International Journal of Food Science and Technology

Running head: Soy fortified sorghum and bread wheat biscuits

ORIGINAL PAPER

Nutritional quality, sensory quality and consumer acceptability of sorghum and bread wheat biscuits fortified with defatted soy flour

Charlotte A. Serrem^{1,2}, Henriëtte L. de Kock¹, John R. N. Taylor^{1,3}

¹Department of Food Science, University of Pretoria South Africa

²Moi University, Kenya

³Correspondence: Prof J R N Taylor
Department of Food Science, University of Pretoria, Pretoria 0002, South Africa
Tel +27 12 4204296, Fax +27 12 4202839
e-mail: john.taylor@up.ac.za

Summary

Biscuits were produced by compositing sorghum or bread wheat flours with defatted soy flour. Compared to the 100% cereal biscuits, sorghum-soy and bread wheat-soy 1:1 ratio composite biscuits had at least double the protein content and the lysine content increased by 500-700%. For the sorghum-soy biscuits, in vitro protein digestibility increased by 170%. Two such biscuits of 28 g each could provide 50% of the recommended daily protein intake for 3 to 10 year olds. Descriptive sensory evaluation revealed that sorghum-soy composite biscuits had crispy and dry texture characteristics associated with biscuits. Sensory evaluation by school children showed that the composite biscuits were rated as acceptable as the cereal only biscuits and this was maintained over four days of evaluation. Hence sorghum- and bread wheat-soy biscuits have considerable potential as protein-rich supplementary foods to prevent Protein Energy Malnutrition in children.

Key words Biscuits, bread wheat, consumer acceptability, defatted soy flour, lysine availability, protein digestibility, sensory evaluation, sorghum.

Introduction

The most serious deficiency diseases resulting from under nutrition among children in developing countries are the various forms of Protein Energy Malnutrition (PEM). Cereal-staples, which are principal dietary sources of protein and energy, are suitable vehicles for delivering protein to prevent PEM (Bulusu *et al.*, 2007). Sorghum is an important cereal crop especially for at-risk communities, providing food for over 500 million people in semi-arid tropics of Africa (ICRISAT, 2009). However, sorghum has poor protein quality because it is

limiting in the indispensable amino acid lysine with only approximately 2 g per 100 g protein (Taylor & Schussler, 1986). The problem is further compounded by the reduction in protein digestibility on wet cooking (Duodu *et al.*, 2003) which is apparently unique to sorghum. Wheat, mainly hard, bread-type wheat is another important cereal staple in the semi-arid tropics of Africa, being cultivated in some 33 countries (Taylor, 2004).

Formulation of foods from low-lysine staples fortified with legumes has been proposed as a practical and sustainable approach to improving the protein nutritional value of foods for young children in developing countries (Young & Pellet, 1999; FAO/WHO, 1994). Soy beans contain 30 to 45% protein with a good source of all indispensable amino acids (Karr-Lilienthal *et al.*, 2006). Visconcelos *et al.* (2001) established that the indispensable amino acid profile for soy beans essentially met the reference pattern for children of 2 to 5 years and above. The true nitrogen digestibility of soy beans is approximately 95% in children (Bodwell & Marable, 1981). Defatted soy flour (DSF) has been used to fortify bread resulting in dramatic improvement of protein content (Mashayekh *et al.*, 2008). However, biscuits are probably a better vehicle of fortification with protein because of their popularity, and high nutrient density and long shelf-life because they are very low in moisture (Sudha *et al.*, 2007).

Soft wheat flours are normally used for biscuit preparation, but they can also be made with nonwheat flours such as sorghum (Dendy, 1993) and bread wheat flour. Fortification of wheat biscuits with 20% isolated soy protein (Mohsen *et al.*, 2009) or 20% DSF (Singh *et al.*, 2000) increased protein content to 20 and 10%, respectively. There are also reports of biscuits made using sorghum flour (Badi & Hoseney, 1976) or pre-gelatinised sorghum flour dough (Dendy, 1993). The biscuits were gritty and fragile. Biscuits of acceptable quality have been made using wheat-sorghum composites with 5% DSF (Mridula *et al.*, 2007), but soy compositing of sorghum only biscuits has not been reported.

Food products developed specifically for children must be tested by children (Guinard, 2001). Children aged 8 to 9 years were found to be consistent when evaluating biscuits using hedonic categorisation (Leon *et al.*, 1999). Additionally, a consumer exposure test of several days can be used to determine long term acceptance of new food products (Wiejzen *et al.*, 2008). There are no reported studies using children in Africa to establish long term acceptability of new foods using the repeated exposure test.

The objectives of this study were to formulate and develop biscuits from sorghum and bread wheat flours composited with defatted soy flour and to determine the effect of compositing with defatted soy flour on the nutritional and sensory quality and long term consumer acceptability.

Materials and methods

Biscuit ingredients

Commercial decorticated red, non-tannin sorghum meal particle size $\leq 1000 \ \mu m$ and white bread wheat flour both obtained from Tiger Consumer Brands (Bryanston, South Africa) and DSF flour from Nedan Oil Mills (Potgietersrus, South Africa) were used. Other ingredients, granulated sugar, sunflower oil, baking powder and vanilla essence, were purchased locally in Pretoria, South Africa.

