Orange-fleshed sweet potato-wheat composite breads: physico-chemical, sensory and nutritional quality

By

Madjaliwa Nzamwita

Submitted in partial fulfilment of the requirements for the degree

MSc (Agric) Food Science and Technology

in the

Department of Food Science

Faculty of Natural and Agricultural Sciences

University of Pretoria

Pretoria, South Africa

August, 2012

Declaration

I declare that the dissertation herewith submitted for the degree of MSc (Agric.) Food Science and Technology at the University of Pretoria, has not previously been submitted by me for a degree at any other University or institution of higher education.

Madjaliwa Nzamwita

Dedication

In loving memory of my late father Mr. Kaganda Djumatano.

To my mother Kadodo Zubeda.

To the Almighty God for the gift of life.

Acknowledgement

I would like to express my sincere gratitude and appreciation to the following people and organizations for their support during the execution of this MSc study:
Prof A. Minnaar my first supervisor for her excellent supervision, constant encouragement, guidance and her commitment to my work.

Dr. K.G. Duodu, my co-supervisor, for his scientific support, constructive criticism and technical support.

I am thankful to all academic staff in the Department of Food Science for their scientific inputs that were extremely important during the discussion of my results. These include food rheology, thermal properties of starch, protein functionality, processing of vegetables, principles of liquid chromatography, food dispersions, descriptive sensory analysis and statistical analysis of data.

My special thanks go to Mr. Bheki Dlamini for training me on the use of various instruments in the laboratory.

The Southern Africa Grain Laboratory and Anchor Yeast for allowing me to do various experiments on dough and bread on their premises.

The Republic of Rwanda, in particular the Ministry of Public Service and Labour and my employer, the Rwanda Agriculture Board, for offering me the opportunity to pursue my MSc studies.

The Department of Plant Production and Soil Science, University of Pretoria for allowing us to do the HPLC analysis of sugars on our samples.

African Development Bank (AfDB) and the Forum for Agricultural Research in Africa (FARA) for providing financial support during this study. I would like to extend my gratitude to the Department of Food Science for covering the cost of various experiments.

Dr Margaret McEwan, the coordinator for DONATA (Dissemination of New Agricultural Technologies in Africa) project and Dr Lydia Kimenye from ASARECA (Association for Strengthening Agricultural Research in Eastern and Central Africa) for their constant encouragement throughout this journey.

I am grateful to my family and friends for their love and support. Thanks to God for blessing and guiding me throughout this lengthy journey.

ABSTRACT

Orange-fleshed sweet potato-wheat composite breads: Physico-chemical, sensory and nutritional quality

By

Madjaliwa Nzamwita

Degree: MSc (Agric) Food Science and Technology

Supervisor: Prof A. Minnaar

Co-supervisor: Dr K.G. Duodu.

Vitamin A deficiency (VAD) is a public health problem especially in developing countries. The main strategy that has been used to fight against VAD was the distribution of high dose capsules of vitamin A to people at risk or already affected with VAD. The orange-fleshed cultivars of sweet potato (OFSP) are known to be a good source of provitamin A carotenoids especially β-carotene. However, fresh OFSP roots are perishable but processing them into flour may extend their shelf-life and could be used as an ingredient in baked products such as bread. It is evident that the consumption of bread in Rwanda, for example, is predominantly higher in urban areas than in rural areas. However, the distribution of vitamin A supplements to people at risk is done in both rural and urban areas. Thus, supplementing wheat flour bread with OFSP flour would contribute, to a certain extent, to the eradication of vitamin A deficiency in those areas. The current study aims at exploring the possibility of using OFSP flour in bread-making and the effect of baking on the β-carotene content of OFSP-wheat composite bread and the contribution of the latter to the vitamin A requirements in different groups of people.

Substituting wheat flour with OFSP flour increased the time required to reach optimum dough development. It also reduced the tolerance of the dough to mixing. The resistance of the dough to extension increased thereby reducing the extensibility of the dough by 26, 41, 64 and 79% at 10, 20, 30 and 40% substitution levels, respectively. Pasting parameters such as peak, breakdown and final viscosities were decreased probably due to the differences in the physical and chemical properties (granular size and shape and amylose/amylopectine ratio) of starches from OFSP flour and wheat flour. Bread containing OFSP flour displayed low oven spring, loaf volume and specific volume presumably due to low gluten content.

The retention of β-carotene, under baking conditions (190^{o}C for 30 min), significantly differed (p < 0.05) among all bread samples studied. An average of 34, 29 and 17% of β-carotene was lost in breads containing 10, 20 and 30 % OFSP flour, respectively. 9-*cis*-β-carotene and 13-*cis*-β-carotene were the main *cis* isomers of β-carotene formed during baking. Breads containing 10 or 20% OFSP would meet 28.9 and 61% respectively of the recommended dietary allowance (RDA) of vitamin A amongst children aged between 3-10 years. In contrast, the same amount of bread (100 g) supplemented with 30% OFSP flour would provide the amount of vitamin A close to the RDA of vitamin A for children aged between 3-10 years but it would meet less than 50% of the RDA of vitamin A for pregnant and lactating women.

The first two principal components (PC1 and PC2) explained about 98% of total variation in the 17 sensory attributes of OFSP-wheat composite breads. PC1 alone accounted for 83.8% of total variation among the breads and separated them based on the amount of OFSP flour they contained. PC2 accounted for 13.9 % of total variation of sensory data. The descriptive sensory panel detected increased sweetness, caramel aroma, sweet potato aroma and flavour in bread samples containing OFSP flour. Breads containing OFSP flour were described as dark (on the crust) probably due to the Maillard reaction and caramelisation.

Partial substitution of wheat flour with OFSP flour adversely affected both rheological properties of the dough and its bread-making performance. These effects tended to increase as the amount of OFSP flour increased. Bread containing 10% OFSP flour had oven spring value, loaf volume and specific volume close to the control (but statistically different, p < 0.05) when compared to its counterparts. The descriptive sensory analysis revealed distinct differences in the sensory profiles of OFSP-wheat composite breads as compared with wheat flour bread. The baking process causes detrimental effects to the nutritional quality of OFSP-wheat composite breads through the degradation of β-carotene by heat. Nevertheless, breads containing 20 and 30% OFSP flour were found to possess the potential to fight against vitamin A deficiency in children, pregnant and lactating women. However, it is not yet

known how consumers would perceive them, suggesting that a consumer acceptability study may be required.

Table of contents

Contents Pages

List of tables **Pages**

List of figures Pages

CHAPTER 1: INTRODUCTION

Vitamin A deficiency (VAD) is a major nutritional concern in poor societies, especially in lower income countries (WHO, 2009). Bovell-Benjamin (2007) reported that more than 230 million of the world's children have inadequate vitamin A intake, with 13 million of them being affected by night blindness. Research conducted by the Rwandan Ministry of Health in collaboration with the National Institute of Statistics in 2007-2008 revealed that 26% of children under the age of 6 months, 21% of children between the age of 6-12 months, 25% of children below the age of 5 years and 7% of women of reproductive age were affected by vitamin A deficiency. The Ministry of Health is currently providing vitamin A supplements to children and pregnant women to try and overcome this problem. The main strategy for combating vitamin A deficiency has been to distribute massive dose capsules to people identified as vitamin A deficient which requires appropriate infrastructure and qualified personnel. It is, therefore, important to find a long term solution to deal with this problem that may involve a cheap and rich source of vitamin A.

Orange-fleshed sweet potatoes (OFSP) are a rich source of vitamin A (in the form of provitamin A carotenoids) (van Jaarsveld, De wet Marais, Harmse, Nestel and Rodriguez-Amaya, 2006) and can be grown in different agroecological zones (Stathers, Namanda, Mwanga, Khisa and Kapinga, 2005). They are resistant to environmental stresses, diseases and are easily propagated through vines (Stathers, Namanda, Mwanga, Khisa and Kapinga, 2005).

Sweet potatoes are a common staple food in many developing countries, including Rwanda, but there are various problems related to storage owing to high perishability (Ahmed, Akter and Eun, 2010) and transport of the fresh roots (Greene and Bovell-Benjamin, 2004). These problems can be solved by processing them into flour, which is less bulky and more stable than the highly perishable roots (Ahmed, Akter and Eun, 2010). Sweet potato flour from β-carotene rich cultivars such as OFSP can be utilised in baked products with extended shelf-life, improved nutritional quality and sensory properties. These include products such as bread, biscuits and cakes.

In Rwanda, wheat flour is used to produce bread and the consumption of bread is high in urban areas compared to rural areas. However, the distribution of vitamin A supplements to people at risk is done in both rural and urban areas. Wheat-orange fleshed sweet potato composite bread could therefore contribute, to a certain extent, to the eradication of vitamin A deficiency in urban areas. The consumption of vitamin A fortified bread has been found to decrease the percentage of Philipino school children with inadequate liver vitamin A stores from 28.6 to 15.3%, a decrease of approximately 50% (Solon *et al*., 2000). The use of OFSP flour in baked products can also contribute to the reduction of amount of wheat flour imported from foreign countries since the latter is relatively expensive.

β-carotene, which is the most abundant of the carotenoids in orange-fleshed sweet potato (Kimura, Kobori, Rodriguez-Amaya and Nestel, 2007) with the highest vitamin A activity, can be negatively affected during heat processing (Chandler and Schwartz, 1988). The unsaturated structure of carotenoids renders them more susceptible to degradation by heat, oxygen and light (Leskova *et al*., 2006). Although raw orange-fleshed sweet potato root may contain high amount of β-carotene, their contribution to vitamin A intake will definitely depend on the amount that has been retained after processing or preparation into food.

During the current study, the effect of substituting different levels of wheat flour with orange-fleshed sweet potato flour on the rheological properties of the doughs, their bread-making performance and sensory properties of the resulting breads will be determined. Sweet potatoes (especially those with coloured flesh) are considered as an excellent model system for quantitatively studying changes in β-carotene since they contain high concentrations of β-carotene and low concentrations of other carotenoids (Chandler and Schwartz, 1988). The effect of baking on the β-carotene content of OFSP-wheat composite bread and the contribution of the latter to the vitamin A requirements will be investigated.

CHAPTER 2: LITERATURE REVIEW

In this review, focus is put on the importance of sweet potato nutritionally and economically in the developing countries especially in sub-Saharan Africa. It highlights the potential impact of OFSP in the fight against vitamin A deficiency with specific cases in different countries where various studies have shown promising results with regard to the use of OFSP in combating VAD. An overview of health benefits of vitamin A, β-carotene and other carotenoids is given. The bread-making process and the effect of heat treatments on β-carotene are also discussed.

2.1 The sweet potato root

Sweet potato, *Ipomoea batatas L. (Lam),* is a dicotyledonous plant which belongs to the family of Convolvulaceae (Woolfe, 1992). It has been suggested (Austin, 1987) that sweet potato originated from Yucatan peninsula of Mexico and the mouth of the Orinoco River in Venezuela. Malnutrition, food insecurity, droughts and limited agricultural technologies are among major problems facing the population in developing countries (Bovell-Benjamin, 2007). Using conventional breeding techniques, sweet potato varieties with high provitamin A carotenoids can be developed (van Jaarsveld *et al.*, 2006). The sweet potato breeding process aims at obtaining sweet potato varieties with high yield, high total carotenoid content with sweet taste and acceptable texture (Laurie, Van Den Berg and Tjale, 2009). Under HarvestPlus, provitamin A-rich sweet potato varieties are identified and further bred using the best traditional methods and modern biotechnology to achieve high concentrations of provitamin A carotenoids (Kimura, Kobori, Rodriguez-Amaya and Nestel, 2007). OFSP is among the biofortified sweet potatoes bred for their high nutritional value in terms of provitamin A carotenoids (Bengtsson, Namutebi, Alminger and Svanberg, 2008).

Sweet potato is globally, the second most economically important root crop after potato and is an important food security crop in many of the poorest regions of the world including sub-Saharan Africa. Sweet potato has the third greatest production after cassava and yams. It is amongst the most widely grown of the major root crops in sub-Saharan Africa and covers an estimated 2.1 million hectares with an annual estimated production of 9.9 million tonnes of roots (Stathers *et al*., 2005). Sweet potato is also an important food crop in eastern and central

Africa. The crop is particularly important in the densely populated countries in the Lake Victoria zone particularly Rwanda, Uganda, and Burundi, where it plays more prominent and diversified roles in the food system (Lemaga, Mwanga and Owori, 2007). In Rwanda, sweet potato is a staple food in rural areas especially amongst the farmers and low income population. It is mainly grown in the mash land and harvested progressively depending on the need for consumption. Part of the harvest can also be sold to the market in order to sustain the family financially.

In monetary terms, sweet potato ranks thirteenth globally in the production value of commodities and is fifth on the list of developing countries' most valuable food crops (Laurie, 2004). The annual world production was 140.9 million tonnes (Mt) in 2000 with 91 % produced in Asia (128.8 Mt), 7% in Africa (9.1 Mt), 1% in Central and North America (1.1 Mt), 1% in South America (1.2 Mt), 0.5% in Oceania (0.59 Mt) and only 46.000 t in Europe. Furthermore, the crop accounts for about one third of the production of root and tuber crops in developing countries. China is by far the largest producer, accounting for 80 % of the supply. The total area under production is 9.6 million hectares and the average yield is 13.5 t/ha (Laurie, 2004).

Sweet potato in the eastern and central African region is mainly used as food for human consumption and to a lesser extent as animal feed. The crop is mainly consumed in the fresh form as boiled/steamed and roasted roots. Boiled roots can also be served as mashed, stewed and fried food (Lemaga *et al.*, 2007). Sweet potato flour, fresh-grated and boiled and mashed sweet potato roots are the types of sweet potato primary products that are used as ingredients for processing of value-added sweet potato processed products (Owori and Hagenimana, 2007). According to Stathers *et al.* (2005), sweet potato consumption in the past was considered as a sign of poverty and was consumed in secrecy by people in the urban areas. With increasing urbanisation and consumer awareness about health benefits associated with sweet potato consumption, sweet potato is becoming increasingly important in urban food systems and there has been a tremendous positive change in attitude towards the crop.

2.2 The potential of orange-fleshed sweet potato for combating vitamin A deficiency in developing countries

Strategies to control vitamin A deficiency include dietary diversification, food fortification and vitamin A supplementation. Dietary diversification includes the production of β-carotene–rich crops, such as orange-fleshed sweet potato (Van Jaarsveld *et al*., 2005). Orange-fleshed sweet potato is an excellent source of β-carotene (Table 2.1) that provides sufficient vitamin A to meet the daily requirements in the diet. The orange-fleshed sweet potato varieties can make important contributions to alleviating vitamin A deficiency, which is currently a major health problem in most developing countries. Sweet potato roots contain lysine, the important amino acid which cereals are deficient in. The leaves and tips are good sources of vitamin A, energy, zinc, calcium and protein. Sweet potato leaves contain twice the amount of protein as the same in weight of roots (Lemaga *et al*., 2007).

The estimates of potential impact of orange-fleshed sweet potato in improving the vitamin A status among the population at risk in sub-Saharan Africa have been made (Low, Walker and Hijmans, 2001). These estimates are applicable when white-fleshed sweet potato varieties will have been fully substituted with orange-fleshed ones. It was shown that in Tanzania, about half of the population at risk attains the full-impact outcome. Full impact is equivalent to an increase in provitamin A intake of 40% of the recommended dietary allowance. In Kenya 72.6% receives partial benefit while in countries like Rwanda, Uganda and Burundi more than 80% of the population at risk receives full benefit. In Ethiopia and South Africa, the mean intake increases by only a modest 2% of the recommended dietary allowance (Low *et al*., 2001).

Increased awareness and sensitisation on the benefits of OFSP have raised the demand in the countries where it was introduced (Kapinga, 2007). Various studies conducted in different countries where OFSP was introduced have proven that it can play a major role in reducing vitamin A deficiency. The efficacy of daily consumption of boiled and mashed OFSP in improving the vitamin A status of primary school children in South Africa has been assessed (van Jaarsveld *et al.*, 2005). Children aged between 5 and 10 years were randomly assigned to two groups. The first group comprised of 90 children consumed 125 g boiled and mashed OFSP for each child. The second group with 90 children consumed an equal amount of white-fleshed sweet potato variety deprived of β-carotene for 53 school days. All children were dewormed to eliminate any helminthic infection. The proportions of children with

Table 2.1 Mean value for the nutrient content (per 100 g, db) of different orange-fleshed sweet potato cultivars (adapted from Leighton, 2007)

Nutrient	**Unit**	**W119** **n=2**	**Jewel** **n=4**	**Resisto** **n=2**	**A15** **n=2**	**Composite** **n=3**
Ash	G	1.20	0.98	1.04	0.97	0.77
Dry matter	G	16.8	20.9	23.1	19.8	23.6
Moisture	G	83.2	79.1	76.9	80.2	76.4
Protein	G	2.02	1.40	0.92	0.70	1.42
Fat	G	0.16	0.25	0.23	0.18	0.27
Carbohydrate	G	13.4	18.2	20.9	17.9	21.1
Energy	kJ	265	339	375	320	389
β-carotene	µg	4323	6853	9230	6880	-
Vitamin C	Mg	5.91	6.37	8.56	7.45	7.58
Calcium	Mg	65.5	115	143	105	80.0
Magnesium	Mg	18.5	19.2	16.0	13.5	26.7
Phosphorus	Mg	57.5	57.5	71.5	60.5	41.0
Potassium	Mg	362	324	328	348	2.19
Iron	Mg	0.64	0.71	0.45	0.74	0.75
Manganese	Mg	0.14	0.14	0.15	0.77	0.03
Zinc	Mg	4.18	3.96	3.95	2.85	3.57
Copper	Mg	0.02	0.11	0.15	0.11	0.13

Db: Dry basis.

normal vitamin A status in the treatment group tended to increase from 78% to 87% (p = 0.096) and did not change significantly (from 86% to 82%) in the control group (p = 0.267).

In Bangladesh, Haskell *et al.* (2004) evaluated the effect of 60 days of daily supplementation with 750 µg retinol equivalents (RE) of either cooked, puréed OFSP, cooked, vitamin A as retinyl palmitate and synthetic β-carotene in Bangladesh men. The plasma retinol concentrations in the sweet potatoes (1.24 ± 0.06 μmol/L), vitamin A (1.24 ± 0.07 μmol/L) and β-carotene (1.49 ± 0.07 μmol/L) groups were significantly higher than the mean concentration in the control group (1.12 ± 0.05 μmol/L) ($p < 0.004$).

The intervention study in Mozambique did not only focus on improving the vitamin A status of the participants but it also enhanced the wellbeing of the farmers in the intervention area through increased OFSP production (Low *et al*., 2007). They pointed out that the percentage of intervention households selling sweet potatoes increased from 13% to 30% and in 2004 OFSP was the cheapest source of vitamin A in the market. Households in farmers' groups expanded the area under production from 33 to 359 m^2 within 2 years with 73% increase in sweet potato production. The median intake of vitamin A was almost 8 times higher in the intervention children (426 μg RAE) compared with control children (56 μg RAE). OFSP contributed 35% to the vitamin A intakes of all children in the intervention area and 90% in those who consumed it the previous day.

2.3 Health benefits of vitamin A and carotenoids

Vitamin A occurs naturally in animal products such as meat, milk and eggs (Potter and Hotchkiss, 1995). Plant food contains no vitamin A but contains its precursors (Fig. 2.1) such as β-carotene (Potter and Hotchkiss, 1995), α-carotene and β-cryptoxanthin (Palace, Qin and Singal, 1999). Vitamin A is fat soluble and its absorption by the body depends on the normal absorption of fat from the diet. Vitamin A deficiency can lead to blindness, failure of normal bone and tooth development in the young. Other diseases include the diseases of the epithelial cells and membranes of the nose, throat and eyes, which can decrease the body's resistance to infection (Potter and Hotchkiss, 1995). Vitamin A is essential for normal maintenance and functioning of body tissues and for the growth and development of the foetus (WHO, 1998). It also plays an important role in the vision cycle (Fig. 2.2).

Within the human body, vitamin A can be found as retinol, retinal and retinoic acid. It is primarily stored as long chain fatty esters and as provitamin A carotenoids in the liver, kidney and adipose tissue (Palace *et al*., 1999). β-carotene, α-carotene, γ-carotene, lycopene, lutein, β-cryptoxanthin, zeaxanthin and astaxanthin are the most common carotenoids in the diet but only β-carotene, α-carotene and β-cryptoxanthin can be converted into retinol (the active form of vitamin A) in the body (Kiss, Kiss, Milotay, Kerek and Markus, 2005). Vitamin A

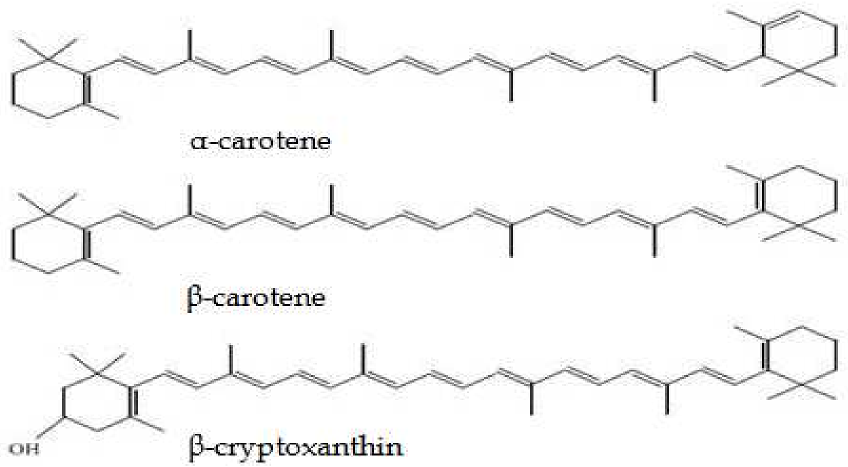


Figure 2.1 Provitamin A carotenoids (Liu, 2007)

and carotenoids are effective antioxidants for inhibiting the development of heart disease (Palace *et al*., 1999). For a carotenoid compound to be denoted as antioxidant, it should demonstrate the ability to prevent oxidative reactions that bring about damage in the organism by reacting with oxidising agents and free radicals (Woodall, Lee, Weesie, Jackson and Britton, 1997).

β-carotene and other carotenoids such as lycopene, α-toccopherol, lutein and zeaxanthin have a certain degree of antioxidant activity (Deshpande, Deshpande and Salunkhe, 1996). Unlike carotenoids such as β-carotene, α-carotene and β-cryptoxanthin, lutein and zeaxanthin are not provitamin A carotenoids. However, they are the only two carotenoids present in the retina and have been reported to play a prominent role in preventing ocular diseases notably age-related macular degeneration by inhibiting the oxidative damage induced by blue light and thus reduce the risk of cataract formation (Krinsky and Johnson, 2005).

The vitamin A derivative, retinoic acid, is a major signal controlling compound that regulates various biological processes (von Lintig *et al*., 2005) through specific nuclear receptors (FAO/WHO, 2002). All-*trans* and 9-*cis*-isomers of β-carotene are precursors for all-*trans*- and 9-*cis*-retinoic acid respectively. All-*trans*-retinoic acid binds preferentially to the retinoic nuclear receptor (RAR), while 9-*cis*-retinoic acid is the preferred ligand for the retinoid X receptor (RXR) (Ferruzzi, Lumpkin, Schwartz and Failla, 2006). Once activated, these receptors bind to DNA response elements located upstream of specific genes to regulate the level of expression of those genes. The retinoid-activated genes are known to regulate the synthesis of a large number of proteins essential to maintaining normal physiological functions (FAO/WHO, 2002). In addition, the binding of these ligands to their respective receptors is necessary for reproduction, growth, maintenance of skin and mucous membranes, and immune functions (Ferruzzi *et al.,* 2006).

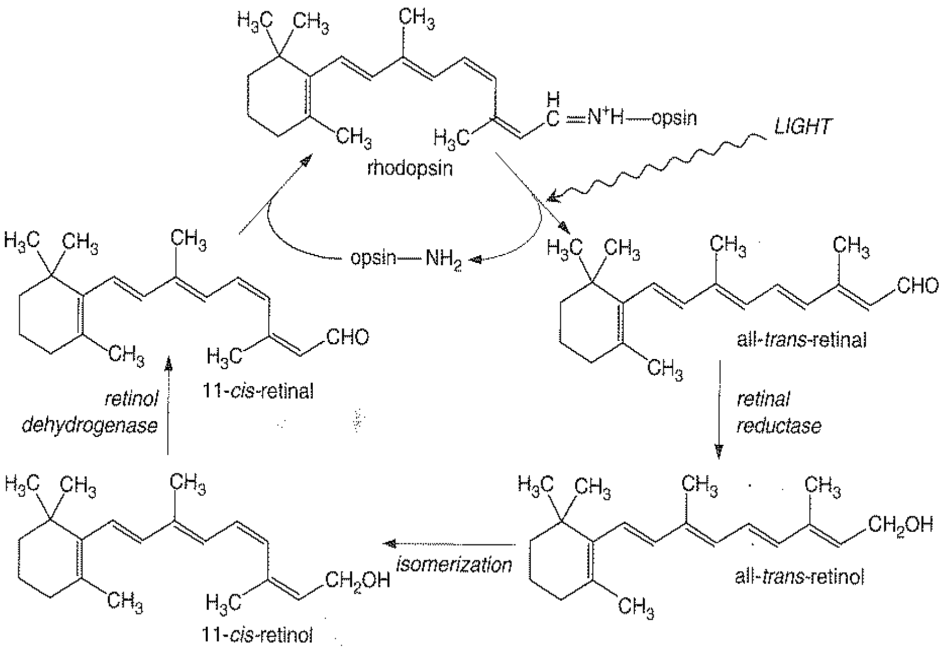


Figure 2.2 Vitamin A in the visual cycle (adapted from Coultate, 2009)

In the dark, 11-*cis*-retinal combines with the protein opsin to form rhodopsin. When this absorbs light (wavelength 500 nm) the *cis* bond isomerises to *trans* and ultimately the all-*trans* retinal detaches from the protein. During the course of this isomerisation the complex processes which lead to the generation of the nerve impulse are initiated. The 11-*cis*-retinal is then regenerated to recombine with opsin in the darkness (Coultate, 2009).

2.4 Sweet potato flour

Sweet potato flour is generally prepared by drying the peeled slices in a hot air drier or by drum drying of cooked sweet potato mash into flakes followed by milling and sieving (Yadav, Guha, Tharanathan and Ramteke, 2006). However, in some parts of East Africa sweet potato is traditionally processed into dried chips and/or flour to preserve the roots for household food security and to a lesser extent for sale in rural markets (Stathers *et al*., 2005). Sweet potato flour can be used as a source of carbohydrates, dietary fibre (Table 2.2), β-carotene, minerals and can also add natural sweetness, colour and flavour to processed products (van Hal, 2000). In countries where wheat is not produced in sufficient amounts to meet domestic demands, sweet potato flour can serve as a low cost alternative to imported wheat flour (Bovell-Benjamin, 2007).

Table 2.2 Chemical composition of sweet potato flour from different sources (adapted from Van Hal, 2000)

	Colour of the flour			
	White	Orange	Cream	Gray
Moisture (%)	6.87-7.7	7.86-8.85	6.5-7.8	5.6-6.01
Protein (% db)	1.66-1.86	4.61-6.48	4.1-8.5	-
Fat (% db)	-	0.48-0.52	-	-
Total carbohydrates/starch (% db)	72.6-82 S	89-91 C	64-74 S	72-90 S
Total sugar (% db)	13.8-23	-	6.9-9.7	-
Reducing sugars (% db)	-	-	-	6.26-24-39
Dietary fibre (% db)	2.53-4.23	2.17-2.26	10.7-13.8	3.63-7.08
Ash (% db)	1.66-1.86	2.11-2.89	1.3-2.53	2.52-3.91

*C= total carbohydrate content and S= total starch content. Db= dry weight basis. -: Not reported.

Besides permitting better preservation, the drying and processing of sweet potato into dried chips and flours offers other opportunities such as (Stathers *et al*., 2005):

- facilitating storage and transport;
- reducing bulkiness and losses due to high perishability of fresh roots (if roots are left in the ground further weevil attack can occur; this damage increases the risk of other pathogens contaminating and destroying the roots);
- increased shelf life;
- greater nutritive value due to the fact that as a great part of the water content is removed, the carbohydrates, pectin, proteins, oils and mineral salts are concentrated in the tissues of dried products;

- creating new income opportunities for farmers such as new markets and new sources of income;
- changing some of the negative attitudes about sweet potato consumption and enabling them to see sweet potato as an important commercial crop with a diverse range of uses and consumers.

Creation of new economic and employment activities for farmers and rural households can be achieved by processing sweet potato roots into flour and can provide nutritional value to food system (Bovell-Benjamin, 2007). The main processes involved in the production of sweet potato flour are shown in Figure 2.3.

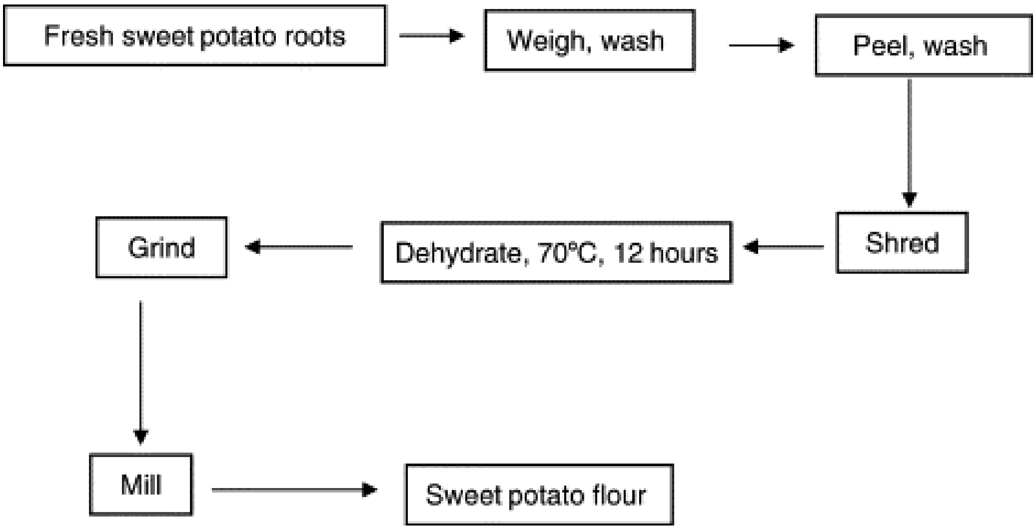


Figure 2.3 Production of sweet potato flour (Dansby and Bovell-Benjamin, 2003)

In some African countries, sweet potato flours have been produced in different ways depending on the traditions and their utilisation in various products. Bovell-Benjamin (2007) reported that women in Uganda crush and sun dry chunks of the fresh root to prepare a coarse *inginyo*. For *amukeke* chips, the men slice up the roots into round, flat pieces, which the women then spread out to dry; both keep for 4–5 months. The *inginyo* or *amukeke* are ground into coarse flour, which is rehydrated with water, boiled, mashed and then eaten directly as a thick porridge known as *atapa*, a starchy staple.

