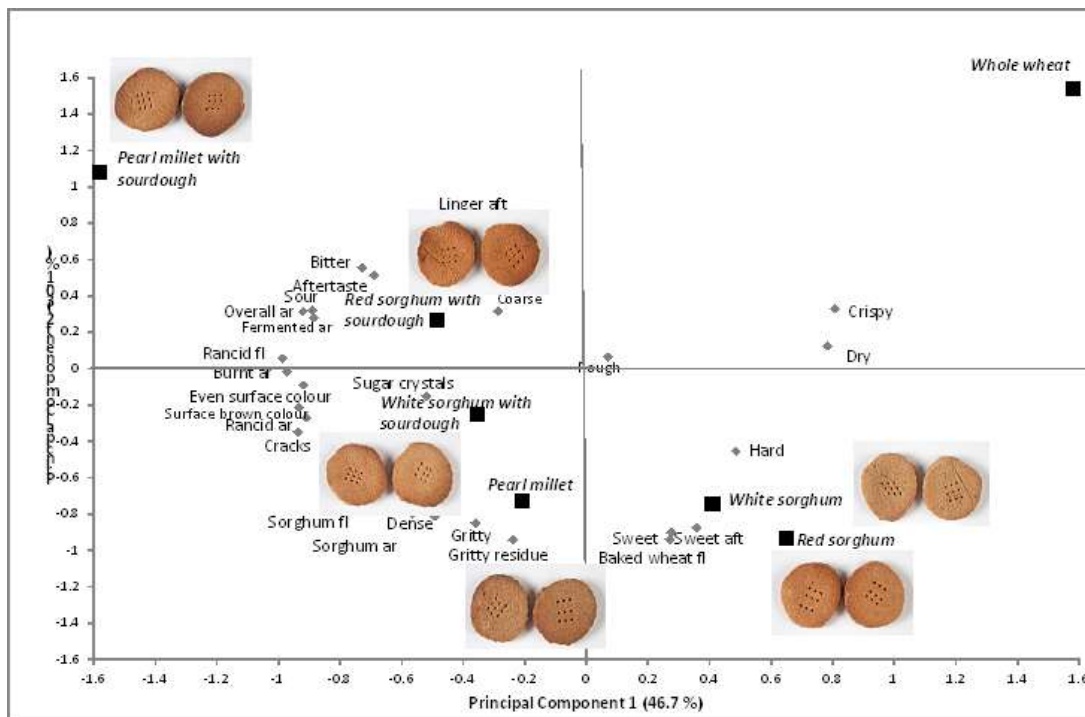
Sensory and nutritive profiles of biscuits from whole grain sorghum and pearl millet plus soy flour with and without sourdough fermentation

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Graphical abstract



Summary

Whole grain sorghum- and pearl millet-soy composite biscuits, with and without sourdough were produced with the aim of developing a ready-to-eat supplementary food for nutritionally at-risk school-age children in Africa. Descriptive sensory profiling revealed that

the biscuits were indistinguishable from a whole wheat biscuit standard in terms of hardness, roughness and coarseness but they were darker, less crisp, less dry and denser with distinctive sorghum flavour. Biscuits containing sourdough were sourer to taste, had more aroma, and a more fermented taste and odour character, other examples were more rancid and bitter with less sorghum-type flavour. Biscuits without sourdough were blander and sweeter. Two biscuits per day will, on average, contribute 13% of the Dietary Reference Intake of fibre for children 4-8 years and 16%, 11% and 8% Mg, Fe and Zn, respectively. The sourdough biscuits had 10-17% less phytate, and phenolic content while antioxidant activity increased.

Keywords Biscuits, sensory, minerals, pearl millet, sorghum

Introduction

Increasing numbers of people in Africa suffer from malnutrition due either to undernutrition caused by inadequate intake of macro- and micronutrients, and increasingly obesity (Zeba *et al.*, 2012). The latter has been related to “nutrition transition”, where there is excessive consumption of refined energy-rich foods that may be deficient in micronutrients (Popkin *et al.*, 2012; Zeba *et al.*, 2012).