Biscuit formulation and preparation

The 100% sorghum, 100% bread wheat and 100% soy biscuits basic formulation comprised 225 g flour, 56 g sugar, baking powder, 66 g sunflower oil, water and 13.5 g vanilla essence (Table 1). Water was dependent on the treatment and ranged from 10.0% (100% sorghum biscuits) to 30.7% (100% soy biscuits) of total weight of ingredients, as was baking powder, 0.25 g in 100% bread wheat biscuits to 1.5 g in the sorghum biscuits. The amounts added were based on results of preceding experiments. In the formulations, 28.6, 50, 71.4, and 100%, DFS replaced sorghum or bread wheat flours on a weight by weight basis. The proportions were based on a basic formulation in which a maximum of 161 g DSF (50 g protein/ 100 g) and 64 g cereal (12 g protein /100 g) in a 225 g composite could produce 12 biscuits each containing approx. 7 g protein. For the consumer study, the particle size of the sorghum flour was reduced to a maximum of 500 μ m to in order to reduce the gritty texture of biscuits. As a consequence, the dough water content had to be doubled to 80 ml and 180 ml/225 g flour for 100% sorghum and sorghum-soy composite biscuits, respectively, to make the dough workable (Table 1).

Flour, sugar and baking powder, were mixed by hand. Oil and water were added gradually and the mixture kneaded for 3 min at medium speed in an electric mixer to a firm dough. The dough was manually rolled out on a steel tray, to a height of 5 mm and cut into circular shapes using a 6.3 cm diameter biscuit cutter. The dough pieces were transferred onto a baking tray lined with aluminium foil. The biscuits were baked in a preheated electric oven at $180^{\circ}C \pm 2^{\circ}C$ for 20 ± 5 min and cooled for 30 min at ambient temperature. Biscuits, were vacuum packed in polyethylene bags and stored at $10^{\circ}C$. Biscuits of each formulation were made three separate

times. For chemical analyses, biscuits were ground using a mortar and pestle to a particle size of ≤ 1 mm before storage.

Proximate analyses

Proximate composition of biscuits and flour was determined according to standard AACC International (2000) methods. Moisture content was determined by the one stage air oven procedure, method 44-15A. Protein content (N x 6.25) was determined by the Dumas combustion method 46-30. Oil and ash (mineral) were determined by soxhlet extraction, and dry ashing, methods 08-01 and 30-25, respectively. Crude fibre was determined using method 32-10. Carbohydrate was calculated by difference and energy was calculated using Atwater conversion factors (FAO, 2003).

Protein quality

Amino acids were determined using the Pico-Tag method (Bidlingmeyer *et al.*, 1984) after acid hydrolysis. Reactive lysine was determined using a rapid dye-binding lysine (DBL) method of Kim *et al.* (2007) using Crocein Orange G dye (70% dye content) (Fluka grade 27965: Sigma-Aldrich, Buchs, Switzerland). In vitro protein digestibility (IVPD) was determined by a pepsin digestion method (Hamaker *et al.*, 1987). Accurately weighed samples (200 mg) were digested with P700-100G pepsin, activity 863 units/mg protein (Sigma-Aldrich, St. Louis, MO) for 2 h at 37°C. Residual protein was determined by Dumas combustion.

Descriptive sensory analysis

The descriptive sensory panel comprised nine women and two men, aged between 20 and 41 years who were students of the University of Pretoria. They were selected after undergoing

screening tests that included the five basic taste (bitter, sweet, sour, salt and umami) test, an aroma identification test and an exercise to describe differences among biscuits. Before engaging in the sensory exercise the panellists signed a consent form informing them of the nature of the biscuit samples they would evaluate. Descriptive sensory profiling of the nine biscuit samples was performed using the generic descriptive method of Einstein (1991). The panel was trained in 10 sessions of 2 hours each per day during which they generated and reached consensus for 28 descriptors with definitions, reference standards (biscuits and other food items available commercially) to anchor the scale ends and the sequence of descriptors on the ballot (Table 2). Evaluation of the biscuits was carried out over a three day period in three sessions of 1.5 hours a day following a randomized complete block design. All nine treatments were randomly presented to each panellist during each session first as a set of 5 biscuits followed by a set of 4 biscuits after a 20 minute break. Each treatment was presented as a ¹/₂ biscuit in a transparent polyethylene zip-lock type bag with a random three digit code. Panellists were seated in individual sensory booths under red light and were provided with a glass of deionised water and raw carrot slices to cleanse the palate. Each of the nine biscuits was rated for appearance, texture, flavour and aftertaste using the 28 descriptors on a 10 point scale. Responses were collected using a computer system and the computer software program (Compusense® Five release 4.6

[1986-2003] Guelph, Ontario Canada).

Consumer evaluation

Four treatments were used, 100% sorghum, 100% bread wheat (control), 50:50 sorghum and DSF and 50:50 bread wheat and DSF. This formulation was selected as one biscuit of 28 g would provide approx. 25% of a child's RDA of protein.

Children (26 boys and 34 girls) aged 8 to 9 years who attended Zakhele primary school in Pretoria, South Africa evaluated the biscuits. Ethical approval was granted by the University of Pretoria Ethics Committee. Permission to conduct the study was granted by the school principal and only children who voluntarily accepted and whose parents signed a consent form that informed them of the nature of the biscuit samples and the activities involved in the study were included. Evaluation was conducted over a period of four days using hedonic categorisation and repeated exposure tests (Wiejzen et al, 2008) to determine children's liking. A five point hedonic facial scale (dislike very much at 1 and like very much at 5) was used. The children were divided into four groups. Two half-hour evaluation sessions were conducted each day. In the first session, responses for the four ¹/₂ biscuits presented to each child in polyethylene zip lock type bags labeled with randomized 3 digit codes were entered by the children into a score sheet A. Each child was then presented with a test biscuit (two whole biscuits of one of the four types), to eat as much as they wished. Immediately after, the children were asked if they would like to eat the test biscuit again, to which they could say yes, no or not sure. In the second session, responses for evaluation of another set of the same four biscuit types were entered into score sheet B. Bottled water to cleanse the palate was provided. The same procedure was repeated for the four days, but the test biscuit was different for each group every day. In four days each child evaluated each type of biscuit 8 times and tasted each test biscuit once.