2.5 Wheat flour

Wheat four is obtained by reducing the particle size of wheat kernels using either a hammer mill or roller mill. It involves the separation of the bran and germ from the starchy endosperm so that a flour, in which the endosperm cells predominate, can be obtained (Dewettinck *et al*., 2008; Belitz and Grosch, 1999). Dewettinck *et al*. (2008) reported that wheat bran and germ are rich in proteins, fat, B vitamins and minerals, while the milled product contains less of these than the original grain. The aleurone layer, which is rich in protein, minerals and vitamins, contributes significantly to the nutritional quality of the bran fraction. However, wheat proteins generally contain less lysine, arginine, aspartic acid, glycine and alanine, but more glutamine and proline, as the outer layer proteins (albumins and globulins) are richer in these amino acids. The effect of extraction rate on the nutrient content of wheat grain is shown in Table 2.3.

Wheat flour and particularly wheat starch is widely used in food industries (Rojas, Rosell and de Barber, 1999). The unique properties of wheat flour to form a viscoelastic dough capable of retaining the gas and expand are due to the characteristics of wheat gluten (Roccia, Ribotta, Perez and Leon, 2009). It has long been established that the bread-making quality of wheat flour is related both to the quantity and the quality of its protein. The viscoelastic properties of gluten are believed to arise mainly from the properties of and interactions between the proteins comprising its two main sub-fractions, gliadin and glutenin (Khatkar, Bell and Schofield, 1995).

Since bread-making involves progressive increase in temperature of the dough, thermal properties of the flour play a major role in the baking process. Complete gelatinisation of starch is considered as the first quality index of soft baked products such as bread and should be taken as a minimum baking index; otherwise, sensory quality of the product would not be ensured (Zanoni, Peri and Bruno, 1995). Blazek and Copeland (2008) stated that in wheat starch, amylopectin contributes to water absorption, swelling and pasting of starch granules, whereas amylose and lipids tend to retard these processes. However, thermal properties of flour are more complex due to the presence of other flour components. Protein forms a complex with starch especially on the surface of starch granules which may interfere with increase in the viscosity of the flour during the gelatinisation process (Olkku and Rha, 1978).

Sugars inhibit the swelling of starch granules thereby retarding the gelatinisation process. Sucrose for example ties up water molecules making them unavailable for starch. Water insoluble pentosans (fibre) absorb 4.8 times their weight of water in a gluten system, 6.9 times in a starch-water system and 6.5 times in reconstituted gluten-prime starch dough. This indicates that owing to their high affinity for water, pentosans can also interfere with water absorption of starch during gelatinisation (Olkku and Rha, 1978).

Table 2.3 Chemical composition (db) of wheat flour as a function of the extraction rate (Adapted from Dewettinck *et al*., 2008)

	Extraction rate (%)						
	100	**95**	**91**	**87**	**80**	**75**	**66**
Starch + sugar (%)	69.9	73.2	75.3	77.2	80.8	82.9	84
Protein ($n \times 6.25$) (%)	14.2	13.9	13.8	13.8	13.4	13.5	12.7
Fat (%)	2.7	2.4	2.3	2	1.6	1.4	1.1
Dietary fibre (%)	12.1	9.4	7.9	5.5	3	2.8	2.8
Ash (%)	1.8	1.5	1.3	1	0.7	0.6	0.5
Energy (kJ/g)	18.5	18.5	18.5	18.5	18.5	18.4	18.3
Phosphorus (mg/g)	3.8	3.3	2.8	2.1	1.5	1.3	1.2
Calcium (mg/g)	0.44	0.43	0.38	0.33	0.27	0.25	0.23
Zinc (ppm)	29	25	21	18	12	8	8
Copper (ppm)	4	3.7	3.4	2.8	2.4	1.6	1.3
Iron (ppm)	35	33	28	23	15	13	10
Thiamine (µg/g)	5.8	5.4	–	4.8	3.4	2.2	1.4
Riboflavin (µg/g)	0.95	0.79	–	0.69	0.46	0.39	0.37
Niacin (µg/g)	25.2	19.3	–	10.1	5.9	5.2	3.4
Pyridoxine (µg/g)	7.5	6.6	–	3.4	1.7	1.4	1.3
Biotin (µg/g)	116	108	–	106	76	46	25
Folic acid (µg/g)	0.57	0.53	–	0.45	0.11	0.11	0.06

Db: Dry basis.

2.6 Composite flours

Composite flour is a mixture of wheat flour with flours from other crops such as cereals, legumes (Shahzadi, Butt, ur Rehaman and Sharif, 2005) and roots. Wheat flour is commonly mixed with flours from different sources (Shittu, Raji and Sanni, 2007) to produce bread and other baked products. The use of composite flours has been encouraged in various countries especially in developing countries to reduce the importation of wheat flour which is more expensive by substituting it partially with flour from locally grown crops (Shittu *et al.*, 2007; Aziah and Komathi, 2009).

Wheat flour has been partially replaced with flours from different crops such as cassava flour in bread (Khalil, Mansour and Dawoud, 2000; Shittu *et al*., 2007), pumpkin flour in crackers (Aziah and Komathi, 2009), sweet potato flour in bread, cakes and cookies (Owori, Nungo, Kapande, Mukantwali and Randrianaivoarivony, 2007) and soya bean flour in bread (Khetarpaul and Goyal, 2009). According to Stathers *et al.* (2005), enriched weaning flour can be made from mixing sweet potato, soya beans and sorghum or millet flours into a porridge.

Compositing wheat flour with OFSP flour may result in bread with increased sweetness, crust browning due to the presence of sugars and sweet potato flavour. The perceived bread flavour depends on the type of bread, ingredients, processing method and shelflife. Bread contains more than 300 volatile compounds that play a key role in the perception of bread flavour (Heenan, Dufour, Hamid, Harvey and Delahunty, 2009a). It is therefore difficult to associate a perceived flavour to a particular flavour compound as it can be a result of a combined effect of various chemical substances present in bread. Bovell-Benjamin (2007) reported that sweet potato flours vary widely in colour depending on genotype and when used in wheat-based composite flours they may impart characteristic colours, which may be favourable or unfavourable for particular food products. The presence of β-carotene may confer bread an orange coloration which would be more pronounced in the crumb. The increase in the firmness may result from an increase of dietary fibres from OFSP flour (van Hal, 2000) which may also interfere with the gluten network resulting in the depression of bread volume (Morrice and Morrice, 2012).

One of the main reasons for substituting wheat flour with other types of flours is to improve the nutritional quality of the final product. Orange-fleshed sweet potato flour can be a good choice for improving the nutritional value of baked products such as bread. Although it has low protein content it can contribute to increasing the vitamin A content in the form of provitamin A carotenoids.

2.7 Bread-making process

The bread-making process involves the cooking or baking of dough obtained by mixing wheat flour (to which other flours can be added as in the case of composite bread), salt (table salt) and potable water (drinking water), fermented by species of budding yeast used in baking such as *Saccharomyces cerevisae* and with or without the inclusion of any special component (Moreno-Alvarez *et al.*, 2009). A mixture of water, flour, yeast, salt and other ingredients are kneaded to make a dough (Alais and Linden, 1991) either by hand or a kneading machine. The fermenting activity of yeast specialised for bread making is optimum at about 30^{o}C and above 47^{o}C yeast dies rapidly. During the fermentation stage β-amylase attacks the starch molecules to form maltose and dextrins. The subsequent cleavage of starch chains by α-amylase from wheat flour (Gray and Bemiller, 2003) produces more dextrins, which give β-amylase a new substrate (Alais and Linden, 1991). The yeast produces carbon dioxide and ethanol that expand the air bubbles (10^{2}-10^{5}/mm^{3}) developed in the dough during kneading. The characteristics of the flour determine the fermentation tolerance, i.e. the maximum or minimum time after which the fermentation has to be stopped and the dough loaded into the oven. Dough fermentation of a weak gluten flour is rapid, but its fermentation tolerance is low (Belitz and Grosch, 1999). At the end of the fermentation process, the dough is baked in the oven at about 250^{o}C (Mondal and Datta, 2008) for 20-30 min (Alais and Linden, 1991).

The following changes take place during baking (Alais and Linden, 1991):

- Expansion in the volume of the bread occurs as a result of a speed-up in CO_2 production at the same time a film forms on the surface of the dough as a precursor of the crust. These two changes stop when the internal temperature rises to about 60^{o}C, whilst the alcohol produced evaporates.

- The gluten proteins denature and coagulate from 70°C and lose their affinity with water, which then moves towards the starch. This water plays an important part, together with CO_2, in the grain structure of bread.

- When the temperature reaches 90-100°C the formation of the crust starts taking place and water inside the crumb is transformed into vapour. The temperature of the crumb remains slightly below 100°C.

- Dextrinisation takes place, then caramelisation also occurs when the temperature reaches 110°C thereby contributing to the browning of the crust. The Maillard reaction between free amino groups of certain amino acids such as lysine and the carbonyl group of reducing sugars also play an important role in the colour formation in the crust.

- During the course of baking, the gelatinisation of starch begins at about 70°C where the structure of starch changes from the semi-crystalline state to the amorphous state and becomes more hydrophilic. The amylolysis stops when the β-amylase is rendered inactive (at about 75°C). Due to the fact that α-amylase is more thermoresistant its activity disappears at about 85°C. When the enzyme activity is high, the excess of small molecules (dextrins and maltose) makes the crumb sticky and the crust becomes deeply coloured (brown).

When the hot bread is cooled it starts to go stale (Alais and Linden, 1991; Coultate, 2009). Starch retrogradation starts taking place when the temperature drops to 60°C whereby starch molecules start to re-associate themselves in a more ordered manner or crystalline state (Alais and Linden, 1991). This is followed by a release of water and the crumb becomes dry and hard whereas the crust becomes soft and leathery (Gellynck, Kühne, Van Bockstaele, Van de Walle and Dewettinck, 2009). The rigidity of the stale bread can be partially overcome by reheating to about 60°C whereby weak bonds become dissociated and the aroma molecules adsorbed on the dried crumb are released (Alais and Linden, 1991).

However, the incorporation of flours from other sources into wheat flour has negative effects on gluten network formation, extensibility and gas retention of the dough as well as the final product quality. The addition of flours devoid of gluten to wheat flour results in the weakening of the gluten network due to the dilution effect on gluten. Hathorn *et al.* (2008) pointed out that lack of gluten limits the amount of sweet potato flour that can be incorporated with wheat flour as it can lead to changes in the texture of bread and reduced loaf volume. Sweet potato is known to be a rich source of dietary fibre (Table 2.2) but the latter may interfere with the formation of gluten network thereby resulting in increased dough mixing time (Peressini and Sensidoni, 2009; Stojceska and Ainsworth, 2008), reduced dough elasticity (Ade-Omowaye, Akinwande, Bolarinwa and Adebiyi, 2008) and loaf volume (Stojceska and Ainsworth, 2008).

2.8 Bread staling

Staling is a process of gradual deterioration of bread that starts after baking. It is characterised by a loss of flavour and an increase in crumb firmness (Pozo-Bayón, Guichard and Cayot, 2006.). Bread loses its sensory quality resulting in a negative perception from the consumer. Bread staling is also responsible for huge economical losses to the baking industry because large quantities of stale bread are discarded (Robotta and Le Bail, 2007)

Bread staling has been studied intensively, but it is still not yet well understood (Robotta and Le Bail, 2007; Gray and Bemiller, 2003). This is probably due to the fact that ingredients present in the dough contain several components each of which may undergo changes during bread-making and during the aging of bread thereby making the staling process more complex (Gray and Bemiller, 2003). Thus, the effect of compositing wheat flour with OFSP flour on bread staling is not well known and needs to be thoroughly investigated.

The use of enzymes can be used to successfully delay the staling process in bread (Armero and Collar, 1998). One of the useful enzymatic approaches to delay bread staling is the use of α-amylase (Pozo-Bayón *et al.*, 2006) which may catalyse a small portion of hydrolysis of starch thereby keeping the freshness of bread for a longer storage time (Giménez, Varela, Salvador, Ares, Fiszman and Garitta, 2007). Hydrocolloids such as alginate and hydroxypropylmethylcellulose have been found to possess an antistaling effect and to delay crumb hardening (Guarda, Rosell, Benedito and Galotto, 2004). The softening effect of hydrocolloids is attributed to their water retention capacity, a possible inhibition of the amylopectine retrogradation (Guarda *et al.*, 2004) and the weakening of starch structure due to the inhibition of the amylose chain association (Pozo-Bayón *et al.*, 2006).

2.9 Effect of heat treatments on β-carotene

Conventional oven baking, boiling and microwaving are the most widely used methods for sweet potato processing at home and in food-service establishments (Padda and Picha, 2008). Thermal processing relies on high temperatures which in turn, may cause detrimental effects on the nutritional quality of food. As a major precursor of vitamin A, it is important to understand the implications of heat treatments on β-carotene. Reduction in the β-carotene content has been observed during processing and has been associated with the production of *cis* isomers of β-carotene (Chandler and Schwartz, 1988; Bechoff, Dhuique-Mayer, Marouzé, Reynes and Westby, 2009).

Carotenoids, including β-carotene, are predominantly found in nature in the all-*trans*-configuration (Lessin, Catignani and Schwartz, 1997). The most predominant isomers of β-carotene include 9-*cis*-β-carotene, 13-*cis*-β-carotene and 15-*cis*-β-carotene (Fig. 2.4). Heat treatments are known to change the configuration of β-carotene through isomerisation to a *cis* configuration (Grabowski, Truong and Daubert, 2008). As the number of *cis* isomers of provitamin A carotenoids increases, there is a decrease in the nutritional value of food, since *cis* isomers have provitamin A activities that are approximately 50% or less of that of corresponding all-*trans* carotenoids (Lessin *et al.*, 1997).

Various thermal processing methods such as roasting (Kidmose, Christensen, Agili and Thilsted, 2007), drying (Bechoff *et al.*, 2009), frying (Kidmose, Yang, Thilsted, Christensena and Brandt, 2006) boiling (Kidmose *et al.*, 2007) canning and microwaving (Chandler and Schwartz, 1988) have been reported to cause the isomerisation of β-carotene in sweet potato. The above mentioned thermal processing methods caused the degradation of β-carotene differently probably due to the duration as well as the mechanism of heating involved in each treatment. Conventional heating (e.g., baking, boiling and frying) requires enough time and temperature to achieve the acceptable doneness of food (Roberts and Pollien, 1997) which in turn, can cause extensive degradation of β-carotene. The mechanism of heating during microwave irradiation involves the collisions of electrolytes in the solution with other ions or molecules in the presence of electromagnetic fields (Yeo and Shibamoto, 1991). These collisions result in the production of enough heat to produce a microwaved product with acceptable doneness within a short period of time at relatively low temperature (Yeo and Shibamoto, 1991; Roberts and Pollien, 1997). This probably caused less loss in β-carotene content after microwave cooking of sweet potato roots (22.7%) than baking (31.13%) (Chandler and Schwartz, 1988).

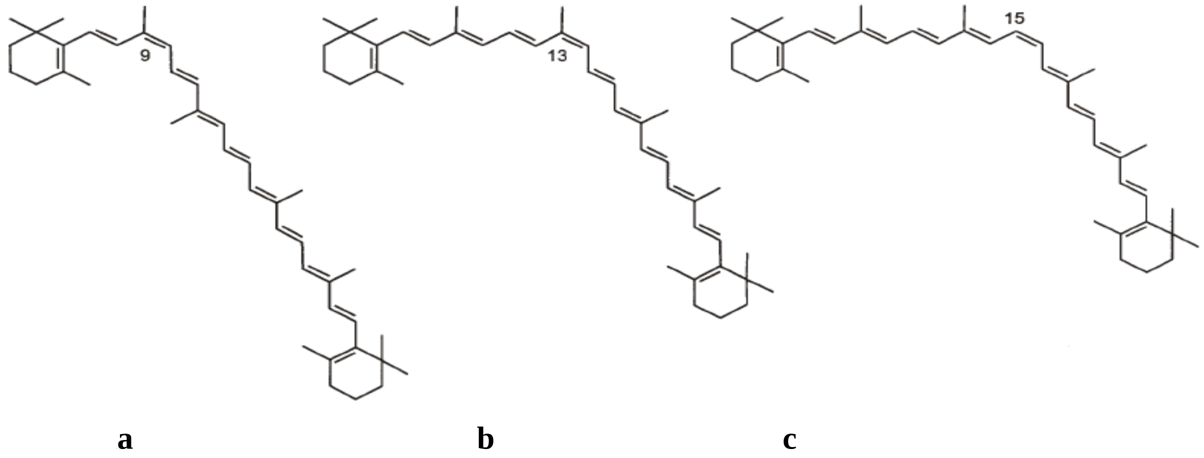


Figure 2.4 Chemical structures of predominant *cis* isomers of β-carotene: a) 9-*cis*-β-carotene, b) 13-*cis*-β-carotene and c) 15-*cis*-β-carotene (Schierle, Pietsch, Ceresa and Fizet, 2004)

Nevertheless, processing of foods containing β-carotene and other carotenoids by heat can determine the extent to which they are released during the digestion process. Thermal treatments break the protein complexes in which carotenoids are enclosed contributing to their availability (Tumuhimbise, Namutebi and Muyonga, 2009). Unaccessible provitamin A carotenoids remain trapped in the matrix (Tumuhimbise *et al.*, 2009), suggesting that the action of heat is required to make sure that they are released and made available. Tumuhimbise *et al.* (2009) assessed the effect of traditional heat processing on in vitro bioaccessibility of β-carotene from orange-fleshed sweet potato. Although heat processing reduced the β-carotene content of the samples, processed OFSP had significantly higher bioaccessible β-carotene ($p < 0.05$) compared to the raw ones. Deep fried OFSP samples were found to have the highest bioaccessible (*in vitro*) amount of β-carotene. Authors concluded that heat processing improved *in vitro* bioaccessibility of β-carotene in OFSP and this was probably due to the disruption of the tissue microstructure as observed under the light microscope.

Compositing OFSP flour with wheat flour would lead to changes in the rheological properties of the doughs, their bread-making performance and sensory properties of the resulting breads. However, the effect of substituting wheat flour with OFSP flour on the above mentioned characteristics is not well known. The current study has been undertaken to explore the effect of partial substitution of wheat with OFSP flour on dough rheology, bread-making performance and sensory properties of the breads. OFSP-wheat composite bread would contribute to adequate intake of vitamin A (in the form of provitamin A carotenoids) among people at risk of vitamin A deficiency. The baking process, however, may cause detrimental effects on β-carotene due to extreme temperatures prevailing in the oven. Thus, the effect of baking on β-carotene in OFSP-wheat composite breads will be investigated in order to determine its retention and the estimated contribution to the vitamin A requirements of different groups of people.

CHAPTER 3: HYPOTHESES AND OBJECTIVES

3.1 Hypotheses

1. Compositing wheat flour with OFSP flour will negatively affect the rheological behaviour of the dough and physical properties of composite breads. Partial replacement of wheat flour in composite dough formulations with any other component devoid of gluten would result in a weak dough (Dhingra and Jood, 2004). The resulting breads would be characterised by low loaf volume, oven spring and specific volume.

2. Compositing wheat flour with OFSP flour will change sensory properties of composite breads. The orange colour of OFSP flour will impart an orange coloration to the breads which would be more pronounced in the crumb. The presence of reducing sugars from OFSP flour (van Hal, 2000) and free amino groups will confer a dark appearance to the breads as a result of the Maillard reaction (Morales and Jimenez-Perez, 2001; Kitts, Wu, Kopec and Nagasawa, 2006). Sugars will enhance the sweetness of the bread but on the other hand, they may also promote the production of bitter compounds such as melanoidins (Pedreschi, Kaack, Granby, Troncoso, 2007) on the crust. The increase in the firmness may result from an increment of dietary fibres from OFSP flour (van Hal, 2000) which would interfere with the gluten network resulting in the depression of bread volume (Morrice and Morrice, 2012).

3. The β-carotene content will decrease during baking of OFSP-wheat composite breads as well as their vitamin A activity. Isomerisation of β-carotene could occur during baking of OFSP-wheat composite breads due to high temperatures (Chandler and Schwartz, 1988) resulting in the formation of *cis* isomers of β-carotene with lower vitamin A activity (Lessin, Catignani and Schwartz, 1997).

3.2 Objectives

1. To determine the effect of substituting wheat flour with different levels of orange-fleshed sweet potato flour on rheological properties of the doughs and their bread-making performance.

2. To determine the effect of substituting wheat flour with OFSP flour on sensory properties of OFSP-wheat composite breads.

3. To determine the stability of β-carotene during baking and the estimated contribution of OFSP-wheat composite breads to vitamin A requirements for different groups of people.

CHAPTER 4: RESEARCH

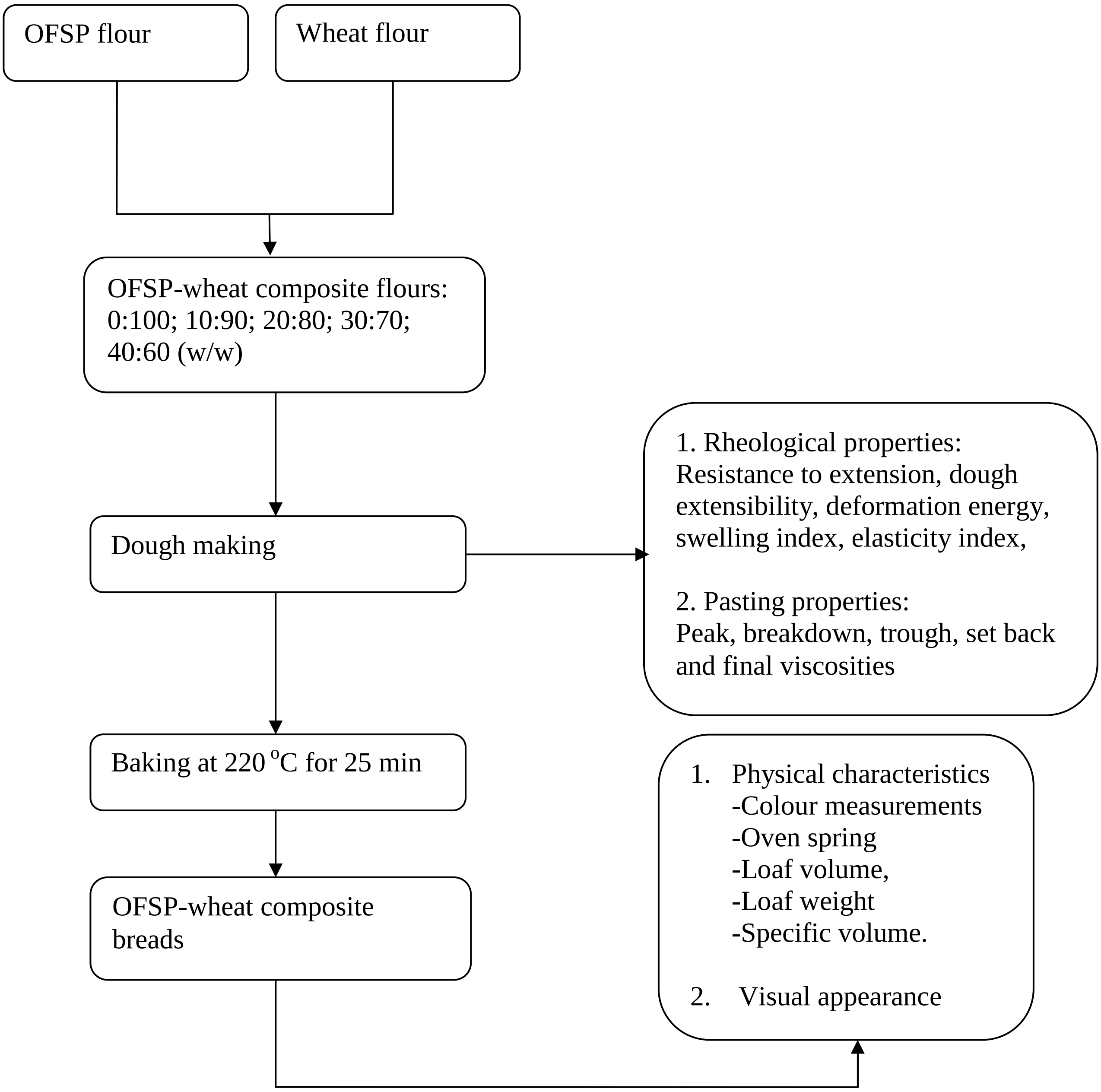
This research chapter is divided into two main chapters. Figures 4.1 and 4.2 show the experimental design of both research chapters.

PHASE I

Effect of substituting wheat flour with orange-fleshed sweet potato flour on rheological, pasting and bread-making properties

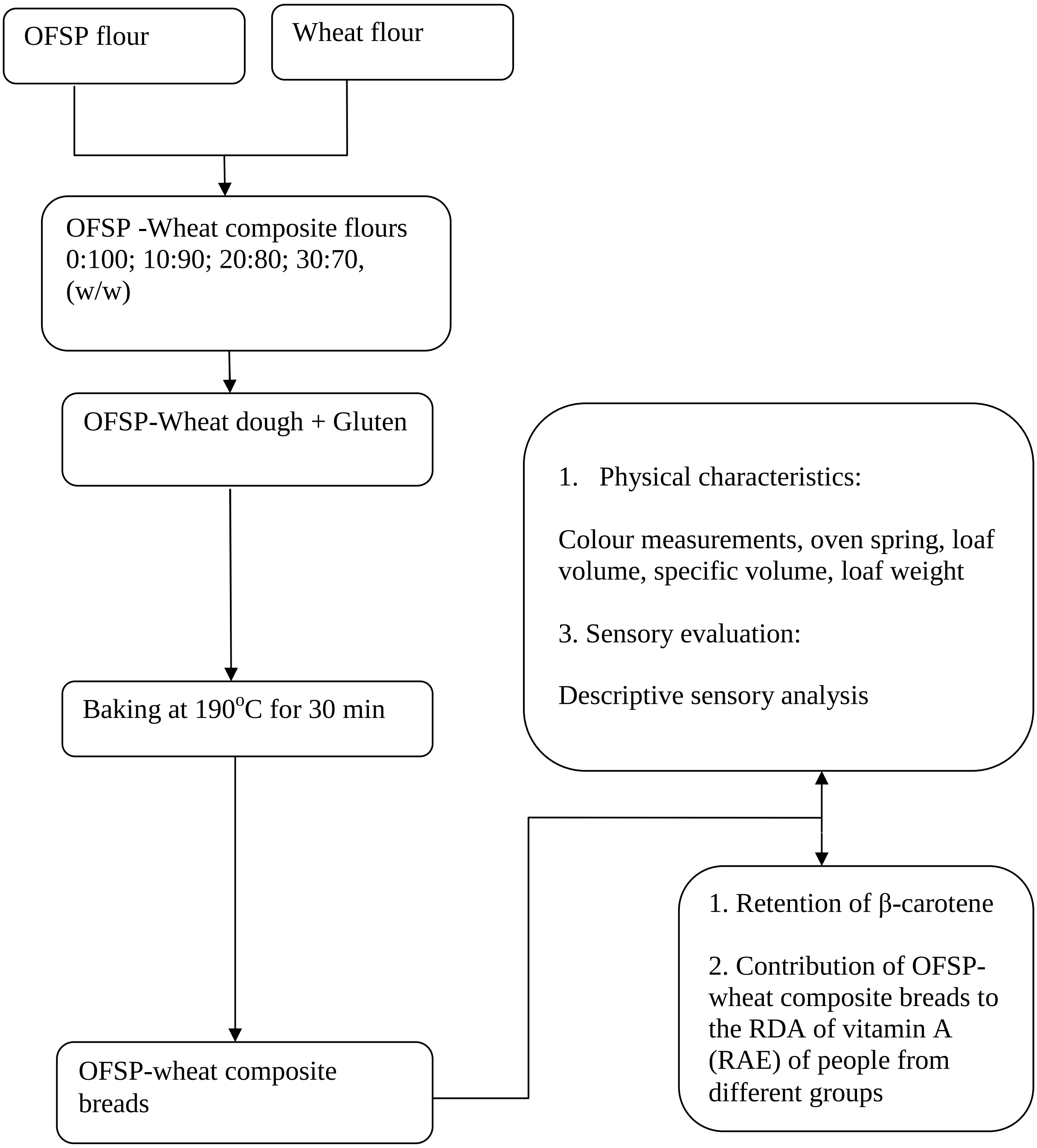
PHASE II

Stability of β-carotene and sensory quality of OFSP-composite breads and their estimated contribution to vitamin A requirements



OFSP: Orange-fleshed sweet potato

Figure 4.1 Experimental design of the study on the effect of substituting wheat flour with orange-fleshed sweet potato flour on rheological, pasting and bread-making properties (Phase I)



RAE: Retinol Activity Equivalent
OFSPF: Orange-fleshed sweet potato flour
WF: Wheat flour.
RDA: Recommended Dietary Allowance

Figure 4.2 Experimental design of the study on the effect of baking on the stability of β-carotene and sensory quality of OFSP-composite breads supplemented with gluten and their estimated contribution to vitamin A requirements (Phase II)

4.1 Effect of substituting wheat flour with orange-fleshed sweet potato flour on dough rheology and bread-making properties

Abstract

White wheat flour was partially substituted with orange-fleshed sweet potato (OFSP) flour at 0, 10, 20, 30 and 40% (w/w) levels. The effect of OFSP flour supplementation on rheological properties of the doughs as well as their bread-making performance was studied. Dough resistance to extension increased with increasing proportion of OFSP flour thereby compromising the extensibility of the doughs in all composites. Pasting parameters of composite flours decreased but their tendency towards retrogradation was slowed down probably due to the interaction between sugars from OFSP flour and starch molecules. The volume of the breads decreased as the level of OFSP flour increased probably due to the dilution effect on gluten and the presence of dietary fibre from OFSP flour. The most pronounced effect was observed at 30 and 40% substitution levels. The colour of the crust became progressively brown presumably due to the Maillard reaction and caramelisation. Bread containing 10% OFSP flour had oven spring value, loaf volume and specific volume close to the control (but statistically different, $p < 0.05$) when compared to its counterparts.

Key words: Rheological properties, loaf volume, Maillard reaction.

4.1.1 Introduction

Migration of the population from rural to urban areas is generally followed by change of lifestyle whereby people from rural areas adjust themselves to the way of life of people living in big cities. Food preferences and eating habits are among the lifestyles that are adopted by the migrants (Delisle, 1990). In Rwanda, for example, bread is more consumed in urban areas than in rural areas. The demand for bread will increase with overgrowing population in urban areas. Globally, the price of wheat in 2010 increased substantially by about 50% compared to the previous year (FAO, 2010) which could eventually lead to an increase of the price of wheat flour bread. Sweet potato grows well under marginal conditions with low production cost (van Hal, 2000). The use of orange-fleshed sweet potato (OFSP) flour in the bread industry may therefore contribute to cutting down on the amount of wheat imported and the reduction of bread price especially in countries where wheat is not produced. This would also enable easy access to OFSP-wheat composite bread for people affected with vitamin A deficiency since OFSP flour is a rich source of provitamin A carotenoids such as β-carotene (Van Hal, 2000).