Epidemiological studies have shown that regular consumption of whole grain foods is protective against certain cancers, cardiovascular disease, type-2 diabetes and obesity (Slavin, 2004). Because of the inclusion of the pericarp and germ components, whole grains are rich sources of micronutrients: vitamins, minerals and phytochemicals including phenolic compounds, carotenoids, vitamin E, β -glucan and sterols, and dietary fibre (Fardet *et al.*, 2008).

Sorghum (*Sorghum bicolor*) and pearl millet (*Pennisetum glaucum*) are important traditional staples of nearly 600 million nutritionally at-risk people in the semi-arid tropics, particularly in Africa (ICRISAT). Previously, we have developed protein-rich biscuits made from refined sorghum flour composited with defatted soy flour (DFS) for use as a supplementary food to prevent Protein Energy Malnutrition in children (Serrem *et al.*, 2011a,b). Biscuits are a good vehicle for nutrition interventions as a supplementary food, due to their popularity, high nutrient density, long shelf-life and are ready-to-eat (Sudha *et al.*, 2007).

Sourness, a primary taste sensation (DeSimone *et al.*, 2001), can be imparted by sourdough fermentation, and is a desirable characteristic of many traditional African cereal foods (Nout, 2009). The sourness may also potentially mask the reported undesirable beany flavour of soy (Serrem *et al.*, 2011a) and mousy flavour of pearl millet foods (Seitz *et al.*, 1993). Further, sourdough fermentation reduces the level of phytate, a potential anti-nutrient, in whole grains (Katina *et al.*, 2005) and may improve the texture of baked goods (Arendt *et al.*, 2007). Phytate (myo inositol hexaphosphate) can adversely influence the bioavailability of essential minerals (Lönnerdal, 2002).

Building on previous work (Serrem *et al.*, 2011a,b) where elevated levels of phenolic phytochemicals, high energy density and improved protein quality of cereal-legume composite biscuits were demonstrated, the objectives of this study were:

- (1) to determine whether sorghum and pearl millet biscuits enriched with soy protein can be produced from whole grain;
- (2) to determine the impact of whole grain and sourdough fermentation on the sensory characteristics, essential minerals, dietary fibre and phytate content of the biscuit.

The aim was to provide the benefits of whole grains in the form of a ready-to-eat supplementary food for children in at-risk communities in Africa.

Materials and methods

Grains and other biscuit ingredients

Red non-tannin sorghum, cultivar PAN 8564 from Free State Maltsters, South Africa, non-tannin white tan-plant sorghum, variety Macia obtained from Botswana and pearl millet, variety SDMV 89004 from ICRISAT, Zimbabwe were used. The whole grains were separately milled using a laboratory hammer mill fitted with a 500 µm opening screen to produce a relatively coarse whole grain flour. It is not practical to mill whole grain sorghum and pearl millet into a finer fine due to these grains having a hard, corneous (vitreous) endosperm component (Munck, 1995). The flour was vacuum-packed and stored at 4°C prior to usage. DFS was obtained from Nedan Oil Mills, Mokopane, South Africa. Other ingredients were purchased at a local supermarket.

Preparation of sourdough

Traditional lactic acid starter cultures were prepared from the respective whole grain sorghum and pearl millet flours by back-slopping as described by Taylor & Taylor (2002). The sourdoughs had a pH of 3.7 and a titratable acidity of 1.76% lactic acid.

Preparation of biscuits

Biscuits (\pm 10.4 g each) were prepared from the sorghum and pearl millet whole grain flours and DFS, with and without sourdough, essentially according to Serrem *et al.* (2011a). The sorghum-soy or pearl millet-soy biscuit formulations comprised by weight, 100 parts whole grain flour, with addition, on flour basis of 40 % DFS, 35 % sugar, 50 % baking margarine and water (158% on flour basis). For the sourdough biscuits, 50% of the flour portion formed part of the sourdough with water and 50% were added during mixing. Biscuits were baked at

180 °C for 20 ± 5 min. Cooled biscuits were vacuum-packed in polyethylene bags and stored at 4°C. The water activity of the biscuits was ≤ 0.4 .

