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Effect Of Supplementing Biofortified Orange-Fleshed Sweet Potato Flour To Wheat Flour On The Nutritional, Physical And Sensory Properties Of Cookies

By

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Acronyms

AIDS- Acquired Immuno-Deficiency Syndrome
α- Alpha
AACC-American Association of Cereal Chemist
AARC-Awassa Agricultural Research Center
AAU-Addis Ababa University
ANOVA-Analysis of Variance
AOAC- Association of Official Analytical Chemists
β- Beta
BD-Bulk Density
BR- Blending ratio
CIP-International Potato Center
DWB-Dry Weight Basis
EHNRI-Ethiopia Health and Nutrition Research Institute
FAO- Food and Agriculture Organization of the United Nations
FWB-Fresh Weight Basis
g- gram (s)
Ha-Hectare
HIV- Human Immuno Virus
HPLC-High Performance Liquid Chromatography
ILSI-International Life Sciences Institute
μg-Micro-gram(s)
OAC-Oil Absorption Capacity
OFSP-Orange-fleshed sweet potato
OFSPF- Orange-fleshed sweet potato flour
OCC- Open Column Chromatography
RAE-Retinol Activity equivalent
RDA- Recommended Dietary Allowance
SF-Spread factor
SMB-sodium Metabisulfite
SWF-soft Wheat flour
TVET-Technical and Vocational Education Training
VAD-Vitamin A Deficiency
VITAA-Vitamin A for Africa
WAC-Water Absorption Capacity
WHO-World Health Organization
WWB-Wet weight Basis

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Abstract

The effect of blending ratio and cultivar on the quality characteristics of cookies made from orange-fleshed sweetpotato and wheat composite flour were studied. A constrained, simplex lattice design was used to blend the composite flour and five different blending ratios of OFSPF: SWF (0:100, 12.5:87.5, 25:75, 37.5:62.5 and 50:50) were found. The OFSPF was processed from two different cultivars of OFSP roots, namely *Tulla* and *Kulfo,* where as the soft wheat flour which has 17% gluten was purchased from the local market. Functional properties of the composite flours as well as physical, sensory and nutritional qualities of cookies developed from various composite flours were analyzed. Consequently, as the ratio of OFSPF increased in the blend, the water absorption capacity of the composite flour was found to be significantly higher ($p< 0.05$). The oil absorption capacity and moisture content were significantly higher ($p< 0.05$) for SWF while bulk density and dispersibility were significantly higher ($p< 0.05$) for OFSPF. Blending ratio also had a significant effect ($p< 0.05$) on the quality characteristics of cookies. Increased ratio of OFSPF in the blend caused significant increase ($p< 0.05$) in moisture, β-carotene, crude fiber, ash and Calcium content of cookies and resulted in a significant decrease ($p< 0.05$) in protein, carbohydrate, iron and energy value of cookies. Blending ratio also significantly affected ($p< 0.05$) the other quality parameters of cookies (spread and sensory acceptability); the more OFSPF in the blend, the less spread and the less sensory acceptability. However, cookies made from 25:75 of OFSPF: SWF had similar overall acceptability with cookies made from 100% SWF. On the other hand, cultivar was found to have effect on the quality parameters of cookies. Cookies made from *Tulla*-OFSPF and SWF blend had significantly higher ($p< 0.05$) spread and better sensory acceptability in terms of color, crispiness, taste, flavor and overall acceptability than cookies made from *Kulfo*-OFSPF and SWF blend. It was also observed that the moisture, fiber, fat, β-carotene, energy and Calcium contents were significantly higher ($p< 0.05$) for cookies made from *Kulfo*-OFSP and SWF blend. However, the ashes, protein, Phosphorus were significantly higher ($P< 0.05$) for Cookies made from *Tulla*-OFSPF and SWF blend. Thus, formulation of composite flour for cookie making was optimized from *Tulla*-OFSPF and SWF blend and found that cookies developed from 71.0655% SWF and 28.935% *Tulla*-OFSPF blend could be the best to attain all the desired properties optimally with prediction value of 0.714 for desirability. In general, the result of this study indicated that supplementation of orange-fleshed sweet potato flour to wheat flour is possible and very promising to develop β-carotene rich cookies.

Key words: - Cookies, OFSP, VAD, β-carotene, Blending ratio, Cultivar, Nutritional quality, Physical property, Sensory property, Optimization.

1. Introduction

1.1 Back ground

Orange-fleshed sweetpotato (OFSP) is one of the biofortified food crops to alleviate micronutrient malnutrition, specifically vitamin A deficiency. It provides high amounts of highly bio-available β-carotene which is a precursor to vitamin A .Therefore, OFSP is a bred for high provitamin A carotenoid content which is obtained through Biofortification (CIP 2006.

Biofortification is the process of generating genetically improved food crops that are rich in bioavailable micronutrients, either through conventional breeding or genetic modification. Biofortification, the focus of the HarvestPlus program, represents a potentially powerful tool to increase dietary intake of essential nutrients in staple foods. HarvestPlus is a global alliance of research institutions seeking to improve human nutrition in developing countries by tackling micronutrient deficiencies in iron, zinc and vitamin A. It focuses on bio fortification of staple food crops such as Cassava, Maize, Rice, Bean, Millet and sweetpotato that are consumed by the poor (Johns and Eyzaguirre 2007).

Orange-fleshed sweet potato varieties represent a recognized benefit for individuals vulnerable to vitamin A deficiency. Efforts of plant breeders and programs to select varieties with enhanced levels of β-carotene using conventional methods and to promote their increased adoption by farmers in sub-Saharan Africa and elsewhere are currently coordinated by the International Potato Center (CIP) in the VITAA program. Besides, in order to tackle vitamin A deficiency in sub-Saharan African countries by increased consumption of OFSP, CIP and the HarvestPlus Challenge Program have launched trials to incorporate OFSP in various recipes of African foodstuffs; and a diversity of products have been tested with promising outcomes. Products from dried OFSP developed for sub-Saharan Africa include porridge, bread, and bakery products such as mandazi (traditional doughnut), chapatti, cake etc. (Owori et al., 2007).

In the case of Ethiopia, despite the efforts made at AARC since 2001, by introducing 20 cultivars of OFSP from CIP-Nairobi and then selecting and breeding them through participatory breeding initiatives with local farmer communities to incorporate agronomic traits that the farmers want and analyzing their nutritional value to release the best cultivars (Tofu et al., 2007), scientific researches (practices and trials) on processing of OFSP and development of value added products by introducing OFSP flour in various foodstuffs are very limited, unlike other sub-Saharan African countries. However, in order for communities to benefit from the high β-carotene content of OFSP via increased consumption of OFSP, evaluation of finished food products made from OFSP composite flour, to meet nutritional requirement in provitamin A, is an important matter.

Among various finished food products made from composite flour, Cookies have been suggested as a better use for composite flour because of their Ready-to-eat form, wide consumption, relatively long shelf life and good eating quality (Vieira et al., 2007). However, the utilization of OFSP flour as part of composite flour in cookie processing depends on, developing suitable processing technology, securing consumer acceptance, developing marketable products, and achieving economic feasibility. The strategy for solving these problems include setting a criteria for processing technology and product development (including energy and labor intensity), consumer appeal and convenience, nutritional and storage qualities, and ease of packaging and transportation (Tekle et al., 2009). Generally, the basis of this research is evaluation of nutritional significance of cookies, especially in terms of β-carotene content made from OFSP and wheat composite flour and indirectly, promotion of OFSP to be more used, as part of composite flour in food formulation and as value added products.

1.2 Statement of the problem

Vitamin A deficiency is a major public health issue in developing countries (FAO/WHO 2002). Sub-Saharan Africa is one of the most affected areas with 33 million pre-school children who are deficient (West 2002). Similarly, VAD is a major public-health problem in Ethiopia (Demisse et al., 2009). According to EHNRI (2006), the national prevalence of vitamin A deficiency among preschool children in Ethiopia is 61.2% which is among the highest in Sub- Saharan Africa and about 32% of child deaths, which is approximately 80,000 per year, are attributed to VAD. Children affected by VAD, consequently, are more susceptible to infectious diseases; VAD increase the risk of morbidity and mortality, and occur when diets are lacking in vitamin A or there is a lack of recommended supplementation (EHNRI 2010). However, the diet and food based approach in combating micronutrient malnutrition is essential for its role in increasing the availability and consumption of micronutrient rich foods (FAO 1997); deficiency of vitamin A therefore can be prevented/corrected by a diet high in β-carotene, which serve as precursor to vitamin A. For this effect, formulation of composite flour from OFSP, which is excellent source of β-carotenes (Rodriguez-Amaya 1997) and wheat that contain far from adequate amount of vitamin A precursor (Berdanier et al., 2008), and then development of cookies can be a good approach to provide OFSP-based finished food product to the consumer and for increased consumption of OFSP, ultimately to meet nutritional requirements of vitamin A in the form of provitamin A. Cookies are characterized by high fat content which is advantage for bioavailability of beta-carotene; the presence of dietary lipid can improve bioefficacy of β-carotenes (Haskell and others 2004). Besides, Cookies are well-known products which represent the largest category of snack items among baked foods all over the world. They can serve as a vehicle for important nutrients if made readily available to the consumer (Dogan 2006). Ideally, a food consumed by most segment of the population should be chosen as a vehicle for micronutrients like vitamin A (Mahmood et al., 2008). OFSP supplemented cookies, for example, can be helpful against VAD menace. Hence, this study is intended to develop cookies, rich in β-carotenes, from wheat and orange fleshed sweet potato composite flour.

1.3 Significance of the study

The study will provide reference materials and research gap to be studied for Researchers, Academicians, Teachers and Students who will work and study in this area. It will also contribute for production of alternative OFSP-based finished food product in order for communities benefit from increased consumption of OFSP and to combat VAD. The other significance of the study are related to public significance, public awareness about the potential nutritional benefit of OFSP, especially against VAD, rather than stigmatizing it as "poor man's" food, and industrial significance by which the findings of the study will help food industries for decision making to use OFSP flour as composite flour in various formulations to develop value-added food products. Moreover, this study will attract the attentions of all stakeholders who involve in promotion of OFSP and/or reduction of VAD and will encourage the agricultural sectors in adoption and cultivation of various cultivars of OFSP roots throughout the country.

1.4 Objectives

General objective

To study the possibility of incorporating OFSP flour to wheat flour, ultimately to develop β-carotene rich cookies and to examine their nutritional, physical and sensory properties.

Specific objectives

1. To analyze functional Properties of the composite flours.
2. To assess the effect of cultivar and blending ratio on physical properties of cookies.
3. To evaluate the effect of cultivar and blending ratio on sensory properties of cookies
4. To determine the effect of cultivar and blending ratio on nutritional value of cookies.
5. To optimize formulation of the composite flour.