Different types of bread are grouped into two main categories. These include wheaten bread comprising white bread, brown bread and whole wheat bread; and composite bread comprising bread made from a mixture of flours (Ellis *et al.*, 1997). Although bread is traditionally made from wheat flour, several studies have indicated the possibility of incorporating flours from other sources such as malted rice flour (Veluppillai, Nithyanantharajah, Vasantharuba, Balakumar and Arasaratnam, 2010) soya bean flour (Khetarpaul and Goyal, 2009), barley flour (Holtekjølen, Bævre, Rødbotten, Berg and Knutsen, 2008), cowpea flour (Hallén, Ibanoğlu and Ainsworth, 2004), banana flour (Mohamed, Xu and Singh, 2010) and cassava flour (Khalil, Mansour and Dawoud, 2000) into wheat flour at varying levels (10-50%). The need for alternative sources of flours for bread-making is generally driven by health and economic reasons. Wheat flour is naturally low in essential amino acids notably lysine and threonine as well as fat-soluble vitamins such vitamin A (in the form of provitamin A carotenoids), D, E and K (Dewettinck *et al*., 2008). In certain cases wheat flour is completely excluded from bread destined for people suffering from gluten intolerance (Gallagher, Gormley and Arendt, 2003).

Supplementing wheat flour with OFSP flour, which is devoid of gluten, is expected to adversely affect the rheological and bread-making properties of the composite flour. The addition of flours lacking gluten has been reported to negatively affect gluten network formation in the dough, gas retention capacity, loaf volume as well as the specific volume of bread (Hallén *et al.,* 2004). The need for using OFSP flour in composite breads, however, is driven by its nutritional value since it is a rich source of β-carotene which may vary from 60 to 39,848 μg/100 g (db) (van Hal, 2000). β-carotene is considered as the predominant carotenoid in OFSP with the highest vitamin A activity (Chandler and Schwartz, 1988).

The objective of this study was to determine the effect of supplementing wheat flour with OFSP flour on dough rheology and bread-making properties of OFSP-wheat composite flours.

4.1.2 Materials and methods

4.1.2.1 Materials

Fresh OFSP roots, Beauregard cultivar, were procured from Tshwane Market, Pretoria West, South Africa in January 2011. They were grown in the town of Tom Burke located in Limpopo, South Africa. White bread flour for bread-making was obtained from Ruto Mills factory (Pretoria West, South Africa). Compressed yeast was supplied by Anchor Yeast (Johannesburg, South Africa). Other ingredients such as salt, fat and sugar were purchased from the local supermarket.

Baking test

The bread-making performance of wheat flour and mixtures containing OFSP flour and wheat flour (10:90; 20:80, 30:70; 40:60, w/w) was determined based on the 100 g baking test. The bread formulation used for each test consisting of 100 g flour (14% flour moisture basis), 0.25 g malt, 3 g fat, 25 ml yeast solution (3% yeast), 5 ml of 0.1% ascorbic acid; 11 ml of sugar and salt solution (1.5% salt and 6% sugar) were transferred into the mixing bowl. Twenty millilitres of water was added to reach the optimum dough consistency. The mixing time was obtained from the mixograms at maximum dough consistency for each sample (3.4-

4.3 min). The dough was mixed with a 100 g mixer (National MFG CC Lincoln, Nebraska, USA), then rounded by hand and placed into a proofing cabinet (National MFG CO. Lincoln, Nebraska, USA). Proofing was performed at 30^{o}C for 90 min. The dough was punched 52 ± 1 min after the end of mixing and the second punch was done 25 min later. Moulding and panning were also done 13 ± 1 min later followed by baking in a Parthon rotary oven (National MFG CC Lincoln, Nebraska, USA) preheated at 225^{o}C for 24 min. Samples were baked in duplicate. Dimensions of the pans used were top inside 14.3 x 8 cm, bottom 13 x 6.5 cm, depth 5.5 cm.

4.1.2.2 Methods

Preparation of OFSP flour

Fresh OFSP roots were washed with clean water, hand peeled using a knife and peeler. Peeled sweet potatoes were diced into approximately 10 mm x 10 mm x 5 mm cubes using a dicing machine (TR21 vegetable cutter, Cappucino) then, dipped into 1% acetic acid solution for 1 h to reduce the enzymatic activity (Krishnan, Padmaja, Moorthy, Suja and Sajeev, 2010). Sweet potato dices were drained and dried in a forced-air cabinet oven dryer at 60^{o}C to a moisture content of 6%. Dehydrated sweet potato dices were milled using a laboratory hammer mill (Falling Number 3100, Huddinge, Sweeden) fitted with a 500 µm screen. The flour was milled again for further reduction of the particle size and then sieved through a 250 µm sieve. The sweet potato flour with water activity (a_w) of 0.3 was packaged in vacuum plastic bags and sealed using a vacuum sealer. The flour was stored at -20^{o}C before use. OFSP-wheat composite flours were prepared in the proportions of 0:100, 10:90, 20:80, 30:70 and 40:60 (w/w).

Protein content

The protein content (N x 6.25) of the flours was determined by the Dumas combustion method (American Association of Cereal Chemists, AACC International, 2000), Method 46-30.

Rheological properties of OFSP-wheat composite doughs

Mixing properties of OFSP-wheat composite doughs

The dough mixing properties were determined using a 35 g mixograph (National MFG CC Lincoln, Nebraska, USA) according to the AACC method 54-40A (AACC, 2000). The flour (35 g) was weighed into a bowl and a plastic scraper was used to create a triangular hole in the centre of the bowl. The required amount of water, based on absorption of the flour corrected to 14% moisture basis, was added. The mixer was stopped after 6 min of mixing.

Viscoelastic behaviour of OFSP-wheat composite doughs

OFSP-wheat composite flours were tested using the Alveolink NG Consistograph (Chopin, S.A, France). The Alveolink NG Consistograph controls both the alveographic and consistographic measurements (Chopin Technologies, 2008). The consistographic measurements were performed to determine the water absorption capacity of the flours. The dough was prepared using the NG mixer and cut into 10 pieces; then each piece was allowed to rest for 20 min in the resting chamber and blown with air until it burst. The following parameters were analysed: T: resistance to extension or tenacity, A: dough extensibility, Ex: swelling index, Fb: deformation energy, T/A: tenacity/extensibility, Iec: elasticity index. The air flow on the alveograph was calibrated every day before starting the tests and all tests were done at room temperature. Samples were analysed in duplicate.

Pasting properties of the flours

The pasting properties of the flours were determined using a 3 D Rapid Visco Analyser (RVA) (Newport Scientific, Warriewood, Australia). A suspension of 10% flour in distilled water (db, w/v) was held at 50^{o}C for 2 min and then heated from 50 ^{o}C to 91^{o}C over 7.5 min at a uniform rate (5.46^{o}C/min) with constant stirring at 160 rpm. The sample was held at 91^{o}C for 5 min, cooled to 50^{o}C over 7.5 min at a uniform rate (5.46^{o}C/min) and then held again for 5 min at 50^{o}C. For each sample the peak viscosity, breakdown, trough setback and final viscosities were recorded. Every test was done in triplicate.

Physical properties of OFSP-wheat composite breads

Oven spring, loaf weight, loaf volume and specific volume

Oven spring was calculated by subtracting the height of the dough after proofing from the height of the bread after baking. Loaf weight (g) was determined using a digital balance while loaf volume (cm^3) was measured by rape seed displacement method. Specific volume (cm^3/g) was determined by dividing loaf volume by loaf weight. Samples were analysed in duplicate.

Colour analysis

After baking, bread samples were cooled at room temperature and colour measurements were done on 11 spots (on top and other sides) of the crust and 6 spots (in the centre and towards the edges) of the crumb using a Chroma Meter CR-400 (Konica Minolta Sensing, Inc. Japan). Colour analysis was done based on L* (measure of lightness ranging from 0-100 indicating black to white), a * (+a, redness and –a, greenness) and b* (+b, yellowness and –b, blueness) values (Shalini and Laxmi, 2007). Bread samples were tested in duplicate.

Statistical analyses

One way analysis of variance (ANOVA) was used to test for differences in effects among the various substitution levels of wheat flour with orange-fleshed sweet potato flour on the dependent variables. Statistica (Version 10, Statsoft, Tulsa, USA) was used for data analysis. Significant differences between means were determined using Fisher's least significant difference test (LSD) at 5% probability level ($p < 0.05$).

4.1.3 Results and Discussion

4.1.3.1 Rheological properties of orange-fleshed sweet potato-wheat composite dough

The results for the mixing properties of doughs containing different amounts of OFSP flour are presented in Table 4.1.1. Compositing wheat flour with OFSP flour at 30 and 40%

substitution levels extended time required to reach optimum or peak dough development but decreased peak height. The difference at 10 and 20% was not statistically significant. Mixing tolerance of the dough decreased significantly ($p < 0.05$) when compared to the control except at 10% substitution. The addition of OFSP also led to a decrease in the protein content and water absorption capacity of composite flours (Fig. 4.1.1).

Table 4.1.1 Effect of substituting wheat flour with orange-fleshed sweet potato flour on the mixing properties of composite flours

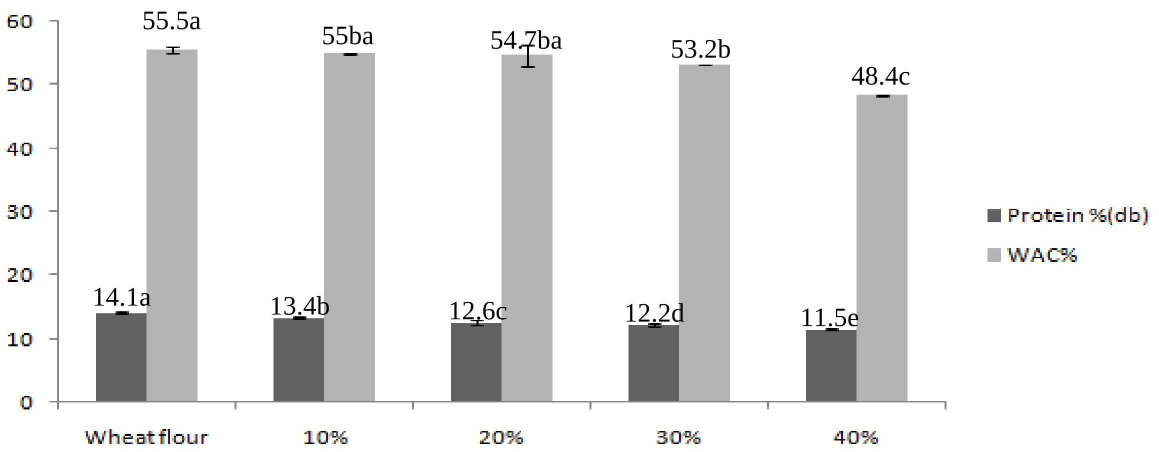
OFSPF:WF (w/w)	Peak time (min)	Peak height (mm)	Mixing tolerance
0:100	$3.4±0.1^{a}$	$55.0±1.4^{a}$	$43.0±1.4^{a}$
10:90	$3.2±0.0^{a}$	$56.0±1.4^{a}$	$41.2±0.3^{a}$
20:80	$3.1±0.2^{a}$	$51.5±0.7^{ab}$	$37.7±0.3^{b}$
30:70	$3.8±0.0^{b}$	$48.0±0.0^{b}$	$38.7±0.3^{b}$
40:60	$4.3±0.0^{c}$	$42.0±3.5^{c}$	$38.7±2.4^{b}$

Different subscripts in the same column indicate that means were significantly different ($p < 0.05$)

OFSPF: Orange-fleshed sweet potato flour, WF: wheat flour.

The increase in mixing time at 30 and 40% substitution levels was probably due to slow gluten network formation following the interference with OFSP flour components such as dietary fibre (Noort, van Haaster, Hemery, Schols and Hamer, 2010). OFSP flour contains dietary fibre ranging from 1.4-10 % (db) (van Hal, 2000). Low water absorption capacity of flours containing 30 and 40% OFSP flour (Fig. 4.1.1) may also have contributed to long mixing time and decrease in peak height (which indicates the consistency of the dough) when compared to the control (Table 4.1.1).

Compositing wheat flour with OFSP flour increased the sugar content of composite flours. Among the sugars detected include sucrose, glucose and fructose (reported in research chapter 4.2). The presence of sucrose has been associated with inhibition of water absorption by starch and gluten. The resulting dough becomes less consistent and the protein network requires long time to unfold (Salvador, Sanz and Fiszman, 2006). The decrease in protein content upon the addition of OFSP flour may also explain the reason for poor hydration capacity of composite flours especially at 30 and 40% substitution levels (Fig. 4.1.1). Lack of fully developed gluten as a result of insufficient amount of water has a negative effect on dough formation, mixing time and bread quality (Mohamed *et al*, 2010).



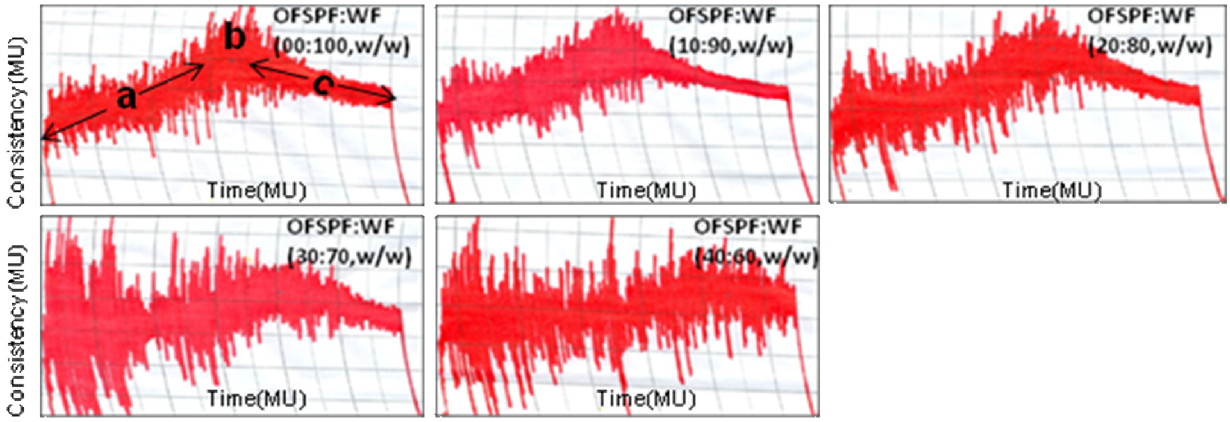
Values in percentage (x-axis) indicate levels of substitution of wheat flour with OFSP flour.

Figure 4.1.1 Effect of substituting wheat flour with orange-fleshed sweet potato flour on protein content and water absorption capacity (WAC) of composite flours

The mixogram tail (designated as C in Fig. 4.1.2) that appears beyond the optimum dough development stage showed that OFSP-wheat composite doughs were subjected to overmixing. Doughs made from the control and 10% substitution levels showed more tolerance to overmixing (Table 4.1.1). The remaining doughs displayed the least tolerance behaviour probably due to the weakening of the gluten network. The weakening of gluten network could be attributed to the depolymerisation of gluten proteins after mixing the doughs for extended period of time (Danno and Hoseney, 1982a). The same observations have been made by Tsen (1967) who found that overmixing increased the amount of glutenin fractions as an indication of disaggregation of gluten proteins.

The addition of OFSP flour up to 30% substitution level significantly increased dough resistance to extension thereby reducing dough extensibility (Table 4.1.2). The swelling and elasticity indices also decreased with increased OFSP flour addition.

The increase in dough resistance to extension as a result of OFSP flour addition, adversely affected the extensibility of the dough. The same observations were made by Mlakar *et al.* (2009) who found that the presence of amaranth flour in spelt wheat flour caused an increase in the resistance of the dough to extension. This resulted in the decrease in dough extensibility as the level of substitution with amaranth increased. It may be hypothesised that the addition of OFSP flour reduces the visco-elastic behaviour of the dough due to a decrease in the gluten content, especially the gliadins, of the composites (Van Der Borght, Goesaert, Veraverbeke and Delcour, 2005). Doughs with higher resistance to extension and deformation energy do not have optimum viscoelastic behaviour. They are also characterised by higher values on curve configuration ratio (T/A) (Hrušková and Šmejda, 2003). Lower values on curve configuration ratio suggest that breads made from such doughs might have better volumes and crumb characteristics (Silva-Sanches, Gonzalez-Castaneda, De Leon-Rodriguez and Barba De La Rosa, 2004). No significant difference was found in the deformation energy of OFSP-wheat composite doughs.



a: Dough development stage

b: Optimum dough development stage

c: Breakdown stage

MU: Mixograph units

OFSPF: Orange-fleshed sweet potato flour, WF: wheat flour.

Figure 4.1.2 Mixograms of doughs made from orange-fleshed sweet potato-wheat composite flours

The swelling index of the dough decreased with increased substitution of wheat flour with OFSP flour in all mixes (Table 4.1.2). Similarly, the elasticity index of the dough was reduced following the incorporation of OFSP flour and the most pronounced effect was observed at 30 and 40% substitution levels. The decrease in the swelling and elasticity indices was presumably related to the reduction in the gluten content of the mixtures due to the dilution effect caused by the addition of OFSP flour. For bread dough to exhibit viscoelastic behaviour that would allow the air bubbles to inflate (expand) and retain the gas,

Table 4.1.2 Effect of substituting wheat flour with orange-fleshed sweet potato flour on alveographic parameters of the dough

OFSPF:WF (w/w)	T (mm H_2O)	A (mm)	Ex	Fb (x10:^{4}J)	T/A	Iec
0:100	49.5 ± 3.6^{a}	86.0 ± 8.6^{a}	20.8 ± 2.4^{a}	133.4 ± 11.1^{a}	0.6 ± 0.1^{a}	51.1 ± 1.3^{a}
10:100	60.5 ± 7.2^{b}	63.5 ± 0.7^{b}	18.2 ± 2.9^{b}	125.8 ± 9.7^{a}	1.0 ± 0.4^{a}	40.4 ± 3.9^{b}
20:80	87.5 ± 6.6^{c}	50.7 ± 7.7^{c}	15.8 ± 1.2^{c}	137 ± 10.1^{a}	2.0 ± 0.3^{b}	33.4 ± 2.4^{c}
30:70	112.3 ± 5.7^{d}	30.9 ± 5.7^{d}	12.3 ± 1.2^{d}	130.5 ± 11^{a}	3.7 ± 1.2^{c}	0 ± 0^{d}
40:60	157.3 ± 0.5^{e}	18.0 ± 1.9^{e}	9.4 ± 0.8^{e}	133.2 ± 6.8^{a}	8.4 ± 1.1^{d}	0 ± 0^{d}

Different subscripts in the same column indicate that means were significantly different ($p < 0.05$)

OFSPF: Orange-fleshed sweet potato flour, WF: wheat flour, WAC: Water absorption capacity; T: Resistance to extension or Tenacity; A: Dough extensibility; Ex: Swelling index; Fb: Deformation energy; T/A: Curve configuration ratio; Iec: Elasticity index.

there should be an interaction between gluten proteins. As more OFSP flour is added, the gluten from wheat flour is diluted to the point where gluten proteins are no longer able to interact with each other to form a cohesive and continuous network.

Pasting properties of OFSP and wheat flours as well as their composites are shown in Table 4.1.3.Wheat flour exhibited high peak viscosity compared to OFSP flour alone and composite flours. The trough, breakdown and final viscosities decreased with increased addition of OFSP flour. However, the setback viscosity decreased as more OFSP flour was added.

Table 4.1.3 Effect of substituting wheat flour with orange-fleshed sweet potato flour on pasting properties of wheat flour, orange-fleshed sweet potato flour and their composites

OFSPF:WF (w/w)	Peak viscosity (RVU)	Trough viscosity (RVU)	Breakdown viscosity (RVU)	Final viscosity (RVU)	Setback viscosity (RVU)
0:100	75.8 ± 1.9^{e}	62.1 ± 1.5^{d}	13.6 ± 0.4^{e}	91.1 ± 1.4^{e}	28.9 ± 0.2^{e}
10:90	53.3 ± 0.3^{d}	43.4 ± 0.5^{c}	9.8 ± 0.2^{d}	66.2 ± 0.1^{d}	22.8 ± 0.6^{d}
20:80	45.0 ± 1.8^{bc}	36.5 ± 1.8^{b}	8.5 ± 0.2^{c}	57.0 ± 1.3^{c}	20.4 ± 0.6^{c}
30:70	43.2 ± 0.3^{b}	35.1 ± 0.0^{b}	8.1 ± 0.2^{c}	54.9 ± 0.2^{b}	19.8 ± 0.1^{c}
40:60	38.8 ± 0.9^{a}	32.5 ± 1.0^{a}	6.3 ± 0.1^{b}	48.6 ± 0.8^{a}	16.0 ± 0.2^{b}
100:0	45.7 ± 0.7^{c}	42.8 ± 0.8^{c}	2.9 ± 0.4^{a}	54.3 ± 1.2^{b}	11.5 ± 1.1^{a}

Different subscripts in the same column indicate that means were significantly different ($p < 0.05$). OFSPF: Orange-fleshed sweet potato flour, WF: wheat flour, RVU: Rapid Visco Units.

High peak viscosity is linked in part to the large granular size and swelling power of the starch (Jangchud, Phimolsiripol and Haruthaithanasan, 2003). Van Der Borght, Goesaert, Veraverbeke and Delcour (2005) reported that wheat starch granular size are of two sizes; large, lenticular A type granules with size ranging from 15-40 µm (mean diameter 20 µm) and spherical B-type granules with size ranging from 1-10 µm (mean diameter 5 µm). Jangchud *et al.* (2003) reported the size of sweet potato starch granules ranging from 3-27 µm with OFSP having small granular size ranging from 7-12 µm, which probably contributed the low peak viscosity. They suggested that larger starch granules are associated with lower pasting temperature and high swelling properties. Therefore, differences in the starch granular size might have played a significant role in the pasting properties of wheat and OFSP flours evaluated in the current study.

Paste containing 40 % OFSP flour was more resistant to disintegration as indicated by low breakdown viscosity. On the other hand, pastes containing 20 and 30% OFSP flour showed moderate sensitivity to disintegration. High degree of swelling of starch granules during heating is related to high value of breakdown (Ragaee and Adbel-Aal, 2006). Paste containing high proportions of wheat flour showed high breakdown viscosity. Breakdown viscosity can give an estimation of the resistance of a paste to disintegration as a result of heat or mechanical shear stress (Waramboi, Dennien, Gidley and Sopade, 2011). The low breakdown viscosity observed in OFSP-wheat composite flours was probably due to the presence of sugars from OFSP flour (research chapter 2). Interaction of sugar molecules with starch chains increases the rigidity of starch granules which confers them more resistance to disintegration (Gunaratne, Ranaweera and Corke, 2007).

Low setback values were recorded in pastes containing OFSP flour and the most pronounced effect was observed at 40% substitution level. Setback values can be used to determine the tendency of a flour-water suspension towards retrogradation (Ragaee and Adbel-Aal, 2006; Waramboi *et al.*, 2011). Sugars such as sucrose, glucose (I'anson *et al.*, 1990; Gunaratne *et al.*, 2007) and fructose (Kohyama and Nishinari, 1991) have been found to act as antistaling agents in the presence of gelatinised starch. It may, therefore, be hypothesised that sugars from OFSP flour, notably sucrose, glucose and fructose (Table 4.2.4), interacted with starch thereby reducing the mobility of starch molecules (Lii, Lai and Liu, 1998) which probably prevented them from rearranging themselves into an ordered manner (crystalline structure).

4.1.3.2 Effect of substituting wheat flour with orange-fleshed flour on the bread-making performance

Increasing the proportion of OFSP flour in the composites led to proportional decreases in height of the dough after proofing, height of bread after baking, oven spring, loaf volume and specific volume but an increase in loaf weight (Table 4.1.4).

Loaf volumes were significantly different among all samples with the control having the highest loaf volume. Failure to retain gases such as CO_2 and steam produced during the baking process may have led to the depression in loaf volume (Dhingra and Jood, 2004) as more wheat flour was replaced with OFSP flour. The decrease in loaf volume was attributed to lower gluten content of the blends following a gradual substitution of wheat flour with

OFSP flour suggesting a dilution effect on gluten. The decrease in loaf volume was more pronounced in breads containing high levels of OFSP flour (Fig. 4.1.3). Protein from OFSP flour was not expected to improve the rheological properties of the dough and subsequent characteristics of the breads such as oven spring, loaf volume and specific volume. Proteins other than gluten may form aggregates inside the gluten fibrils thereby interfering with continuous gluten network (Roccia, Ribotta, Perez and Leon, 2009) which may result in low loaf volume as observed in breads containing OFSP flour. Similarly, the presence of high amounts of dietary fiber from OFSP flour (1.4 - 10%, db) (Van Hal, 2000) may interfere with the formation of gluten network thereby resulting in low loaf volume (Stojceska and Ainsworth, 2008). The decrease in loaf volume was also followed by a significant decrease in specific volume probably due to increase in loaf weight.

Table 4.1.4 Effect of substituting wheat flour with orange-fleshed sweet potato flour on physical properties of the bread

OFSPF:WF (w/w)	Height of the dough after proofing (mm)	Height of the bread after baking (mm)	Oven spring (mm)	Loaf weight (g)	Loaf volume (cm^3)	Specific volume (cm^3/g)
0:100	80.5±0.7^{a}	104.0±8.4^{a}	23.5±7.7^{a}	145.7±0.3^{a}	877.5±3.5^{a}	5.3±0.9^{a}
10:90	82.0±0^{a}	102.5±2.1^{a}	20.5±2.1^{a}	145.7±3.5^{a}	760.0±14^{b}	5.0±0.3^{b}
20:80	77.0±0^{b}	77.5±0.7^{b}	0.5±0.7^{b}	150.0±1.0^{ab}	580.0±0.0^{c}	3.8±0.0^{c}
30:70	71.5±2.1^{c}	65.0±1.4^{c}	-6.5±0.7^{b}	154.8±2.4^{bc}	397.5±31.8^{d}	2.5±0.1^{d}
40:60	58.5±0.7^{d}	50.0±0.0^{d}	-8.5±0.7^{b}	158.5±0.6^{c}	275.0±0.0^{e}	1.7±0.0^{e}

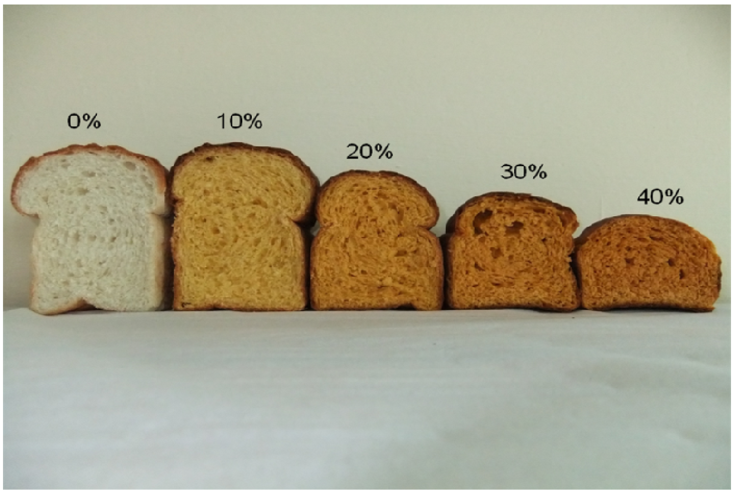
Different subscripts in the same column indicate that means were significantly different ($p < 0.05$). OFSPF: Orange-fleshed sweet potato flour, WF: wheat flour

The height of breads containing 30 and 40% OFSP flour was lower compared to the height of the dough (after proofing) which gave negative oven spring values (Table 4.1.4). The decrease in the height of the breads as opposed to the height of the doughs was attributed to the rupture of air bubbles in bread dough which is directly linked to the thickness of the gluten-starch matrix surrounding them (Sroan, Bean and MacRitchie, 2009). It may be hypothesised that the amount of gluten inside the gluten-starch matrix decreased as more OFSP flour was supplemented. Consequently, the gluten-starch matrix lost the ability to hold gases produced during baking and ruptured. Presumably, steam produced during baking (Mills, Wilde, Salt and Skeggs, 2003) contributed to further increase in pressure inside air bubbles which caused the bread to collapse. This was probably the reason why bread containing 40% OFSP flour did not show clear sign of gas cell formation (Fig. 4.1.4).

The crumb structure of OFSP-wheat composite breads is shown in Figure 4.1.4. Bread containing 30% OFSP flour was characterised by large gas cells in the crumb while bread containing 40% OFSP flour was dense and compact. In contrast, other breads (i.e., 100% wheat flour bread, breads containing 10 and 20 % OFSP flour) had less compact crumbs.

OFSPF: Orange-fleshed sweet potato flour, WF: wheat flour.

Figure 4.1.3 Orange-fleshed sweet potato-wheat composite breads



Values in percentage indicate levels of substitution of wheat with OFSP flour.

Figure 4.1.4 Crumb structures of orange-fleshed sweet potato-wheat composite breads

The formation of large gas cells could be explained by the fact that during proofing and baking the size of gas cells increased and reached a point where adjacent gas cells started to press against one another (Dobraszczyk and Roberts, 1994) and probably fused due to high pressure that ruptured the cell walls. This implies that more gas cells merged to form large ones following the rupture of the cell walls (Shah, Cambell, McKee and Rielly, 1998; van Vliet, 2008; Bremond, Thiam and Bibette, 2008). Large gas cells were not formed in bread containing 40% OFSP flour probably because there was not enough gluten that would allow further expansion of the cell walls. In order to ensure their stability, gas cell walls need to be sufficiently extensible to respond to gas pressure but also strong enough to resist collapse (Sroan *et al.*, 2009). This may also explain the reason why bread containing 40% OFSP flour was dense and compact while other breads (i.e. 100% wheat flour bread, breads containing 10 and 20 % OFSP flour) were relatively soft (they did not collapse as shown by positive oven spring values, Table 4.1.4).

4.1.3.3 Relationship between rheological properties of the dough and bread quality characteristics

The increased resistance to extension (Table 4.1.2) resulted in a decrease in dough extensibility and elasticity index which probably led to low loaf volume. Dough resistance to extension negatively correlated (Table 4.1.5) with dough extensibility (r= -0.966, $p < 0.001$) and swelling index (r= -0.985, $p < 0.001$). The same relationship was found between dough resistance to extension and loaf volume (r= -0.977, $p < 0.001$). However, the latter positively correlated with dough extensibility (r= 0.997, $p < 0.001$), elasticity (r= 0.965, $p < 0.001$) and swelling (r= 0.966, $p < 0.001$) indices.