Statistical analyses

Data were analysed as one way analysis of variance. Data for consumer evaluation were analysed using repeated measures analysis of variance. All means were compared using Fischer's least significant difference test. Principal Component Analysis (PCA) of the significant sensory attributes was performed using a correlation matrix with biscuit samples in rows and descriptors in columns. Box and whisker plots were used to illustrate consumer hedonic score distribution for the biscuits.

Results and discussion

Proximate composition

Fortification of sorghum or bread wheat flours with DSF at a 1:1 ratio substantially increased the mineral, oil, crude fibre and protein content compared to unfortified biscuits, whereas there was substantial reduction in carbohydrate content (Table 3). Mineral content was two and five fold higher in composited sorghum and bread wheat biscuits, respectively. Defatted soy flour has high mineral content, particularly Ca, Mg, K and P (USDA, 2007). The fat content was the same in soy composite and 100% cereal biscuits, as a result of the inclusion of 20% sunflower oil in the biscuits formulae. The fat content of the biscuits was within the range recommended of 10 to 25 g oil per 100 g according to the guidelines for formulation of supplementary foods for young children (FAO/WHO, 1994). The substantially lower carbohydrate content of soy composite biscuits was due to the low carbohydrate content of DSF (30%), because soy bean stores energy as oil (USDA, 2007). The crude fibre contents of 3.7 g and 3.3 g in 1:1 ratio sorghum- and bread wheat-soy composite biscuits, respectively, was probably within the recommended range of not more than 5 g dietary fibre and other non-absorbable carbohydrates per 100 g dry matter (FAO/WHO, 1994). The soy composite sorghum and bread wheat biscuits contained 1924 and 1880 kJ/100 g energy, respectively and hence conformed to the (FAO/WHO, 1994) recommended minimum energy content of 1674 kJ/100 g. Hence, two biscuits of 28 g would provide some 14% of the energy requirements of a of 5-7 year old child, which are approx. 7500 kJ (FAO/WHO/UNU, 1985).

Protein content increased by 168% and 139%, respectively in 1:1 ratio sorghum- and bread wheat-soy composite biscuits compared to 100% cereal biscuits. The increase is due to the 50% protein content of DSF. Other studies have reported a similar increase in protein in sorghum-soy composite flours (Bookwalter *et al.*, 1987; Awadelkareem *et al.*, 2008), 20% DSF composite wheat biscuits (Singh *et al.*, 2000) and 7% DSF composite wheat bread (Mashayekh *et al.*, 2008). The biscuits with a 1:1 ratio of sorghum or bread wheat with soy flour met the target of providing 7 g protein in a 28 g biscuit thus consumption of one, two and three biscuits would provide approximately half the protein intake for children aged 1 to 2, 3 to 10, and 10 to 18 years, respectively (IOM, 2005).

Lysine and reactive lysine content

The protein lysine content of 1:1 ratio sorghum- and bread wheat-soy biscuits was markedly improved by 231 and 152%, respectively compared to 100% cereal biscuits (Table 4). The increase is due to DSF protein having three times higher lysine than the cereals because of the lysine-rich globulin fraction in soy bean (Marcone & Kakuda, 1999). Consequently, the biscuit protein lysine scores improved from 29% to 95% and 39% to 98% of the WHO pattern of 48 mg lysine/100 g protein for 3-10 year old children (WHO, 2007) in the sorghum and bread wheat biscuits, respectively. Similar results were reported for sorghum-soy composite chapattis (Lindell & Walker, 1984), which exhibited a 27% increase in lysine content and represented 77% of the

WHO (2007) pattern. The improvement in the lysine content of the biscuits themselves was even more dramatic. At a 1:1 cereal to soy ratio, there was a 500-700% increase.

Compositing with DSF at a 1:1 ratio increased the reactive (chemically available) lysine content of the sorghum and bread wheat biscuits by 261 and 4%, respectively (Table 4). Bookwalter *et al.* (1977) obtained similar results for dry milled sorghum flour in which reactive lysine increased by 115% as lysine from DSF increased. The reactive lysine content of these biscuits was substantially lower than their total lysine content. It is probable that there was substantial loss of reactive lysine during baking due to the Maillard reaction which was enhanced by the lysine contributed by the soy proteins. Similar losses (27 to 47%) were noted during the production of dietetic biscuits (Hovatic & Eres, 2006).

In vitro protein digestibility

Compositing with soy substantially increased the IVPD of the sorghum biscuits (Table 4). At the 1:1 ratio IVPD was 170% higher than the sorghum only biscuits. The increase can be attributed to dilution of the less digestible sorghum kafirins with the more available soy globulins. These findings agree with the results of Bookwalter *et al.* (1987) for sorghum-soy grits with 13% higher IVPD than 100% sorghum grits. Table 4 also shows that compositing with soy relieved the reduction in IVPD that occurred when sorghum only biscuits were made. This reduction in IVPD of sorghum foods on cooking is due primarily to disulphide bonding of the sorghum kafirin proteins (Duodu *et al.*, 2003).

As a consequence of the dramatic improvement in lysine score in both the sorghum and bread wheat biscuits and improvement in the IVPD of the sorghum biscuits as a result of compositing with soy, Protein Digestibility Corrected Amino Acid Score (PDCAAS) was 8 and 2 times higher in the 1:1 ratio sorghum- and bread wheat-soy biscuits, respectively compared to the 100% cereal biscuits. PDCAAS is the internationally accepted measure of food protein quality and is used to assess the protein quality of both dietary mixtures and individual protein food sources (WHO, 2007).