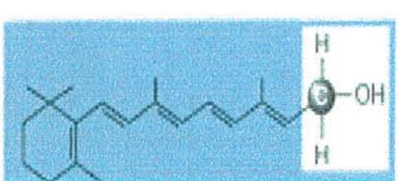
2. Review of Literature

2.1 Vitamin A: An Overview

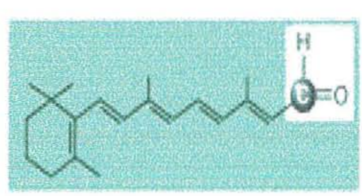
Vitamin A is fat soluble vitamin which can be found in body in three main active forms; retinol; the alcohol form, retinal; the aldehyde form, and retinoic acid; the carboxylic form, collectively these compounds are called retinoids. The cells in the body can convert retinol and retinal to the other active forms of vitamin A as needed. The conversion of retinol to retinal is reversible, but the further conversion of retinal to retinoic acid is irreversible. This irreversibility is significant because each forms of vitamin A performs a function that the others cannot; retinol supports reproduction, retinal participates in vision, and retinoic acid regulates growth (Ellie and Sharon, 2008).

There are two types of vitamin A available in the foods. Foods derived from animals (liver, fish, fish liver oil, whole milk, meat, egg yolk, meat and other dairy products) provide compounds called Retinyl esters that are readily digested and absorbed as retinol in the intestine. Whereas foods derived from plants contain no retinoids but provide carotenoids in which some have vitamin A activity (Ellie and Sharon, 2008). Plant foods such as dark green leafy vegetables, yellow and orange vegetables and fruits, and orange fleshed sweetpotato are good sources of carotenoids (McLaren and Frigg, 2001).

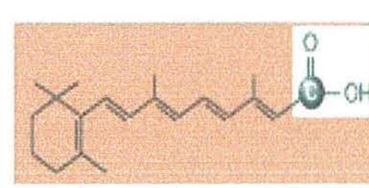
The carotenoid with the greatest vitamin A activity is β-carotene and this β-carotene is the major and the dominant carotenoid in orange-fleshed sweetpotato (Low and others 2009). Despite the fact that preformed vitamin A from animal sources is well absorbed and used efficiently in the body as compared to carotenoids from plants, 82% of vitamin A consumption in developing countries is provided by plants. This is because meat is generally not affordable for most poor people (Rodriguez-Amaya, 1996).



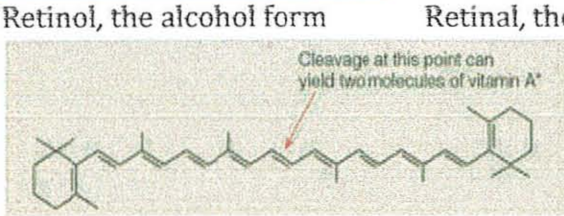
Retinol, the alcohol form



Retinal, the aldehyde form



Retinoic acid, the acid form



Beta-carotene, A precursor

Figure 2.1 Forms of vitamin A and β-carotene (Ellie and Sharon, 2008)

2.1.1 Physiological benefits

Vitamin A and its precursor, β-carotene, have diverse roles and profound effects on health. Vitamin A is essential for human metabolism. Its main function is in the visual cycle in the retina of the eye, but it also plays an important role in growth, development, and reproduction (FAO/WHO 2002). Moreover, Vitamin A is commonly known as the anti-infective vitamin, because it is required for normal functioning of the immune system. Retinol and its metabolites are required to maintain the integrity and function of the skin and mucosal cells which function as a barrier and form the body's first-line of defense against infection (Yeung and Laquatra 2003). According to Ellie and Sharon (2008), the three forms of Vitamin A perform specific tasks. Retinol supports reproduction and it is a major transport and storage form of the vitamin. Retinal is active in vision and it is also an intermediate in the conversion of retinol to retinoic acid. Retinoic acid acts like a hormone, regulating cell differentiation, growth, and embryonic development.

The other benefit of β-carotene is, it's antioxidant property; β-carotene is an antioxidant nutrient. Antioxidants have proven effective in fighting free radicals, highly unstable compounds that are formed when oxygen combines with certain substances. Free radicals can damage the basic structure of cells and thus lead to chronic diseases (notably cancer and heart disease) and accelerate the aging process. Thus, β-carotene protects oxidation and free radical damage by quenching singlet oxygen, radicals. Beta- carotene is actually the most effective singlet oxygen quencher known; A mole of beta-carotene may quench 250- 1000 singlet oxygen before being irreversibly oxidized (Bradley and min 1992; Roomi et al., 2005)

According to some epidemiological studies, individuals who eat more fruits and vegetables rich in carotenoids and/or who have higher levels of serum β-carotene have a lower risk of cancer, particularly lung cancer and high-carotenoids diets are associated with a reduced risk of heart disease (both coronary events and stroke). Furthermore, in HIV-infected patients, plasma carotenoid concentrations are reduced by 50% and T-helper (CD4) cells are destroyed, thereby impairing the immune response. However, large doses of β-carotene increase the CD4/CD8 ratio, which is usually depressed in HIV infection, and improve the response to vaccines. Hence, carotenoids seem to ameliorate the condition of AIDS patients, probably, at least in part, by enhancing the immune response (Rucker 2001).

2.1.2 Requirements of vitamin A

In order to tackle vitamin A deficiency, daily nutritional needs in vitamin A for different class-ages were evaluated by FAO/WHO. The mean requirement intake is the minimum intake to prevent xerophthalmia in the absence of clinical or sub-clinical infection whereas the recommended safe intake or recommended dietary allowance (RDA) is the average intake of vitamin A to permit adequate growth and other vitamin A dependent functions and to maintain an acceptable total body reserve of the vitamin (FAO/WHO, 2002). Since the body can derive Vitamin A from various retinoids and carotenoids, it's content in the foods and it's recommendations are expressed as retinol activity equivalent (RAE). A μ-gram of retinol counts as 1 RAE as does 12 μ-grams of dietary β-carotene or 24 μ-grams for other carotenoids; α-carotene or β- cryptoxanthin (Ellie and Sharon, 2008). On the other hand, a recent study by Haskell et al. (2004), in mashed sweet potato, reported a conversion factor of 13:1; 13 μg of β-carotene in mashed sweet potato is counts as 1 RAE.

Table 2.1 Estimated mean requirements and safe level of intake for vitamin A, by group (FAO/WHO, 1998).

Class- Age Group	Mean Requirements (μ g RE/day)	Recommended safe intake (μ g RE/day)
Infants and children		
0-6 months	180	375
7-12 months	190	400
1-3 years	200	400
4-7 years	200	450
8-9 years	250	500
Adolescents		
10-18 years	330-400	600
Adults		
Females, 19-65 years	270	500
Males, 19-65 years	300	600
65+ years	300	600
Pregnant women	370	800
Lactating women	450	850

2.2 Vitamin A Deficiency (VAD)

Vitamin A deficiency, which is among the micronutrient malnutrition, is considered as hidden hunger; while people are generally quite aware when their calorie consumption is inadequate, the opposite is true for diets deficient in trace nutrients, micronutrients. Hence, the problem of poor diet quality is often referred to as hidden hunger (WHO 1995). WHO (1996) defines VAD as tissue concentrations of vitamin A low enough to have adverse health consequences; even if there is no evidence of clinical xerophthalmia. Globally, magnitude of VAD in preschool children was estimated by WHO in 1995; about 3 million children have some form of clinical xerophthalmia and, on the basis of low blood levels of retinol, another 250 million are sub clinically deficient; South-East Asia and Africa have the highest prevalence, 69% and 49% respectively (FAO/WHO 2002). VAD can occur in individuals of any age. However, it is a disabling and potentially fatal public health problem for children under 6 years of age and VAD related blindness is most prevalent in children less than 3 years of age because this period of life is characterized by high requirements for vitamin A to support rapid growth, and the transition from breastfeeding to dependence on other dietary sources of the vitamin (Sommer 1994). Regarding the health consequences of VAD in school-age children, the prevalence of Bitot's spots (i.e. white foamy patches on the conjunctiva) is highest in this age group but their occurrence may reflect past more than current history of VAD. Women of reproductive age are also vulnerable to VAD during pregnancy and lactation (FAO/WHO 2002). Generally, VAD is a global public health challenge, particularly in infants, children, and pregnant and lactating women because of the extra requirements to support fetal, infant, and child growth (WHO 2003)

2.2.1 Causes, Consequences and Interventions

There are two principle causes of Vitamin A deficiency. Vitamin A deficiency is largely due to chronic dietary insufficiency of preformed vitamin A and proactive carotenoids (Klemm et al., 2010) which are considered as primary causes. Whereas, those factors that affect the availability, consumption, and absorption of preformed vitamin A and proactive carotenoids such as high frequency of infection, dependence of communities on few food components, socioeconomic status, lack of knowledge regarding the importance of vitamin A, poverty and malnutrition (Gibson, 1990) are considered as secondary causes and these contribute their share for major outburst of VAD. The absorption of vitamin A is interfered by diarrhea, parasitic infections and other intestinal disorders where the acute and chronic infections speed up its catabolism and excretion (West et al., 1992).Moreover, VAD is most common in populations consuming most of their vitamin A needs from provitamin carotenoid sources and where minimal dietary fat is available. 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 and the fat content of the accompanying meal. Where possible, an increased intake of dietary fat is likely to improve the absorption of vitamin A in the body (Haskell et al., 2004).

Vitamin A deficiency is responsible for Xerophthalmia, increased susceptibility to infections and impaired growth and development. Xerophtalmia includes all manifestations of visual deficiency caused by vitamin A from the mild and reversible form of night blindness, conjunctival xerosis, and Bitot's spots to irreversible form of cornea ulceration where the eye can irreversibly be damaged or lost (Sommer, 1998). In addition to the specific signs and symptoms of xerophthalmia and the risk of irreversible blindness, nonspecific symptoms include increased morbidity and mortality, poor reproductive health, increased risk of anemia, and contributions to slowed growth and development. However, these nonspecific adverse effects may be caused by other nutrient deficits as well, making it difficult to attribute non-ocular symptoms specifically to VAD in the absence of biochemical measurements reflective of vitamin A status (FAO/WHO 2002). Depression of the immune system increases the severity of measles and diarrhea, leading to increased child mortality, which is apparent even before the appearance of xerophthalmia (Berdanier et al., 2008). The chances of death from measles or diarrheal disease are greatly increased by VAD (FAO/WHO 2002). South Asia and Africa are the parts of the world most affected by vitamin A deficiency and a large proportion (20-24%) of child mortality from measles, diarrhoea and malaria can be attributed to vitamin A deficiency (WHO 2004). Children are at risk but so are pregnant and lactating women and immuno-deficient persons, such as those suffering from HIV and AIDS (Sommer, 1998). Mother to fetus transmission of HIV is elevated during VAD. Low serum retinol level is common in HIV infection and linked with viral load, increased progression to disease (Coutsoudis, 2000). VAD also interacts with other nutrients; for example, iron metabolism is negatively affected and iron is not incorporated effectively into hemoglobin (Hodges et al., 1978).