Descriptive sensory analysis

An experienced 10-member sensory panel was selected after screening tests that included tastes (bitter, sweet, sour, salt and umami) identification and exercises to describe differences among biscuits. Sensory profiling of the biscuits was performed using the generic descriptive analysis method (Einstein, 1991). The panel was trained in ten sessions of 2 hours per day during which 26 appearance, aroma, texture, flavour, and aftertaste sensory properties with definitions, reference standards and methodology of evaluation were developed (Table 1). Each attribute was evaluated on a 10 point scale (1-10) anchored with verbal descriptions.

Evaluation of the six biscuit treatments and a commercial whole grain wheat biscuit (McVitie's Wholewheat Digestives, United Biscuits, United Kingdom) was replicated three times in sessions of 1.5 h a day for 3 days. This was done in a sensory laboratory with individual booths following standard good sensory practices (Lawless & Heymann, 1999). Filtered tap water and raw carrot slices were provided as palate cleansers. Biscuits were presented in transparent polyethylene zip-lock bags with random three-digit codes. Responses were collected using Compusense® *five* release 4.6 (Compusense Guelph, ON, Canada).

Total dietary fibre

Total dietary fibre content was determined in duplicate by an enzyme-gravimetric method (Prosky *et al.*, 1988).

Total phenolic content and antioxidant activity

A modified Folin-Ciocalteu method was used to measure total phenolic content (Waterman & Mole, 1994) with catechin as a standard. Antioxidant activity was determined using the 2,2'-azino-bis-3-ethylbenzothiazoline-6-sulphonic acid (ABTS) radical scavenging assay according to Awika *et al.* (2003) with Trolox as standard.

Major mineral composition and phytate contents

Samples were prepared by dry-ashing, method 935.13 (AOAC, 2000). Ca, Fe, Zn and Mg quantified using atomic absorption spectrophotometry using a single or multi-element lamp with hollow cathode and an air-acetylene burner (Giron, 1973). Phytate was determined as described by Fruhbeck *et al.* (1995).

Statistical analyses

Separate two-way analyses of variance (ANOVA) were used to determine the effects of cereal types, sourdough application (excluding the whole wheat biscuit) and biscuit type on sensory attributes. For each ANOVA, session was included as an additional main effect. The means of significant effects ($p < 0.05$), were compared by Fisher's least significant difference (LSD). Principal component analysis (PCA) of the sensory data for attributes that showed significant differences, was performed using Panelcheck V1.4.0, option original PCA. Nutritional measures and phytate data were analysed with one-way ANOVA to determine the effect of treatment.

Results and discussion

Descriptive sensory analysis of biscuits

Descriptive sensory analysis is a standard tool that gives comprehensive insight on the nature and size (quantity or intensity) of sensory attributes as perceived by humans while consuming food products. The product profiling information gathered with the method assist in understanding the effects of ingredients on sensory properties of products that is useful for predicting potential consumer responses to new products (Lawless & Heymann, 1999). Table 1 summarises the effect of cereal type and sourdough addition on biscuit sensory properties and Table 2 gives the detailed sensory profiles. Choice of cereal significantly influenced the appearance, aroma, some of the texture and flavour attributes of the biscuits. Pearl millet biscuits, in particular, received higher ratings compared to sorghum biscuits for appearance, aroma and flavour. White sorghum biscuits were generally drier and crispier than the red sorghum and pearl millet biscuits. Addition of sourdough increased ratings for colour, fermented aroma, overall aroma intensity, sour taste, rancid flavour and bitterness (of pearl millet-soy biscuit in particular) while it decreased sweetness and sorghum aroma. Rancidity and bitterness of pearl millet can be reduced by thermal pre-treatment (including toasting) of the grain (Nantanga et al. 2008). Sour taste is a desirable attribute of many traditional African cereal foods (Nout, 2009). Sourdough did not affect sorghum and pearl millet biscuit texture. This is unlike the effect of sourdough on the texture of bread (Edema *et al.*, 2013). The difference presumably due to biscuits not having open, cellular crumb structure.