Descriptive Sensory Analysis

Analysis of variance *F*- values of the biscuits profile data of the 28 attributes scored by the descriptive sensory panel showed significant differences (P \leq 0.05) between the treatments for 26 attributes (data not shown). The data were further analyzed by PCA to determine the systematic variation and underlying relationships between attributes and biscuits made from composite flours of varying cereal-legume ratios. The first two principal components explained 92% of the total variation (Fig 1). PC1 explained 59% of the total variation and separated the biscuits based on their cereal component with the wheat biscuits to the right and the sorghum biscuits to the left. PC2 accounted for 33% of the total variation and separated soy and cereals with high soy content biscuits at the top and high cereal biscuits at the bottom (Fig 1a).

Figure 1(b), shows that the 100% DSF and the sorghum and bread wheat biscuits substituted with 71.4% DSF were associated with the roasted soy bean flavour, aroma and aftertaste. The beany flavour is commonly associated with food legumes. In soy beans, enzymatic breakdown by lipoxygenases or autoxidation of linoleic and linolenic acid produces hydroperoxides such as ketones, aldehydes and alcohols that may be responsible for the beany, flavour which

discourages soy consumption (Boge, Boylston and Wilson, 2009). Examples of such compounds were identified by Mohsen *et al.* (2009) who categorized furans in wheat biscuits fortified with isolated soy protein as 2-ethyl-5methylfurans and 2-pentylfurans.

Sorghum biscuits with DSF content above 50% also had burnt flavour and odour notes described by panelists as baked biscuit. The protein contributed by DSF probably readily reacted with sugar in the Maillard reaction, hence the negative correlation with sweet flavour and aftertaste. Similarly, Mohsen *et al.* (2009) reported biscuit-like and burnt odour in biscuits when 2-ethyl-5methylpyrazine increased after substituting wheat with 20% soy protein isolate. The roasted cereal flavour and aroma associated with sorghum biscuits with DSF replacement of 50% and below may be attributed to derivatives of the Maillard reaction similar to findings by Bredie *et al.* (1998) who noted the development of roasted/toasted flavour in thermally treated maize and wheat flours when pyrazines increased. The high intensity of browning perceived in biscuits with high DSF was probably also caused by the Maillard reaction that produce brown polymers, which contribute to the surface coloration of biscuits similar to findings by Ameur *et al.* (2008).

The gritty, coarse and rough characteristics for sorghum biscuits may be due to the hard vitreous endosperm cells of sorghum grain that remain intact during milling (Rooney & Miller, 1982) which is related to the fact that the sorghum starch granules are encapsulated by hydrophobic cross-linked kafirins (Ezeogu *et al.*, 2008). Grittiness in cookies was also reported (Badi & Hoseney, 1976; Chiremba *et al.*, 2009) for sorghum biscuits made from tannin sorghums. The white specks may have been the bran fragments that were not completely milled, as reported by Kebakile *et al.* (2008) for porridge made using sorghum flour. The gritty texture of the biscuits indicated the need to mill the sorghum meal more finely.

All the sorghum biscuits were further associated with dry and crisp texture. The dry texture may be ascribed to sorghum doughs that absorbed the least amount of water compared to all other composite doughs (Table 1). This may be due to the hydrophobic nature of kafirin proteins of the endosperm (Duodu *et al.*, 2003). Crispness in sorghum biscuits was probably due to the absence of gluten. In contrast, the doughy, dense and hard texture of bread wheat biscuits was probably caused by the damaged starch, pentosans and high protein content of bread wheat absorbing high amounts of water during dough mixing (Kent and Evers, 1994). The hard texture was probably due to the stronger gluten of hard wheat flours which impart toughness to biscuits (Hoseney, 1994).

Consumer acceptability

All biscuits had a high hedonic rating of above 80% and the soy biscuits were equally liked to the whole wheat biscuits (Figure 2). Mashayekh, *et al.* (2008) also reported high sensory scores for wheat bread with 3% soy addition. Repeated exposure did not change the children's liking with the hedonic scores of 80% being sustained throughout the study period. This indicates that the children did not become bored with the biscuits (Zandstra *et al.*, 2004) and hence would eat them daily over a long period of time. Previous studies have demonstrated that time preference curves for staple foods which are moderately liked such as bread and butter (Hetherington *et al.*, 2002) and dairy products (Siegel & Pilgrim, 1958) are flat because there is no significant decrease following repeated servings, compared to chocolate and vegetables, respectively which are not staples and declined in liking. It is also likely that the children sustainably liked the biscuits similar to the study by Stubenitsky *et al.* (1999). The relative short distribution of scores along the bar graphs indicates general agreement among the children over scores for all biscuits and that results were consistent. This is in agreement with the findings of Leon *et al.* (1999) for biscuits which showed that 8 to 9 year old children are consistent when using hedonic categorization.

Conclusions

Fortifying sorghum or bread wheat with DSF dramatically improves the protein content and quality of biscuits. At a 1:1 cereal: DSF ratio, two biscuits of 28 g each could provide 50% of the protein requirements for 3-10 year old children, with greatly improved PDCAAS. Additionally two biscuits would provide approx. 14% of their energy requirement. Fortifying sorghum biscuits with defatted soy flour imparts positive sensory characteristics such as crisp texture and roasted cereal flavour and reduces hard and dense texture. Additionally, the composite biscuits were highly acceptable to school children and appear to retain their acceptability over time. Hence, sorghum- or bread wheat-soy fortified biscuits have considerable potential for use as a protein-rich supplementary food in semi-arid tropical countries to prevent PEM in preschool and school children.

Acknowledgement

Charlotte Serrem acknowledges a scholarship from VLIR-UOS, Belgium and Moi University, Kenya.

References

AACC International (2000) Approved Methods of the AACC, 10th edn. Methods 10-50D, 44-15A, 46-30, 08-01, 30-25 and 32-10. St Paul, MN: The Association.