Reduction in micronutrient deficiencies can contribute significantly to improve health, productivity and well-being of human. There is dire need of time to address these malnourishments before they could play havoc with human health. Micronutrients deficiencies can be overcome by adopting multifarious strategies (Adu-Afarwuah et al., 2008). For this effect, international agencies active in micronutrient deficiencies have advocated different strategies to eliminate the VAD problems in developing countries. These include food fortification, dietary supplementation, dietary diversification, and use of bio-fortified staple foods. Administrating vitamin A capsules in every six months or every year is also part of interventions, medical intervention.

2.2.2 Magnitude of VAD in Ethiopia

In Ethiopia, vitamin A deficiency of public health significance was identified in 1958 and cognizant of the wide-scale prevalence and enormous health impacts, interventions were initiated as early as in 1960; with nutrition education and vitamin A capsule distribution. The nationwide vitamin A supplementation began in 1995 as a component of Expanded Program on Immunization (EPI), and starting from 1997, vitamin A supplementation was effected through campaigns either integrated with the National Immunization Days or as a stand-alone activity (Demisse et al., 2009). Although WHO has advocated the routine administration of vitamin A supplement for children aged 6-59 months every four to six months and mothers soon after delivery since 1987, the coverage of vitamin A supplementation of the country in 2005 among preschool children and women in the postpartum period was below satisfactory level. According to Demographic and Health Survey (DHS) Ethiopia 2005, among preschool children aged 12-59 months, only 45.8% received the supplement in the preceding 6 months of the survey and among women in the postpartum period, only 20.6% received the supplement in the first two months after their delivery (FAO 2008).

On the same year, Clinical vitamin A deficiency among children and women of childbearing age was assessed in a national survey by the Ethiopian Health and Nutrition Research Institute (EHNRI). According to this survey, the prevalence of preschool children (6-71months) night blindness was 0.7% at national level and the prevalence of Bitot's spots was 1.7%, more than 3 fold the WHO threshold (0.5%) indicating that vitamin A deficiency is a public health problem. The highest prevalence was recorded in Amhara region (3.2%). Also prevalence of maternal night blindness at national level was 1.8%. The situation was particularly alarming in Tigray region, where 14.1% of women were affected. But Sub-clinical vitamin A deficiency among mothers (based on low level of serum retinol or retinol in breast milk) was not documented (FAO 2008). Additionally, the gravity of the problem in the country is not limited to preschool children and pregnant and lactating women, rather extends to other age-group (Kassaye et al., 2001). According to their study the prevalence of Clinical xerophthalmia in northern Ethiopia in children aged 6-9 years was 5.8 % whereas the prevalence of sub clinical VAD was 59.4 %.

Generally, in Ethiopia, Vitamin A deficiency is a major public health problem. The main causes of vitamin A deficiency in the country are a very low intake of foods of animal origin, which contain high amounts of preformed retinol, and very low availability and intake of fruit and vegetables rich in carotenoids. Chronic malnutrition is also a contributing factor. Furthermore, there is a lack of knowledge among mothers regarding the importance of vitamin A and foods sources of the vitamin. Survey findings indicated that in all regions children consumed dark green leafy vegetables, red/yellow vegetables and fruits less than three times a week. According to WHO, when less than 75% of children aged 6-71 months consume vitamin A rich foods less than three times a week, the community must be considered at risk of vitamin A deficiency (FAO 2008).

2.3 Orange-Fleshed Sweet Potato (OFSP)

Sweetpotato (*Ipomoea batatas L.*), which belongs to the morning glory family *Convolvulaceae*, is a root crop that is grown in many countries predominantly for human food. Sweetpotato has long been an important food crop to many peoples, particularly to the poor, and is the sixth most important food crop in the world and fourth most important in the tropics (ILSI 2004). It is considered as a staple and co-staple in many Asian and African countries (Woolfe 1992). Sweetpotato is starchy and sweet testing root vegetable with different varieties having their own unique flavor profiles. They are often grouped into two categories depending on texture; some are firm, dry, and mealy when cooked, while others are soft and moist when cooked (ILSI 2004). Sweetpotato tuberous roots vary in color (white, yellow, orange, red, or purple fleshed), with orange-fleshed types being particularly rich in β-carotene, the most important provitamin A carotenoid. Irrespective of their color, all sweetpotato are low in protein quantity and quality (ILSI 2004).

According to ILSI (2004), two different nutritional improvements have been made to sweetpotato. One involves conventional breeding and selection of orange-fleshed sweetpotato as a crop biofortified with β-carotene (Biofortification through conventional selective breeding) that results sweetpotato with enhanced β-carotene content to control vitamin A deficiency (VAD). However, there are also some orange-fleshed varieties which are found naturally. In both cases, selected orange-fleshed varieties are being further developed through participatory breeding initiatives with local farmer communities to incorporate agronomic traits that farmers want. The other improvement involves the use of modern biotechnology to increase both the quality and quantity of protein in sweetpotato through the introduction of the synthetic *asp-1* gene (Biofortification through Biotechnology) that results sweetpotato with increased protein quantity and quality (improved amino acid profile) to address both PEM and poor quality food.

Biofortification of sweetpotato through conventional breeding, which includes the selection of orange-fleshed varieties, is being done with the intent of controlling vitamin A deficiency in developing countries. This is because β-carotene is the most important provitamin A carotenoid and the predominant carotenoid found in orange-fleshed sweetpotato, and is more bioavailable than that in carrot or green leafy vegetables for instance (Van Jaarsveld et al., 2005; Haskell et al., 2004) even if the bioefficacy depend on method of preparation; the presence of dietary lipid, can improve bioefficacy (Haskell et al., 2004). Besides β-carotene, orange-fleshed (including both white and yellow fleshed) sweetpotato roots are an important source of carbohydrate, energy, vitamin C, vitamin B6, copper, potassium, iron, and fiber. The vitamin C and β-carotene may work as antioxidants to help eliminate free radicals, molecules that contribute to the damage of both cells and cell membranes and are associated with the development of conditions such as colon cancer, arteriosclerosis, and heart disease (Roomi et al., 2005).

OFSP is an example of a biofortified crop in which the micronutrient status of staple foods is enhanced through plant breeding to the point where impact on micronutrient status can be achieved (Low et al., 2009). Under a special project called Reaching end-users of orange-fleshed sweet potato in east and southern Africa, the Harvest Plus program is popularizing orange-fleshed sweet potato varieties amongst poor populations of crop growers and consumers. Since the poorest households typically obtained over 60% of their energy needs from food staples, this strategy is particularly suited to poor rural households that cannot access purchased fortified food products but could grow OFSP (Low et al., 2009). The aim of HarvestPlus is to make available varieties that can contribute significantly to meeting the recommended daily intake of vitamin A for preschool children. A number of food science and nutrition studies have been carried out, showing that most of the provitamin A carotenoid is retained after simple processing and that consumption of these products can improve the vitamin A status of deficient children. Beta-carotene has not been reported to be toxic at intake levels observed for orange-fleshed sweetpotato (ILSI, 2004). Currently the bio fortified sweet potato varieties have shown to be capable of reducing vitamin A deficiency in studies on children in sub-Saharan Africa (van Jaarsveld et al., 2005; Low et al., 2007).

2.3.1 Carotenoids

Carotenoids are pigments which are found naturally in plant foods and their colors may vary from yellow to red. The carotenoids group structurally includes carotenes (non-polar) and xanthophylls (polar). They are usually C_{40} that include more than 600 structures synthesized by plants. Of the 600 carotenoids found in nature, about 50 carotenoids are known to have a provitamin A activity and only 3 are important precursors of vitamin A in humans; α-carotene, β-carotene, and β- cryptoxanthin (Rodriguez-Amaya 1997). The carotenoids in orange-fleshed sweetpotato comprise almost exclusively *trans*-β-carotene (Rodrigues-Amaya and Kimura 2004). Similarly, Bengsston et al. (2008) reported that trans-β-carotene represents about 80-90% of the total carotenoids in OFSP. Trans-β-carotene is the carotenoid with the highest provitamin A activity (100%) because the β-carotene molecule can be entirely converted into two molecules of vitamin A or retinol (Rodriguez-Amaya 1997).

2.3.2 Degradation of Carotenoids

Carotenoids are lipophilic unsaturated structures that are unstable (Gayathri et al., 2004). The main causes for degradation of carotenoid are light, heating, and oxygen. These could play a role in the breakdown of carotenoid in food products such as OFSP exposed to environment (heat, air and light) during processing and storage. The chemical degradation occurs by two phenomena; isomerization and oxidation (Bechoff et al., 2010b).

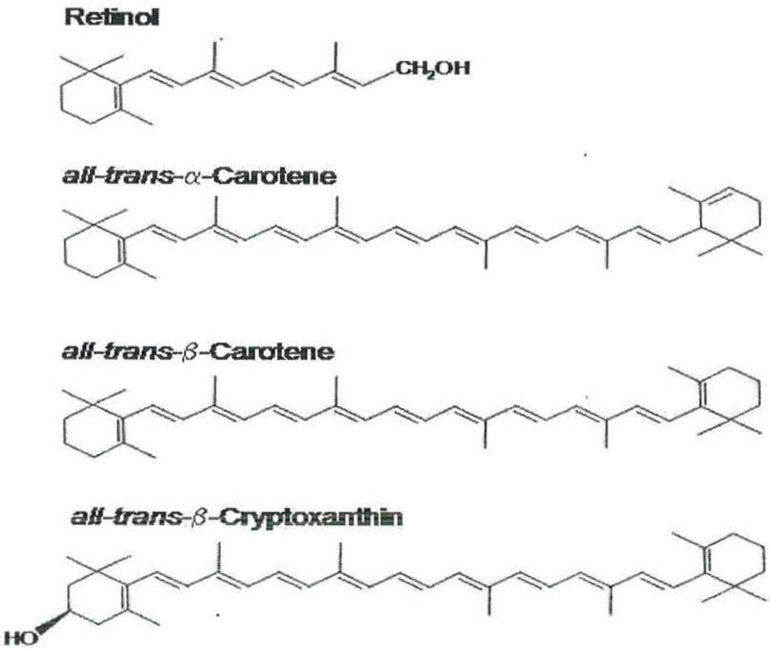


Figure 2.3.2.1 Structures of the most common provitamin A carotenoids and retinol (Bechoff et al., 2010a).

❖ Isomerization

Trans-carotenoids, carotenoids that are found in nature, isomerize in to cis-carotenoids when there is stressful condition like heat and UV-light exposure. For instance, trans-β-carotene isomerizes to 9-cis, 13-cis and 15-cis-β-carotene and these Cis-isomers have less provitamin activity (about half) than trans-β-carotene (Bechoff et al., 2010a). Moreover, cis-β-carotene is less bioavailable than trans-β-carotene (Rodriguez-Amaya and Kimura 2004). The quantity of isomer formed in processed products is related to the heat and length of treatment. Isomerization can occur in provitamin A carotenoids at temperatures above 35°C. 9-cis is predominantly formed above 100°C whereas 13-cis and 15-cis are formed below 100°C (Bechoff et al., 2010a).