By PCA, the first two PCs accounted for 90.5 % of the total variation in the sensory properties of the biscuits (Fig. 1). PC1 separated the biscuits according to whether sourdough was added or not, with the wheat biscuit to the left and the sorghum and pearl millet biscuits (without sourdough) in the centre and the biscuits with sourdough more to the right. The sorghum and pearl millet biscuits with sourdough was identified as more aroma intense with

more fermented and sour aroma. The lack of aroma and flavour attributes on the left of the plot show that the standard whole wheat biscuit, in contrast, was generally perceived as very bland. Colour and visual attributes, crispy and dry texture are also distinguishing factors of PC1. PC2 separated the sorghum and pearl millet biscuits without sourdough added, at the top, from the sourdough containing biscuits and whole wheat biscuit towards the bottom. Biscuits without sourdough were sweeter, with more sorghum aroma and cooked sorghum-type flavour.

The flavour of pearl millet biscuits with sourdough, in particular was more bitter, but also the red and white sorghum biscuits with sourdough were sourer with more intense overall and fermented aroma. The more intense bitter taste might be due to partial conversion of phenolic acid esters to phenolic acids during lactic acid fermentation (Svensson *et al.*, 2010). Phenolic acids have been proposed to be responsible for sour, bitter, and astringent sensory characteristics in protein rich plant foods (Huang and Zayas, 1991).

The more intense aroma of biscuits with sourdough can be attributed to the ability of lactic acid bacteria and yeasts to liberate aroma precursors such as free amino acids (Hansen & Schieberle, 2005). The browner colour of biscuits with sourdough can be attributed to browning reactions, possibly due to the amylolytic action of sourdough fermenting microorganisms (Gänzle, 2014). In general biscuits from red sorghum and pearl millet were less dry and crispy compared to the white sorghum biscuits. Serrem *et al.* (2011a) noted dry and crispy texture in sorghum biscuits and attributed it to the hydrophobic nature of the kafirin prolamin proteins (Duodu *et al.*, 2003).

Dietary fibre, phenolics and antioxidant activity

The whole grain sorghum and pearl millet had a high dietary fibre content of 15-16% (Table 3), even in comparison with whole wheat (14.3%) and whole wheat crackers (10.6%)

(USDA National Nutrient Database for Standard Reference). Thus, the biscuits could be labelled as high-fibre (>6 g dietary fibre/100 g) (European Commission, 2007). The high dietary fibre content of the biscuits is not just because of the fibre from the whole grain cereals, up to 12.1% in non-white sorghum (USDA National Nutrient Database for Standard Reference) and approximately 8.5% in pearl millet (Taylor, 2004), but also because DFS is very rich in fibre (18.9%) (USDA National Nutrient Database for Standard Reference). Consumption of two biscuits (20.8 g) would provide 13% of a 4-8 year old's dietary fibre Dietary Reference Intake (DRI) (USDA Food and Nutrition Information Center).

The total phenolic content of the cereal-soy biscuits (78-94 mg catechin equiv./100 g) (Table 3) was some three times higher than reported for whole grain sorghum biscuits

Table 3: Dietary fibre contents, total phenolics and antioxidant activity of wholegrain sorghum- and pearl millet-soy biscuits¹ with and without sourdough

	Red non-tannin sorghum	White tan-plant sorghum	Pearl millet
Dietary fibre (g/100 g db)			
Cereal-soy composite biscuits ²	16.0±1.6	Not determined	15.0±1.7
Dietary fibre contribution to DRI of two biscuits ³	13%	Not determined	13%
Total phenolics (mg catechin equiv./100 g db)			
Cereal-soy composite biscuits (flour)	95 ^a ±0	78 ^a ±1	82 ^a ±4
Cereal-soy composite biscuits (flour +sourdough)	105 ^b ±2	95 ^b ±5	114 ^b ±18
% increase due to sourdough	11%	22%	39%
ABTS antioxidant activity (µMol Trolox equiv./100 g db)			
Cereal-soy composite biscuits (flour)	168 ^a ±16	46 ^a ±9	276 ^a ±14
Cereal-soy composite biscuits (flour +sourdough)	269 ^b ±28	171 ^b ±4	470 ^b ±23
% increase due to sourdough	60%	272 %	70%

¹Mean and standard deviation, n = 2

²Dietary fibre contents of the flour and flour+sourdough biscuits were the same.