Flour-water suspensions that displayed high peak viscosities produced breads with higher loaf volume and specific volume compared to the ones that exhibited low values on these parameters. These included composite flours containing 10 and 20% OFSP flour. Loaf volume and specific volume positively correlated with peak viscosity with coefficients of correlation of 0.884 and 0.828 respectively.

4.1.3.4 Effect of substituting wheat flour with orange-fleshed sweet potato flour on colour of bread

Colour of the crust got progressively darker as more wheat flour was substituted with OFSP flour (Table 4.1.6). Orange and yellow colourations could also be observed on the crust but they were dominated by the brown colour. The lightness of the crust of bread at 10%

Table 4.1.5 Correlation coefficients between the viscoelastic characteristics of the doughs, pasting properties of the flour-water suspensions and bread characteristics [a]

Parameters	WAC	T	A	Ex	Fb	T/A	Iec	PV	TV	BDV	SV	PT	OS	LW	LV	SV
Water Absorption capacity (WAC)	1															
Resistance to extension (T)	-0.941*	1														
Dough extensibility (A)	0.840	-0.966*	1													
Swelling index (Ex)	0.890*	-0.985*	0.994*	1												
Deformation energy (Fb)	-0.348	0.311	-0.128	-0.165	1											
Curve configuration ratio (T/A)	-0.995*	0.966*	-0.878*	-0.922*	0.347	1										
Elasticity index (Iec)	0.802	-0.921*	0.961*	0.963*	-0.032	-0.847	1									
Peak viscosity (PV)	0.647	-0.817	0.916*	0.879*	0.081	-0.682	0.813	1								
Trough viscosity (TV)	0.619	-0.798	0.905*	0.865	0.101	-0.656	0.803	0.999*	1							
Breakdown viscosity (BDV)	0.758	-0.885*	0.946*	0.926*	0	-0.784	0.844	0.987*	0.98*	1						
Setback viscosity (SV)	0.796	-0.909*	0.954*	0.940*	-0.051	-0.820	0.852	0.975*	0.966*	0.997*	1					
Oven spring (OS)	0.728	-0.915*	0.96*	0.940*	-0.263	-0.783	0.915*	0.871	0.865	0.881*	0.887*	0.910*	1			
Loaf weight (LW)	-0.901*	0.985*	-0.963*	-0.978*	0.310	0.937*	-0.954*	-0.777	-0.760	-0.837	-0.860	-0.990*	-0.937*	1		
Loaf volume (LV)	0.857	-0.977*	0.997*	0.996*	-0.187	-0.896*	0.965*	0.884*	0.872	0.921*	0.933*	0.954*	0.966*	-0.980*	1	
Specific volume (SV)	0.874	-0.981*	0.982*	0.989*	-0.250	-0.914*	0.968*	0.828	0.813	0.875	0.892*	0.980*	0.959*	-0.995*	0.994*	1

[a] Significance level :*=$p < 0.001$

Table 4.1.6 Effect of substituting wheat flour with orange-fleshed sweet potato flour on the colour of bread

	Colour of the crust[1]			Colour of the crumb		
OFSPF:WF (w/w)	L*	a*	b*	L*	a*	b*
0:100	58.5±6.8^{a}	10.8±3.2^{a}	19.0±0.5^{a}	64.2±1.6^{a}	0.4±0.2^{a}	5.6±0.8^{a}
10:90	48.2±6.0^{b}	13.5±1.3^{b}	12.5±1.5^{b}	61.9±1.7^{b}	1.6±0.4^{b}	25.7±1.0^{b}
20:80	43.0±2.8^{c}	13.2±1.8^{b}	5.6±2.1^{c}	58.7±1.1^{c}	5.9±0.6^{c}	27.0±1.0^{c}
30:70	41.2±2.9^{cd}	12.5±2.1^{b}	5.6±1.7^{c}	58.4±1.6^{c}	9.0±0.4^{d}	28.6±0.7^{d}
40:60	38.7±8.2^{d}	12.7±3.4^{b}	5.0±1.8^{c}	56.8±0.7^{d}	11.0±0.2^{e}	27.0±1.0^{bc}

[1] : Standard deviations are slightly large because colour of the crust was not evenly distributed on all sides of the breads.
Different subscripts in the same column indicate that means were significantly different ($p <$ 0.05)
OFSPF: Orange-fleshed sweet potato flour, WF: wheat flour.

substitution level was significantly higher than its counterparts (breads containing OFSP flour). Crumb colour was predominantly orange and yellow.

Supplementing wheat flour with OFSP flour led to an increase in the sugar content of the composites which probably contributed to the browning of the crust as shown by the decrease in the L* value. The reducing sugars detected in the flours, such as glucose and fructose (research chapter 2), were probably involved in the darkening of the crust through the Maillard reaction (Michalska, Amigo-Benavent, Zielinskiand del Castillo, 2008). Sucrose, which is not a reducing sugar, was also detected in the composite flours and might have contributed to the browning of the crust through caramelisation (Kitts, Wu, Kopec and Nagasawa, 2006). The a* value of the crust of the control (100% wheat flour bread) was significantly lower than in breads containing OFSP flour probably because of the presence of carotenoids from OFSP flour. The latter did not differ significantly when compared to each other. The b* value on the crust of breads containing OFSP flour was lower compared to the control. Presumably, the yellowness of the crust of breads containing OFSP flour was dominated by extensive browning following increased addition of OFSP flour. The latter was associated with crust browning due to the presence of sugars.

Compositing OFSP flour with wheat flour increased the darkness of the crumb significantly (Table 4.1.6). The darkness of the crumb decreased probably because areas close to the crust were slightly darker when compared to the central part of the crumbs and the increase in OFSP flour was related to increase in the browning of the crust. The a* value that denotes the greenness to redness component of the colour is not usually emphasised for baked products (Shittu, Aminu and Abulude, 2009). However, different researchers (Bengtsson, Namutebi, Alminger and Svanberg, 2008; Jangchud *et al.*, 2003; Takahata, Noda and Nagata, 1993) who worked on sweet potato used it to describe the orange colour in sweet potato products. The a* value of the crumbs increased with increased OFSP flour supplementation and the orange colour of OFSP flour was certainly responsible for the orange colouration observed in the crumb. The b* value also increased significantly in breads containing OFSP flour when compared to the control and was associated with the presence of OFSP flour as well.

4.1.4 Conclusions

Supplementing wheat flour with OFSP flour changes rheological properties of the doughs and their bread-making performance differently based on the amount of OFSP flour added. Dough development time and dough consistency remains unchanged up to 20% substitution level. Dough extensibility decreases as a result of increase in dough resistance to extension. The former is reduced from 21.8 to 79.5% and the most pronounced effect is observed at 30 and 40% substitution levels. OFSP flour lowers pasting properties of the flours but reduces their tendency towards retrogradation in all composites. Bread containing 10% OFSP flour had oven spring value, loaf volume and specific volume close to the control (but statistically different, $p < 0.05$) when compared to its counterparts.

4.2 Effect of substituting wheat flour with orange-fleshed sweet potato flour and baking on β-carotene content and sensory quality of orange-fleshed sweet potato-wheat composite breads

Abstract

Orange-fleshed sweet potato (OFSP) flour was incorporated into wheat flour at 0, 10, 20 and 30% (w/w) substitution levels. The stability of β-carotene during baking, sensory properties and the contribution of OFSP-wheat composite breads to vitamin A requirements were evaluated. The descriptive sensory analysis revealed distinct differences in the sensory profiles of OFSP-wheat composite breads as compared with wheat flour bread. Breads containing OFSP flour were described as sweet with sweet potato flavour, caramel aromas and had dark crusts. The retention of all-*trans*-β-carotene under baking conditions (190°C for 30 min) significantly differed ($p < 0.05$) among all bread samples studied. 63, 71 and 83% of all-*trans*-β-carotene were retained in breads containing 10, 20 and 30% OFSP flour respectively. 9-*cis*-β-carotene and 13-*cis*-β-carotene were the main *cis* isomers of β-carotene formed during baking. The amount of 13-*cis*-β-carotene was five to eight times greater than that present in the doughs. In contrast, 9-*cis*-β-carotene found in the breads containing OFSP flour was formed during baking as it was not found in the doughs. One portion of bread, approximately 2 slices (100 g), containing 30% OFSP would meet 89, 45 and 42% of the recommended dietary allowance (RDA) of vitamin A for children (3-10 years), pregnant and lactating women respectively. The same amount of bread containing 20% OFSP flour can provide 61% of the RDA of vitamin A for children but less than 50% of the RDA for other groups. OFSP-composite breads can potentially contribute to the eradication of vitamin A deficiency amongst people at risk or already living with it.

Key words: All-*trans*-β-carotene, retinol activity equivalent, gluten, descriptive sensory analysis.

4.2.1 Introduction

Vitamin A deficiency (VAD) is considered a serious nutritional problem in developing countries (Padula and Rodriguez-Amaya, 1986) especially among vulnerable groups of the population such as children, pregnant and lactating women (Kidmose, Yang, Thilsted, Christensena and Brandt, 2006). In countries where vitamin A deficiency is endemic, women experience deficiency symptoms such as night blindness that may continue during the early period of lactation (WHO, 1998). Vitamin A is essential for maintaining the integrity of epithelial tissues (skin, cornea, gastrointestinal tract, lungs and urinary tract), growth, proper functioning of the retina and the immune system (Johnson, 1994).

Food-based strategies, including the use of orange-fleshed sweet potato (OFSP) as a rich source of provitamin A carotenoids such as β-carotene, have been found to be a promising tool to eradicate VAD in developing countries (Jalal, Nesheim, Agus, Sanjur and Habicht, 1998; Haskell *et al*., 2004; van Jaarsveld *et al.*, 2005; Low *et al*., 2007). However, thermal treatments such as baking may have negative effects on β-carotene which may compromise their nutritional quality.

Wheat (*Triticum aestivum*) is the most important crop for bread-making owing to its supreme baking performance as compared to other cereals (Dewettinck *et al*., 2008). The unique ability of wheat flour to form a viscoelastic dough capable of retaining gas and expand are due to the characteristics of gluten proteins (Van Der Borght, Goesaert, Veraverbeke and Delcour, 2005; Roccia, Ribotta, Perez and Leon, 2009). Gliadin is believed to confer viscous properties to the dough, while glutenin imparts the strength and elasticity required to hold the gases that are produced during fermentation and baking processes (Khatkar, Bell and Schofield, 1995). Since sweet potato flour lacks the gluten proteins (Limroongreungrat and Huang, 2007), it is difficult to use it in baked products, especially breads, in high quantities. This is because it may result in poor quality products thereby affecting their sensory properties and acceptability by the consumers.

Supplementing wheat flour with OFSP flour may change sensory properties of the resulting bread such as texture, colour, flavour and aroma. The fact that total carbohydrate of the sweet potato root is comprised of 20% simple sugars (van Hal, 2000) may contribute to the browning of the crust. Surface colour that results from the Maillard reaction and caramelisation (Martinez-Anaya, 1996; Ajandouz, Tchiakpe, Ore, Benajiba and Puigserver, 2001) contributes to consumer preference and is considered as a critical baking index (Zanoni, Peri and Bruno, 1995). Flavours developed by those reactions are highly desirable and are associated by consumers with a delicious and high grade product (Lindenmeier and Hofman, 2004). β-carotene is also an important source of flavour and aroma compounds such as β-ionone, 5,6,-epoxy-β-ionone and dihydroactinidiolide (DHA). These compounds are associated with fruity, floral and woody notes and are produced upon the degradation of β-carotene through an attack by enzyme-generated free radicals and a cleavage at the C9-C10 bond (Waché, Bosser-DeRatuld, Lhuguenot and Belin, 2003).

The objective of this study was to determine the effect of baking on the stability of β-carotene and sensory quality of OFSP-wheat composite breads supplemented with gluten and their estimated contribution to vitamin A requirements.

4.2.2 Materials and methods

4.2.2.1 Materials

Fresh orange-fleshed sweet potato roots, Beauregard cultivar, were procured from Tshwane Market, Pretoria West, South Africa in January 2011. They were grown in the town of Tom Burke located in the Northern Province of South Africa. OFSP flour was produced as described previously (research chapter I) except that OFSP dices were not treated with acetic acid in order to prevent any effect it might have on carotenoids.White bread flour was obtained from Ruto Mills (Pretoria West, South Africa). Exogenous gluten (wheat gluten) was obtained from Sigma (Sigma-Aldrich, South Africa). Compressed yeast and EN85 (bread improver) were provided by Anchor Yeast (Johannesburg, South Africa). Other ingredients such as salt, fat and sugar were purchased from the local super market.

Bread-making

OFSP-wheat composite flours were prepared in the proportions of 10:90, 20:80 and 30:70 (w/w). Other ingredients (expressed on flour weight basis) such as sugar (3%), salt (1%), fat (1%), compressed yeast (2%) and EN85 (0.1%) were added to composite flours and the control (100% wheat flour). The weight of gluten (wheat gluten) equivalent to the quantity lost during the substitution of wheat flour with OFSP flour was added to composite flours to improve the quality of breads. The quantity of water added was based on the amount required to reach 500 FU. Flours (2500 g) and other ingredients were mixed in a Tauro dough mixer (model: 2221, Italy) at speed 1 for 1 min without water. Water was added and the speed increased to no. 2 for 8 min. The dough was then divided into 700 g pieces to simulate commercial breads, moulded and placed into bread pans of dimensions: top inside 230 x 80 mm, bottom 235 x 80 mm, depth 95 mm. They were proofed at 40^{o}C and 90% relative humidity for 60 min and baked at 190^{o}C for 30 min. Four replicates for each type of bread were prepared and physical properties of bread were analysed on all four replicates.

4.2.2.2 Methods

Gluten content

Gluten content of the flours was determined according to the AACC method 38-10 (AACC, 2000). Flour (25 g) was mixed with 10 ml of water and hand kneaded in a mortar. More water (approximately 5 ml) was added gradually until a stiff dough was formed and allowed to stand in water for about 20 min. The dough was kneaded gently under running water to remove starch and other soluble matter. Gluten obtained was allowed to stand in water for 1 h, then pressed between the hands and rolled into a ball. The gluten ball was transferred to the oven, dried at 100^{o}C for 24 hr and weighed as dry gluten after cooling.

Determination of β-carotene content in wheat-OFSP flour composite breads

Carotenoid extraction

Sample extraction was done according to van Jaarsveld, De wet Marais, Harmse, Nestel and Rodriguez-Amaya (2006) with slight modifications. Two slices (approx. 1 cm thick) were taken from each sample of bread and homogenised for about 1 min using a blender. Bread slices were in a frozen state (-20^{o}C) to prevent the degradation of carotenoids due to heat generated by the rotating blades. A portion of ground sample, weighing 1 g, was extracted with 10 ml acetone containing 0.1% butylated hydroxytoluene (BHT) using a mortar and pestle. The extraction with acetone was repeated for about 3 to 4 times until most of the pigments were removed from the residue. The extracts were collected and filtered through a Buchner funnel with filter paper (Whatman no.1). More solvent was used to rinse the residue, funnel and filter paper. The extracts were combined and the carotenoid solution was made up to volume (30-70 ml depending on the amount of carotenoids in the sample) with the extracting solvent. An 8 ml aliquot of extract was collected and dried in a rotary evaporator at 35^{o}C. The volume of extract collected was decided based on preliminary tests. The dry sample was dissolved in 1 ml of methanol:methyl-*tert*-butyl ether (80:20, v/v) containing 0.1% butylated hydroxytoluene (BHT), filtered through a 0.2 µm PTFE syringe filter directly into amber sample vials and 10 µL were injected onto the chromatograph. The analysis of carotenoids from breads was carried out under dim light and aluminium foil was used to cover all glassware to minimise the chances of direct contact with light. Three breads samples were analysed twice to obtain 6 values for each type of bread. The results were reasonably repeatable as shown by the coefficients of variation (2.8-1.2%) and the response of the detector was checked regularly before the analysis.

Chromatographic analysis of carotenoids

The quantitative analysis of carotenoids was carried out using a Prominence Ultra Fast Liquid Chromatograph (Shimadzu, Tokyo, Japan) equipped with a SIL-20A Prominence auto-sampler, a DGU-20A$_3$ Prominence degasser, a CTO-10AS VP Shimadzu column oven and a SPD-M20A Prominence diode array detector. The detection was done at 450 nm and UV/Vis spectra of carotenoids were recorded between 200 to 600 nm. The separation of carotenoids was performed at 25^{o}C on a C30 YMC carotenoid column (250 x 4.6 mm, i.d., 5 µm particle size) by isocratic elution with a mobile phase consisting of methanol:methyl-*tert*-butyl ether (80:20, v/v) at a flow rate of 0.8 ml/min (Kimura, Kobori, Rodriguez-Amaya and Nestel, 2007). The quantification of carotenoids was done using a calibration curve of β-carotene standard (Merck, South Africa). The purity of the standard β-carotene was determined and the concentrations of standard solutions were corrected accordingly (Rodriguez-Amaya and Kimura, 2004).

Estimation of the contribution of OFSP-wheat composite breads to vitamin A requirements

Vitamin A values of breads was calculated as retinol activity equivalents (RAE) using an RAE conversion factor of 12 µg β-carotene to 1 µg retinol (FAO/WHO, 2005). The vitamin A activity of *cis*-isomers of β-carotene was estimated to be one-half of that of all-*trans*-β-carotene (Haskell *et al.*, 2004). The contribution of each type of bread to the vitamin A requirements for different groups of people was determined based on one portion of bread weighing 100 g (approximately 2 slices). Those groups were children (3-10 years), adolescents (10-18 years), adult males and females (19-65 years) as well as pregnant and lactating women.

Physical properties of OFSP-wheat composite breads

Oven spring, loaf weight, loaf volume and specific volume

Oven spring was calculated by subtracting the height of the dough after proofing from the height of the bread after baking. Loaf weight (g) was determined using a digital balance while loaf volume (cm^3) was measured by rape seed displacement method. Specific volume (cm^3/g) was determined by dividing loaf volume by loaf weight. Four samples were analysed.

Colour analysis

After baking, bread samples were cooled to room temperature and colour measurements were done on 4 different locations (on top and sides) of the crust and 3 locations (in the centre and towards the edges) of the crumb using a Chroma Meter CR-400 (Konica Minolta Sensing, Inc. Japan). Colour analysis was done based on L* (measure of lightness ranging from 0-100 indicating black to white), a * (+a, redness and –a, greenness) and b* (+b, yellowness and –b, blueness) values (Shalini and Laxmi, 2007). The results obtained were used to determine the chroma and hue angle according to the method of Little (1975) as follows:

Chroma=$\sqrt{a^2+b^2}$

Hue angle=$\tan^{-1}(b/a)$

HPLC analysis of sugars in OFSP -wheat composite flours

The extraction of sugars was performed according to Liu, Robinson, Madore, Witney and Arpaia (1999) with slight modifications. The sample (0.1 g) was mixed with 10 mL 80% (v/v) ethanol and homogenised by vortexing for 1 min. The mixture was incubated in a water bath at 80°C for 60 min to extract the soluble sugars. After centrifugation at 12000 g for 15 min, the supernatant was taken to dryness in a rotary evaporator at 60°C. Dried sample was dissolved in 2 ml of 50% acetonitrile, filtered through a 0.45 μm nylon filter and 100 μl of the sample was injected onto the chromatograph. Sugars were analysed using an HPLC (Hewlett Packard Agilent 1100 series) with a refractive index detector. A ZORBAX carbohydrate analysis column (150 x 4.6 mm, i.d.) was used for the separation of sugars. An isocratic run was performed with acetonitrile:water (75 : 25, v/v) at a flow rate of 2 ml/min. The identification of the sugars was achieved by comparing their retention times with known sugar standards (glucose, sucrose and fructose). The amount of each identified compound was expressed in μg/mg of dry sample.

Descriptive sensory analysis

Recruitment of the panel

Students from the University of Pretoria, who were willing to consume OFSP-wheat composite bread and did not suffer from any food allergies and had participated on at least two other descriptive sensory panels, were invited to apply for the descriptive sensory analysis of OFSP-wheat composite breads. Eight individuals responded and attended the introduction session but only seven confirmed their availability during the evaluation of breads.

Training of the panel

The training of the panel was done during 5 sessions of about 2 h each per day following the generic descriptive analysis method as described by Einstein (1991). During the training, panellists described the differences between OFSP-wheat composite bread samples at least two times. Seventeen descriptive terms (Table 4.2.1) and scale (1-10) were developed, defined and used during the evaluation. Before the actual evaluation the panellists' performance was checked using Compusense software (Compusense five ® release 4.6, Guelph, Ontario, Canada). Reference samples used are provided in Table 4.2.1.

Sample evaluation

The descriptive sensory analysis of OFSP-wheat composite breads was conducted in a sensory evaluation laboratory with individual booths equipped with computers for direct data entry. The evaluation of OFSP-wheat composite breads was performed three times in three different sessions (five samples were analysed during each session) whereby one sample (1/2 slice) representing each type of bread was randomly presented to the panellist in a transparent polyethylene zip-lock bag with a random three-digit code. Panellists were seated in separate sensory booths under red light to minimise the influence of the orange colour imparted by OFSP flour and a glass of water was provided in order to cleanse their palates. Each of the four bread samples as well as a commercial white bread (Sasko) was rated for appearance, aroma, texture and flavour characteristics using the descriptors generated during the training session. Ten-point line scales were used to measure the intensity of each attribute for a given sample. The minimum value was 1 denoting not perceived and the maximum point was 10 denoting strongly perceived. Responses were recorded directly into the computers and analysed using Compusense software (Compusense five ® release 4.6, Guelph, Ontario, Canada).

Statistical analyses

One way analysis of variance (ANOVA) was used to test for differences in the effects among the various substitution levels of wheat flour with orange-fleshed sweet potato flour on the dependent variables (e.g. physical characteristics of breads and sensory attributes). Significant differences between means were determined using Fisher's least significant difference test (LSD) at 5% probability level ($p < 0.05$). Statistica (Version 10, Statsoft, Tulsa, USA) was used for data analysis. Furthermore, Principal Component Analysis (PCA) was performed to determine the variations between samples based on sensory attribute loadings.

Table 4.2.1 Descriptive sensory lexicon developed by the descriptive sensory panel to evaluate sensory properties of OFSP-wheat composite breads

Sensory attributes	Definition	Reference	Rating scale
Appearance/visual attributes			
Visual moistness	The extent of surface wetness of the crumb from dry to wet.	Pick 'n Pay double chocolate muffin (rated 10)	0=very dry, 10=very wet
Crust darkness	Degree of darkness in the crust ranging from a light brown to a dark brown colour.	Pick 'n Pay double chocolate muffin (rated 10)	0=not a dark brown colour at all, 10=very dark brown colour
Crumb denseness	The extent of compactness/denseness of the crumb.	Woolworths 100% rye bread (rated 10)	0=Not compact; 10=very compact
Uniformity of gas cells or holes in the crumb	Homogeneity of the size of the gas cells or holes in the crumb.	Sasko white bread (rated 10)	0=Not uniform; 10-very uniform
Aroma			
Sweet potato aroma	Fundamental aroma sensation associated with cooked sweet potatoes.	Boiled sweet potatoes (rated 9)	0=no sweet potato aroma, 10=intense sweet potato aroma
Wheat aroma	Fundamental aroma sensation associated with white wheat bread.	Pick 'n Pay poppy seed bread (rated 10)	0=no wheat aroma, 10=intense wheat aroma
Yeasty (fermented) aroma	Odour associated with aroma compounds with regard to yeast fermentation	Wheat flour dough (50g wheat flour, 2% compressed yeast, 3% sugar and 60% water) (rated 10)	0=yeasty/fermented aroma, 10=intense yeasty/fermented aroma
Heated sugar aroma	Typical aroma sensation associated with heated cane sugar.	Caramelised sugar (rated 4). Sugar was heated at 190^{o}C for 45 min to obtain a caramel aroma.	0=no heated sugar aroma, 10=intense heat sugar aroma

Table 4.2.1 Descriptive sensory lexicon developed by the descriptive sensory panel to evaluate sensory properties of OFSP-wheat composite breads (continued)

Sensory attributes	Definition	Reference	Rating scale
Flavour (taste and aroma)			
Sweetness	Basic taste sensation for which sucrose is typical	Spar fresh white hamburger bun (rated 7)	0=Not sweet, 10=very sweet
Saltiness	Fundamental taste sensation elicited by sodium chloride	Pick ‘n Pay poppy seed bread (rated 8)	0=Not salty, 10=very salty
Sweet potato flavour	Typical taste sensation associated with cooked sweet potatoes	Boiled sweet potato (rated10)	0=No sweet potato flavour, 10= intense sweet potato flavour
Wheat-based bread flavour	Typical taste sensation associated with freshly baked commercial wheat bread	Spar fresh white hamburger bun (rated 7)	0 = No freshly-baked wheat bread flavour; 10 = intense freshly-baked wheat bread flavour
Bitter taste	Fundamental taste sensation of which caffeine or quinine are typical	Burnt cooked sweet potato (rated 10)	0=not bitter, 10=very bitter
Texture			
Firmness	Force required to compress the crumb between fingers	Commercial rye bread (rated 10) Pick ‘n Pay rye bread (rated 7) Sasko white bread (rated 3)	0=not firm, 10=very firm
Springiness	Degree to which the crumb returns to its original shape after compression with the flat top part of the second finger	Sasko white bread (rated 10)	0=not springy, 10=very springy
Chewiness (crumb)	Number of chews required to masticate the sample (1 chew/sec) to the consistency suitable for swallowing.	White Albany bread (rated 7) N.B: Crumb alone	0=not chewy, 10 very chewy
Sticky	The extent to which bread material stick on the teeth and hard palate after swallowing	Rye bread (rated 10) Albany white bread (rated 7)	0=not sticky, 10= 0=very sticky

4.2.3 Results and Discussion

4.2.3.1 Effect of substituting wheat flour with OFSP flour and gluten supplementation on bread-making performance of OFSP-wheat composite flour

Increasing the proportion of OFSP flour in composite flours led to decreases in height of the dough after proofing, height of bread after baking, oven spring, loaf volume and specific volume but resulted in increase in loaf weight (Table 4.2.2).

Table 4.2.2 Effect of substituting wheat flour with OFSP flour and gluten supplementation on bread-making performance of OFSP-wheat composite flour

OFSPF:WF (w/w)	Gluten added (%) [1]	Height of dough after proofing (cm)	Height of bread after baking (cm)	Oven Spring (cm)	Loaf volume (cm^3)	Loaf weight (g)	Specific volume (cm^3/g)
0:100	0	14.3 ± 0.3^{a}	18.4 ± 0.2^{a}	4.1 ± 0.5^{a}	3370 ± 71^{a}	620 ± 4^{a}	5.4 ± 0.1^{a}
10:90	1.2	12.5 ± 0.2^{b}	14.1 ± 0.5^{b}	1.6 ± 0.7^{b}	2990 ± 14^{b}	631 ± 5^{b}	4.8 ± 0.0^{b}
20:80	2.3	10.8 ± 0.5^{c}	12.8 ± 0.3^{c}	2.1 ± 0.6^{b}	2605 ± 21^{c}	641 ± 3^{c}	4.2 ± 0.0^{c}
30:70	3.5	8.7 ± 0.3^{d}	8.8 ± 0.3^{d}	0.1 ± 0.1^{c}	1794 ± 33^{d}	647 ± 1^{c}	2.9 ± 0.0^{d}

Different subscripts in the same column indicate that means were significantly different ($p < 0.05$) ; n=4

OFSPF: Orange-fleshed sweet potato flour, WF: wheat flour.

[1] The weight of gluten equivalent to the quantity lost during the substitution of wheat flour with OFSP flour was added to composite flours.

Although all composite flours contained the same amount of gluten as compared to the control, the addition of exogenous gluten did not increase the height of the doughs after proofing, the height of the breads after baking and oven spring. Additionally, the loaf volume also decreased with increasing substitution of wheat with OFSP flour. The combination of gluten proteins from different sources (endogenous and exogenous) might have resulted in a gluten structure with different properties from its main constituents (Kontogiorgos, 2011) presumably because both gluten proteins (endogenous and exogenous) were different in terms of glutenin and gliadin composition (Wieser, 2000). Uthayakumaran, Gras, Stoddard and Bekes (1999) demonstrated that increase in the glutenin-to-gliadin ratio increased the loaf height and loaf volume while the decrease in the former reduced them. The gliadin, that promotes viscous flow and extensibility of the dough, may interact with one another or with glutenin through hydrophobic interactions and hydrogen bonding (Van Der Borght *et al.*, 2005). It is believed that differences in the way gluten proteins interact may also play a key role in determining the network formation and rheological properties of gluten (Kontogiorgos, 2011) which may eventually affect the rheology of the dough and its bread-making performance.

The starch-gluten matrix formed during the initial stage of dough expansion reinforces the gas cells thereby contributing to the increase in loaf volume (Ohm and Chung, 1999) and loaf height. The membrane surrounding the gas cell should be able to sustain large extensions without rupturing. The disruption of the exterior membranes would lead to a loss of gas and a decrease in loaf volume (Kim, Morita, Lee and Moon, 2003). The presence of dietary fiber from OFSP flour (1.4 - 10%, db) (van Hal, 2000) may also interfere with the formation of gluten network thereby negatively affecting the loaf volume (Stojceska and Ainsworth, 2008).

The specific volume is described as the most reliable measure of loaf size (Shittu, Raji and Sanni, 2007) as it indicates the volume of bread per unit weight. The specific volume of breads differed significantly with the control having the highest value. Breads containing OFSP flour showed a decrease in specific volume as more wheat flour was substituted. This was due to increase in weight of breads as more OFSP flour was supplemented (indicating that less water evaporated from these samples) while their volume decreased gradually.

4.2.3.2 Crust colour and descriptive sensory analysis of OFSP-wheat composite bread

The lightness (L* value) of the crust of breads containing OFSP flour was lower compared with wheat flour bread (Table 4.2.3). The a* and b* values decreased with increased OFSP flour addition. Colours of the crust were not evenly distributed on all sides of the crust (top and other sides) which resulted in slightly large standard deviations. Presumably, the intense browning in samples containing more OFSP flour camouflaged, to a certain extent, the orange and yellow colours on the crust. The darkness of the crust may be attributed to the Maillard reaction which is a non-enzymatic browning reaction (Martinez-Anaya, 1996; Ajandouz *et al.*, 2001). During this reaction, carbonyl groups of reducing sugars (e.g., glucose and fructose) condense with the free amino group of proteins (Michalska, Amigo-Benavent, Zielinskiand del Castillo, 2008; Hallén, Ibanoğlu and Ainsworth, 2004) leading to a series of chemical reactions which result in the formation of nitrogenous polymers and co-polymers known as melanoidins with brown pigmentation (Martins, Jongen and Van Boekel, 2001).