❖ Oxidation

Both type of Oxidation, Autoxidation (non-enzymatic oxidation) and Enzymatic oxidation are considered to be the major causes of loss of provitamin A activity during processing and storage and the mechanism in both cases is a free radical reaction (Bechoff et al., 2010). Oxidation changes carotenoids in to other type of degraded products in chain radical reaction (Rodriguez-Amaya and Kimura 2004).

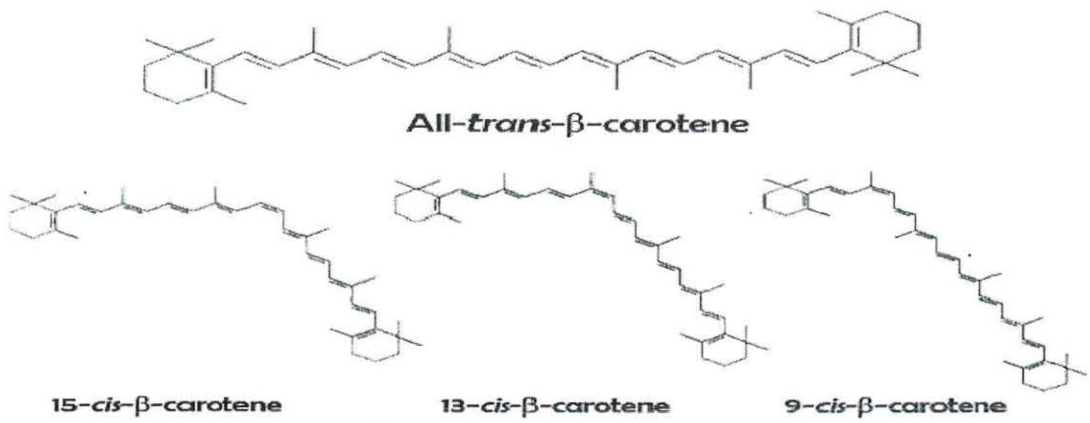


Fig 2.3.2.2 Isomers of β-carotene (Rodriguez-Amaya and Kimura, 2004)

- **Autoxidation (Non-enzymatic oxidation)**

The unsaturated and conjugated double bond system of carotenoids is the electron rich systems that induce instability toward oxidation because of attraction of electrophilic molecules (Siems et al., 2008). Autoxidation occurs through free radical reactions when unpaired and highly unstable electron or singlet oxygen is induced. Singlet oxygen is an excited state of molecular oxygen (in air) that can initiate free radical reactions with a variety of substrates having double bonds and ultimately causes oxidation of organic compounds found in a food. These oxidation reactions damage components such as fatty acids or other unsaturated compounds found in plant food cells (Britton et al., 2008). However, β-carotene has rich electron system to quench singlet oxygen and this quenching property means that b-carotene can protect cell components from free radical oxidation which is responsible for its autoxidation. In addition to this, Loss of water during drying has proved to be a risk factor in a free radical process (Bechoff et al., 2010a).

- **Enzymatic oxidation**

Enzymatic oxidation easily happens during food preparation, such as cutting, peeling or low temperature heating, because tissue disruption frees enzymes that isomerizes and oxidize carotenoids (Rodriguez-Amaya and Kimura 2004). Enzymatic oxidation of carotenoids occur when carotenoid cleavage dioxygenases cleave specifically the 9 and10 or 11 and 12 double bonds of carotenoids leading to apocarotenoids products (Huang et al., 2009)

2.4 Composite flour

Composite flours are a mixture of flours from tubers rich in starch (e.g. Cassava, yam, Sweetpotato, etc) and/or protein-rich flours (e.g. Soy, peanut etc.) and/ or cereals (e.g. maize, rice, millet, sorghum, etc.), with or without wheat flour (Shittu et al., 2007). The presence and type of wheat flour in the composite flour depend on the nature of products to be produced. For instance, soft wheat flour is the most suitable flour for the production of cookies. Because, soft wheat flour has unique properties of relatively lower protein content (8-11%) and more mellow gluten properties than other flours including hard wheat flour(Sumnu and Sahin 2008). The use of composite flour is considered advantageous in developing countries, with the progressive increase in the consumption of baked products and confectioneries, as it reduces the importation of wheat flour (save hard currency), encourages the use of locally grown crops as flour, widens the utilization of locally grown crops in food formulation (Ade-Omowaye et al., 2008), promotes the nutritional value of locally grown crops, provides nutritious food to more people, and brings a better overall use of domestic agriculture production (Hugo et al., 2000).

In the last few years, satisfactory baked products such as bread, cookies, pasta etc. have been made from composite flours. Considering previous studies and trials made on the incorporation of tubers in composite flour, root vegetables such as Taro, cassava, potato and sweetpotato were blended with wheat flour for cookie making (Singh et al., 2008; Ojinnaka et al., 2009; Tekle et al., 2009). When bakery products made from composite flour like wheat flour and vegetable flour, their overall quality like odor, flavor, appearance and shelf life, should be as similar as possible to those of products made from wheat flour only. Because the success of using composite flour is only expected if all the sensory attributes of baked products made from composite flour differ only slightly from those of the products with which they are compared (Singh et al., 2008). Similarly, cookies developed from composite flour such as mixture of wheat flour (cereal) and OFSP flour (root vegetable) should have acceptable sensory quality and physical properties, even though the purpose of the composite flour was to compensate what is nutritionally absent in wheat flour.

2.4.1 Quality of OFSP flour during Processing and storage

Processing sweetpotato in to the form of flour improves the shelf life and makes it easier to incorporate it into food products. Sweet potato flour can easily be promoted as a substitute for wheat flour in sweet baked products and can also be used for its high carotenoid content. However, the quality of the sweetpotato flour should be considered. The quality characteristics of Sweet potato flour include moisture content where storage is concerned, nutritional value, microbiological quality, functional properties and organoleptic quality. Sweetpotato flour can serve as a source of energy and nutrients (carbohydrates, β-carotene (provitamin A), minerals (Ca, P, Fe, and K), and can add natural sweetness, color, and flavor to processed food products (Van Hall 2000).

Despite the fact that, processing provides an alternative to the difficulties associated with storage of fresh roots by reducing their water content (Woolfe 1992) and by transforming bulky and perishable sweetpotato roots into compact, easily stored and transportable material, such as flour (Van Hall 2000), the removal of water affects the internal cell structure of the fresh roots leading to higher losses of micronutrients such as provitamin A (Bechoff et al., 2009). Disruption of the food's cellular matrix by processing makes carotenoids more vulnerable to oxidative degradation (Kosambo et al., 1998). Loss of water during drying has proved to be a risk factor in a free radical process and β-carotene oxidation (Bechoff et al., 2010a). There are various factors that affect β-carotene content in sweet potato flour, especially from OFSP tubers, like initial levels of carotenoids which are in turn influenced by cultivar type, root maturation and location (Kosambo et al., 1998), processing methods, and deteriorative changes during storage (Van Hall 2000). Generally, raw materials, practices during processing and storage conditions are very important matter to be considered to have better b-carotene in the flour.

Drying is the most critical step in processing sweet potato flour in terms of the final flour quality (Van Hall, 2000). There are two basic types of dryers; Natural dryers (solar or sun) which uses the cheapest, free and non-polluting energy with a minimum investment in equipment and artificial dryers (air-oven, cabinet, tunnel, etc) where air is heated by a fuel or electricity (Bechoff et al., 2009). Artificial dryers have advantages over natural dryers by which temperature, drying time and air-velocity are controlled leading to consistent, high quality products. In this respect, natural drying has a number of disadvantages; poor control of energy input and product quality, the interruption of drying caused by cloud, rain, and nightfall as well as frequent contamination of food by microorganisms, dust, and insects. Therefore, the quality of naturally dried foods is often considered inferior to that of foods dried by Artificial drying methods (Van Hall 2000). Moreover, Traditional open air sun drying involves in damaging provitamin A by UV-sun radiation because of poor control over environmental factors. Even though, shade drying is an alternative to sun drying to limit degradation by UV-radiation, it has disadvantage. The disadvantage of this dryer is the requirement longer drying times that can lead to off-odors (fermentation), if the temperature is low or humidity high (Bechoff et al., 2009).

- **Raw materials** (Food type, Cultivar type)

Types of foods have effect on retention of carotenoid after processing. In a study on carotenoid retention of OFSP and carrot in solar drying, levels of loss were found to be significantly different, 10% and 59% respectively. The author suggested that the difference in initial moisture contents of these food products, being 75.8% and 90.5%, would have influenced the level of carotenoid lost after drying (Mdziniso et al., 2006). According to Kosambo (2004), cultivar types also have effect on β-carotenoid retention level after processing. Based on the study, chips from thirteen different Kenyan OFSP cultivars with initial β-carotene content between 1.64 and 422 μg.g-1 (DWB) had different retention level, between 6 and 57%, after drying by electric cabinet dryer at 58°C for 4 hrs.

Similarly, twenty three cultivars with a total carotenoid content of between 2 and 632 μg.g-1 (DWB) had losses ranging from 0 to 80% in dried and processed sweet potato products (Hagenimana et al., 1999). In both studies, the reductions observed were greater in provitamin A carotenoids rich cultivars than in ones containing low amounts. Therefore, sweet potato cultivars have a significant effect on carotenoid losses in drying and the carotenoid loss appeared to be related to the initial carotenoid and moisture content. Cultivars with higher initial moisture and carotenoid contents of cultivars occurred to be related to higher levels of carotenoid losses after drying (Bechoff et al., 2009).

- **Processing** (Peeling, Slicing, Pre-treatments and Drying methods)

- **Peeling**

Peeling of the roots is included in the process to increase the quality of flour; avoids discoloration of the flour that can be caused by the skin of tubers (Van Hall, 2000). Color serves as a basis for the identification and quantification of carotenoids by Spectrophotometric reading, High Performance Liquid Chromatography (HPLC) and colorimetric methods (Bechoff et al., 2010); loss or change of color during the analysis can clearly be related to degradation of carotenoids (Rodriguez-Amaya 2001). Besides, sweet potato is reported as an excellent source of peroxidase activity, which is mostly situated in the peel. Peroxidase can cause enzymatic cleavage of b-carotene in the presence of phenolic compounds. Peroxidase can also generate free radical species to degrade carotenoids during storage (Castillo-Leon et al., 2002).

- **Slicing**

The fresh roots of OFSP can be cut in to slices, cubes, or shred to speed up drying. The thinner the pieces, the shorter the drying time; the size of the slices most commonly used is with thickness of 2-3mm (Van Hall 2000). Slices with a larger surface area were less exposed to carotenoid loss (Bechoff et al., 2009) during drying by sun light; high degree of shrinkage may have protected them from the sun's rays and oxidation damage.

- **Pre-treatments** (Blanching and Soaking with or without additives)

Blanching and soaking are treatments before drying to keep the quality of flour. Blanching inhibits enzymes that cause discoloration of products and degrade provitamin A such as lipoxygenases and peroxidases. Similarly, soaking can prevent discoloration without deterioration of other quality characteristics if carried out in water with SMB (0.5%) for some minutes (Van Hall 2000). But soaking chips in water without additives lead to high loss of carotenoids; after drying, unsoaked samples have higher retention than the soaked samples. Therefore, the use of additives such as sulphite (SMB) is important because they act as an antimicrobial agent and inhibitor of enzymes (Bechoff et al., 2010).