³Based on US Dietary Reference Intake (DRI) for children 4-8 years (USDA National Nutrient Database for Standard Reference) for biscuits, each of 10.4 g (db)

^{ab} For a specific parameter, different superscript letters within a column denote a significant difference (p<0.05)

(Chiremba *et al.*, 2009). Similarly, the cereal-soy biscuits had much higher antioxidant activity (46-276 $\mu\text{Mol Trolox equiv./100 g}$). This is due to the high iso-flavone content of DFS, approximately 60 mg/100 g (Wang and Murphy, 1996). The biscuits made with pearl millet had the highest antioxidant activity. Major phenolics in pearl millet are ferulic acid, apigenin and myricetin (Shahidi & Chandreskara, 2013) with antioxidant activity being concentrated in the bran. Addition of sourdough increased the total phenolic content of the biscuits somewhat (11-39%) but increased their antioxidant activity to a much greater extent (60-272%), especially in the biscuits made with the white tan-plant sorghum, which contains low levels of phenols (Dykes and Rooney, 2007). Thus, the increase in antioxidant activity with sourdough addition was probably not primarily due to phenolics but also because of other substances, particularly Maillard reaction products (Amarowicz, 2009). However, despite their antioxidant activity, there is evidence that dietary Maillard reaction products may be deleterious to health (Tessler and Birlouez-Aragon, 2010).

Major minerals and phytate content

With the exception of calcium, the mineral contents of the biscuits (Table 4) were very similar to those of sorghum grain (USDA National Nutrient Database for Standard Reference) and pearl millet grain (Taylor, 2004). Calcium content was somewhat higher, probably due to the very high content in DFS (260 mg/100 g) (USDA National Nutrient Database for Standard Reference). Despite this, only the magnesium, iron and zinc in the biscuits would make a significant contribution (7-17%) to the DRIs of 4-8 year olds (USDA Food and Nutrition Information Center) if two biscuits were consumed daily. Importantly, however, as shown in Table 4, whole cereal grains flour and soy are rich in phytate. Sourdough fermentation of the millet flours reduced their phytate content substantially, by more than 50%. The low pH due to lactic acid production promotes the dephosphorylation of

phytate by the cereal and/or bacterial phytases (Feil, 2001). However, phytate was only reduced 10-17% in the biscuits due to the sourdough making up only 35% of the flour. Thus, the mineral bioavailability of the whole grain biscuits, is likely to be low as very substantial reductions (> 50%) in phytate in whole sorghum foods are required to improve mineral bioavailability (Kruger *et al.*, 2012).

Conclusions

High fibre biscuits that contain substantial levels of magnesium, iron and zinc can be developed from whole grain sorghum and pearl millet composited with DFS that are similar in texture but different in flavour to commercial whole wheat biscuits. The inclusion of sourdough increases phenolic content slightly and antioxidant activity substantially and slightly reduces the level of phytate. It strongly influences their sensory characteristics, including sourer taste, more intense aroma and more fermented flavour. These whole-grain biscuits have potential as a supplementary food for school-age children in nutritionally at-risk communities in Africa where sorghum and/or pearl millet are consumed. Optimising the use of locally relevant food supplies and producing the biscuits locally, can contribute to food security in Africa as proposed by Mwaniki (2006) of the United Nations Office of the Special Advisor on Africa. Further research is, however, needed to determine whether the target consumers would find the biscuits acceptable and practical to include in their diet.

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