Table 4.2.3 Effect of substituting wheat flour with OFSP flour on the colour of the crust

OFSPF:WF (w/w)	L*	a*	b*
0:100	34.3 ± 4.8^{a}	5.5 ± 2.7^{a}	8.4 ± 1.0^{a}
10:90	29.5 ± 3.9^{b}	7.2 ± 0.9^{ab}	9.6 ± 1.8^{a}
20:80	24.7 ± 2.5^{c}	6.8 ± 1.2^{ab}	6.0 ± 1.6^{b}
30:70	23.8 ± 2.4^{c}	6.1 ± 1.9^{c}	4.3 ± 1.7^{b}

Different subscripts in the same column indicate that means were significantly different ($p < 0.05$) n=3
OFSPF: Orange-fleshed sweet potato flour, WF: wheat flour.

Sugar degradation reactions in the absence of amino groups (caramelisation) of proteins may have also contributed to the browning of the crust. The sugar profile of composite flours revealed increased amount of sugars upon gradual substitution of wheat flour with OFSP flour (Table 4.2.4). Intense browning was more evident in bread containing 30% OFSP flour probably because it was made from composite flours with the highest amount of sugars especially sucrose which is a non reducing sugar.

Table 4.2.4 Sugar profile of OFSP-wheat composite flours

OFSP:WF (W/W)	Fructose (mg/100g, db)	Glucose (mg/100g, db)	Sucrose (mg/100g, db)
0:100	ND	121 ± 2^{a}	633 ± 18^{a}
10:90	534 ± 29^{a}	650 ± 12^{b}	2223 ± 50^{b}
20:80	959 ± 35^{b}	1215 ± 33^{c}	3841 ± 82^{c}
30:70	1288 ± 28^{c}	1618 ± 38^{d}	5328 ± 63^{d}

Different subscripts in the same column indicate that means were significantly different ($p < 0.05$); n=3
OFSPF: Orange-fleshed sweet potato flour, WF: wheat flour. ND: Not detected.

Results obtained from the descriptive sensory analysis (Table 4.2.5) on the darkness of the crusts of breads agreed with L* value obtained from the colorimeter (Table 4.2.3). Crusts of breads made with OFSP-wheat composite flours were described as darker when compared with bread samples made from wheat flour alone probably because the former contained more sugars (Fig. 4.2.1).

OFSPF: Orange-fleshed sweet potato flour, WF: wheat flour.
Figure 4.2.1 Orange-fleshed sweet potato-wheat composite breads

Crumbs of bread samples containing OFSP flour were found to be denser (visual appearance, Table 4.2.5) compared to bread made from wheat flour alone. The denseness may be attributed to inability of the gas cells to expand properly in order to provide enough open spaces in the crumb. This probably affects the movement of the moisture or water vapour from the crumb since breads containing OFSP flour appeared to be moister compared to the control. The inability of the gas cells to expand did not only affect the movement of water vapour but it also affected textural properties of the crumb since they were described to be relatively firm due to the denseness of the crumb. The increase in the firmness may also result from an increment of dietary fibres from OFSP flour (1.4 - 10%, db) (Van Hal, 2000) which would interfere with the gluten network resulting in the depression of bread volume (Morrice and Morrice, 2012).

Breads containing OFSP flour were also characterised as having a sweet potato aroma, heated sugar aroma, more sweetness and less salty taste. The sweet potato aroma may be attributed to the presence of sweet potato flour in the breads. Bread containing 30% OFSP flour was regarded as having the most intense sweet potato aroma and heated sugar aroma compared to other types of breads. The heated sugar aroma may be associated with the caramelisation of sugars (glucose, fructose and sucrose) from OFSP flour. Aromatic volatile compounds such as ethylbenzene and trimethylbenzene, for example, can be produced by heating glucose during caramelisation (Kitts, Wu, Kopec and Nagasawa, 2006). These sugars could also explain the reason for the sweetness perceived in breads containing OFSP flour which probably masked the saltiness in those samples.

Bitterness could be detected in the samples containing OFSP flour but this was perceived only in their crusts. The bitterness was presumably due to the melanoidins which are bitter compounds from the Maillard reaction (Pedreschi, Kaack, Granby and Troncoso, 2007). The bitterness in the crust could also be attributed to high molecular weight colour bodies with bitter taste formed during the caramelisation of sugars (Kamuf, Nixon, Parker and Barnum, 2003; Pozo-Bayó, Guichard and Cayot, 2006).

Principal component analysis (PCA) was used to summarise the variation in the descriptive sensory attributes of OFSP-wheat composite breads. The first factor of the PCA explained 83.8% of the total variation among the breads and separated them based on the amount of OFSP flour they contained. Breads containing 20 and 30% OSFP flour were located to the right axis of the PC1 while breads containing 10% OFSP flour and the control were located to the left side (Fig. 4.2.2a). PC2 accounted for 13.9 % of total variation of the sensory data. OFSP flour was the most important component that changed sensory properties of the breads as shown by the pattern in which they were distributed on the PCA plot. The clustering of the bread samples (Fig. 4.2.2a) was similar to that of the sensory attributes that significantly discriminated among them (Fig. 4.2.2b). All the attributes related to sweet potato such as sweetness, sweet potato flavour, sweet potato aroma, bitterness and heated sugar aroma, were loaded across the positive axis of the PC1. Samples containing 100% wheat flour (control and commercial wheat flour bread, Sasko) contrasted with those supplemented with OFSP flour by displaying a negative loading along the PC1 axis.

Table 4.2.5 Effect of substituting wheat flour with OFSP flour on sensory characteristics of the bread

Sensory attributes	0-100	10-90	20-80	30-70	Sasko
Appearance/visual attributes					
Crust darkness	3.6 ± 1.3^{a}	4.6 ± 1.0^{b}	6.5 ± 1.2^{c}	6.7 ± 1.2^{c}	2.2±1^{d}
Crumb denseness	3.0 ± 1.5^{a}	4.0 ± 1.7^{b}	5.4 ± 1.6^{c}	7.0 ± 1.6^{d}	4.3±1.7^{c}
Uniformity of gas cells	4.3 ± 2.0^{a}	3.7 ± 1.8^{a}	4.6 ± 1.8^{a}	5.0 ±2.0^{a}	6±2^{a}
Visual moistness	2.4 ± 1.3^{a}	3.1 ± 1.0^{a}	4.2 ± 1.3^{b}	5.4 ± 2.0^{c}	3.3±1.5^{bc}
Aroma					
Sweet potato aroma	0 ± 0^{a}	3.8 ± 1.5^{b}	5.2 ± 1.4^{c}	6.1 ± 1.5^{d}	0±0^{d}
Wheat aroma	5.3 ± 1.2^{a}	3.0 ± 1.4^{b}	2.5 ± 1.5^{bc}	2.0 ± 1.8^{c}	5±1.3^{a}
Yeasty or fermented aroma	2.1 ± 1.3^{a}	2.3 ± 1.4^{a}	2.8 ± 1.5^{ab}	3.8 ± 2.0^{b}	2.1±1.5^{b}
Heated sugar aroma (caramel)	1.0 ± 1.6^{a}	2.0 ± 1.2^{b}	3.4 ± 1.0^{c}	4.6 ± 1.0^{d}	0.5±1.1^{d}
Flavour					
Sweetness	2.0 ± 1.0^{a}	3.8 ± 0.8^{b}	5.0 ± 1.4^{c}	6.1 ± 1.5^{d}	1.5±08^{d}
Saltiness	3.0 ± 1.3^{a}	1.6 ± 1.0^{b}	1.7 ± 1.0^{b}	1.1 ± 0.7^{b}	4±1^{a}
Sweet potato flavour	0.1 ± 0.2^{a}	4.0 ± 1.3^{b}	5.3 ± 1.7^{c}	6.6 ± 1.4^{d}	0±0^{d}
Wheat based bread flavour	5.0 ± 1.0^{a}	2.5 ± 1.5^{b}	2.3 ± 1.5^{bc}	1.6 ± 1.4^{c}	5.1±1.2^{a}
Bitterness (crust)	2.4 ± 1.3^{a}	3.2 ± 1.0^{ab}	4.2 ± 1.4^{b}	4.3 ± 1.6^{b}	2±1.3^{c}
Texture					
Firmness	2.4 ± 1.4^{a}	3.5 ± 1.6^{b}	4.0 ± 1.5^{c}	6.5 ± 1.7^{d}	2.7±1.4^{d}
Springiness	6.4 ± 1.8^{a}	5.0 ± 1.7^{b}	4.0 ± 1.5^{b}	2.5 ± 1.3^{c}	6.8±1.6^{a}
Chewiness (crumb)	4.2 ± 1.7^{a}	4.4 ± 1.3^{a}	5.0 ± 1.7^{ab}	5.6 ± 2.0^{b}	5±1.4^{ab}
Stickiness	4.1 ± 1.6^{a}	4.2 ± 1.4^{a}	4.4 ± 1.5^{a}	5.0 ± 1.6^{a}	4.7±1.6^{a}

Different subscripts in the same row indicate that means were significantly different ($p < 0.05$). 0:100, 10:90; 20:80 and 30:70 represent breads containing 0, 10, 20, 30% OFSP flour respectively. OFSP: Orange-fleshed sweet potato. Sasko= Commercial wheat flour bread.

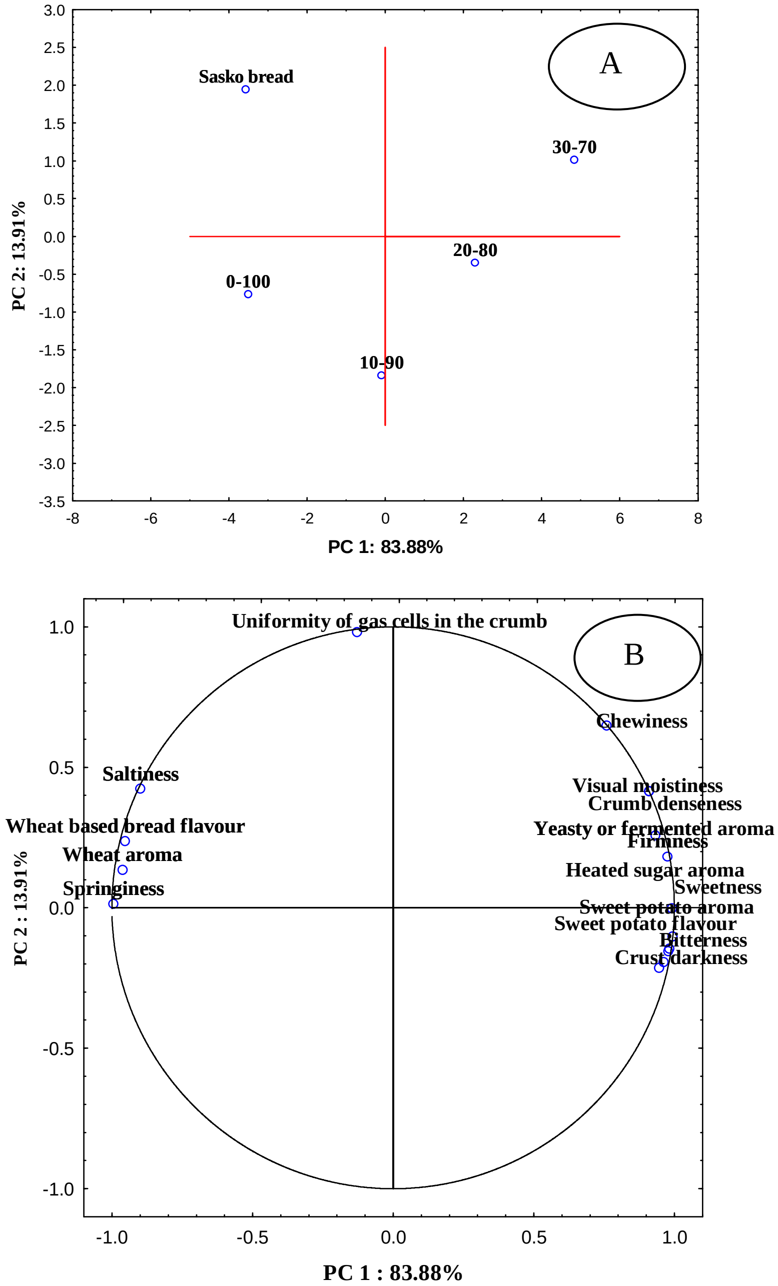


Figure 4.2.2 Principal component analysis of the breads (A) Plot of the first two principal component scores of the breads (B) Plot of the first two principal component loadings projections of the sensory attributes. 00:100; 10:90; 20:80 and 30:70 represent breads supplemented with 0, 10, 20 and 30% orange-fleshed sweet potato flour. Sasko= Commercial wheat flour bread.

4.2.3.3 Effect of baking OFSP-wheat composite breads on the retention of all-trans-β-carotene and colour

Carotenoids detected in doughs and breads are shown in Figure. 4.2.3. Compositing wheat flour with OFSP flour resulted in an increase in the amount of all-*trans*-β-carotene in composite breads (Table 4.2.6). Neither all-*trans*-β-carotene nor other forms of carotenoids were detected in bread containing 100% wheat flour. The baking process led to the production of *cis* isomers of β-carotene at the expense of all-*trans*-β-carotene. These include 9-*cis*-β-carotene and 13-*cis*-β-carotene. The identification of all-*trans*-β-carotene was achieved by co-chromatography using authentic all-*trans*-β-carotene standard and the visible absorption spectrum. The *cis* isomers of β-carotene were identified by the wavelengths of maximum absorption and the appearance of a *cis* peak (Fig. 4.2.3).

The retention of all-*trans*-β-carotene differed significantly among all types of breads despite the fact that they were subjected to the same baking conditions (190°C for 30 min). The retention of all-*trans*-β-carotene in breads containing 10, 20 and 30% OFSP flour was 63, 71 and 83% respectively, suggesting that the loss of all-*trans*-β-carotene did not follow the same rate. The degradation of all-*trans*-β-carotene during heating follows first-order kinetics (Chen, Chen and Chien, 1994; Aparicio-Ruiz, Mínguez-Mosquera, Gandul-Rojas, 2011) that is, the reaction rate is proportional to the concentration under isothermal conditions (Dhuique-Mayer *et al.*, 2007). The degradation rate of carotenoids depends on temperature and increases as the temperature increases (Chen, Peng and Chen, 1995). The differences in the degradation kinetics of carotenoids in OFSP-wheat composite breads were therefore attributed to differences in the temperatures prevailing inside the breads as explained in the subsequent paragraph. The influence of time was not taken into consideration because all bread samples were baked for the same period of time (30 min).

The moisture inside the breads seemed to have played a critical role with regard to heat transfer throughout the breads. The weight of breads containing OFSP flour, taken immediately after baking, was found to be significantly higher (Table 4.2.2) as the amount of OFSP flour increased suggesting that less water evaporated from those samples. It is important to recall that the weight of the dough was initially the same for all samples, i.e., 700 g. The water absorption capacity (required to reach 500 FU) of the flours containing 10, 20 and 30% OFSP flour was 55, 55.5 and 57.5% respectively (Appendix 1). However, high moisture in the bread does not allow achievement of extremely high temperature in the crumb (Capuano, Ferrigno, Acampa, Ait-Ameur, Fogliano, 2008) which would be more destructive.

The aqueous environment may also have had a protective effect against oxidative degradation of all-*trans*-β-carotene (Goldman, Horev and Saguy, 1983; Tsimidou and Tsatsaroni, 1993; Dhuique-Mayer *et al.*, 2007). Carotenoids act as antioxidants in the presence of free radicals and get easily degraded (Jadhav, Nimbalkar, Kulkani, Madhavi, 1996; Hidalgo, Brandolini and Pompei, 2010). Carotenoid stability towards oxidation is a function of both the number of conjugated double bonds and the presence of functional groups (Henry *et al*., 2000) that can be used by oxidising agents to interact with carotenoids. Hydrogen bonding between hydroperoxides and water, metal catalyst inactivation by water and the reduction of free radical content by interaction with water are some of the mechanisms by which water might have, in part, contributed to the stability of all-*trans*-β-carotene (Glória, Vale and Bobbio, 1995).

Baking OFSP-wheat composite breads in the conventional oven led to the formation of new carotenoid compounds at the expense of all-*trans*-β-carotene. The prevailing temperature inside the oven was enough to bring about the isomerisation of all-*tans*-β-carotene molecule (Chandler and Schwartz, 1988). All-*trans*-β-carotene, being the most abundant carotenoid in OFSP (Kimura *et al*., 2007; van Jaarsveld *et al*., 2006; Bechoff *et al*., 2009), was the main carotenoid detected in the doughs and breads containing OFSP flour. However, it was not detected in the dough and bread made exclusively from wheat flour as previously reported (Ranhotra, Gelroth, Langemeier and Rogers, 1995; Rogers, Malouf, Langemeier, Gelroth and Ranhotra, 1993). Apart from durum variety of wheat, which may contain up to 7.3 mg/kg of total carotenoids (Belitz and Grosch), wheat grain may contain traces or no β-carotene (Rogers *et al*., 1993; van Hal, 2000).

9-*cis*-β-carotene and 13-*cis*-β-carotene were the main *cis* isomers of β-carotene formed during baking. The amount of 13-*cis*-β-carotene formed was five to eight times greater than the amount initially available in the doughs. In contrast, 9-*cis*-β-carotene found in the breads containing OFSP flour was formed during baking since it was not found in the doughs. The reason for the formation of large amounts of 13-*cis*-β-carotene than 9-*cis*-β-carotene was probably due to the fact that the activation energy for *trans*-*cis* isomerisation about the central double bond is less than about the other double bonds (Zechmeister, 1944). The formation of high quantities of 13-*cis*-β-carotene has been reported in thermally treated sweet potato (Chandler and Schwartz, 1988; Bechoff, Dhuique-Mayer, Marouzé, Reynes and Westby, 2009).

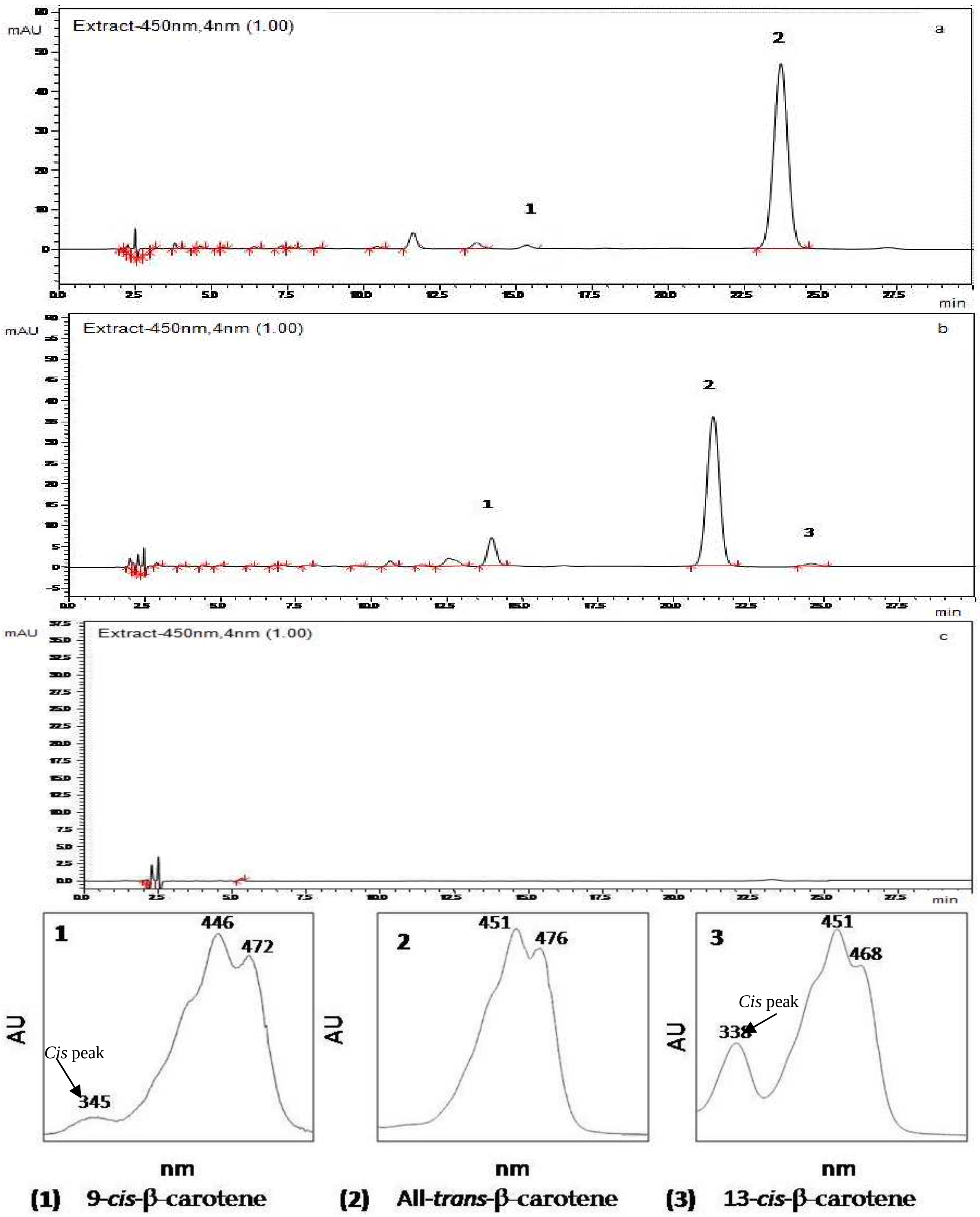


Figure 4.2.3 HPLC chromatograms and UV/Vis spectra of carotenoids from (a) dough containing 30% OFSP flour (b) bread containing 30% OFSP flour and (c) bread containing 100% wheat flour

Colour analysis revealed a drop in a* value and an increase in b* value when the doughs were baked into breads, especially in samples containing OFSP flour (Table 4.2.7). The change in colour was accompanied by a detrimental loss of all-*trans*-β-carotene (Table 4.2.6). Thermo-isomerisation has been reported to decrease the intensity of colour imparted by β-carotene (Chen *et al.*, 1995). The a* value was found to strongly correlate positively (r = 0.889, $p < 0.001$) with the amount of all-*trans*-β-carotene retained (Table 4.2.8) after baking. However, it showed a weak correlation with the amount of all-*trans*-β-carotene lost. In contrast, the correlation between the b* value and the amount of all-*trans*-β-carotene retained was low (r = 0.858, $p < 0.001$) as compared to the a* value but strongly correlated with the amount of all-*trans*-β-carotene lost (r = 0.929, $p < 0.001$). The latter weakly correlated (r = 0.268) with a* value.

The L* value was negatively correlated with all colour parameters. With regard to the all-*trans*-β-carotene levels in the breads, L* value showed a significant but negative correlation with the amount of all-*trans*-β-carotene retained (r = -0.892, $p < 0.001$) indicating that breads containing a high proportion of wheat flour would be expected to have lower β-carotene content. A negative correlation (r = -0.885) between the L* value and β-carotene content has been reported by Takahata, Noda and Nagata (1993) in OFSP roots. Furthermore, it also showed a negative correlation with colour parameters that positively correlated with the amount of β-carotene retained in breads notably the a* value (r = -0.985, $p < 0.001$), b* value (r = -0.553, $p < 0.01$) and chroma (r = -0.710, $p < 0.001$) but positively correlated with the hue angle (r = 0.981, $p < 0.001$). The hue angle showed an inverse and strong correlation (r = -0.822, $p < 0.001$) with the amount of all-*trans*-β-carotene retained. The latter positively correlated with chroma (r = 0.941, $p < 0.001$). These findings indicated that colour parameters such as a* value and chroma may, in part, be used to predict the β-carotene content in breads or any other products containing OFSP flour if the OFSP cultivar used does not contain other natural colorants.

The present observations are in agreement with the results obtained from previous studies on OFSP products (Takahata, Noda and Nagata, 1993; Bengtsson, Namutebi, Alminger and Svanberg, 2008). However, they did not consider other colour parameters such as chroma and hue angle. Chroma is a measure of the intensity (Lewis, Bloor and Schwinn, 1998) or purity of a colour (Adamson, 2010). High chroma colours look rich and full while low chroma colours look dull and grayish (Adamson, 2010). Hue angle relates colour to a position on a

Table 4.2.6 Effect of substituting wheat flour with OFSP flour and baking on β-carotene content (db) in OFSP-wheat composite breads

	Dough		Bread				
OFSPF:WF (w/w)	All-*trans*-β-carotene (µg/100g of dough)	13-*cis*-β-carotene (µg/100g of dough)	All-*trans*-β-carotene (µg/100g of bread)	9-*cis*-β-carotene (µg/100g of bread)	13-*cis*-β-carotene (µg/100g of bread)	Retention of All-*trans*-β-carotene (%) [1]	Vitamin A activity (µg RAE /100g of bread) [2]
0:100	ND	ND	ND	ND	ND	ND	0.0
10:90	3100.0 ± 57.0^{a}	39.6 ± 1.0^{a}	2037.4 ± 46.0^{a}	47.6 ± 1.2^{a}	349.6 ± 8.8^{a}	65.7 ± 1.5^{a}	186.3
20:80	5956.0 ± 120.0^{b}	82.3 ± 1.2^{b}	4251.7 ± 85.0^{b}	120.3 ± 2.3^{b}	703.5 ± 13.0^{b}	71.4 ± 1.4^{b}	388.6
30:70	8020.0 ± 167.0^{c}	114.7 ± 3.0^{c}	6657.1 ± 101.0^{c}	148.4 ± 4.1^{c}	862.6 ± 14.0^{c}	83.0 ± 1.2^{c}	596.9

[1] The retention was determined based on the remaining amount of all-*trans*-β-carotene after baking because other carotenoids were formed at its expense.

[2] The standard deviations were not provided because the vitamin A activity (µg RAE /100g of bread) of each type of bread was determined based on different forms of provitamin A carotenoids (n=3)

Different subscripts in the same column indicate that means were significantly different ($p < 0.05$)

OFSPF: Orange-fleshed sweet potato flour, WF: wheat flour. RAE: Retinol activity equivalent.

Db: Dry basis

ND: Not detected

$$\text{Retention of all-}\textit{trans}\text{-}\beta\text{-carotene (\%)} = 100\text{g} - \left(\frac{\text{D-B}}{\text{D}}\right) \text{x } 100$$

D: all *trans*-β-carotene content (%, db) in the dough

B: all-*trans*-β-carotene (%, db) in bread.

colour circle or wheel with red at 0°, yellow at 90°, green at 180° and blue at 270° (Lewis *et al.,* 1998).

However, colour parameters may not always be used to predict the β-carotene content in sweet potato products especially when the sweet potato cultivars used contain other types of natural colorants. A study conducted by Ameny and Wilson (1997) on sweet potato revealed that a* value could not always reflect the β-carotene content in sweet potato. Kidmose, Christensen, Agili and Thilsted (2007) also reported that the β-carotene content in OFSP was not reflected in the intensity of the orange colour. Presumably, the presence of high amounts of anthocyanins in sweet potato (Oki *et al.*, 2002; Philpott, Gould, Markham, Lewthwaite and Ferguson, 2003) can also contribute to the colour of sweet potato products (Bovell-Benjamin, 2007). Anthocyanins possess various colours such as orange, red, purple and blue which can be used as natural colourants in soft drinks, confectioneries and bakery products (Bovell-Benjamin, 2007). Resorting to chemical analysis, however, remains the most reliable way of estimating the β-carotene content in OFSP products.

4.2.3.4 Contribution of OFSP-wheat composite bread to vitamin A requirements

The contribution of breads to the recommended dietary allowance (RDA) of vitamin A largely depended on the proportion of OFSP flour in the breads (Table 4.2.9). It also varied based on the physiological requirements of each group of individuals. One portion of bread weighing 100 g (approximately 2 slices) containing 10 or 20% OFSP would meet 28.9 and 61% respectively of the RDA of retinol amongst children aged between 3-10 years. On the other hand, the same portion of bread would provide far less amount of vitamin A for an adult male and female (19-65 years old). In contrast, the same portion of bread supplemented with 30% OFSP flour would provide the amount of vitamin A close to the RDA of vitamin A for children aged between 3-10 years and more than a half for adolescents, adult males and females. Pregnant and lactating women may still need additional amounts of retinol to fulfil both their needs and those of their children. This is because all types of bread studied would meet less than 50% of the RDA of vitamin A for those groups of people.

The formation of *cis* isomers of β-carotene led to a decrease in the nutritional value of OFSP-wheat composite breads. This was, possibly, due to the fact that *cis* isomers have vitamin A activity that is approximately 50% or less of that of corresponding all-*trans* carotenoids (Lessin, Catignani and Schwartz, 1997, Kidmose *et al*., 2007). 9-*cis*-β-carotene provided 1.2, 3.1 and 3.7 µg of vitamin A as RAE (µg/100 g) in breads containing 10, 20 and 30% OFSP flour respectively while 13-*cis*-β-carotene contributed 9, 18.4 and 21.5 µg of vitamin A as RAE (µg/100 g) in the same types of breads (Table 4.2.10). The contribution of all-*trans*-β-carotene was greater when compared to the contribution of *cis* isomers as it provided 105.4, 222.5 and 331.7 µg of vitamin A as RAE (µg/100 g) in breads supplemented with 10, 20 and 30% OFSP flour respectively.

All-*trans*-β-carotene was the major contributor to the RDA of vitamin A in all groups studied (Table 4.2.10). 13-*cis*-β-carotene was the second major contributor to the RDA of vitamin A followed by 9-*cis*-β-carotene probably because the latter was formed in low amounts. The contribution of each provitamin A carotenoid depended on the amount of OFSP flour available in composite breads since it increased as more OFSP flour was supplemented.

Table 4.2.7 Effect of baking on the retention of colour in OFSP-wheat composite bread

OFSPF:WF (%)	Colour of the dough					Bread colour*				
	L*	a*	b*	Chroma	Hue angle	L*	a*	b*	Chroma	Hue angle
0:100	66.0±2.3^{a}	-0.2±0.1^{a}	11.7±0.7^{a}	11.7±0.7^{a}	-0.8±0.3^{a}	59.6±0.8^{a}	1.4±0.4^{a}	11.8±0.5^{a}	11.8±0.5^{a}	83.3±1.8^{a}
10:90	57.5±2.4^{b}	10.2±0.6^{b}	25±1.6^{b}	27±1.7^{b}	22.3±0.7^{b}	50.7±0.8^{b}	6.3±0.2^{b}	25.4±0.6^{b}	26.2±0.6^{b}	76.1±0.2^{b}
20:80	50.2±1^{c}	15.2±0.4^{c}	22.3±0.5^{c}	27±0.6^{b}	34.2±0.5^{c}	48.6±0.6^{c}	10.7±0.1^{c}	25.6±0.8^{b}	27.8±0.8^{c}	67.2±0.4^{c}
30:70	48.8±0.8^{c}	18.4±0.5^{d}	22.7±1^{c}	29.2±1^{c}	39±0.4^{d}	44±1.3^{d}	13.1±0.2^{d}	23±1.5^{c}	26.4±1.4^{bc}	60.1±1.4^{d}

*Crust and crumb blended together

Different subscripts in the same column indicate that means were significantly different ($p < 0.05$); n=3

OFSPF: Orange-fleshed sweet potato flour, WF: wheat flour.