Bechoff et al., (2010) confirmed that most pre-treated and soaked samples (salt-treated; ascorbic acid and SMB-treated) have higher retention than control that have been dipped in deionized water; the addition of chemicals to the water reduced the loss of carotenoids. SMB was the most significant in terms of reducing losses. Combining the chemicals (salt-ascorbic acid; salt-citric acid and citric acid-SMB) did not appear to have a synergistic effect. All in all, the use of pre-treatments successfully reduced the carotenoid degradation in dried products but not in stored ones. Blanching should be as a form of incomplete cooking that enables the slices to extend on tray easily and dry rapidly, otherwise, the slices will be easily eroded, very difficult to handle, takes more time to be dried. Blanching should be followed by immediate cooling to avoid further cooking (Van Hall 2000).

- **Drying methods** (Artificial drying or Natural drying)

According to Bengtsson et al., (2008), who studied effects of various traditional processing methods on the all-trans-b-carotene content of several Ugandan OFSP cultivars, the average losses during drying were only 12% and 9% in oven drying and solar drying respectively. However, the retention of all-trans-b-carotene in OFSP after open-air sun drying was somewhat lower due to the exposure to direct sunlight (84 %). The significant finding was that both solar and oven drying can be considered as appropriate drying methods, resulting in high retention values. On the other hand, Hagenimana et al., (1999), found an average total carotenoid loss of 30% in 23 sweet potato varieties with various initial carotenoid contents by drying in an oven at 65°C for 12 h. In general, in the different food commodities studied, artificial drying (fan-operated) has been reported to retain more provitamin A than natural drying (shade, solar and sun) (Kosambo, 2004).

- **Storage** (packaging and Temperature)

Although flour is less vulnerable to spoilage during storage than the fresh roots, it has a capacity for absorbing moisture. Dried sweetpotato products have a higher sugar content, which favors the growth of microorganisms. For prolonged storage, sweetpotato flour must be put in sealed containers or packaging while it still has low moisture content. The packaging material must be impermeable to vapor and gas (Van Hall 2000) and during storage, auto-oxidation of carotenoids may take place, leading to a loss of color and an undesirable decline in nutritional value. The stability of beta-carotene proved to be strongly and adversely affected by storage temperature and light, with low temperature (4°C) and storage in the dark to be optimal for stability (Woolfe 1992). Storage at low temperatures slow down enzymatic activities and other chemical reactions in general and therefore help maintain product quality. A simple fridge (4°C) would help to significantly extend the shelf life of dried sweet potato (Cinar 2004). But during storage, oxygen has a greater impact on b-carotene breakdown compared to light, clear or opaque packaging (Bechoff et al., 2010b). Quality changes during storage of sweetpotato flour are protein quality, b-carotene content due to auto-oxidation, browning, and microbiological contamination. However quantitative data are very scarce (Van Hall, 2000).

According to Bechoff et al., (2010a), Packaging is one of the factors which have effect on loss of provitamin A carotenoids during storage. A good packaging material should isolate the product (OFSP chips or flour) from the three main degrading elements (water, oxygen and light). Hagenimana et al., (1999) further noted that carotenoid losses during storage also depend on the type of samples. Losses in Chips/slices were lower than of flour; consequently it would be therefore more judicious to store chips/slices and make flour as needed. He found that there was only average 10% loss of total carotenoid in dried slices from 24 cultivars in opaque paper bags under ambient conditions for 11 months.

2.4.2 The use of composite flour for cookie making

Cookies, one of bakery products, are ideal product for food fortification purpose due to their higher nutritive value, palatability, compactness and convenience. Lower moisture contents make them more suitable as they are usually free from microbial spoilage thus being more stable (Wade, 1988). Bakery products are varied by addition of value added ingredients. Experience gained in the use of composite flours has clearly demonstrated that for reasons of both product technology and consumer acceptance, wheat is an essential component in many of these flours. However, the extent to which percentage of wheat flour required to achieve a certain effect in composite flours depends heavily on the quality and quantity of wheat gluten and the nature of the product involved (Mepba et al., 2007); the nature of the products to be baked affect the amount of wheat flour to be replaced by other non-wheat flours (e.g. vegetable flours).

Satisfactory cookies have been made from composite flour through a blend of wheat flour with other cereals, legumes, tubers etc. Singh et al., (2008) studied the nutritional, textural, and sensory properties of cookies by supplementing various proportion of sweet potato flour (0-100%) at 20% interval with an objective of developing cookies with good taste, texture and appearance, which resembles as closely as possible to the wheat flour based product and concluded that incorporation of 40% sweetpotato flour yielded approximately similar results compared with wheat flour cookies in terms of nutritional value and texture. Tesfaye et al., (2010) have also developed cookies from wheat, Quality protein maize (QPM) and carrot composite flour with an objective of producing cookie rich in β-carotene and tryptophan and acceptable and nutritious cookies were obtained. Contrarily, there was no published work about cookies developed from OFSP However, Kosambo (2004) has developed porridge, mandazi, and bread from OFSP and losses of all-trans-β-carotene on processed products made from sweet potato flour were high in all cases; losses were greater than 80% in mandazi and bread and greater than 65% in porridge.

2.5 Cookies

Cookies or biscuits are chemically leavened baked products which are characterized by a formula high in sugar and fat and low in water (Sumnu and Sahin, 2008). They are stable foods and have advantages such as long shelf life and good eating quality (Dogan, 2006). Besides, cookies are nutritive, ready-to-eat, convenient and inexpensive snacks which are a rich source of fat and carbohydrate. Hence, they are energy giving foods and they are also a good source of protein and minerals (Oladele et al., 2007).

Cookies and biscuits are products made from soft and weak flours. Soft wheat flour is the main ingredient in cookie dough formula (Tesfaye et al., 2010) which provides the matrix around which other toughening or tenderizing ingredients in varying proportions are mixed to form dough (Pareyt and Delcour, 2008). Low content of protein (8 to 10% in the grain), low water absorption, and low resistance to deformation are the characteristics that describe the suitability of wheat for biscuit production (Pedersen et al., 2004).

2.5.1 Soft Wheat Flour

Wheat is categorized as hard or soft based on kernel texture. Compared to wheat with a softer texture, hard wheat requires more energy to be milled into flour and produces coarser flour, and also one with more starch damage. Conversely, wheat kernels with softer texture produce finer flour with less starch damage, both important attributes of high-quality soft wheat flour. Hard wheat is generally bred to have higher protein content (11%) than soft wheat (8 to 10%), although protein content and hardness are not necessarily linked (Sumnu and Sahin, 2008). Soft wheat flour is the most suitable flour for the production of cookies because it has unique properties of relatively lower protein content (8-11%) and more mellow gluten properties than other flours including hard wheat flour.

Wheat cookie flours can be specified as soft wheat flours with a moisture content of about 14% and a starch content of about 70-75%. They have lower absorption, finer granulation, and less starch damage than hard wheat flours (Pareyt and Delcour, 2008). The main use of hard wheat flours is in bread, where strong and high levels of protein are needed. Soft wheat flours on the other hand are used in products where weaker protein (i.e., weaker dough strength and weaker viscoelastic properties) is desired. Products like cookies however, generally require doughs that are weaker (Sumnu and Sahin, 2008). The most important flour constituents in relation to flour functionality include the proteins, starches, and lipids where all affect the size or spread of cookies as well as their texture and appearance (Sumnu and Sahin, 2008).

Wheat proteins have the unique ability to form a viscoelastic network that allows for the production of products such as bread. The proteins mainly responsible for the viscoelastic properties of flour are the gliadins (prolamins) and glutenins (glutelins). Glutenins are large polymeric proteins that give dough strength and elasticity. Gliadins are smaller monomeric proteins that are responsible for dough extensibility. Together these proteins form the gluten proteins. Both the quantity (amount) and quality (type) of protein are important to flour characteristics. The strong gluten proteins found in hard wheat flour are able to form a network with good gas-retaining properties vital for yeast-leavened products. Soft wheat flours are typically low in protein content (8 to 10%) and the proteins are weak in strength, characteristics better suited to making more tender products such as cookies. Generally, gluten is responsible for the rheological properties of dough. Increasing the protein content of flour increases dough viscosity and elasticity. Highly elastic dough is not desirable in cookie making because it shrinks after lamination. Cookie dough tends to spread (become larger and wider) as it bakes rather than to shrink. Otherwise, diameter and spread of cookie will be reduced; Spread is an important quality parameter for cookies (Sumnu and Sahin, 2008).

Wheat flour contains over 70% starch that is composed of approximately 25% amylose and 75% amylopectin. Starch granules can be physically damaged during flour milling, increasing their water-holding ability and susceptibility to attack from the enzyme α-amylase. Soft wheat flour, in general, is lower in damaged starch content than hard wheat flour, due to the softer kernel texture and higher break flour yield. In bread flour, a controlled amount of damaged starch is needed because the enzymatic breakdown of starch provides some food for the yeast. However, in soft wheat products, the increased water absorption associated with increased levels of damaged starch can be detrimental to product quality (Sumnu and Sahin, 2008).

Flour lipids are important for quality attributes of soft wheat products such as cookie spread. Studies involving the removal and reconstitution of flour lipids have shown that they are important to cookie spread, top grain (an "islanding" pattern formed on the surface of cookies), and internal structure (Sumnu and Sahin, 2008).

2.5.2 Effect of Ingredients on cookie processing

Apart from soft wheat flour, Sugar, fat, water, leavening agent and salt are the common ingredients in formulation of cookie dough. These ingredients have a major effect on rheological properties of doughs and quality of cookies. With regard to wheat flour, rheology is the measure of the flow and deformation of dough. These dough properties can affect product qualities such as geometry (e.g., cookie spread), texture, and handling during processing (Sumnu and Sahin, 2008).

❖ **Sugar (Sucrose)**

Sugar restricts the development of gluten network by competing for water that otherwise would have been absorbed by gluten. The limited amount of water used in cookie formulation, and also its non availability to protein and starch, partially contributes to the crispness of cookie (Pareyt and Delcour, 2008). Sucrose acts as a hardening agent by crystallizing as the cookie cools and making the product crisp. Fine grain size and a high concentration of sugar contribute to significant spreading of the biscuit.

An increase in sugar concentration in cookie dough reduces its consistency and cohesion. Sugar makes the cooked product fragile, because it controls hydration and tends to disperse the protein and starch molecules, thereby preventing the formation of a continuous mass. The addition of sugar to the formula decreases dough viscosity and relaxation time. Sugar promotes cookie length and reduces thickness and weight. Biscuits rich in sugar are characterized by high cohesive structure and crisp texture. Increasing the amount of sugar generally increases the spread and reduces the thickness of biscuits (Sumnu and Sahin, 2008).