- Ameur, L. A., Rega, B., Giampaoli, P., Trystram, G., & Birlouez-Aragon, I. (2008). The fate of furfurals and other volatile markers during the baking process of a model cookie. *Food Chemistry*, **111**, 758-763.
- Awadelkareem, A. M., Mustafa, A. I. & El Tinay, A. H. (2008). Protein, mineral content and amino acid profile of sorghum flour as influenced by soybean protein concentrate supplementation. *Pakistan Journal of Nutrition*, **7**, 475-479.
- Badi, S. M. & Hoseney, R.C. (1976). Use of sorghum and pearl millet flours in cookies. *Cereal Chemistry*, **53**, 733-738.
- Bidlingmeyer, B.A., Cohen, S. A. & Tarvin, T. L. (1984). Rapid analysis of amino acids using pre-column derivatization. *Journal of Chromatography*, **336**, 93-104.
- Bodwell, C. E. & Marable, N. L. (1981). Effectiveness of methods for evaluating the nutritional quality of soy bean protein. *Journal of the American Oil Chemists Society*, **58**, 475-483.
- Boge, E. L., Boylston., T. D. & Wilson., L. A. (2009). Effect of cultivar and roasting method on composition of roasted soy bean. *Journal of the Science of Food and Agriculture*, **89**:829-826.
- Bookwalter, G.N., Kirleis, A.W. & Mertz, E.T. (1987). In vitro digestibility of protein in milled sorghum and other processed cereals with and without soy-fortification. *Journal of Food Science*, **52**, 1577-1579.
- Bookwalter, G.N., Warner, K. & Anderson, R. A. (1977). Fortification of dry-milled sorghum with oilseed proteins. *Journal of Food Science*, **42**, 969-97.
- Bredie, W. L. P., Mottram, D. S., Hassel, G. M. & Guy, R. C. E. (1998). Sensory characteristics of the aromas generated in extruded maize and wheat flour. *Cereal Science*, **28**, 96-106.

Bulusu, S., Laviolette, L., Mannar, V. & Reddy, V. (2007). Cereal fortification programmes in developing countries. In: *Issues in Complementary Feeding* (edited by C. Agostoni & O. Brunser). Pp. 91-105. Nestec, Basel.

Chiremba C., Taylor, J. R. N., and Duodu, K. G. 2009. Phenolic content, antioxidant activity and consumer acceptability of sorghum cookies. *Cereal Chemistry*, **86**, 590-594.

Dendy, D.A.V. (1993). Drought and urbanization in Africa: The future role of sorghum and millets. In: *Cereal Science and Technology: Impact on a Changing Africa* (edited by J. R. N. Taylor., P. G. Randall. & J. H. Viljoen). Pp 299-317. Pretoria, South Africa: CSIR.

Duodu, K. G., Taylor, J. R. N., Belton, P. S. & Hamaker, B. R. (2003). Factors affecting sorghum protein digestibility. *Journal of Cereal Science*, **38**, 117-131.

Einstein, M. A. (1991). Descriptive techniques and their hybridization. In: *Sensory Science Theory and Applications in Foods* (edited by H. T. Lawless and B. P. Klein). Pp. 317-338. New York: Marcel Dekker.

Ezeogu, L. I., Duodu, K. G., Emmambux, N. M. &Taylor, J. R. N. (2008). Influence of cooking conditions on the protein matrix of sorghum and maize endosperm flours. *Cereal Chemistry*, **85**, 397-402.

FAO. (2003). Food Energy: Methods of Analysis and Conversion Factors. FAO Food and Paper Nutrition Paper 77. Pp. 18-37. Rome FAO., Rome pp18-37.

FAO/WHO. (1994). Codex Alimentarius: *Foods for Special Dietary Uses (including Foods for Infants and Children)*, 2nd edn. Rome: FAO.

FAO/WHO/UNU Expert Consultation. (1985). Energy and Protein Requirements, Report of a Joint FAO/WHO/UNU Expert Consultation. World Health Organization Technical Report Series724. Geneva: WHO.

Guinard, J. (2001). Sensory and consumer testing with children. *Trends Food Science and Technology*, **11**, 263-273.

Hamaker, B. R., Kirleis, A. W., Butler, L. G., Axtell, J. D. & Mertz, E. T. (1987). Improving the in vitro protein digestibility of sorghum with reducing agents. *Proceedings of the National Academy of Sciences of the United States of America*. **84**, 626-628.

Hetherington, M. M., Pirie, L. M. & Nabbs, S. (2002). Stimulus satiation: effects of repeated exposure to foods on pleasantness and intake. *Appetite* **38**, 19-28.

Horvatic M. & Eres, M. (2006). Protein nutrititive quality during production and storage of dietetic biscuits. *Journal of the Science of Food and Agriculture*, **82**, 1617-1620.

Hoseney, R. C. 1994. *Principles of Cereal Science and Technology*. 2nd edn. St. Paul, M. N. American Association of Cereal Chemists. St. Paul.

Institute of Medicine, Food and Nutrition Board (IOM). (2005). *Dietary Reference Intakes for Energy Carbohydrates, Fibre, Fat, Fatty Acids, Cholesterol, Protein and Amino Acids (Macronutrients)*. Washington, DC: National Academies Press.

International Crops Research Institute for the Semi Arid Tropics (ICRISAT) (2009) Sorghum. Available online @ <u>http://www.icrisat.org/sorghum/sorghum.htm</u>.

Karr-Lilienthal, L. K., Bauer, L. L., Zinn, K. E., Frazier, R. L., Parsons, C. M. & Fahey, G.C. (2006). Chemical composition and nutritional quality of soy bean meals prepared by extruder/expeller processing for use in poultry diets. *Journal of Agricultural and Food Chemistry*, **54**, 8108-8114.