Table 4.2.8 Relationship between the stability of all-*trans* β-carotene during baking and colour parameters of OFSP-wheat composite bread [1]

Parameters	L*	a*	b*	Chroma	Hue angle	Amount of all-*trans*-β-carotene retained	Amount of all-*trans*-β carotene lost
L*	1						
a*	-0.985*	1					
b*	-0.553**	0.575**	1				
Chroma	-0.710*	0.729*	0.978*	1			
Hue angle	0.981*	-0.987*	-0.444	-0.617**	1		
Amount of all-*trans*-β-carotene retained	-0.892*	0.889*	0.858*	0.941*	-0.822*	1	
Amount of all-*trans*-β carotene lost	-0.237	0.268	0.929*	0.842*	-0.123	0.633**	1

[1] Crust and crumb blended together. OFSP: Orange-fleshed sweet potato

Significance level: *=$p < 0.001$ and **= $p < 0.01$

Table 1 Table 4.2.9 Contribution (%) of OFSP-wheat composite breads (100 g portion, wb) to the RDA of vitamin A in different groups [1]

OFSPF:WF (w/w)	3-10 years children (RDA=400*)	Adolescents 10-18 years (RDA=600**)	Adult female 19-65 years (RDA=500**)	Adult male 19-65 years (RDA=600**)	Pregnant women (RDA=800**)	Lactating women (RDA=850**)
0:100	0.0	0.0	0.0	0.0	0.0	0.0
10:90	28.9	19.2	23.1	19.3	14.5	13.6
20:80	61.0	40.7	48.8	40.7	30.5	28.7
30:70	89.2	59.5	71.4	59.5	44.6	42.0

[1] The standard deviations were not provided because the contribution of each type of bread was determined based on different forms of provitamin A carotenoids.
** RDA adapted from FAO/WHO (2005).
* RDA adapted from FAO (1988), reviewed by Woolfe (1992).
RDA: Recommended Dietary Allowance.

Table 2 Table 4.2.10 Contribution of individual provitamin A carotenoids to the RDA of vitamin A in different groups

OFSPF:WF (w/w)	Provitamin A carotenoids	Vitamin A (µg RAE/100g of bread)	3-10 years children (RDA=400*)	Adolescent 10-18 years (RDA=600**)	Adult Female (RDA=500**)	Adult male (RDA=600**)	Pregnant women (RDA=800**)	Lactating women (RDA=850**)
0:100	9-*cis*-β-carotene	0.0 ± 0.0^{a}	0.0 ± 0.0^{a}	0.0 ± 0.0^{a}	0.0 ± 0.0^{a}	0.0 ± 0.0^{a}	0.0 ± 0.0^{a}	0.0 ± 0.0^{a}
	13-*cis*-β-carotene	0.0 ± 0.0^{a}	0.0 ± 0.0^{a}	0.0 ± 0.0^{a}	0.0 ± 0.0^{a}	0.0 ± 0.0^{a}	0.0 ± 0.0^{a}	0.0 ± 0.0^{a}
	All-*trans*-β-carotene	0.0 ± 0.0^{a}	0.0 ± 0.0^{a}	0.0 ± 0.0^{a}	0.0 ± 0.0^{a}	0.0 ± 0.0^{a}	0.0 ± 0.0^{a}	0.0 ± 0.0^{a}
10:90	9-*cis*-β-carotene	1.2 ± 0.0^{a}	0.3 ± 0.0^{a}	0.2 ± 0.0^{a}	0.2 ± 0.0^{a}	0.2 ± 0.0^{a}	0.2 ± 0.0^{a}	0.1 ± 0.0^{a}
	13-*cis*-β-carotene	9.0 ± 0.2^{b}	2.3 ± 0.1^{b}	1.5 ± 0.0^{b}	1.8 ± 0.0^{b}	1.5 ± 0.0^{b}	1.1 ± 0.0^{b}	1.1 ± 0.0^{b}
	All-*trans*-β-carotene	105.4 ± 2.4^{c}	26.4 ± 0.6^{c}	17.6 ± 0.4^{c}	21.1 ± 0.5^{c}	17.6 ± 04^{c}	13.2 ± 0.3^{c}	12.4 ± 0.3^{c}
20:80	9-*cis*-β-carotene	3.1 ± 0.1^{a}	0.8 ± 0.0^{a}	0.5 ± 0.0^{a}	0.6 ± 0.0^{a}	0.5 ± 0.0^{a}	0.4 ± 0.0^{a}	0.4 ± 0.0^{a}
	13-*cis*-β-carotene	18.4 ± 0.3^{d}	4.6 ± 0.1^{d}	3.1 ± 0.1^{d}	3.7 ± 0.1^{d}	3.1 ± 0.1^{d}	2.3 ± 0.0^{d}	2.2 ± 0.0^{d}
	All-*trans*-β-carotene	222.5 ± 4.4^{e}	55.6 ± 1^{e}	37.1 ± 0.7^{e}	44.5 ± 1^{e}	37.1 ± 0.7^{e}	27.8 ± 0.6^{e}	26.2 ± 0.5^{e}
30:70	9-*cis*-β-carotene	3.7 ± 0.1^{a}	0.9 ± 0.0^{a}	0.6 ± 0.0^{a}	0.7 ± 0.0^{a}	0.6 ± 0.0^{a}	0.5 ± 0.0^{a}	0.4 ± 0.0^{a}
	13-*cis*-β-carotene	21.5 ± 0.3^{d}	5.4 ± 0.1^{d}	3.6 ± 0.1^{d}	4.3 ± 0.1^{d}	3.6 ± 0.1^{d}	2.7 ± 0.0^{d}	2.5 ± 0.0^{d}
	All-*trans*-β-carotene	331.7 ± 5^{f}	83 ± 1.3^{f}	55.3 ± 0.8^{f}	66.3 ± 1^{f}	55.3 ± 0.8^{f}	41.5 ± 0.6^{f}	39 ± 0.6^{f}

Different subscripts in the same column indicate that means were significantly different ($p<0.05$)

** RDA adapted from FAO/WHO (2005).
* RDA adapted from FAO (1988), reviewed by Woolfe (1992).
RDA: Recommended Dietary Allowance.
The retinol activity equivalency factors of 12:1 (FAO/WHO, 2005) and 24:1 (Haskell, *et al.*, 2004) were used for all-*trans*-β-carotene and *cis* isomers respectively.

4.2.4 Conclusions

Baking causes the degradation of all-*trans*-β-carotene which results into the formation of new forms of carotenoids in the *cis* configuration with lower vitamin A activity. The formation of *cis* isomers of β-carotene notably 13-*cis*-β-carotene and 9-*cis*-β-carotene occurs at the expense of all-*trans*-β-carotene which is the major contributor to vitamin A activity of OFSP-wheat composite breads. Nevertheless, the latter can potentially contribute to the eradication of vitamin A deficiency amongst people at risk such as children, pregnant and lactating women. Breads containing 20 and 30 % OFSP flour can provide more than a half of the RDA of vitamin A for children but consumer preference test needs to be conducted to determine the perception consumers towards these breads. OFSP flour confers different sensory properties to the breads such as sweet potato flavour, caramel aroma and sweetness and increased firmness. The presence of sugars, both reducing and non-reducing sugars, from OFSP flour imparts a brown appearance to the breads especially on the crust as a result of the Maillard reaction and caramelisation.

5 General Discussion

This chapter comprises two main sections. The first section critically discusses the experimental and methodologies used during this study. The second section evaluates the rheological properties of OFSP-wheat composite doughs, bread-making performance of the flours and other chemical, sensory and nutritional properties of the breads.

5.1 Critical discussion of experimental design and methodologies used

The current study has been conducted in two phases. The first phase focussed on determining the possibility of using OFSP flour in composite breads by substituting wheat flour with OFSP flour at various levels (0, 10, 20, 30 and 40% substitution levels). The effect of compositing OFSP flour with wheat flour on the rheological properties of the dough was evaluated, which gave an idea of what would happen to the breads. The bread-making performance of the composite flours was done using 100 g baking test. This experiment helped to determine the right substitution levels that could result in breads with characteristics close to wheat flour bread. Statistical analysis showed that none of the composite breads had the same characteristics as the control sample. For that reason, the second phase was initiated to try to improve the quality of OFSP wheat composite breads by adding exogenous gluten. Bread containing 40% OFSP flour was not taken into consideration because of low quality in terms of firmness, loaf volume and oven spring values. Additionally, because OFSP contains β-carotene, the effect of substituting wheat flour with OFSP flour and baking on β-carotene content of OFSP-wheat composite breads and sensory properties was evaluated. The contribution of composite breads to the RDA of vitamin A in different groups of people was also determined.

Sweet potato processing is generally linked with loss of quality following the action of enzymes that may bring about undesirable changes such as browning (van Hal, 2000). Before the preparation of the flour from OFSP it was necessary to minimise the enzymatic activity in sweet potato dices. OFSP dices were therefore treated with acetic acid (1%) for one hour since it was found to effectively minimize the enzymatic activity in sweet potato (Krishnan *et al*., 2010). However, the use of ascorbic acid was found to cause the loss of more orange colour from dices, which would be an indication of the loss of β-carotene. Thus, during the study on the effect of baking on the stability of β-carotene, untreated OFSP dices were used to prevent losses that might be caused by acetic acid. The loss of β-carotene during the drying process was 17.6% of the original amount present in the fresh roots.

The rheological properties of wheat flour dough and doughs made from mixtures of wheat flour and OFSP flour were studied using a mixograph and alveo-consistograph. The mixograph was used to determine the resistance of the dough to the mixing process, optimum dough development, dough consistency and tolerance to overmixing. Alveographic analysis is normally carried out on dough that has been prepared by mixing the flour and water containing salt (AACC, 2000). However, it is evident that other ingredients used for bread-making such as sugar (Singh, Bajaj, Singh and Gujral, 2002), yeast (Salvador, Sanz and Fiszman, 2006) and fat (Singh, Gujral and Singh, 2002) can also influence the rheological properties of the dough as well as its bread-making performance. There is a need to evaluate the characteristics of the dough when all ingredients are incorporated.

The RVA has been used to study pasting properties of sweet potato starches (Noda, Kobayashi and Suda, 2001), sweet potato flours (Waramboi, Dennien, Gidley and Sopade, 2011) and mixtures of wheat flour and starches from different crops including sweet potato (Zaidul, Norulaini, Omar, Yamauchi and Noda, 2007). It was used during the present study because baking is a thermal process that involves progressive increase of temperature, starch gelatinisation and protein denaturation that play a major role during the bread-making process (Zanoni, Peri and Bruno, 1995). Starch gelatinisation and protein denaturation, that take place during the same temperature interval of 60-85^{o}C (Thorvaldsson and Skjöldebrand, 1998), contribute, in part, to the increase in volume and the formation of the crumb and crust of bread (Zanoni, Peri and Bruno, 1995).

OFSP-wheat composite breads were baked in a conventional oven during both phases (i.e. phase I and phase II). Breads made during the 100 g baking test (phase I) were baked at 225^{o}C for 24 min, which is the normal baking conditions for wheat flour bread (AACC, 2000). However, these conditions were not favourable for the retention of β-carotene then another baking temperature and time were used to minimise the losses (i.e. 190^{o}C for 30 min) during the second phase. Breads obtained were well baked since no raw sensation was perceived during the descriptive sensory analysis.

Descriptive sensory analysis makes use of trained human subjects to identify, describe and quantify sensory attributes of a food material (Einstein, 1991). Before descriptive sensory evaluation of OFSP-wheat composite breads could be done, the panel underwent training in order to familiarise themselves with the products. They developed the sensory vocabulary used to describe sensory properties of the breads. Each panellist had already participated on more than one descriptive sensory panel of various products. The outcome of the current study relied upon the ability of the panel to identify sensory attributes and to detect the differences between samples. The evaluation took place during three different sessions; two of them were conducted within one day with about 10 minutes rest to avoid fatigue among the panellists. The last session took place on another day and mean scores were determined. The performance of the panel was checked using the Compusense software (Compusense five® release 4.6, Guelph, Ontario, Canada) and was found to be acceptable since their results were repeatable on different occasions. The limitation encountered during the descriptive sensory analysis of the breads was that the panel was not able to determine the colour of the crumb because of the colour (red) of light used in the sensory laboratory. The red light was chosen to minimise the influence of the orange colour on the performance of the panellists.

Carotenoid extraction was carried out with acetone but re-extraction (residue) was necessary for complete extraction as previously reported (van Jaarsveld, De Wet Marais, Harmse, Nestel and Rodriguez-Amaya, 2006; Kimura *et al.*, 2007). In the current work, the use of acetone alone gave acceptable results probably because the extraction process was repeated for about 3 to 4 times until most of the pigments were removed from the residue. Kimura *et al.* (2007) found no difference in the results obtained from the extraction of carotenoids from sweet potato and cassava using different solvents notably acetone or tetrahydrofuran:methanol (1:1). However, although tetrahydrofuran has excellent solubility for carotenoids such as β-carotene and lutein, it easily accumulates peroxides and there is time limit to its use (Rodriguez-Amaya, 2010).

Carotenoids are susceptible to degradation by oxygen, heat and light due to their unsaturated structure (Leskova *et al.*, 2006). A number of measures were therefore taken to minimise losses that would occur during the extraction process. These include working under low light conditions in the laboratory by putting the lights off, covering glassware with aluminium foil (Teow *et al.*, 2007, Lee and Chen, 2001) and the use of butylated hydroxytoluene, an antioxidant commonly used during the analysis of carotenoids (Liu, Lin and Yang, 2009; Kimura *et al.*, 2007). Additionally, after evaporating the solvent and dissolving the dried sample into the mobile phase, the carotenoid solution was immediately transferred to amber vials (brown vials) which provided further protection against light.

The quantitative analysis of carotenoids was performed using a reversed-phase HPLC coupled with a diode array detector (the absorbance was read at 450 nm). The diode array detector provided the absorption spectra that were used during the identification of different forms of carotenoids. Their retention times were not considered because they may vary during the course of a day (Rodriguez-Amaya and Kimura, 2004). One of the major challenges common to various chromatographic methods used during the analysis of carotenoids is the separation of different geometric isomers of carotenes (Carvalho *et al.*, 1995). The separation of geometric isomers of β-carotenes during our study was done on a C30 column due to its ability to separate the isomers of carotenoids (Rodriguez-Amaya and Kimura, 2004).

The first polymeric C30 column that achieved the separation of carotenoid isomers was developed by Sande, Sharpless, Craft and Wise (1994). Quantitative data on the geometric isomers of carotenoids using a C30 column were published later by Lessin, Catignani and Schwartz (1997) for the first time. They used the new C30 HPLC methods to quantify the provitamin A carotenoid isomers in fresh and processed fruits and vegetables. Subsequently, the research done on isomers of carotenoids using the C30 column has increased substantially (Schierle, Pietsch, Ceresa and Fizet, 2004; van Jaarsveld, De Wet, Harmse, Nestel and Rodriguez-Amaya, 2006; Kidmose, Christensen, Agili and Thilsted, 2007; Kimura *et al*., 2007; Bechoff, Dhuique-Mayer, Marouzé, Reynes and Westby, 2009; Liu, Lin and Yang, 2009).

Alternatively, the analysis of carotenoids in OFSP-wheat composite breads could also be done spectrophotometrically because their absorbance is directly proportional to their concentration. This implies that when in solution carotenoids obey the Beer-Lambert law (Rodriguez-Amaya and Kimura, 2004). This technique does not require a constant supply of carotenoid standards since separated fractions are directly quantified using published coefficients of absorption (Kimura and Rodriguez-Amaya, 2002). However, the main disadvantage of using this method is that it does not allow proper identification of different types of carotenoids present in the sample. Thus, the results are expressed as total carotenoid content (µg/ml) and when the sample contains high levels of all-*trans*-β-carotene, this value may only give an estimation of the amount of this compound in the sample. It may not be recommended to use those results for the estimation of the RAE.

The identification of all-*trans*-β-carotene was achieved by co-chromatography (i.e. spiking) using authentic all-*trans*-β-carotene standard and the visible absorption spectrum. The *cis* isomers were identified by the wavelengths of maximum absorption which were slightly lower compared to those of all-*trans*-β-carotene as well as the appearance of a *cis* peak at a lower absorbance. The location of the *cis* double bond was indicated by the ratio of the height of the *cis* peak designated as A_B and that of the middle main peak designated as A_{II} multiplied by 100 (%A_B/A_{II}) (Rodriguez-Amaya and Kimura, 2004; Kimura *et al.*, 2007). %A_B/A_{II} is an indicator of the intensity of the *cis* peak which increased as the *cis* double bond was closer to the centre of the molecule. Therefore, the 15-*cis*-β-carotene, in which the *cis* double bond is at the centre of the chromophore, has a prominent *cis* peak (Chen, Chen and Chien, 1994; Rodriguez-Amaya and Kimura, 2004). The %A_B/A_{II} of 10, 45 and 56 for 9-*cis*-β-carotene, 13-*cis*-β-carotene and 15-*cis*-β-carotene respectively (Mercadante, Steck and Pfander, 1999) helped during the identification of *cis* isomers of β-carotene. During the current study, the values of %A_B/A_{II} for 9-*cis*-β-carotene and 13-*cis*-β-carotene were 10 and 45 respectively exactly as reported by Mercadante *et al.* (1999). Carotenoids from OSFP-wheat composite breads eluted exactly in the same order as previously reported in OFSP (Kimura *et al.*, 2007) on the same type of stationary phase (C30 column). 15-*cis*-β-carotene could not be detected probably because its levels were extremely low.

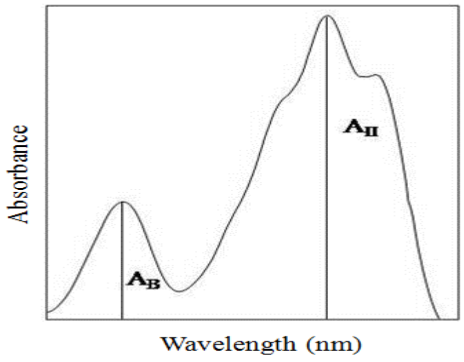


Figure 5.1 Illustration of the height of the *cis* peak designated as A_B and that of the middle main peak designated as A_{II}. Adapted from: Rodriguez-Amaya and Kimura (2004)

The volume of extract collected for drying (with the rotary evaporator), before the chromatographic analysis of carotenoids in breads, was also critical. The peak areas of carotenoids that were present in small quantities, especially the 9-*cis*-β-carotene, could not be recorded when 5 ml of extract was collected for drying and sometimes they remained undetectable in 7 ml of extract. However, 8 ml was found to be enough when the extract from 1 g sample was diluted up to 60 ml during the extraction process. This can be explained by the fact that the only source of carotenoids in the breads was mainly the OFSP flour. The levels of OFSP flour ranged from 0-30% suggesting that the amount of *cis* isomers formed during baking was relatively low. The repeatability of this method was good and the response of the detector was excellent as shown by the coefficients of variation obtained (1.2-2.8%).

Each provitamin A carotenoid detected in OFSP-wheat composite breads contributed to the RDA of vitamin A for different groups of people studied. However, although a serving of bread may weigh up to 300 g (Škrbic, Milovac, Dodig and Filipsev, 2005), estimates made in the current study were based on a portion of 100 g of bread, which is approximately 2 slices. The reason was that the targeted groups included children and young people who may not necessarily take large portions of bread. It is obvious that this amount may also increase based on individual desire or preference.

It is estimated that about 90% of ingested preformed vitamin A is absorbed but the absorption efficiency of provitamin A carotenoids varies widely depending on the type of plant source used (FAO/WHO, 2005), which can also affect the retinol activity equivalency factor. The β-carotene to vitamin A conversion factor previously used was 6:1 (Palace *et al*., 1999; Haskell *et al.*, 2004). However, it has been found that the bioavailability of β-carotene from plant sources, including sweet potato, may be lower than previously assumed (Haskell *et al.*, 2004). The most accepted vitamin A equivalency factor for β-carotene is 12:1 (FAO/WHO, 2005; Haskell *et al.*, 2004; van Jaarsveld *et al.*, 2006) which was used during this study.

5.2 Critical evaluation of the rheological properties of OFSP-wheat composite doughs, pasting behaviour, bread-making performance of the flours and chemical, sensory and nutritional properties of the breads

Mixing OFSP-wheat composite doughs beyond the optimal dough development stage decreased their tolerance to mixing. Prolonged mixing stresses disulphide bonds and reaches the point where they break (Danno and Hoseney, 1982a). However, breaking the disulphide bonds alone would not be enough to explain dough weakening during extremely long mixing times (Danno and Hoseney, 1982b). The presence of compounds containing an activated double bond such as ferulic acid, generally found in wheat flour (Jensen *et al*., 2011), is essential for rapid breakdown (Danno and Hoseney 1982a). The rapid breakdown is probably due to a reaction between these substances and thiyl radicals formed from disulphide bonds during mixing (Graveland, Bosveld, Lichtendonk and Moonen, 1984). Presumably, this reaction caused the glutenin molecule to denature and become hydrophilic on the exterior and hydrophobic in the interior which gave the dough excess water, previously hidden in the glutenin molecule, resulting in a wet, sticky dough with no elasticity (Danno and Hoseney 1982a). Lack of elasticity may have led to the weakening of OFSP-wheat composite doughs upon extended mixing times. It may also be attributed to insufficient amount of gluten as a result of increased substitution of wheat flour with OFSP flour.

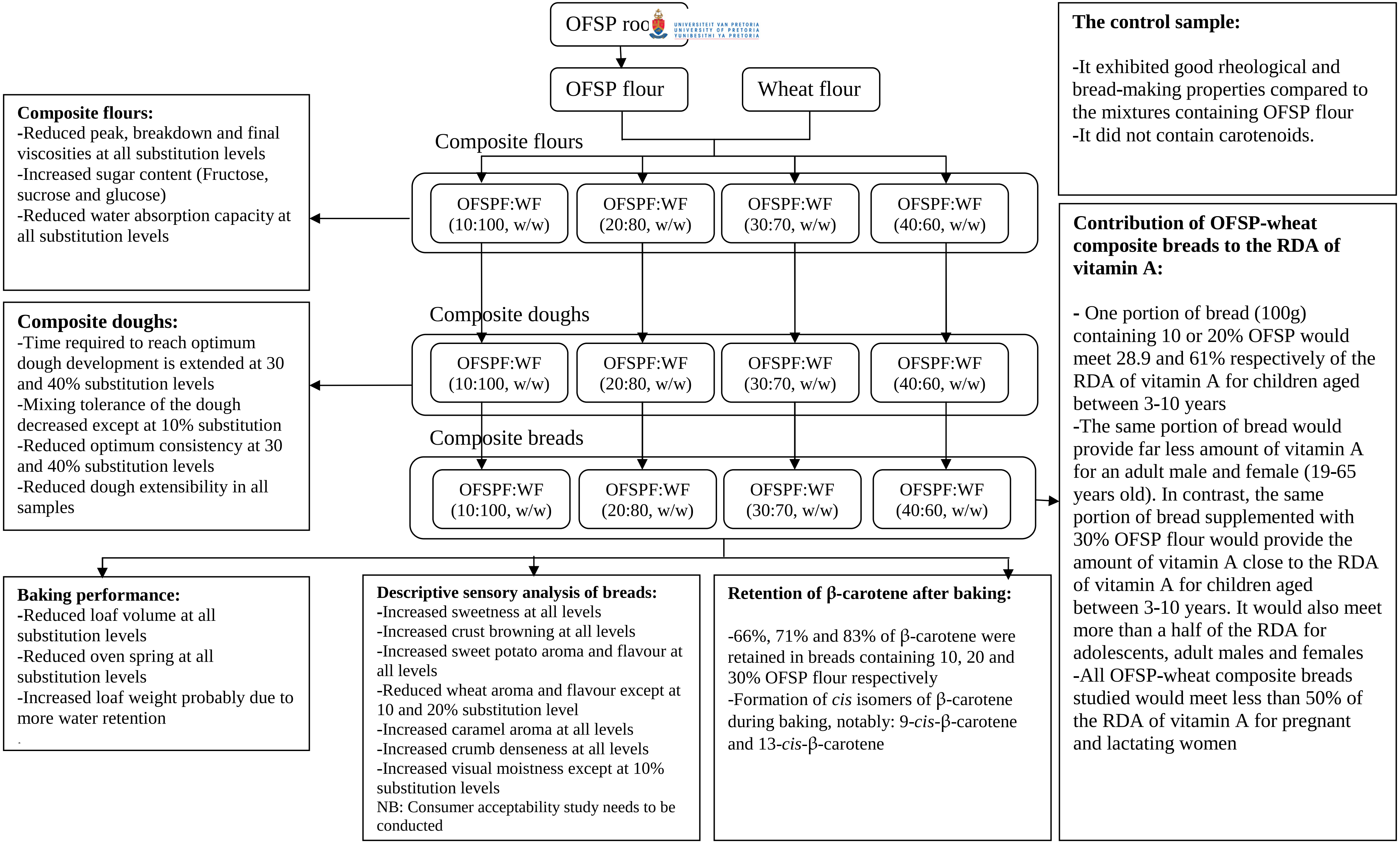
With gradual substitution of wheat flour with OFSP flour, a point was reached where the dough was no longer able to support the weight of bread and collapsed. Subtracting the height of the dough from the height of the bread resulted in negative values, especially at 30 and 40% substitution levels as the height of the breads became lower compared to the doughs. This happened in breads where no exogenous gluten was added (research chapter I) but it did not happen in breads where exogenous gluten was added (research chapter II). The collapse of the doughs may be attributed to various destabilising factors such as Ostwald ripening. Ostwald ripening, also known as disproportionation (Rouillé, Bonny, Valle, Devaux and Renou, 2005; Kokelaar and Prins, 1995; Mills, Wilde, Salt and Skeggs, 2003), is the growth of large gas bubbles at the expense of small ones due to high pressure inside the small cells (Walstra, 1996; van Vliet, 2008).

Coalescence of gas bubbles may be another factor that probably caused the destabilisation of the doughs. It results from the rupture of the thin dough films thereby promoting the loss of gas and irregular crumb structure (Kokelaar and Prins, 1995). In contrast to Ostwald ripening, gas bubbles involved during coalescence need to come close to each other until they merge when the films between the adjacent cells get ruptured (Bremond, Thiam and Bibette, 2008). Presumably, as the amount of large gas cells increased either due to coalescence or Ostwald ripening, the dough was no longer able to support the weight of the bread and collapsed. Consequently, other bread characteristics such as loaf volume and specific volume were also adversely affected. The addition of exogenous gluten probably prevented the collapse of the dough during baking since the height of bread remained higher compared to the dough in all bread samples. However, no significant increase in loaf volume was observed. The same amount of gluten lost during the substitution of wheat flour with OFSP flour was added. This was probably not sufficient enough to allow the increase in loaf volume of breads containing OFSP flour. A study on the effect of increased gluten supplementation and hydrocolloids on the baking performance of OFSP-wheat composite flour may be recommended.

Mohamed *et al.* (2010) supplemented wheat-banana composite flours (10, 15, 25 and 30% substitution levels) with gluten up to 25% (in all blends) and found that loaf volume was significantly higher than the control except for bread containing 30% banana flour. Hydrocolloids have been used in gluten-free breads as gluten substitutes (Lazaridou, Duta, Papageorgiou, Belc and Biliaderis, 2007; Marco and Cristina, 2008) but they have also been used in wheat-cassava composite bread (10% substitution level) (Shittu, Rashidat, Aminu and Abulude, 2009) with good results.

The darkness of the crust increased gradually as more OFSP flour was supplemented presumably as a result of increased amounts of reducing sugars. Additionally, lysine is known to be more reactive than other amino acids such as alanine and glycine with regard to browning during the Maillard reaction (Morales and Jimenez-Perez, 2001). Therefore, intense browning of breads containing high quantities of OFSP flour might not only be attributed to the presence of sugars but also to increased levels of lysine in the blends since sweet potato protein contains 3.5-4.5 g lysine/100 g protein (van Hal, 2000) while prolamins are known to be deficient in lysine (Hallén, Ibanoğlu and Ainsworth, 2004; Dewettink *et al.*, 2008).

In addition to Maillard browning which requires the presence of reducing sugars and proteins, the browning of the crusts in breads containing OFSP flour may also have occurred without the interaction between sugars and proteins. Direct heating of sugars in the absence of proteins brings about complex reactions commonly known as caramelisation (Purlis and Salvadori, 2007). The caramelisation of sugars that occurs simultaneously with the Maillard reaction (Zanoni, Peri and Bruno, 1995; Ajandouz *et al*., 2001; Kitts *et al.*, 2006) contributes to nonenzymatic browning that takes place during baking (Ajandouz *et al*., 2001, Purlis



The control sample:

-It exhibited good rheological and bread-making properties compared to the mixtures containing OFSP flour
-It did not contain carotenoids.

Composite flours:
-Reduced peak, breakdown and final viscosities at all substitution levels
-Increased sugar content (Fructose, sucrose and glucose)
-Reduced water absorption capacity at all substitution levels

Composite doughs:
-Time required to reach optimum dough development is extended at 30 and 40% substitution levels
-Mixing tolerance of the dough decreased except at 10% substitution
-Reduced optimum consistency at 30 and 40% substitution levels
-Reduced dough extensibility in all samples

Contribution of OFSP-wheat composite breads to the RDA of vitamin A:

- One portion of bread (100g) containing 10 or 20% OFSP would meet 28.9 and 61% respectively of the RDA of vitamin A for children aged between 3-10 years
-The same portion of bread would provide far less amount of vitamin A for an adult male and female (19-65 years old). In contrast, the same portion of bread supplemented with 30% OFSP flour would provide the amount of vitamin A close to the RDA of vitamin A for children aged between 3-10 years. It would also meet more than a half of the RDA for adolescents, adult males and females
-All OFSP-wheat composite breads studied would meet less than 50% of the RDA of vitamin A for pregnant and lactating women

Baking performance:
-Reduced loaf volume at all substitution levels
-Reduced oven spring at all substitution levels
-Increased loaf weight probably due to more water retention
.