❖ **Fat (Shortening)**

Fat acts as a lubricant and contributes to the plasticity of the cookie dough. It prevents excessive development of the gluten network during mixing. The presence of fat contributes to the reduction of the elastic nature of dough and therefore the shrinking of the dough during molding. The addition of fat influences the texture and taste of cookies, making the cookies crispier because this allows the dough to spread as it cooks on the hot cookie sheet (Sumnu and Sahin, 2008). Fat functionality is very versatile in baked products which include providing of flavor and mouth feel and also contributes to the appearance, palatability and texture of the cookies (Zoulias et al., 2002). When fat is mixed with the flour before its hydration, the fat prevents the formation of a gluten network and produces less-elastic dough. High-elastic dough is not desirable in biscuit making, because it shrinks after lamination. Starch swelling and its gelatinization are also reduced at high levels of fat, giving a crisp texture (Sumnu and Sahin, 2008).

❖ **Leavening agent (Baking Powder)**

Leavening is a raising action that aerates dough during mixing and baking so that the finished products are greater in volume, lighter and superior in texture. Cookies are mainly leavened by chemical leavening, baking powder, that makes cookies porous and crisp (Edwards, 2007). The dough used in soft wheat products is usually rich in fat and sugar but usually low in water content. Therefore, yeast is not suitable as a leavener in most of the soft wheat products. Baking powder is a dry chemical leavening agent used in baking which contain an alkali, typically sodium bicarbonate, and an acid together with starch to keep it dry. When dissolved in water, the acid and alkali react and emit carbon dioxide gas, which expands existing bubbles to leaven the mixture (Sumnu and Sahin, 2008).

- **Water (H_2o)**

Water is necessary for solubilizing other ingredients, for hydrating proteins and carbohydrates and for the development of gluten network. The doughs with low water are not consistent because they lack hydration. Doughs with high water content are extremely soft and sticky, making it impossible to work because an increase in water content lead to a decrease in dough viscosity and a slight reduction of the relaxation time, indicating reduction of elasticity. Therefore, the cookie expanded lengthwise, with a smaller thickness (Sumnu and Sahin, 2008).

- **Salt (NaCl)**

Salt is used in all biscuit recipes for its flavor and flavor-enhancing properties. Salt also toughens the gluten and hence reduces stickiness (Sumnu and Sahin, 2008).

3. Materials and Methods

3.1 Location of the study

The experiment was conducted in Addis Ababa, Ethiopia, which lies at an altitude of 2,500 meters and is located at 9.03°N 38.74°E. Cookies were baked at the department of Hotel Management in Entoto Technical and Vocational Education Training (TVET). Analysis for functional properties, physical properties, sensory properties and nutritional values were done at the laboratory of Food Science and Nutrition Program (AAU), laboratory of Food Technology laboratory (AAU) and laboratory of Ethiopian Health and Nutrition Research Institute (EHNRI). The OFSP roots were grown at Wondo Genet field located at 7°1'N and 38°35'E at an altitude of 1723 meters.

3.2 Sources of raw materials

Raw materials, two cultivars of orange-fleshed sweet potatoes (OFSP), namely *Tulla* and *Kulfo* which are 6 months old, optimum maturity period based on total yield/ha, were obtained from Awassa Agricultural Research Center (AARC); Sweet potato roots at optimum time of harvest can produce flour with better nutritional value (Van hall 2000). Soft wheat flour (SWF) which has 17% gluten was bought from kality food Share Company. The other ingredients such as powdered salt, baking powder, powdered sugar and shortening were purchased from the local market in Addis Ababa.

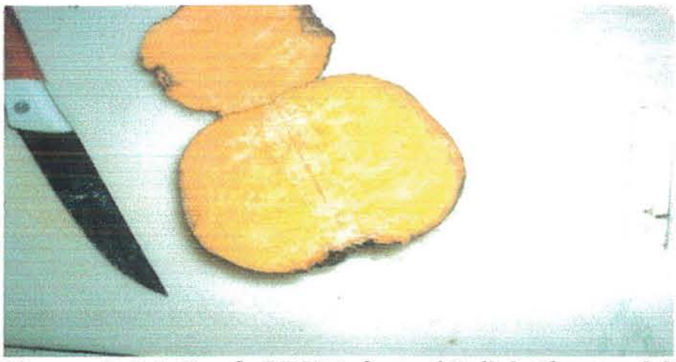


Figure 3.1.1 Fresh OFSP tubers (*Tulla*) Obtained from AARC.

Figure 3.1.2 Boiled OFSP tubers (*Tulla*) Obtained from AARC

3.3 Preparation of OFSP flour

Flour from two cultivars of OFSP was prepared according to the method described by Van Hal (2000) and as shown in figure 3.3.2. Briefly; fresh sweet potato tubers (3-7 days after harvest, maintained under ambient air condition) were thoroughly sorted to avoid rotten and excessively damaged tubers. The sorted tubers were washed with tap water to remove the adhered soil and dirt. The tubers were thereafter peeled manually using a stainless steel kitchen knife. The Peeled tubers were submerged in water to avoid enzymatic browning and then sliced in to thin slices, approximately similar thickness, to facilitate fast rate of drying. The sliced tubers were blanched in boiling water for 10 min, in order to inactivate enzymes that may cause browning reaction and degradation of beta-carotene, followed by immediate cooling in cold water to avoid further cooking. The cooled slices were then drained on perforated plastic tray and evenly spread on stainless steel tray to allow maximum surface area for drying. The slices were dried in an Air- oven (model DHG-9055A) provided with a motor fan set at 60°C until the chips were brittle and easy to be milled (10 hrs). The OFSP chips were then milled into flour using a laboratory disc mill (Tecator, CYLOTEC 1093 Sweden) fitted with sieve having circular opening 100μm diameter at EHNRI. Finally, the flour obtained was packed in double package, polyethylene plastic bag and aluminum foil to avoid moisture absorption and destruction of β-carotene by light and oxygen. Packed flour was stored at 4°C until analyzed.

Figure 3.3.1 OFSP flour obtained from Tulla cultivar.

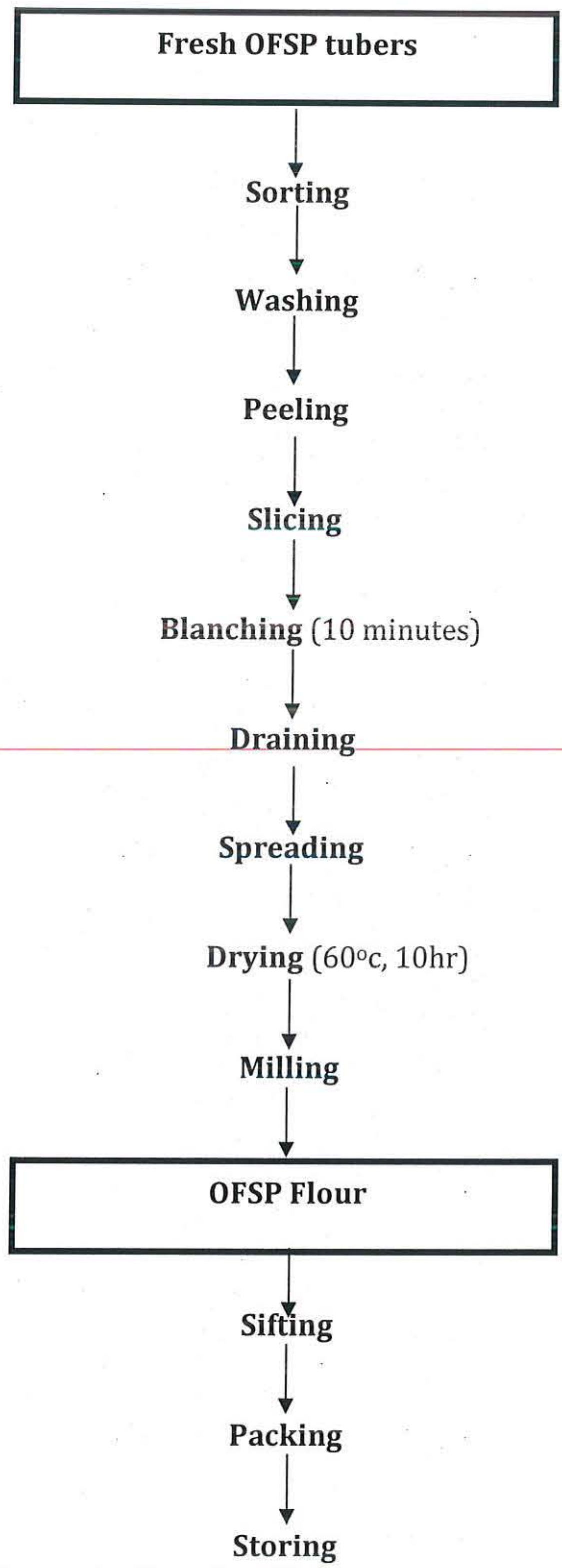


Fig 3.3.2 Flowchart for the production of OFSP flour

3.4 Formulation of composite flour

Formulation of composite flour was done by mixture design, simplex lattice design, using design expert 8 soft ware (version 8.0.5.2). The basis of simplex-lattice design is the distribution of experimental combinations to be tested evenly and regularly in the simplex region, experimental space. Therefore, points on a simplex form a lattice, an orderly arrangement consisting of an evenly spaced distribution of points (Lazic', 2004). Following the quality parameters required by the cookie, the lower limit of soft wheat flour in the simplex region has been selected to be 50% and five different blending ratios (BR), including control, were formulated by the constrained simplex lattice design.

	BR1	BR2	BR3	BR4	BR5	
SWF	(100:0)	(87.5:12.5)	(75:25)	(62.5:37.5)	(50:50)	**OFSPF**

Fig 3.3 Geometrical description of constrained Simplex lattice design

3.5 Cookie Processing

Cookie doughs were formulated, prepared and baked based on the methods used by Tesfaye et al., (2010), with the following recipes; composite flour that contain different proportion of OFSP flour and soft wheat flour (100g), Sugar (34g), shortening (28g), Baking powder (1.12g), Salt (0.93g), and 48 ml and various proportions of water until the required consistency of doughs were obtained (thc amount of water required for dough making was increased as more OFSP flour in the composite increased). Creaming (sugar + shortening + salt) was done in a laboratory mixer with a flat beater for 2 min at low speed (60 rpm) by scraping in the middle of the process then water was added to the cream and mixed for 5 min at high speed (130 rpm). In the second step of dough preparation, the composite flour which had been mixed well with baking powder was added to the premix, the formed cream, and mixed for 3 min at medium speed (90 rpm) by scraping in the middle of the process to obtain homogenous doughs. The doughs were rested for 10 min before shaping.

The doughs were then sheeted to a thickness of 4 mm with the help of a manual sheeting machine and cut with a cookie die of 35 mm in diameter on the baking tray. The cookies were baked in pre-heated (50°c for 1 hour) baking oven. Then, the baking temperature was set at 160°c for 8 minutes, based on preliminary test; Sweet goods are baked at temperature zone of 160, 180 and 200°c (Manely 1998) and the spread ratio decreases with increasing temperature (Tesfaye et al., 2010; Tekle et al., 2009). The baked cookies were finally cooled for 30 minutes at room temperature as recommended by AACC (2000). The cookies were then packed in high density polyethylene bags and aluminum foil until analyzed.