Kent, N. L., Evers, A. D. 1994. Technology of Cereals, 4th edn. Elsevier Science. Oxford.

Kebakile, M. M., Rooney L. W., de Kock, H. L. & Taylor, J. R. N. (2008). Effects of sorghum type and milling process on the sensory characteristics of sorghum porridge. *Cereal Chemistry*, 85: 307-313

Kim, J. S., Kim, K. J., Ma, W. C. J & Chung, H. Y. (2007). Development of a method to quantify lysine in small amounts of rice grain. *Korean Journal of Sanitation*, **22**, 75-84.

Leon, F., Couronne, T., Marcuz, M. C. & Koster, E. P. 1999. Measuring food liking in children: a comparison of non-verbal methods. *Food Quality and Preference*, **10**, 93-100.

Lindell, M. J & Walker, C. E. (1984). Soy enrichment of chapattis made from wheat and nonwheat flours. *Cereal Chemistry*, **61**, 435-438.

Marcone, F. M. & Kakuda, Y. (1999). A comparative study of the functional properties of amaranth and soy bean globulin. *Nahrung*, **43**, S368-373.

Mashayekh, M., Mahmoodi, M. R. & Enterazzi, M. H. (2008). Effect of fortification of defatted soy flour on sensory and rheological properties of wheat bread. *International Journal of Food Science and Technology*, **43**, 1693-1698.

Mohsen, M.S., Fadel, H. H. M, Bekhit, M.A., Edris, A. E. & Ahmed, Y. S. (2009). Effect of substitution of soy protein isolate on aroma volatiles, chemical composition and sensory quality of wheat cookies. *International Journal of Food Science and Technology* **44**, 1705-1712.

Mridula, D., Gupta, R.K. & Manikantan, M. R. (2007). Effect of incorporation of sorghum flour to wheat flour on quality of biscuits fortified with defatted soy flour. *American Journal of Food Technology*, **2**, 428-434.

Rooney, L. W., & Miller, F. R. (1982). Variation in the structure and kernel characteristics of sorghum. in: *Proc. Int. Symp. on Sorghum Grain Quality* (edited by L. W. Rooney & D. S. Murty). Pp. 143-162. ICRISAT: Pantacheru, A. P., India

Siegel, P. S. & Pilgrim F. J. (1958). The effect of monotony on the acceptance of food. *American Journal of Psychology*, **4**, 756-759.

Singh, R., Singh, G. & Chauhan, G. S. (2000). Nutritional evaluation of soy fortified biscuits. *Journal of Food Science and Technology*, **37**, 162-164.

Stubenitsky, K., Aaron, J. I., Catt, S. L., and & Mela, D. J. (1999). Effect of information and extended use on the acceptance of reduced-fat products. *Food Quality and Preference*, **10**, 367-376.

Sudha, M. L., Vetrimani, R. & Leelavathi, K. (2007). Influence of fibre from different cereals on the rheological characteristics of wheat flour dough and on biscuit quality. *Food Chemistry*, **100**, 1365-1370.

Taylor, J. R. N. & Schüssler, L. (1986). The protein compositions of the different anatomical parts of sorghum grain. *Journal of Cereal Science*, **4**, 361-369.

Taylor, J. R. N. (2004). Grain production and consumption: Africa. In: *Encyclopedia of Grain Science* (edited by C. Wrigley C, H., Corke H &, C. E. Walker). Pp. 70-78. London: Elsevier.

USDA. (2007). National Nutrient Database for Standard Reference, Release 20. Available @ http://www.nal.usda.gov/fnic/foodcomp/cgi-bin/list_nut_edit.pl

Visconcelos, I. M., Maia, A. A. B., Siebra, E. A., Oliveira, J. T. A., Carvalho, A. F. F. U., Melo,
V. M. M., Carlini, C. R. & Castelar, L. I. (2001). Nutritional study of two Brazilian soybean (*Glycine max*) cultivars differing in contents of antinutritional and toxic proteins. *Journal of Nutritional Biochemistry*, **12**, 55-62.

Wiejzen, P. L. G., Zanstra, E. H., Alfieri, C. &, and de Graaf, C. (2008). Effects of complexity and intensity on sensory specific satiety and food acceptance after repeated exposure. *Food Quality and Preference*, **19**, 349-359.

World Health Organization. (2007). Protein and Amino Acid Requirements in Human Nutrition.WHO Technical Report Series No. 935. Geneva: World Health Organization.

Young, R. Y. & Pellet, L. P. (1999). Plant protein in relation to human protein and amino acid nutrition. *American Journal of Clinical Nutrition*, **59**, 1203S-12S.

Zandstra, E. H., Weegels, M. F., Van Spronsen, A. A. & Klerk, M. (2004). Scoring or boring? Predicting boredom through repeated in-home consumption. *Food Quality and Preference*, **15**, 549-557.

Ingredients	Sorghum: Soy			Soy	Wheat: Soy				
	100:0	71.4:28.6	50:50	28.6:71.4	100:0	100:0	71.4:28.6	50:50	28.6:71.4
Defatted soy flour (g)	0	64	112.5	161	225	0	64	112.5	161
Sorghum flour* (g)	225	161	112.5	64	0	0	0	0	0
Wheat flour (g)	0	0	0	0	0	225	161	112.5	64
Sugar (g)	56	56	56	56	56	56	56	56	56
Sunflower oil (g)	66	66	66	66	66	66	66	66	66
Baking powder (g)	1.5	1.5	1.5	1.5	1.5	0.25	0.5	1.0	1.0
Vanilla essence (g)	13.5	13.5	13.5	13.5	13.5	13.5	13.5	13.5	13.5
Water (g)	40	70	90	125	160	60	90	100	125
	(80)*		(180)*						
Total dough weight (g)	402	432	452	487	522	421	451	462	487
Individual biscuit weight (g)	25.9	23.7	22.4	21.2	18.2	30.7	27.4	24.2	21.2

Table 1 Formulation of the sorghum, bread wheat, soy and composite biscuit doughs

*Maximum particle size for sorghum flour reduced to 500 µm for sorghum: soy biscuits ratios 100:0 and 50:50 for consumer study and water content of dough doubled to make it workable.