Descriptive sensory analysis of breads:
-Increased sweetness at all levels
-Increased crust browning at all levels
-Increased sweet potato aroma and flavour at all levels
-Reduced wheat aroma and flavour except at 10 and 20% substitution level
-Increased caramel aroma at all levels
-Increased crumb denseness at all levels
-Increased visual moistness except at 10% substitution levels
NB: Consumer acceptability study needs to be conducted

Retention of β-carotene after baking:

-66%, 71% and 83% of β-carotene were retained in breads containing 10, 20 and 30% OFSP flour respectively
-Formation of *cis* isomers of β-carotene during baking, notably: 9-*cis*-β-carotene and 13-*cis*-β-carotene

*Due to poor quality, bread containing 40% OFSP flour was not involved in carotenoid analysis, contribution to the RDA of vitamin A and descriptive sensory analysis

Figure 5.2 Summary of the findings on the use of OFSP flour in bread-making, stability of β-carotene during baking, sensory quality and the contribution of the resulting breads on vitamin A requirements

and Salvadori, 2007). Heating of sugars leads to the formation of brown compounds known as caramel. These compounds result from a series of chemical reactions that involve hydrolysis, dehydration and polymerisation, collectively referred to as caramelisation (Kitts *et al.*, 2006). During baking, sucrose (non reducing sugar) can be hydrolysed to form reducing sugars such as glucose and fructose that might also be involved in both browning processes, notably the Maillard reaction (Kitts *et al.*, 2006) and caramelisation (Purlis, 2010).

Among all the breads studied, the lightness of the crumbs was higher compared with that of the crusts in all bread samples. This may be attributed in part to high temperatures prevailing on the crust that allowed the formation of the Maillard reaction products, hence, intense brown colour (Capuano *et al*., 2008). Low water content and high temperature on the surface of the bread are the main factors that make the crust different from the crumb (Thorvaldsson and Skjöldebrand, 1998). Since browning of bread depends on heat transfer and diffusion of water during baking (Ahrné, Andersson, Floberg, Rosén and Lingnert, 2007), it may be hypothesised that high moisture and low temperature did not provide optimal conditions for the formation of brown compounds in the crumb.

Compositing wheat flour with OFSP flour increased the sweetness of the resulting breads. Sucrose, glucose and fructose were the predominating sugars found in the mixtures and their quantities increased with increased supplementation of OFSP flour. The sweetness detected by the descriptive sensory panel in the breads was probably due to the presence of sugars from OFSP flour. Nevertheless, the same sugars may also have contributed to the bitter taste in the crust through the production of bitter compounds during the Maillard reaction and caramelisation (Kamuf, Nixon, Parker and Barnum, 2003; Pozo-Bayó, Guichard and Cayot, 2006; Pedreschi, Kaack, Granby and Troncoso, 2007). The firmness of breads increased gradually at all substitution levels probably because they were more compact and did not rise properly as compared to the control. The presence of dietary fibre from OFSP flour may also have contributed to the increase in bread firmness.

Baking induced change in other colour parameters especially the a* and b* values, that was followed by a loss of all-*trans*-β-carotene. The change in colour was probably due to structural modifications within the chromophore of all-*trans*-β-carotene molecule through isomerisation which also alters its biological activity (Rodriguez-Amaya and Kimura, 2004) with regard to the vitamin A value. The light absorbing chromophore of carotenoids is not only responsible for their attractive colours (Rodriguez-Amaya and Kimura, 2004) but also for their instability in relation to oxygen, light and heat (Meléndez-Martínez, Vicario and Heredi, 2007). The orange colour, which is a mixture of red and yellow colours (Beltràn-Gonzàlez, Pérez-López, López-Nicolàs and Carbonell-Barrachina, 2008), in breads containing OFSP flour was due to the presence of carotenoid pigments especially the β-carotene and a more pronounced orange colour could be an indication of high β-carotene content (van Hal, 2000). This was confirmed by a substantial increase in the a* and b* values in both doughs and breads as the amount of OFSP flour was increased from 10% to 30% despite the losses that occurred during baking.

The amount of *cis* isomers (9-*cis*-β-carotene and 13-*cis*-β-carotene) appeared to increase when doughs containing OFSP flour were subjected to the prevailing temperature in the oven (190^{o}C) while the amount of all-*trans*-β-carotene decreased. The same observations were made by Chen *et al.* (1994) who found that the concentration of all-*trans*-β-carotene decreased during heating at 150^{o}C while the amount of *cis* isomers increased. The highest proportion of *cis* isomers was found in breads containing 30% OFSP flour. This indicates that *cis* isomers detected in samples containing OFSP flour originated exclusively from structural modifications that took place within the all-*trans*-β-carotene compound. Thermal treatments, like baking, may change the configuration of all-*trans*-β-carotene through isomerisation from *trans* to *cis* typically at the 9th, 13th, and 15th carbon positions (Grabowski *et al.*, 2008).

The results of this work showed that pregnant and lactating women would need more of the OFSP-wheat composite breads to fulfil their vitamin A requirements (Table 4.2.9). This is probably because the foetus in the womb depends on the mother’s vitamin A stores and during the postpartum period when the newborn is growing rapidly. Breast milk from deficient mothers is likely to contain insufficient vitamin A to build or even to maintain vitamin A stores in nursing infants (WHO, 1998). Depletion of stored vitamin A may occur over time when the diet contains too little to replace the amount used by tissues or reduced by breast-feeding (WHO/UNICEF, 1995).Vitamin A deficiency affects not only the immune system but may also lead to malformation during embryogenesis (von Lintig *et al*., 2005). The fact that vitamin A deficiency is known to be associated with increased child mortality after six months of age, presents a convincing argument for improving maternal vitamin A nutrition as a child survival strategy (WHO, 1998). Improving vitamin A status of children aged between 6 months and 6 years may reduce the risk of dying by 20-30% on average

(FAO/WHO, 2005) and probably OFSP-wheat composite bread may contribute to lower the number of deaths caused by VAD.

It is evident that all vitamin A requirements for a pregnant or lactating woman might not be achieved by consuming bread made with OFSP-wheat composite flour alone. However, when coupled with other provitamin A-rich sources in the diet, it could contribute to reduce the incidences of VAD amongst pregnant and breast-feeding women as well as their infants. Apart from OFSP, green leafy vegetables such as spinach, amaranths; yellow vegetables such as pumpkin, squash, carrots, and fruits like mango, apricot and papaya (FAO/WHO, 2005) can also be used as plant sources of provitamin A carotenoids.

This work also showed that an adult male would need more of OFSP-wheat composite bread than an adult female of the same age to attain their RDA of vitamin A. It is normal that during childhood, the nutritional needs for males and females of the same age differ slightly but after puberty the needs are higher for males than females (WHO, 2011).

In certain cases, one portion of 100 g (about 2 slices) of OFSP-wheat composite bread (i.e. bread containing 30% OFSP flour) would provide vitamin A levels close to the recommended safe intake of vitamin A (FAO/WHO, 2005). However, some individuals may prefer to eat more than two slices of bread per day suggesting that the intake of vitamin A would be higher as far as the consumption of OFSP-wheat composite breads is concerned. Nevertheless, the ingestion of high amounts of provitamin A carotenoids might not present adverse health effects when consumed at levels comparable to those recommended for vitamin A supplementation (IVACG, 1998). β-carotene is much less toxic than preformed retinol (Hathcock, *et al.*,1990) since overconsumption of carotenoids does not result in hypervitaminosis (Bendich and Langseth, 1989; Rogers, Malouf, Langemeier, Gelroth and Ranhotra, 1993). They are converted in a regulated manner to retinol in required amounts (Sharman, 1985). However, overconsumption of provitamin A carotenoids can lead to hypercarotenaemia which is a condition characterised by a yellow pigmentation on human skin but is harmless (Sharman, 1985). Thus, normal consumption of OFSP-wheat composite bread may not cause adverse health effects presumably because the human body is well equipped to overcome them. The mechanism that prevents excessive bioconversion of carotenoids into retinol is still not well known (Chandrika, Svanberg and Jansz, 2006).

From the nutritional point of view breads containing 20 and 30% OFSP flour would potentially contribute to the eradication of VAD in developing countries. However, based on the results from the descriptive sensory analysis these breads were described as dark (on the crust) and sweet but it is not yet known how consumers would perceive them, suggesting that a consumer acceptability study may be required. Bread characteristics, such as loaf volume, of breads containing OFSP flour were different from the control sample but the characteristics of bread containing 10% OFSP flour were close to bread made with wheat flour alone. The contribution of this bread to the RDA of vitamin A is relatively low compared to those containing 20 and 30% OFSP flour. Creating awareness about the nutritional benefits of OFSP-wheat composite breads would play a big role in the overall acceptability amongst the consumers.

6 Conclusions and Recommendations

Partial substitution of wheat flour with OFSP flour adversely affects rheological properties of the doughs and their bread-making performance due to the lack of gluten proteins in OFSP flour. It increases dough resistance to mixing by extending time required to reach the optimal dough development and decreases dough extensibility as a result of increase in dough resistance to extension. The peak viscosity of OFSP-wheat composite flours decreases with increased addition of OFSP flour but decreases their tendency towards retrogradation. Bread characteristics, such as loaf volume, of all breads samples containing OFSP flour were different from the control sample but the characteristics of bread containing 10% OFSP flour were close to bread made with wheat flour alone. It is recommended that an evaluation on the reduced tendency towards retrogradation as a result of OFSP flour addition be performed using OFSP-wheat composite bread stored under different conditions.

The use of OFSP flour in bread-making imparts a number of sensory properties to bread such as sweet potato flavour, caramel aroma, sweetness, crust browning and increased firmness that might influence their acceptance by the consumers either positively or negatively. Consumer acceptability study has not been carried out during the current study due to time limitations. It is therefore recommended that consumer acceptability study, probably in Rwanda, be performed for better understanding of consumer perception towards OFSP-wheat composite breads.

Baking adversely affects the β-carotene content in OFSP-wheat composite breads due to high temperatures prevailing in the oven. The retention of β-carotene in OFSP-wheat composite breads varied between 63 to 83%. Low baking temperature and short time (e.g., 190^{o}C for 30 min) can be recommended to minimise losses of β-carotene during baking of bread containing OFSP flour. Although the focus of this study was on the losses of β-carotene during baking of OFSP-wheat composite breads, it is important to mention that 17.6% of β-carotene was lost during the production of OFSP flour (drying process).

OFSP-wheat composite bread can contribute more than a half or less of the RDA of vitamin A for people in different age groups based on the amount of OFSP flour supplemented. Composite bread (100 g portion) containing 30% OFSP, for example, would meet 89, 45 and

42% of the recommended dietary allowance (RDA) of vitamin A for children (3-10 years), pregnant and lactating women respectively. It can therefore be recommended to be used as a tool for combating vitamin A deficiency amongst people at risk or already affected with it. Breads containing 10 and 20% OFSP flour can also contribute to the RDA of vitamin A in different groups but they may be recommended for children because their vitamin A requirements are low.

This study has established that OFSP-wheat composite bread is a potential alternative source of vitamin A (in the form of provitamin A carotenoids) that can be used to eradicate VAD deficiency. However, the choice of an OFSP cultivar rich in provitamin A carotenoids is vital for obtaining a processed product, like bread, that would contribute adequate intake of vitamin A. It is, thus, recommended that breeding programs focus on developing OFSP cultivars with high provitamin A carotenoids. Additionally, since compositing OFSP flour with wheat flour results in breads with increased crust browning, which can be objectionable to certain consumers, it is also suggested OFSP cultivars with low amounts of reducing sugars be developed without compromising other important micronutrients (i.e. β-carotene). All these efforts should also be coupled with increased awareness about the nutritional benefits of OFSP amongst the consumers to facilitate rapid change in their nutritional status with regard to vitamin A.

7 References

AACC International, 2000. Approved Methods of the American Association of Cereal Chemists, 10th Ed. Methods 10-09, 38-10, 46-30 and 54-40A. St. Paul, MN: AACC International.

Adamson, J.C., 2010. Physics and physiology of colour. Available at: http://www.greatreality.com/colour/ColourHVC.htm. Accessed on 22nd January 2012.

Ade-Omowaye, B.I.O., Akinwande, B.A., Bolarinwa, I.F. and Adebiyi, A.O. 2008. Evaluation of tigernut (*Cyperus esculentus*) – wheat composite flour and bread. *African Journal of Food Science 2,* 87-91.

Ahmed, M., Akter, M.S., Eun, J., 2010. Peeling, drying temperatures and sulphite-treatment affect physicochemical properties and nutritional quality of sweet potato flour. *Food Chemistry 121*, 112–118.

Ahrné, L.andersson, C. G., Floberg, P., Rosén, J. and Lingnert, H., 2007. Effect of crust temperature and water content on acrylamide formation during baking of white bread: Steam and falling temperature baking. *Lebensmottel-Wissenschaft und-Technologie – Food Science and Technology 40,* 1708–1715.

Ajandouz, E. H., Tchiakpe, L. S., Ore, F. D., Benajiba, A. and Puigserver, A., 2001. Effects of pH on caramelisation and Maillard reaction kinetics in fructose-lysine model systems. *Journal of Food Science, 66,* 926–931.

Alais, C. and Linden, L., 1991. Food Biochemistry, pp. 222. Ellis Horwood , Chichester, England.

Ameny, M. A. and Wilson, P. W., 1997. Relationship between Hunter colour values and β-carotene content in white-fleshed African sweet potatoes (*Ipomonea batatas* Lam). *Journal of the Science of Food and Agriculture, 73,* 301−306.

Ames, B.N.,1998. Micronutrients prevent cancer and delay ageing. *Toxicology Letters, 5-18.*

Anderssen, R.S., Bekes, F., Gras, P.W., Nikolov, A. and Wood, J.T., 2004. Wheat-flour dough extensibility as a discriminator for wheat varieties. *Journal of Cereal Science 39,* 195–203

Aparicio-Ruiz, R.M.I., Mínguez-Mosquera, M.I. and Gandul-Rojas, B., 2011. Thermal degradation kinetics of lutein, β-carotene and β-cryptoxanthin in virgin olive oils. *Journal of Food Composition and Analysis 24,* 811–820.

Armero, E., Collar, C., 1998. Crumb firming kinetics of wheat breads with antistaling additives. *Journal of Cereal Science 28*, 165–174.

Aruoma, O. I., 1994. Nutrition and health aspects of free radicals and antioxidants. *Food and Chemical Toxicology 32*, 671-683.

Austin, D.F. 1988. The taxonomy, evolution and genetic diversity of sweet potatoes and related wild species. In: *Exploration, Maintenance, and Utilization of Sweet potato Genetic Resources.* Gregory, P. (Ed.), pp. 27–60. CIP, Lima, Peru.

Awika, J.M., Rooney, L.W.,Wu,X., Prior, R.L. and Cisneros-Zevallos, L., 2003. Screening methods to measure antioxidant activity of sorghum (*Sorghum bicolor*) and sorghum products. *Journal of Agricultural and Food Chemistry 51*, 6657–6662.

Awika, J.M., McDonough, C.M. and Rooney L.W., 2005. Decorticating sorghum to concentrate healthy phytochemicals. *Journal of Agricultural and Food Chemistry 53,* 6230-6234.

Aziah, N. A.A. and Komathi, C. A., 2009. Acceptability attributes of crackers made from different types of composite flour. *International Food Research Journal 16*, 479-482.

Bechoff, A., Dhuique-Mayer, C., Marouzé, C., Reynes, M. and Westby, A., 2009. Effect of hot air, solar and sun drying treatments on provitamin A retention in orange-fleshed sweet potato. *Journal of Food Engineering 92,* 164–171.

Belitz, H. D. and Grosch, B.,1999. Food Chemistry , 2nd edition. Springer-Verlag Berlin Heidelberg, Germany.

Beltràn-Gonzàlez, F., Pérez-López, A. J., López-Nicolàs, J.M. and Carbonell-Barrachina, À. A., 2008. Effects of agricultural practices on instrumental colour, mineral content, carotenoids composition and sensory quality of mandarin orange juices, cv. Hernandina. *Journal of the Science of Food and Agriculture 88*, 1731-1738.

Bendich, A. and Langseth, L., 1989. Safety of vitamin A. *American Journal of Clinical Nutrition 49,* 358-371.

Bengtsson, A., Namutebi, A., Alminger, M. L. and Svanberg, U., 2008. Effect of various traditional processing methods on the all-trans-β-carotene content of orange-fleshed sweet potato. *Journal of Food Composition and Analysis 21*, 134–143.

Blazek, J. and Copeland, L., 2008. Pasting and swelling properties of wheat flour and starch in relation to amylose content. *Carbohydrate Polymers 71,* 380–387.

Bovell-Benjamin, A. C., 2007. Sweet Potato: A review of its past, present and future role in human nutrition. *Advances in Food and Nutrition Research 52,* 1-59.

Bremond, N., Thiam, A. R. and Bibette, J., 2008. Decompressing emulsion droplets favours coalescence. *Physical Review Letters 100, 024501.*

Capuano, E., Ferrigno, A., Acampa, I. Ait-Ameur, L. and Fogliano, V., 2008. Characterisation of the Maillard reaction in bread crisps. *Eur Food Res Technol 228,* 311-319.

Carvalho, C.R., Carvalho, P.R. and Collins, C.H., 1995. High-performance liquid chromatographic determination of the geometrical isomers of β-carotene in several foodstuffs. *Journal of Chromatography A, 697,* 289-294.

Castro, I., A., Moraes, B. S., Lanfer, M. U., Motizuki, M. and Higashi, S. T., 2005. Optimisation of the antioxidative capacity of a mixture of carotenoids and ά-tocopherol in the development of a nutritional Supplement. *Food Research International 38,* 861-866.

Chandler, L. A. and Schwartz S.J., 1988. Isomerisation and losses of *trans*-β-carotene in sweet potatoes as affected by processing treatments. *Journal of Agricultural and Food Chemistry 36,* 129-133.

Chandrika, U.G., Svanberg, U. and Jansz, E.R., 2006. In vitro accessibility of β-carotene from cooked Sri Lankan green vegetables and their estimated contribution to vitamin A requirement. *Journal of the Science of Food and Agriculture 86,* 54-61.

Chen, B.H., Chen, T.M. and Chien, J.T., 1994. Kinetic model for studying the isomerisation of α and β-carotene during heating and illumination. *Journal of Agricultural and Food Chemistry 42,* 2391-2397.

Chen, B.H, Peng, H.Y. and Chen, H.E,. 1995. Changes of carotenoids, colour and vitamin A contents during processing of carrot juice. *Journal of Agricultural and Food Chemistry 43,* 1912-1918.

Chopin Technologies, 2008. Alveolink NG Consistograph Instruction Manual. Villeneve Lagarenne cedex, France.

Cinar, I., 2005. Stability studies on the enzyme extracted sweet potato carotenoproteins. *Food Chemistry 89*, 397–401.

Coultate, T.P., 2002. Food: The Chemistry of its Components, Fourth Edition, pp.432. Royal Society of Chemistry, Cambridge, UK.

Coultate, T.P., 2009. Food: The Chemistry of its Components, Fifth Edition, pp. 501. Royal Society of Chemistry, Cambridge, UK.

Danno, G. and Hoseney, R.C., 1982a. Effect of sodium chloride and sodium dodecyl sulphate on mixograph properties. *Cereal Chemistry 59,* 202-204.

Danno, G. and Hoseney, R.C., 1982b. Effect of dough mixing and rheologically active compounds on relative viscosity of wheat protein. *Cereal Chemistry 59,* 196-198.

Dansby, M.Y. and Bovell-Benjamin, A.C., 2003. Production and proximate composition of a hydroponic sweet potato flour during extended storage. *Journal of Food Process Preservation 27,* 153–164.

Deshpande, S. S., Deshpande, U.S. and Salunkhe, D. K.1996. Nutritional and health aspects of food antioxidants. In: *Food antioxidants: Technological, Toxicological and Health Perspectives*. Madhavi, D. L., Deshpande, S. S., Salunkhe, D. K. (Eds.) Marcel Dekker, New York. pp. 361-469.

Dansby, M.Y. and Bovell-Benjamin, A. C., 2003. Production and proximate composition of a hydroponic sweet potato flour during extended storage. *Journal of Food Process Preservation 27,* 153–164.

Delisle, H., 1990. Patterns of urban food consumption in developing countries: perspective from the 1980’s. Food and Agriculture Organisation of the United Nations, Rome, Italy. Available at: ftp://ftp.fao.org/es/esn/nutrition/urban/delisle_paper.pdf. Accessed on 15th May, 2012.

Dewettinck, K., Van Bockstaele, F., Kühne, B., Van de Walle, D., Courtens, T.M. and Gellynck, X., 2008. Nutritional value of bread: Influence of processing, food interaction and consumer perception. *Journal of Cereal Science 48,* 243-257.

Dhingra, S. and Jood, S. 2004. Effect of blending on bread-making characteristics. *International Journal of Food Science and Technology 39,* 213-222.

Dhuique-Mayer, C., Tbatou, M., Carail, M. Caris-Veytrat, C., Dornier, M. and Amiot, M.J., 2007. Thermal degradation of antioxidant micronutrients in citrus juice-kinetics and newly formed compounds. *Journal of Agricultural and Food Chemistry 55,* 4209-4216.

Dobraszczyk, B.J. and Roberts, C.A., 1994. Strain hardening and dough gas cell-wall failure in biaxial extension. *Journal of Cereal Science 20*, 265–274.

Einstein, M. A., 1991. Descriptive techniques and their hybridisation. In: *Sensory science theory and applications in foods*. Lawless, H. T. and Klein, B. P., (Eds) Marcel Dekker: New York. pp 317-338.

Ellis, W.O., Obubuafo, A.K., Ofosu-Okyere, A., Marfo, E.K., Osei-Agyeman, K. and Odame-Darkwah, J.K., 1997. A survey of bread defects in Ghana. *Food Control 8*, 77-82.

FAO, 1988. Requirements of vitamin A, iron, folate and vitamin B_{12}, Report of a joint FAO/WHO Consultation, FAO Food and Nutrition series. No. 23. FAO, Rome.

FAO, 2010. Crop prospects and food situation. Food and Agriculture Organisation of the United Nations, Rome. Available at: http://www.fao.org/docrep/013/al972e/al972e00.pdf. Accessed on 15th May, 2012.

FAO/WHO, 2002. Vitamin A in human vitamin and mineral requirements. Report of joint FAO/WHO expert consultation Bangkok, Thailand. pp.87–107. Available at: http://whqlibdoc.who.int/publications/2004/9241546123_chap2.pdf
Accessed on 6th May, 2010.

FAO/WHO, 2005. Vitamin and mineral requirements in human nutrition, 2nd ed. Geneva, World Health Organisation. Available at: http://whqlibdoc.who.int/publications/2004/9241546123.pdf . Accessed 17th July, 2010.

Ferruzzi, M.G., Lumpkin, J.L., Schwartz, S.J. and Failla, M., 2006. Digestive stability, micellarisation and uptake of β-carotene isomers by Caco-2 human intestinal cells. *Journal of Agricultural and Food Chemistry. 54*, 2780-2785.

Gallagher, E., Gormley, T.R., Arendt, E.K., 2003. Crust and crumb characteristics of gluten-free breads. *Journal of Food Engineering 56*, 153-161.

Gellynck, X., Kühne, B., Van Bockstaele, F., Van de Walle, D. and Dewettinck, K., 2009. Consumer perception of bread quality. *Appetite 53,* 16–23.

Giménez, A., Varela, P., Salvador, A., Ares, G., Fiszman, S. and Garitta, L., 2007. Shelf-life estimation of brown pan bread: A consumer approach. *Food Quality and Preference 18, 196–204.*

Glória, M.B., Vale, S.R. and Bobbio, P.A., 1995. Effect of water activity on the stability of bixin in an annatto extract-microcrystalline cellulose model system. *Food Chemistry 52,* 389-391.

Goldman, M., Horev, B. and Saguy, I., 1983. Decolourisation of β-carotene in model systems simulating dehydrated foods. Mechanism and kinetic principles. *Journal of Food Science 48,* 751-754.

Grabowski, J.A., Truong, V.-D., Daubert , C.R., 2008. Nutritional and rheological characterisation of spray dried sweet potato powder. *Lebensmottel-Wissenschaft und-Technologie – Food Science and Technology 41,* 206–216.

Graveland, A., Bosveld, P., Lichtendonk, W. J. and Moonen, J. H. E., 1984. Isolation and characterisation of (3-methoxy-4-hydroxyphenyl)-β-cellotrioside from wheat flour; a substance involved in the reduction of disulphide-linked glutenin aggregates. *Journal of Cereal Science 2,* 65-72.

Gray, J.A. and Bemiller, J.N., 2003. Bread Staling: Molecular Basis and Control. *Comprehensive Reviews in Food Science and Food Safety 2,* 1-21.

Greene, J. L. and Bovell-Benjamin, A. C., 2004. Macroscopic and sensory evaluation of bread supplemented with sweet potato flour. *Journal of Food Science 69,* 167–173.

Guarda, A., Rosell, C.M., Benedito, C. and Galotto, M.J., 2004. Different hydrocolloids as bread improvers and antistaling agents. *Food Hydrocolloids 18,* 241–247.

Gunaratne, A., Ranaweera, S. and Corke, H., 2007. Thermal, pasting and gelling properties of wheat and potato starches in the presence of sucrose, glucose, glycerol and hydroxypropyl β-cyclodextrin. *Carbohydrate Polymers 70,* 112-122.

Guo, Q. and Mu, T.H., 2011. Emulsifying properties of sweet potato protein: Effect of protein concentration and oil volume fraction. *Food Hydrocolloids 25,* 98-106.

Hallén, E., Ibanoğlu, S. and Ainsworth , P., 2004. Effect of fermented/germinated cowpea flour addition on the rheological and baking properties of wheat flour. *Journal of Food Engineering 63,* 177–184.

Harrison, H.F., Peterson, J.K., Snook, M.E, Bohac, J.R. and Jackson, D.M., 2003. Quantity and potential biological activity of caffeic acid in sweet potato storage root periderm. *Journal of Agricultural and Food Chemistry 51,* 2943−2948.

Haskell, M.J, Jamil, K.M., Hassan, F., Peerson, J.M., Hossain, M.I., Fuchs, G.J. and Brown, K.H., 2004. Daily consumption of indian spinach (Basellaalba) or sweet potatoes has a positive effect on total body vitamin A stores in Bangladesh men. *American Journal of Clinical Nutrition 80,* 705–714.

Hathcock, J.N., Hattan, D.G., Jenkins, M.Y., McDonald, J.T., Sundaresan, P.R. and Wilkening, V.L., 1990. Evaluation of vitamin A toxicity-Review. *American Journal of Clinical Nutrition 52,* 183-202.

Hathorn, C. S., Biswas , M.A., Gichuhi, P. N. and Bovell-Benjamin, A. C., 2008. Comparison of chemical, physical, micro-structuraland microbial properties of breads supplemented with sweet potato flour and high-gluten dough enhancers. *Lebensmottel-Wissenschaft und-Technologie – Food Science and Technology 41,* 803–815.

Henry, L.K., Puspitasari-Nienaber, N.L., Jarén-Galán, M., van Breemen, M.B., Catignani, G.L. and Schwartz, S.J., 2000. Effects of ozone and oxygen on the degradation of carotenoids in an aqueous model system. *Journal of Agricultural and Food Chemistry 48,* 5008-5013.

Hidalgo , A., Brandolin, A. and Pompei, C., 2010. Carotenoids evolution during pasta, bread and water biscuit preparation from wheat flours. *Food Chemistry 121,* 746-751.

Holtekjølen, A.K., Bævre, A.B., Rødbotten, M., Berg, H. and Knutsen, S.H., 2008. Antioxidant properties and sensory profiles of breads containing barley flour. *Food Chemistry 110,* 414-421.

Hrušková, M. and Šmejda, P., 2003. Wheat flour dough alveograph characteristics predicted by NIRSystems 6500. *Czech Journal of Food Science 21,* 28-33.

I'anson, K.J., Miles, M.J., Morris, V.J., Besford, L.S., Jarvis, D.A. and Marsh, 1990. The effects of added sugars on the retrogradation of wheat starch gels. *Journal of Cereal Science 11*, 243-248.

IVACG, 1998. Safe doses of vitamin A during pregnancy and lactation. *International Vitamin A Consultative Group, Washington DC 20036-4810, USA.*

Jadhav, S.J., Nimbalkar, S.S., Kulkani, A.D. and Madhavi, D.L., 1996. Nutritional and health aspects of food antioxidants. In: *Food Antioxidants: Technological, Toxicological and Health Perspectives.* Madhavi, D., L., Deshpande, S. S., Salunkhe, D. K., (Eds.) Marcel Dekker, New York. pp. 5-63.

Jalal, F., Nesheim, M.C., Agus, Z., Sanjur, D. and Habicht, J.P., 1998. Serum retinol concentrations in children are affected by food sources of β-carotene, fat intake and anthelmintic drug treatment. *American Journal of Clinical Nutrition 68,* 623–629.

Jangchud, K., Phimolsiripol, Y. and Haruthaithanasan, V., 2003. Physicochemical properties of sweet potato flour and starch as affected by blanching and processing. *Starch/Stärke* 55, 258–264.

Jensen, S., Oestdal, H., Skibsted, L.H., Larsen, E., Anette K. and Thybo, A.K., 2011. Chemical changes in wheat pan bread during storage and how it affects the sensory perception of aroma, flavour and taste. *Journal of Cereal Science 53,* 259-268.

Johnson, L.E, 1994. The emerging role of vitamins as antioxidants. *Archives of Family Medecine 3,* 809-818.

Kamuf, W., Nixon, O., Parker, O. and Barnum, G.C., 2003. Overview of caramel colours. *Cereal Foods World 48,* 64-69.

Kapinga, R., 2007. Promotion of orange-fleshed sweet potato to combat vitamin A deficiency in sub-Saharan Africa region: VITA experiences. In: *Proceedings: Orange-fleshed sweet potato symposium.* Faber, M., Laurie, S., van Jaarsveld, P., (Eds.), pp 14-18. Pretoria, South Africa.

Kashlan, N.B., Srivastava, V.P., Mohanna, N.A., and Motawa, Y.K., 1991. The proximate elemental composition of wheat flour and major types of bread consumed in Kuwait. *Food Chemistry 39,* 205-210.

Khalil, A.H., Mansour, E.H. and Dawoud, F.M., 2000. Influence of malt on rheological and baking properties of wheat-cassava composite flours. *Lebensmottel-Wissenschaft und-Technologie – Food Science and Technology 33,* 159-164.

Khatkar, B.S., Bell, A.E. and Schofield, J.D., 1995. The dynamic rheological properties of glutens and gluten sub-fractions from wheats of good and poor bread making quality. *Journal of Cereal Science 22,* 29-44.

Khetarpaul, N. and Goyal, R., 2009. Effect of composite flour fortification to wheat flour on the quality characteristics of unleavened bread. *British Food Journal 111,* 554-564.

Kidmose, U., Yang , R.-Y., Thilsted, S.H., Christensena, L.P. and Brandt, K., 2006. Content of carotenoids in commonly consumed Asian vegetables and stability and extractability during frying. *Journal of Food Composition and Analysis 19,* 562–571.

Kidmose, U., Christensen, L.P., Agili, S.M. and Thilsted, S.H., 2007. Effect of home preparation practices on the content of provitamin A carotenoids in coloured sweet potato varieties (*Ipomoea batatas Lam*.) from Kenya. *Innovative Food Science and Emerging Technologies 8,* 399–406

Kim, H.J., Morita, N., Lee, S.H. and Moon, K.D., 2003. Scanning electron microscopic observations of dough and bread supplemented with Gastrodia elata Blume powder. *Food Research International 36,* 387–397.