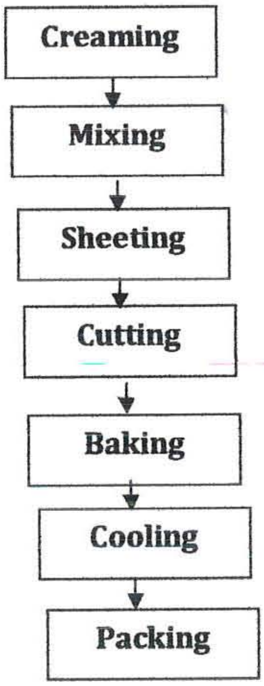


Fig 3.5.1 Cookie Processing flow Diagram (Sumnu and Sahin, 2008)

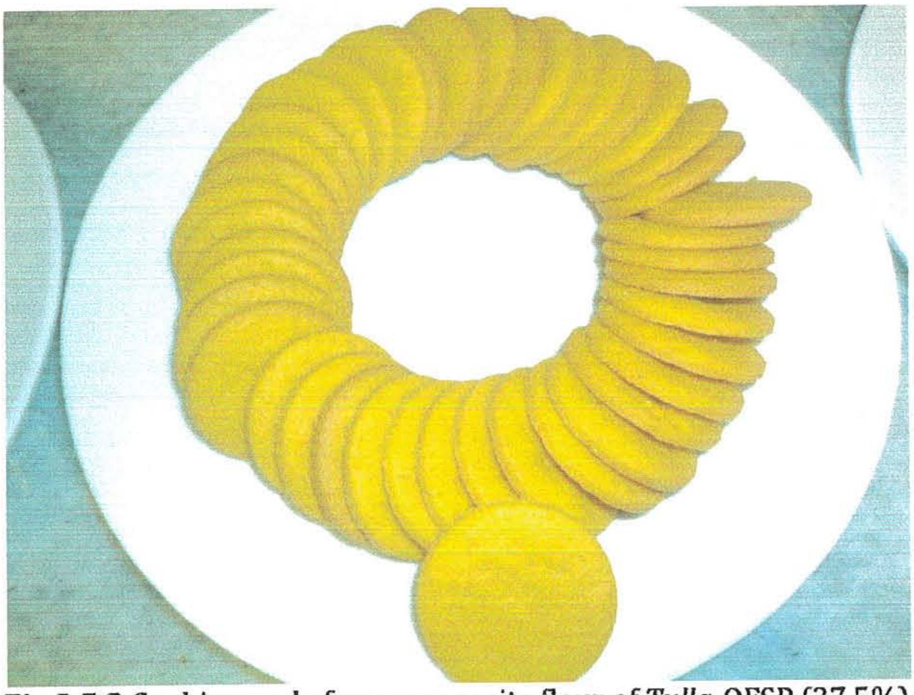


Fig 3.5.2 Cookies made from composite flour of *Tulla*-OFSP (37.5%) and SWF (62.5%)

3.6 Method of Analysis

3.6.1 Sampling and sample preparation

The sampling and sample preparation methods for determination of functional properties and nutritional value of the flours, physical and sensory properties of cookies, and nutritional value of cookies, and β-carotene and dry matter content of the fresh OFSP roots are described below.

- **Determination of functional properties of composite flours**

Representative sample for determination of functional properties of composite flour was prepared according to AOAC (2000, 925.08) and the method used by Emanuel et al., 2008. Both components of the composite flours, each cultivars (*Tulla and Kulfo*) of orange-fleshed sweet potato flours obtained through the method described in section 3.2 and soft wheat flour after purchased, were sieved to pass sieve size having circular opening of 100μm in diameter, mixed thoroughly, packed in polyethylene bag, tagged as SWF, *Kulfo*-OFSPF and *Tulla*-OFSPF for soft wheat flour, orange-fleshed sweet potato flour from *Kulfo* cultivar and Tulla cultivar respectively and then stored at $4^{0}c$ in simple fridge. For analysis the required amount of flours from each tagged samples was taken randomly, weighed using the same measuring apparatus. For determination of water absorption capacity (WAC) of composite flours, the weighed samples were mixed together, based on their ratio in the blend, and homogenized. Finally, the wheat-OFSPF (*Tulla*) composite flours (CF) were sealed in polyethylene bags and tagged as TCF1, TCF2, TCF3, TCF4 and TCF5 whereas and the wheat-OFSPF (*Kulfo*) composite flours (CF) were tagged KCF1, KCF2, KCF3, KCF4 and KCF5 for Kulfo cultivar; the composite flours were then stored at room temperature. TCF1 and KCF1 were similar samples; the control. All the above different composite flours were used for analysis in duplicates.

- **Determination of physical and sensory properties of Cookies**

The baked cookies were cooled for 30 minutes at room temperature (AACC 2000). The cookies retained the normal shape was subjected to physical measurements and to panelists for sensory evaluation.

❖ Determination of nutritional value of cookies and flours

Representative sample for determination of β-carotene content, proximate composition and mineral content of OFSP flour and cookies were prepared according to AOAC (2000, 926.04) and Rodriguez-Amaya and Kimura (2004). The entire representative cookie samples were milled to pass sieve size of 100μm diameter, mixed thoroughly, and kept in polyethylene bag and stored at 4^0c until analyzed. Similarly, the flour samples were also sieved through sieve having circular opening of 100μm in diameter, mixed thoroughly, and kept in polyethylene bag and stored at 4^0c until analyzed. All samples were tagged before storage. These operations were carried out under low light to prevent isomerization or photo oxidation of carotenoids.

❖ Dry matter determination of fresh OFSP roots

Sampling and sample preparation to determine dry matter content of fresh OFSP roots was done according to AOAC (2000, 922.01) and Bechoff (2010a). For each cultivars of OFSP, five average-sized roots were randomly selected and the whole of the roots were combined. Then, all the adhered soils from the roots were removed thoroughly by washing; then the wetted roots were dried with absorbent paper after washing and the roots were then cut in to slices and thoroughly mixed to get homogeneous representative sample. Finally, the slices were put in drying oven to determine dry matter content of fresh tubers.

❖ Beta-carotene determination of fresh OFSP roots

Representative sample for determination of β-carotene content of fresh OFSP was prepared according to Rodriguez-Amaya and Kimura (2004) and Bechoff (2010). For each cultivars of OFSP, five average-sized roots were randomly selected from different parts of the entire lot; random sampling is very important because beta-carotene content is heterogeneously distributed in sweet potato roots from the same batch and cultivar (Bechoff et al., 2010). Then, the roots were washed, peeled, sliced and mixed together. The mixed slices were blended to a fine pulp using blender. The sample was thoroughly mixed, packed in to plastic bag and aluminum foil and stored at 4^oc until analyzed. These operations were carried out under low light to prevent isomerization or photo oxidation of carotenoids.

3.6.2 Determination of functional properties of composite flours

- **Moisture content determination**

Metal dishes, with their covers, previously used for the moisture determination were washed with water and dried at 130⁰C for 1 hr to a constant weight in drying oven provided with a motor fan (model DHG-9055A). The dishes were uncovered during drying in the oven but at the end of drying, immediately before transferring to desiccator. Then the dishes provided with cover placed in the desiccator were cooled for 30 minutes to room temperature. Using digital balance, the weight of each covered dishes were weighed (W1) after tarring and about 2 g of sample was weighed (W2) in to each of the dishes. The dishes with loosen cover together with the sample taken were then transferred into an oven set at 130°C for 6 hrs to dry until a constant weight reading was achieved. At the end of the drying, the dishes (covered while it was in the oven) plus sample were removed from the oven, transferred to desiccator, cooled and the mass was weighed soon after (W3). Loss in weight was reported as moisture content (AOAC 2000, 925.10). The moisture content was calculated from the subsequent formula.

$$\text{Moisture (\%)} = \frac{W_4 - W_3}{W_2} \times 100\%$$

Where,
W_1 = Weight of empty dish
W_2 = Weight of sample before drying
W_3 = Weight of dish and sample after drying
W_4= Weight of dish and sample together before drying

- **Bulk density (BD)**

Bulk density (packed bulk density) was determined with the method reported by Adeleke and Odedeji (2010). A mass of 50 g of the sample was put in to a 100 ml dry and clean measuring cylinder. The cylinder was tapped on a laboratory bench continuously until a constant volume was obtained. Then the volume of sample was recorded. The bulk density was calculated as weight of the grounded flour (g) divided by its volume (ml).

$$\text{Bulk Density (g/ml)} = \frac{\text{Weight of Sample (g)}}{\text{Volume of sample after tapping (ml)}}$$

3.6.2.3 Oil absorption capacity (OAC)

The method of Sosulki et al., 1976 was used with some modification as cited by Emanuel et al., 2010 to determine oil absorption capacity. Instead of water being used, 10 ml (v_1) of refined corn oil with density of 0.92 g/ml was added to one gram of flour in a 25 ml centrifuge tube. The content of the centrifuge tube was stirred for 5 min and then centrifuged at 4000rmp for 20 min. The amount of oil separated as supernatant was decanted and measured using 10 ml cylinder (v_2). The difference in volume was taken as the oil absorbed by the sample and was expressed as ml of oil bound by 1 g dried flour.

Oil Absorption Capacity (ml/g) = (V1 – V2)* 100

3.6.2.4 Bulk density (BD)

Bulk density (packed bulk density) was determined with the method reported by Adeleke and Odedeji (2010). A mass of 50 g of the sample was put in to a 100 ml dry and clean measuring cylinder. The cylinder was tapped on a laboratory bench continuously until a constant volume was obtained. Then the volume of sample was recorded. The bulk density was calculated as weight of the grounded flour (g) divided by its volume (ml). Dispersibility of flour in water was determined by the method of Kulkarni et al., 1991 as cited by Ikpeme et al., 2010.

$$\text{Bulk Density (g/ml)} = \frac{\text{Weight of Sample (g)}}{\text{Volume of sample after tapping (ml)}}$$

3.6.2.5 Dispersibility

Dispersibility of flour in water was determined by the method of Kulkarni et al., 1991 as cited by Emanuel et al., 2010. About 10g of each flour sample was weighed into a 100 ml measuring cylinder. Distilled water was added up to 100 ml volume. The sample was vigorously stirred and mixed and allowed to settle for 3h. The volume of settled particles was recorded and subtracted from 100 to give a difference that was taken as percentage dispersibility.

3.6.3 Determination of Physical properties of cookies

Cookies containing different proportion of wheat flour and OFSP flour from two different cultivars were analyzed for Diameter (mm), Thickness (mm) and Spread factor after the cookies were allowed to cool for 30 min according to the method described in AACC (2000).