Table. 2. Lexicon used by the descriptive	e sensory panel to evaluate fortified biscuits
Sensory Attributes and their definitions	Rating scales

Appearance

Surface colour intensity: From light cream to dark brown. Visible white specks: Quantity on biscuit surface. Rough surface: Degree perceived on surface of a biscuit. Evenness of surface: Degree on top surface.

Aroma/smell

Overall aroma strength: Intensity of aroma of the biscuit. Baked biscuit aroma: Intensity associated with basic sugar biscuit.

Roasted cereal aroma: Intensity associated with cereals sufficiently heated to caramelize sugars and starches. Roasted soy aroma: Intensity associated with roasted soy.

Heated oil aroma: Intensity associated with heated oil.

Texture

Roughness: Degree of abrasiveness perceived by the lips and tongue before chewing.

Bumpy texture: Overall amount of large bumpy areas on surface perceived by lips and tongue.

Hardness: Force required to compress a biscuit between molar teeth.

Crispness: Force and sound with which the sample raptures Denseness: Degree of compactness of cross section of sample after biting completely through

Dry: Degree to which the sample feels dry or absorbs saliva in mouth

Graininess: Amount of small particles perceived by the tongue when the mass is gently compressed between the tongue and palate

Coarseness: Degree to which the mass feels coarse or abrasive during product mastication

Chewiness: Number of chews (at 1 chew/sec) needed to masticate the sample to consistency suitable for swallowing:

Flavour

Sweet: Fundamental taste sensation associated with sugars. Roasted soy flavour: Intensity of flavour associated with roasted soy

Roasted cereal flavour: Intensity associated with cereals sufficiently heated to caramelize some starches and sugars. Doughy flavour: Intensity associated with uncooked dough.

Heated oil flavour: Intensity associated with heated oil

Baked biscuit flavour: Intensity of flavour associated with basic sugar biscuit.

Aftertaste

Sweet aftertaste: as described in sweet flavour Gritty residue in mouth: Degree to which mouth contains small particles after all of the sample has been swallowed Roasted soy flavor aftertaste: as described earlier.

Heated oil flavour: (as above)

Not dark = 0; very dark = 10. No white specks = 0; many white specks = 10. Not rough = 0; very rough = 10 Not uneven = 0; very uneven = 10.

No aroma = 0; Intense aroma = 10 No baked aroma = 0; Intense baked aroma = 10. No roasted cereal aroma = 0; Intense roasted cereal aroma = 10. No roasted soy bean aroma = 0; Intense roasted soy aroma = 10. No heated oil aroma = 0; intense heated oil

Not rough (smooth) = 0; very rough = 10.

Not bumpy = 0; very bumpy = 10.

Not hard = 0; very hard = 10.

aroma = 10.

Not crispy = 0; very crispy = 10. Not dense = 0; very dense = 10.

Not dry (moist) = 0; very dry = 10.

Not grainy = 0; very grainy = 10.

Not coarse = 0; very coarse = 10.

Not chewy = 0; very chewy = 10.

No sweet taste = 0: intense sweet taste = 10. No roasted soy flavour = 0; Intense roasted soy flavour = 10 No roasted cereal flavour = 0; Intense roasted cereal flavour = 10. No doughy flavour = 0; Intense doughy flavour = 10. No heated oil flavour = 0; Intense heated oil flavour = 10. No baked flavour = 0; Intense baked flavour = 10.

No sweet taste = 0; intense sweet taste = 10. Not gritty = 0; very gritty = 10

No roasted soy flavor = 0; Intense roasted soy flavour = 10. (As above)