Kimura, M. and Rodriguez-Amaya, D.B., 2002. A scheme for obtaining standards and HPLC quantification of leafy vegetable carotenoids. *Food Chemistry 78,* 389–398.

Kimura, M., Kobori, C.N., Rodriguez-Amaya, D.B. and Nestel, P., 2007. Screening and HPLC methods for carotenoids in sweet potato, cassava and maize for plant breeding trials. *Food Chemisty 100,* 1734-1746.

Kiss, A. S., Kiss, J., Milotay,. P., Kerek, M.M. and Markus, T. M., 2005. Difference in anthocyanin and carotenoid content of fruits and vegetables. *Food Research International 38,* 1023-1029.

Kitts, D.D, Wu, C.H., Kopec, A. and Nagasawa, T., 2006. Chemistry and genotoxicity of caramelised sucrose. *Molecular Nutrition and Food Research 50,* 1180 – 1190.

Kokelaar, J.J. and Prins, A., 1995. Surface rheological properties of bread dough components in relation to gas bubble stability. *Journal of Cereal Science* 22, 53-61.

Kohyama, K. and Nishinari, K., 1991. Effect of soluble sugars on gelatinisation and retrogradation of sweet potato starch. *Journal of Agricultural and Food Chemistry 1001, 39,* 1406-1410.

Kontogiorgos, V., 2011. Microstructure of hydrated gluten network. *Food Research International 44, 2582-2586.*

Krinsky N.I. and Johnson E.J., 2005. Carotenoid actions and their relation to health and disease. *Molecular Aspects of Medicine 26,* 459–516.

Krishnan, J. G., Padmaja, G., Moorthy, S.N., Suja, G. and Sajeev, M.S. 2010. Effect of pre-soaking treatments on the nutritional profile and browning index of sweet potato and yam flours. *Innovative Food Science and Emerging Technologies 11*, 387-393.

Laurie S.M., 2004. Sweet potato in perspective. In: Niederwieser, J.G., (ed). Guide to sweet potato production in South Africa, pp.57-75. *Agricultural Research Council, Pretoria.*

Laurie, S.M., Van Den Berg, A.A. and Tjale, S.S., 2009. Initiation and first results of a biofortification program for sweet potato in South Africa. *Journal of Crop Improvement 23,* 235-251

Lazaridou, A., Duta, D., Papageorgiou, Belc, N. and Biliaderis, C.G, 2007. Effects of hydrocolloids on dough rheology and bread quality parameters in gluten-free formulations. *Journal of Food Engineering 79, 1033-1047.*

Lee , M.T. and Chen, B.H., 2001. Separation of lycopene and its cis isomers by liquid chromatography. *Chromatographia, 54*, 613-617.

Leighton, C.S., 2007. Nutrient and Sensory Quality of Orange-fleshed sweet potatoes. *Masters thesis. University of Pretoria.*

Lemaga, B., Mwanga, R.O.M. and Owori, C., 2007. Introduction. In: *Sweet potato recipe book: Sweet Potato Processed Products from Eastern and Central Africa*. Owori, C., Lemaga, B., Mwanga, R.O.M., Namutebi, A., Kapinga, R., (Eds.) pp. 1-2. Kampala-Uganda.

Leskova, E., Kubíkova, J., Kovacikova, E., Kosicka, M., Porubska, J. and Holčíková, K., 2006. Vitamin losses: Retention during heat treatment and continual changes expressed by mathematical models. *Journal of Food Composition and Analysis 19,* 252-276.

Lessin, W.J., Catignani, G.L. and Schwartz, S.J., 1997. Quantification of *cis-trans* isomers of provitamin A carotenoids in fresh and processed fruits and vegetables. *Journal of Agricultural and Food Chemistry 45,* 3728–3732.

Lewis, D.H., Bloor, S.J. and Schwinn, K.E., 1998. Flavonoid and carotenoid pigments in flower tissue of *Sandersonia aurantiaca* (Hook). *Scientia Horticulturae 72,* 179–192.

Lii, C. Y., Lai, M. F. and Liu, K. F., 1998. Factors influencing the retrogradation of two rice starches in low-molecular-weight saccharide solutions. *Journal of Cereal Science 28,* 175–185.

Limroongreungrat, K. and Huang, Y., 2007. Pasta products made from sweet potato fortified with soy protein. *Lebensmottel-Wissenschaft und-Technologie – Food Science and Technology 40*, 200–206.

Lindenmeier, M. and Hofman, T., 2004. Influence of baking on conditions and precursor supplementation on the amounts of the antioxidant pronyl-L-lysine in bakery products. *Journal of Agricultural and Food Chemistry 52,* 350–354.

Lindley, 1998. The impact of food processing on antioxidants in vegetable oils, fruits and vegetables. *Trends in Food Science and Technology 9*, 336-340.

Little, A.C., 1975. Off on a tangent. *Journal of Food Science 40, 410-411*.

Liu, R. H., 2007. Whole grain phytochemicals and health. *Journal of Cereal Science 46,* 207–219.

Liu, S., Lin, J. and Yang, D., 2009. Determination of cis- and trans- α and β-carotenoids in Taiwanese sweet potatoes (*Ipomoea batatas* (L.) Lam.) harvested at various times. *Food Chemistry 116,* 605–610.

Liu, X., Robinson, P.W., Madore, M.A., Witney, G.W. and Arpaia, M.L., 1999. ‘Hass’ avocado carbohydrate fluctuations. I. Growth and phenology. *Journal of Amererican Society for Horticultural Science 124,* 671–675.

Low, J., Arimond, M., Osman, N., Cunguara, B., Zano, F. and Tschirley, D., 2007. A food-based approach introducing orange-fleshed sweet potatoes increased vitamin A intake and serum retinol concentrations in young children in rural Mozambique. *Journal of Nutrition 137,* 1320-1327.

Low, J., Walker,T. and Hijmans, R., 2001. The potential impact of orange-fleshed sweet potatoes on vitamin A intake in Sub-Saharan Africa. The VITAA Project, vitamin A and orange-fleshed sweet potatoes in Sub-Saharan Africa, Nairobi Kenya. Available at: *http://www.cipotato.org/vitaa/Publications/Paper_vitaa.pdf* . Accessed: 08 June 2010.

Lupo, M. P., 2001. Antioxidants and vitamins in cosmetics. *Clinics in Dermatology 19,* 467–473.

Marco, C. and Rosell, C.M., 2008. Breadmaking performance of protein enriched, gluten-free breads. *European Food Research and Technology 227, 1205-1213.*

Martinez-Anaya, M. A., 1996. Enzymes and bread flavour. *Journal of Agricultural and Food Chemistry 44,* 2469-2480.

Martins, S.I.F.S., Jongen, W.M.F. and Van Boekel, M.A., 2001. A review of Maillard reaction in food and implication of kinetic modelling. *Trends in Food Science and Technology* 11, 364-373.

Meléndez-Martínez, A.J., Vicario, I.M., and Heredi, F.J., 2007. Analysis of carotenoids in orange juice- Review. *Journal of Food Composition and Analysis 20,* 638–649.

Mercadante, A.Z., Steck, A. and Pfander, H., 1999. Carotenoids from guava (*Psidium guajava L.):* isolation and structure elucidation. *Journal of Agricultural and Food Chemistry 47,* 145–151.

Michalska, A., Amigo-Benavent, M., Zielinski, H. and del Castillo, M.D., 2008. Effect of bread making on formation of Maillard reaction products contributing to the overall antioxidant activity of rye bread. *Journal of Cereal Science 48,* 123–132.

Mills, E.N.C., Wilde, P.J., Salt, L.J. and Skeggs, P., 2003. Bubble formation and stabilisation in bread dough. *Food and Bioproduct Processing 81,* 189-193.

Mlakar, S.G., Bavec, M., Turinek M. and Bavec, F., 2009. Rheological properties of dough made from grain amaranth-cereal composite flour based on wheat and spelt. *Czech Journal of Food Science 27,* 309-319.

Mohamed, A., Xu, J. and Singh, M., 2010. Yeast leavened banana-bread: Formulation, processing, colour and texture analysis. *Food Chemistry 118,* 620–626.

Mondal, A., and Datta, A.K., 2008. Bread baking-A review. *Journal of Food Engineering 86,* 465–474.

Morales, F.J. and Jimenez-Perez,S., 2001. Free radical scavenging capacity of Maillard reaction products as related to colour and fluorescence. *Food Chemistry 72, 119–125.*

Moreno-Álvarez, M.J., Hernández, R., Belén-Camacho, D.R., Medina-Martinez, C.A., Ojeda-Escalona, C.E. and García-Pantaleón, D. M., 2009. Making of bakery products using composite flours: Wheat and cactus pear (*Opuntia boldinghii Britton et Rose*) stems (cladodes). *Journal of the Professional Association for Cactus Development 11,* 78-87.

Morrice, C. and Morrice, G.A., 2012. The effect of inulin and fructo-oligosaccharide supplementation on the textural, rheological and sensory properties of bread and their role in weight management: A review. *Food Chemistry 133,* 237–248.

Noda, T., Kobayashi, T. and Suda, I., 2001. Effect of soil temperature on starch properties of sweet potatoes. *Carbohydrate Polymers 44,* 239-246.

Noort, M.W.J., van Haaster, D., Hemery, Y., Schols, H.A. and Hamer, R.J., 2010. The effect of particle size of wheat bran fractions on bread quality – Evidence for fibre–protein interactions. *Journal of Cereal Science 52,* 59-64.

Oki, T., Masuda, M., Furuta, S., Nishiba, Y., Terahara, N. and Suda, I., 2002. Involvement of anthocyanins and other phenolic compounds in radical-scavenging activity of purple-fleshed sweet potato cultivars. *Journal of Food Science 67,* 1752-1756

Ohm, J.B. and Chung, O.K., 1999. Gluten, pasting and mixograph parameters of hard winter wheat flours in relation to bread-making. *Cereal Chemistry 75,* 606-613.

Olkku, J. and Rha, C. 1978. Gelatinisation of starch and wheat flour starch (a review). *Food Chemistry 3,* 293–311.

Owori, C. and Hagenimana, V., 2007. Sweet potato primary products and processing procedures. In: *Sweet potato recipe book: Sweet Potato Processed Products from Eastern and Central Africa* . Owori, C., Lemaga, B., Mwanga, R.O.M., Namutebi, A., Kapinga, R., (Eds.) pp. 3-7. Kampala-Uganda.

Owori, C., Nungo, R., Kapande, A., Mukantwali, C. and Randrianaivoarivony, 2007. Sweet potato secondary processed products from new recipes. In: *Sweet potato recipe book: Sweet potato processed products from eastern and Central Africa* . Owori, C., Lemaga, B., Mwanga, R.O.M., Namutebi, A., Kapinga, R., (Eds.) pp. 11-60. Kampala-Uganda.

Padda, M. S. and Picha, D. H., 2008. Phenolic composition and antioxidant capacity of different heat-processed forms of sweet potato cv.‘Beauregard’. *International Journal of Food Science and Technology 43,* 1404–1409.

Padula, M. and Rodriguez-Amaya, D.B., 1986. Characterisation of the carotenoids and assessment of the vitamin A value of Brasilian guavas (*Psidium guajava L.*). *Food Chemistry 20,* 11-19.

Palace, V. P., Qin, N. K. Q. and Singal, P. K., 1999. Antioxidant potentials of vitamin A and carotenoids and their relevance to heart disease. *Free Radical Biology and Medicine 26,* 746–761

Pedreschi, F., Kaack, K., Granby, K. and Troncoso, E., 2007. Acrylamide reduction under different pre-treatments in French fries. *Journal of Food Engineering 79,* 1287–1294.

Philpott, M., Gould, K.S., Markham, K.R., Lewthwaite, S.L. and Ferguson, L.R., 2003. Enhanced colouration reveals high antioxidant potential in new sweet potato cultivars. *Journal of the Science of Food and Agriculture, 83,* 1076–1082.

Peressini, D. and Sensidoni, A., 2009. Effect of soluble dietary fibre addition on rheological and bread-making properties of wheat doughs. *Journal of Cereal Science 49,* 190–201.

Potter, N.N. and Hotchkiss, J.H., 1995. Food Science, 5th ed. Chapman and Hall, New York. Pp. 46-68.

Pozo-Bayón, M.A., Guichard, E. and Cayot,N., 2006. Flavour control in baked cereal products. *Food Reviews International 22,* 335-379.

Purlis, E., 2010. Browning development in bakery products – A review. *Journal of Food Engineering 99,* 239–249.

Purlis, E. and Salvadori, V. O., 2007. Bread browning kinetics during baking. *Journal of Food Engineering 80,* 1107-1115.

Ragaee, S. and Adbel-Aal, E.M., 2006. Pasting properties of starch and protein in selected cereals and quality of their food products. *Food Chemistry 95,* 9–18.

Ranhotra, G. S., Gelroth, J. A., Langemeier, J. and Rogers, D. E., 1995. Stability and contribution of beta carotene added to whole wheat bread and crackers. *Cereal Chemistry 72*, 139-141.

Roberts, D.D. and Pollien, P., 1997. Analysis of aroma release during microwave heating. *Journal of Agricultural and Food Chemistry 45,* 4388-4392.

Robotta, P.D. and Le Bail, A., 2007. Thermo-physical assessment of bread during staling. *Lebensmottel-Wissenschaft und-Technologie -Food Science and Technology 40,* 879-884.

Roccia, P., Ribotta, P. D., Perez, G.T. and Leon, A. E., 2009. Influence of soy protein on rheological properties and water retention capacity of wheat gluten. *Lebensmottel-Wissenschaft und-Technologie - Food Science and Technology 42,* 358-362.

Rodriguez-Amaya, D.B., 2010. Quantitative analysis, in vitro assessment of bioavailability and antioxidant activity of food carotenoids-A review. *Journal of Food Composition and Analysis 23*, 726–740.

Rodriguez-Amaya, D.B. and Kimura, M., 2004. HarvestPlus handbook for carotenoid analysis. Copyright HarvestPlus c/o International Food Policy Research Institute, Washington DC, USA. p. 58.

Rogers, D. E., Malouf, R. B., Langemeie, Gelroth, J. A. and Ranhotra, G. S., 1993. Stability and nutrient contribution of β-carotene added to selected bakery products. *Cereal Chemistry 70*, 558–561.

Rojas, J. A., Rosell, C. M. and de Barber, C. B.,1999. Pasting properties of different wheat flour-hydrocolloid system. *Food Hydrocolloids 13*, 27-33.

Rouillé, J., Bonny, J,-M., Valle G.D., Devaux, M.F. and Renou, J.P., 2005. Effect of flour minor components on bubble growth in bread dough during proofing assessed by magnetic resonance imaging. *Journal of Agricultural and Food Chemistry 53*, 3986-3994.

Sander, L.C., Sharpless, K.E., Craft, N.E. and Wise, S.A., 1994. Development of engineered stationary phases for the separation of carotenoid isomers. *Analytical Chemistry 66*, 1667-1674.

Salvador, A., Sanz, T. and Fiszman, S.M., 2006. Dynamic rheological characteristics of wheat flour-water doughs. Effect of adding NaCl, sucrose and yeast. *Food Hydrocolloids 20*, 780–786.

Schierle, J., Pietsch, B., Ceresa, A. and Fizet, C., 2004. Method for the determination of β-carotene in supplements and raw materials by reversed-phase liquid chromatography: single laboratory validation. *Journal of AOAC International 87*, 1070-1082.

Shah, P., Cambell, G.M., McKee, S.L. and Rielly, C.D., 1998. Proving of bread dough: modelling the growth of individual bubbles. *Trans IChemE, Vol 76, Part C*.

Shahzadi, Butt, M.S., ur Rehaman, S. and Sharif, K., 2005. Chemical characteristics of various composite flours. *International Journal of Agriculture and Biology 7,* 105-108.

Shalini, K.G. and Laxmi, A., 2007. Influence of additives on rheological characteristics of whole-wheat dough and quality of chapatti (Indian unleavened flat bread), part I-hydrocolloids. *Food Hydrocolloids 21*, 110-117.

Sharman, I.M., 1985. Hypercarotenaemia. *British Medical Journal 290*, 95-96.

Shibanuma, Y., Takedab ,Y. and Hizukurinv ,S. 1996. Molecular and pasting properties of some wheat starches. *Carbohydrate Polymers 29*, 253-261.

Shittu, T.A., Aminu, R.A. and Abulude, E.O., 2009. Functional effects of xanthan gum on composite cassava-wheat dough and bread. *Food Hydrocolloids 26,* 2254-2260.

Shittu, T.A., Raji, A. O. and Sanni, L.O., 2007. Bread made from cassava-wheat flour: I. Effect of baking time and temperature on some physical properties of bread loaf. *Food Research International 40,* 280-290.

Silva-Sanches, C., Gonzalez-Castaneda, J., De Leon-Rodriguez, A. and Barba De La Rosa, A.P., 2004. Functional and rheological properties of amaranth albumins extracted from two Mexican varieties. *Plant Foods for Human Nutrition 59,* 169–174.

Škrbić, B., Milovac, S., Dodig, D. and Filipčev, B., 2005. Effects of hull-less barley flour and flakes on bread nutritional composition and sensory properties. *Food Chemistry 115,* 982-988.

Singh, N., Bajaj, I. K., Singh, R.P. and Gujral, H.S., 2002. Effect of different additives on mixograph and bread-making properties of Indian wheat flour. *Journal of Food Engineering 56*, 89-95.

Singh, N., Gujral, H.S. and Singh, J., 2002. Effect of baking ingredients and mixing duration on dough development, gas release and bread-making properties. *Journal of Food Quality 25*, 305-315.

Solon, F.S, Klemm, R. D.W., Sanchez, L., Darnton-Hill, I., Craft, N. E., Christian, P. and West, K.P, 2000. Efficacy of a vitamin A–fortified wheat-flour bun on the vitamin A status of Filipino school children. *American Journal of Clinical Nutrition 72,* 738–744.

Sroan, B.S., Bean, S.R. and MacRitchie, F., 2009. Mechanism of gas cell stabilisation in bread making. I. The primary gluten–starch matrix. *Journal of Cereal Science 49*, 32–40.

Stathers, T., Namanda, S., Mwanga, R.O.M., Khisa, G. and Kapinga, R., 2005. *Manual for sweet potato integrated production and pest management farmer field schools in sub-Saharan Africa.* International Potato Center, Kampala, Uganda pp 168 +xxxi. ISBN 9970-895-01-X.

Stojceska, V. and Ainsworth, P., 2008. The effect of different enzymes on the quality of high-fibre enriched brewer’s spent grain breads. *Food Chemistry 110*, 865–872.

Laurie, S.M., Van Den Berg, A.A. and Tjale, S.S., 2009. Initiation and first results of a biofortification program for sweet potato in South Africa. *Journal of Crop Improvement 23, 235-251*

Takahata, Y., Noda, T. and Nagata, T., 1993. HPLC determination of β-carotene content of sweet potato cultivars and its relationship with colour values. *Japanese Journal of Breeding 43,* 421–427.

Teow, C.C., Truong, V., McFeeters, R.F., Thompson, R.L., Pecota, K.V. and Yencho, G.C., 2007. Antioxidant activities, phenolic and β-carotene contents of sweet potato genotypes with varying flesh colours. *Food Chemistry 103,* 829–838.

Tsai, P., Yu, T., Chen, S., Liu, C. and Sun, Y., 2009. Interactive role of colour and antioxidant capacity in caramels. *Food Research International 42,* 380-386.

Thorvaldsson, K. and Skjöldebrand, C., 1998. Water diffusion in bread during baking. *Lebensmottel-Wissenschaft und-Technologie -Food Science and Technology 31*, 658–663.

Tsen, C.C., 1967. Changes in flour protein during dough mixing. *Cereal Chemistry 44, 308-317*.

Tsimidou, M. and Tsatsaroni, E., 1993. Stability of saffron pigments in aqueous extracts. *Journal of Food Science 58*, 1073-1075.

Tumuhimbise, G.A., Namutebi, A. and Muyonga, J. H., 2009. Microstructure and in vitro β-carotene bioaccessibility of heat processed orange-fleshed sweet potato. *Plant Foods for Human Nutrition 64*, 312–318.

Turkmen, N., Sari, F. and Velioglu, Y. S., 2005. The effect of cooking methods on total phenolics and antioxidant activity of selected green vegetables. *Food Chemistry 93*, 713-718.

Uthayakumaran, S., Gras, P.W., Stoddard, F.L. and Bekes, F., 1999. Effect of varying protein content and glutenin-to-gliadin ratio on the functional properties of wheat dough. *Cereal Chemistry 76*, 389-394.

van Boekel, M.A.J.S., 2006. Formation of flavour compounds in the Maillard reaction. *Biotechnology Advances 24*, 230-233.

Van Der Borght, A., Goesaert, H., Veraverbeke, W.S. and Delcour, J.A., 2005. Fractionation of wheat and wheat flour into starch and gluten: overview of the main processes and the factors involved. *Journal of Cereal Science 41*, 221–237.

van Jaarsveld, P.J., Faber, M., Tanumihardjo, S.A, Nestel, P., Lombard, C.J. and Benadé, A.J.S., 2005. β-carotene-rich orange-fleshed sweet potato improves the vitamin A status of primary school children assessed with the modified-relative-dose-response test. *American Journal of Clinical Nutrition 81*,1080-1087.

van Jaarsveld, P.J., De wet Marais, Harmse, E., Nestel, P. and Rodriguez-Amaya, D.B., 2006. Retention of β-carotene in boiled, mashed orange-fleshed sweet potato. *Journal of Food Composition and Analysis 19,* 321–329.

van Hal, M., 2000. Quality of sweet potato flour during processing and storage. *Food Reviews International 16,* 1–37.

van Vliet, T., 2008. Strain hardening as an indicator of bread-making performance: A review with discussion. *Journal of Cereal Science 48,* 1–9.

Veluppillai, S., Nithyanantharajah, K., Vasantharuba, S., Balakumar, S. and Arasaratnam, V., 2010. Optimisation of bread preparation from wheat flour and malted rice flour. *Rice Science, 17,* 51−59.

von Lintig, J., Hessel, S., Isken, A., Kiefer, C., Lampert, J.M., Voolstra, O. and Vogt, K., 2005. Towards a better understanding of carotenoid metabolism in animals. *Biochimica et Biophysica Acta 1740,* 122– 131.

Waché, Y., Bosser-DeRatuld, A., Lhuguenot, J. and Belin, J., 2003. Effect of cis/trans isomerism of β-carotene on the ratios of volatile compounds produced during oxidative degradation. *Journal of Agricultural and Food Chemistry 51,* 1984-1987.

Walstra, P., 1996. Dispersed systems:basic considerations. In: Fennema OR (ed). Food Chemistry. Marcel Dekker, New York.

Waramboi, J.G., Dennien, S.,Gidley, M.J. and Sopade, P.A., 2011. Characterisation of sweet potato from Papua New Guinea and Australia: Physicochemical, pasting and gelatinisation properties. *Food Chemistry 126,* 1759-1770.

WHO, 1998. Safe vitamin A dosage during pregnancy and lactation. World Health Organisation, the micronutrient initiative pp.34. Available at:
http://whqlibdoc.who.int/hq/1998/WHO_NUT_98.4.pdf
Accessed on 05th May, 2010

WHO, 2009. Global prevalence of vitamin A deficiency in populations at risk 1995–2005. WHO global database on vitamin a deficiency. World Health Organization, Geneva. Available at: http://whqlibdoc.who.int/publications/2009/9789241598019_eng.pdf. Accessed on 18th June, 2012.

WHO, 2011. Adolescent Nutrition: A Review of the situation in selected south-east Asian countries. WHO regional office for south-east Asia, New Dehli, India. Available at: http://www.searo.who.int/en/Section13/Section38_11624.htm. Accessed on 03rd September, 2011.

WHO/UNICEF, 1995. Global prevalence of vitamin A deficiency. Micronutrient deficiency information system working paper no. 2, pp. 116. Available at: http://www.who.int/nutrition/publications/micronutrients/vitamin_a_deficieny/WHO_NUT_95.3_intro_background.pdf
Accessed on 05th May, 2010.

Wieser, H, 2000. Comparative investigations of gluten proteins from different wheat species: I. Qualitative and quantitative composition of gluten protein species. European Food *Research and Technology 211, 262-268.*

Woodall , A. A., Lee, S. W., Weesie, R. J., Jackson, M. J. and Britton, G., 1997. Oxidation of carotenoids by free radicals: relationship between structure and reactivity. *Biochemica et Biophysica act (BBA)-General subjects 1336*, 33-42.

Woolfe, J. A. 1992. Sweet potato: An untapped food resource. Cambridge University Press, Cambridge, UK.

Yadav, A. R., Guha, M., Tharanathan, R.N. and Ramteke , R. S., 2006. Changes in characteristics of sweet potato flour prepared by different drying techniques. *Lebensmottel-Wissenschaft und-Technologie –Food Science and Technology 39*, 20-26.

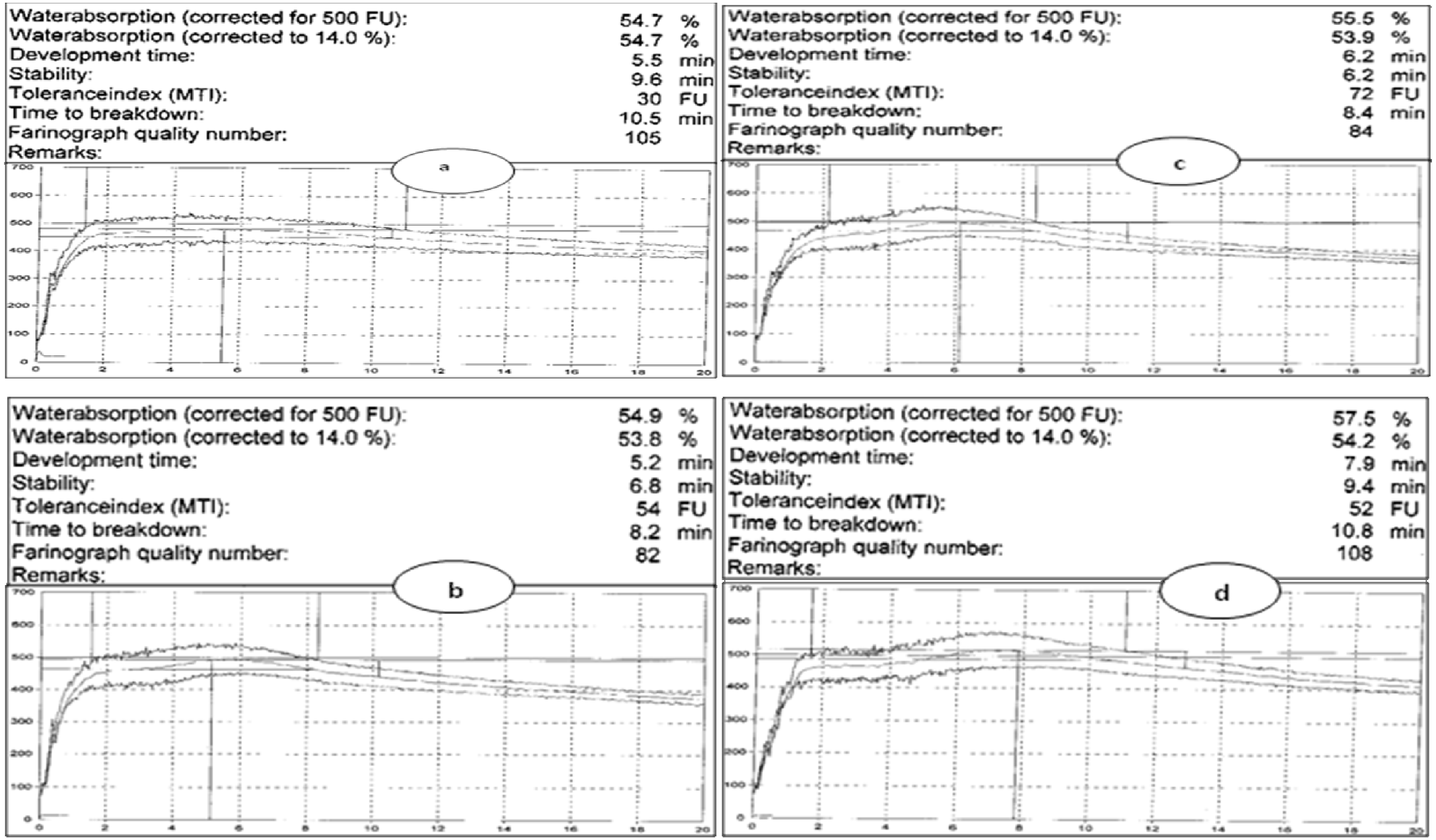
Yeo, H.C. and Shibamoto, T., 1991. Chemical comparison of flavours in microwaved and conventionally heated foods. *Trends in Food Science and Technology 2*, 329-332.

Zaidul, I. S.M., Norulaini, N.A., Mohd. Omar A.K., Yamauchi, H. and Noda, T., 2007. RVA analysis of mixtures of wheat flour and potato, sweet potato, yam and cassava starches. *Carbohydrate Polymers 69,* 784–791.

Zanoni, B., Peri, C. and Bruno, D. 1995. Modelling of starch gelatinisation kinetics of bread crumb during baking. *Lebensmottel-Wissenschaft und-Technologie – Food Science and Technology 28*, 314–318.

Zechmeister, L., 1944. *Cis-trans* isomerisation and stereochemistry of carotenoids and diphenylpolyenes. *Chemical Reviews 34*, 267-322.

Appendix 1



Waterabsorption (corrected for 500 FU):	54.7	%
Waterabsorption (corrected to 14.0 %):	54.7	%
Development time:	5.5	min
Stability:	9.6	min
Toleranceindex (MTI):	30	FU
Time to breakdown:	10.5	min
Farinograph quality number:	105	
Remarks:		

Waterabsorption (corrected for 500 FU):	55.5	%
Waterabsorption (corrected to 14.0 %):	53.9	%
Development time:	6.2	min
Stability:	6.2	min
Toleranceindex (MTI):	72	FU
Time to breakdown:	8.4	min
Farinograph quality number:	84	
Remarks:		

Waterabsorption (corrected for 500 FU):	54.9	%
Waterabsorption (corrected to 14.0 %):	53.8	%
Development time:	5.2	min
Stability:	6.8	min
Toleranceindex (MTI):	54	FU
Time to breakdown:	8.2	min
Farinograph quality number:	82	
Remarks:		

Waterabsorption (corrected for 500 FU):	57.5	%
Waterabsorption (corrected to 14.0 %):	54.2	%
Development time:	7.9	min
Stability:	9.4	min
Toleranceindex (MTI):	52	FU
Time to breakdown:	10.8	min
Farinograph quality number:	108	
Remarks:		

Figure 1. Farinograms of (a) wheat flour and composite flours containing (a) 10, (b) 20 and (c) 30% OFSP flour

FU: Farinograph units.