Flours / Biscuits Mo	oisture	Protein	Fat	Ash (Crude fibre	Carbohydrate ¹	Energy ²
	()	V x 6.25)					(kJ/g 100 g)
Flours							
Sorghum flour 11.	$8^{g}\pm 0.3$ 1	$1.4^{c}\pm 0.1$ 3	$1^{\circ}\pm0.6$ 1	$.4^{c}\pm 0.0$	$2.0^{\circ}\pm0.0$	$70.3^{j}\pm0.2$	1504 ^b
Bread wheat flour 11.	$3^{g}\pm 0.2$ 1	$3.4^{d} \pm 0.0$ 1	.4 ^b ±0.1 0	$0.7^{b} \pm 0.0$	$0.2^{a}\pm0.0$	$73.2^{k}\pm0.1$	1520 ^c
Soy flour 6.2	$^{de}\pm 0.4$ 5	$0.1^{1}\pm0.1$ 0	$0.5^{a}\pm0.0$ 6	$5.2^{k}\pm0.1$	$7.5^{j}\pm0.1$	$29.5^{b}\pm0.1$	1372 ^a
Sorghum/Soy biscuits							
100:0 3.2	2 ^a ±0.4 9	$0.2^{a}\pm0.1$ 2	$1.0^{e} \pm 0.3$ 1	$.4^{c}\pm 0.0$	$1.7^{b}\pm0.4$	$63.5^{i}\pm0.4$	2013 ^j
71.4: 28.6 4.9	$bc \pm 0.2$ 1	$7.9^{e} \pm 0.4$ 20	$0.5^{e} \pm 0.1$ 2	$2.1^{e}\pm0.1$	$2.8^{d} \pm 0.1$	$51.8^{g}\pm0.4$	1943 ^h
50:50 3.8	^{ab} ±0.6 2	$4.7^{g}\pm0.4$ 19	$9.7^{d} \pm 0.6$ 2	$2.8^{g}\pm0.1$	$3.7^{f}\pm0.2$	$45.6^{e} \pm 0.3$	1924 ^{gh}
28.6:71.4 4.9	$bc \pm 0.2$ 3	$0.7^{i} \pm 0.3$ 19	$9.7^{d} \pm 0.2$ 3	$3.3^{i}\pm0.0$	$4.7^{h}\pm0.2$	$36.6^{\circ} \pm 0.2$	1873 ^{ef}
Wheat/Soy biscuits							
100:0 7.2	$e^{ef} \pm 0.7$ 1	$0.8^{b}\pm0.2$ 20	$0.7^{e} \pm 0.2$ 0	$0.5^{a}\pm0.0$	$0.2^{a} \pm 0.0$	$60.6^{h}\pm0.4$	1980 ⁱ
71.4: 28.6 7.4	l ^f ±1.5 1	$9.5^{f} \pm 0.2$ 19	$9.8^{d} \pm 0.9$ 1	$.5^{d} \pm 0.0$	$2.0^{\circ}\pm0.0$	$49.8^{f} \pm 0.8$	1910 ^g
50:50 6.4	$^{def}\pm 0.4$ 2	$5.8^{h}\pm0.4$ 19	$9.3^{d} \pm 0.5$ 2	$2.4^{f}\pm0.0$	$3.3^{e}\pm0.1$	$42.8^{d} \pm 0.5$	1880 ^f
28.6:71.4 6.0	^{cd} ±0.4 3	$1.9^{j} \pm 0.2$ 19	$9.4^{d} \pm 0.1$ 3	$5.1^{h}\pm0.0$	$4.4^{g}\pm0.2$	$35.2^{b}\pm0.3$	1859 ^{de}
Soy biscuit 100% 4.6	$b^{b} \pm 1.2$ 3	$9.9^{k}\pm 0.3$ 19	$9.3^{d} \pm 0.5$ 4	4.2 ^j ±0.1	$5.5^{i}\pm0.0$	$26.5^{a}\pm0.4$	1842 ^d

Table 3. The effect of compositing sorghum and bread wheat with defatted soy flour on biscuit proximate composition (g/100 g)

Values are means ±standard deviations. Values in a column with different letter superscripts are significantly different at P \leq 0.05 ¹Calculated as total carbohydrate by difference]. 100-(weight in grams [moisture + fat + protein + ash + crude fibre] in 100 g of food ²Calculated using the following factors: protein 17 kJ/g, fat 37 kJ/g, and carbohydrates 17 kJ/g

Flour/Biscuits	Lysine	Reactive lysine	IVPD ¹	Lysine ²	PDCAAS ³
	(g/100 g protein)	(g/100 g protein)	(%)	score	
Flours					
Sorghum flour	$1.98^{b}\pm0.05(0.23)^{4}$	$1.46^{b}\pm0.20$	$56.0^{b} \pm 1.1$	0.41	0.22
Bread wheat flour	$2.48^{\circ} \pm 0.00 (0.33)$	$1.89^{\circ} \pm 0.17$	$97.3^{h} \pm 1.5$	0.52	0.50
Soy flour	$8.28^{g} \pm 0.39 (4.15)$	$3.67^{g} \pm 0.08$	$97.5^{h}\pm0.0$	1.73	1.00
Sorghum/Soy biscuits					
100:0	$1.37^{a}\pm0.12$ (0.13	$0.69^{a}\pm0.40$	$30.0^{a} \pm 3.3$	0.29	0.09
71.4: 28.6	$3.93^{d} \pm 0.38 (0.70)$	$2.10^{cd} \pm 0.17$	$74.3^{\circ} \pm 1.6$	0.82	0.61
50:50	$4.54^{de} \pm 0.38 (1.12)$	$2.49^{de} \pm 0.44$	$81.1^{d} \pm 2.3$	0.95	0.77
28.6:71.4	$4.78^{\text{ef}} \pm 0.26 (1.47)$	$2.83^{\rm ef} \pm 0.07$	$87.3^{e} \pm 3.5$	1.00	0.87
Wheat/Soy biscuits					
100:0	$1.86^{b} \pm 0.19 (0.20)$	$2.25^{cd} \pm 0.05$	$96.7^{h} \pm 1.5$	0.39	0.37
71.4: 28.6	$4.20^{d} \pm 0.17 (0.82)$	$2.29^{d} \pm 0.18$	$95.5^{h}\pm1.1$	0.88	0.84
50:50	$4.69^{e} \pm 0.19 (1.21)$	$2.34^{d} \pm 0.56$	$94.2^{gh}\pm0.5$	0.98	0.92
28.6:71.4	$4.92^{\text{ef}} \pm 0.09 (1.57)$	$2.42^{d} \pm 0.15$	$91.4^{fg}\pm 2.5$	1.03	0.93
Soy biscuit 100%	$5.13^{\rm f} \pm 0.03 \ (2.05)$	$3.00^{\rm f} \pm 0.13$	$88.1^{ef} \pm 3.7$	1.06	0.93

Table 4. The effect of compositing sorghum and bread wheat with defatted soy flour on the protein quality of flours and biscuits

Values are means \pm standard deviations. Values in a column with different letter superscripts are significantly different at P \leq 0.05; ¹In vitro protein digestibility (IVPD)

²Lysine score= mg lysine in 1 g protein of test sample/48 mg lysine requirement for 3-10 year old child (WHO 2007)

³Protein Digestibility Corrected Amino Acid Score= lysine score x IVPD/100.

⁴Figures in brackets are g lysine/100 g flour or biscuit.