- **Diameter (D)**

Six cookies were placed edge to edge and total diameter was measured in mm by using a ruler. The cookies were rotated at an angle of 90° for duplicate reading. This act was repeated twice and average diameter was reported in mm.

- **Thickness (T)**

Six Cookies were stacked on top of each other and total height was measured in mm with ruler. This practice was repeated twice to get an average value and results were recorded in mm as thickness.

- **Spread Factor (SF)**

SF was determined from D and T by employing the subsequent formula

$$SF = D/T$$

3.6.4 Sensory Evaluation of cookies

Various sensory attributes of nine cookies including Color, Crispiness, Taste, Flavor and Overall Acceptability were carried out by a panel of 16 members comprising staff and students from food science and nutrition department (AAU), Department of chemical engineering (AAU) and Ethiopia Health and Nutrition Research Institute (EHNRI) using nine-point category scales ranging from like extremely, through neither like nor dislike, to dislike extremely (Appendix-I) as described by Meilgaard et al., 2007. Samples were evaluated on a desk placed in the air-conditioned laboratory, which provided a quiet and comfortable environment (Singh et al., 2008).

Samples were served on identical white plastic tray coded with 3-digit random number; the numbers were different for each sample. The coded samples were presented in random order for each panelist simultaneously to administer and allow panelists to re-evaluate the samples if desired and make comparisons between the samples. Panelists were asked to indicate their degree of liking for each attributes of the coded samples by choosing the appropriate category. Panelists were given distilled water for rinsing to neutralize their mouth between the samples. Covered expectoration cups were also provided when panelists didn't wish to swallow the samples (Watts et al., 1989)

3.6.5 Determination of Nutritional Value

Proximate analysis (moisture, ash, crude protein, crude fat, crude fiber and carbohydrate), mineral analysis (P, Ca, and Fe) and β-carotene analysis for the flours and cookies were done at Food science and nutrition laboratory of AAU and EHNRI. Dry matter content and β-carotene content of the fresh OFSP tubers (two cultivars) were also determined.

3.6.5.1Proximate Analysis

- **Total Solids determination (Air-oven Method)**

Metal dishes, with their covers, used for the moisture determination were washed with water and dried at 130°C to a constant weight (1 hr) in drying oven provided with a motor fan (model DHG-9055A). The dishes were uncovered during drying in the oven but at the end of drying, immediately before transferring to desiccator. Then the dishes provided with cover placed in the desiccator were cooled until room temperature (30 minutes). Using digital balance, the weight of each covered dishes was weighed (W1) after tarring and about 2 g of sample was weighed (W2) in to each of the dishes, after tarring the weight of the covered dishes. The dishes with loosen cover together with the sample taken were then transferred into an oven set at 130°C to dry until a constant weight reading was achieved (6hrs). At the end of the drying, the dishes (covered while it was in the oven) plus sample were removed from the oven, transferred to desiccator, cooled and the mass was weighed soon after (W3). Weight of flour residue was reported as total solids (AOAC 2000, 925.10).

$$\textbf{Total solids (\%)} = \frac{W_3 - W_1}{W_2 - W_1} \times 100\%$$

Where,
W_1 = Weight of empty dish
W_2 = Weight of dish and sample before drying
W_3 = Weight of dish and sample after drying

- **Ash content determination (Direct Method)**

Porcelain Crucibles were washed, with water and HCl, and dried at 130°C (30 minutes) in drying oven provided with a motor fan (model DHG-9055A). The crucibles were then ignited at 550°C in a furnace to a constant weight (2 hours), placed in to the desiccators (1 hour) to cool in to room temperature and their mass were weighed (W1) soon after reaching room temperature using digital balance. About 3 g of each samples was measured (W2) and transferred into the crucibles. The crucibles with the sample were placed on the hot plate at low temperature, to char the sample until it turned black. Then the charred samples were placed in the furnace at 550°C until light gray ash was obtained (6 hours). Then the crucibles were cooled (1 hour) and finally the ash and the crucibles were measured together (W3) (AOAC 2000, 923.03).

The amount of ash was calculated from the following formula.

$$\textbf{Ash (\%)} = \frac{(W_3 - W_1)}{(W_2 - W_1)} * 100$$

Where,
W_1 = Mass of empty Crucible
W_2 = Mass of Crucible and sample before ash
W_3 = Mass of Crucible and ash

- **Crude Fat content determination (Soxhlet-Ether extract Method)**

The cleaned aluminum cups, used for the extraction of fat, with boiling chips were placed in drying oven at 130°c for 1 hr and cooled to room temperature in a desiccator (30 minutes). Then about 2 g of samples were measured (w_1) in to an extraction thimble, with porosity permitting rapid passage of ether, lined with fat free cotton. Then 50 ml of diethyl ether was added to in to each weighed aluminum cups with boiling chips (w_2). Then the thimbles with the samples were attached to soxhlet extraction apparatus and set up of the extraction apparatus was done. The samples contained in the thimbles were soaked at 55°c by lifting down the thimble in to the cup for 3 hrs which was started when the hot plate temperature reached at 55°c. After soaking the thimbles were lifted up and extraction process was taken place for 4 hrs. Then the recycling process made by the diethyl ether was stopped to let the solvent to evaporate from the aluminum cup with the extract, in the process the evaporated solvent was recovered in the apparatus. Then aluminum cup and the content were dried in the oven for 30 min at 100°C to evaporate the remaining solvent in the cup. Finally, the aluminum cups and the content were dried in the oven for 30 min at 100°C to evaporate the remaining solvent in the cup, cooled in desiccator for 30 minutes, and weighed (w_3) using digital balance AOAC 2000, 920.39).

$$\textbf{Fat (\%)} = \frac{W_3 - W_2}{W_1} * 100$$

Where
W1=Weight of sample
W2=weight of cup and it's content before extraction
W3=weight of cup and it's content after extraction

- **Crude protein content determination (Kjeldahl method)**

Crude protein content was determined based on the method described by AOAC (2000, 955.04). About 0.5 g of sample was weighed (W), using digital balance, in to cleaned digestion flask. Then 6 ml of concentrated sulphuric acid and 3.5 ml of 30% hydrogen peroxide solution, as oxidizing agent, were added in to the digestion flask. The tubes were shaked until the violent reaction disappeared. Then about 3g of the catalyst mixture made of 10 g of $CuSo_4$, to facilitate digestion, and 150 g of K_2So_4 potassium sulphate, to increase boiling point of H_2So_4, were added in to the digestion flask.

The same procedure was followed for blank sample too. Then, the mixtures were digested in the digester at a temperature of 370^{0}C for 4 hrs and allowed to cool in the digester for 30 min. Therefore, digestion was the first reaction that occurred during protein determination that converts any nitrogen in the sample in to ammonia and other organic matter to CO_2 and H_2O. In acidic solution, ammonia is not librated as gas because it exists as an ammonium sulfate salt.

N (in a food) + H_2SO_4 $\longrightarrow$ $(NH_4)_2SO_4$

After digestion was completed, distilled water (25ml) and 40% NaOH (25ml) were added to the digested sample in order to neutralize the acid and to make the solution slightly alkaline.

$(NH_4)_2SO_4$+2NaOH $\longrightarrow$ $2NH_3$ + $2H_2O$+Na_2SO_4

Distillation was the next step followed by titration. Therefore, the ammonia was distilled in to receiving conical flask that consist solution of 25 ml H_3BO_3 (4%), 25 ml distilled H_2O and indicator (methyl red). Distillation was stopped when the volume of liquid reached 250 ml.

NH_3 + H_3BO_3 (boric acid) $\longrightarrow$ NH_4^+ + $H_2BO_3^-$ (borate ion)

Then the borate ion was directly titrated with standard acid (0.1N HCl) and the volume of consumed HCl at the end of titration (color change) was recorded for all samples including blank sample.

$H_2BO_3^-$ + H^+ $\longrightarrow$ H_3BO_3

The nitrogen content was calculated from the following equation

$$\text{Nitrogen (\%)} = \frac{V * N * 14}{W} * 100$$

Where:
V (ml) = volume of HCl consumed to the end point of titration
N (0.1) = the normality of HCl used often is about 0.1N
14= the atomic weight of nitrogen
W (g) =Sample weight

The protein content was calculated from the following equation

% Protein = %N * F

Where, F is the conversion factor which is 5.7

❖ Crude fiber content determination

Crude fiber analysis was conducted using the method of AOAC (2000, 962.09)). About 2 g of samples were measured and transferred analytically in to the 600ml beaker. Then 200ml of H_2SO_4 (1.25%) was added in to the beaker the level of the volume was marked on the beaker, then it was boiled for 30min on the hot plate by stirring periodically. During boiling the level was kept constant by addition of hot distilled water.. After exactly 30 mints of boiling 20ml of 28% KOH was added with occasional stirring using glass rod for additional 30min of boiling. The level of the mixture was still kept constant by adding hot distilled water. After boiling was completed the solution was poured in to wetted sintered glass crucible which is filled with 10 mm sand and the, mixture was filtered by turning the vacuum pump on. The beaker wall was rinsed several times with hot distilled water and transfer to crucible. Then the residue in crucible was washed, with hot distilled water , and H_2SO_4(1%),and again with hot distilled water, then with NaOH (1%), and again with hot water, then with 1%H_2SO_4 , then with hot water, filtration was there in every washing process. Finally it was washed using water free acetone. The solution in each beaker was then filtered through crucibles containing sand by placing each of them on Buchner funnel fitted with No.9 rubber stopper. During filtration the sample was washed with hot distilled water. The final residue was washed with 1% sulphuric acid solution, hot distilled water, 1% sodium hydroxide solution and finally with acetone. Each of the crucibles with their contents was dried for 2 hr at 130 0C and cooled in desiccators and weighed (M1). Then again they were ashed for 30 min at 550 0C in furnace and were cooled in desiccators. Finally the mass of each crucible was weighed (M2).The washed crucible with the digested sample was dried for 2hr in the oven at temperature of 130^{0}C. Then it was cooled in the desiccators for 30 min then it was weighed immediately after taking out from the desiccators and the mass was taken as W_1(crucible + fiber + ash). After that the crucible was transferred to the furnace for 30min at a temperature of about 550^{0}C for ashing then it was cooled in the desiccators and weighed, the mass was taken as W_2 (crucible + ash).

$$\textbf{Crude fiber (\%)} = \frac{W_1 - W_2}{\text{Wt of sample}} * 100$$

Where, W_1 crucible wt after drying

W_2 crucible weight after ashing

❖ Carbohydrate content determination (By difference)

The carbohydrate content of cookies was determined by difference method; by subtracting the sum of other components of proximate composition from 100.

3.6.5.2 Energy value determination

Energy values were calculated applying Atwater factors 4, 9 and 4 for each gram of crude protein, crude fat and carbohydrate respectively (Shrestha and Noomhorm, 2002).

Energy value = protein*4+ carbohydrate*4+ fat*9

3.6.5.3 Beta-carotene Analysis

Beta-carotene extraction and analysis were based on the methods described by Rodriguez-Amaya and Kimura (2004) under low light conditions. Extraction, partition, and open column chromatography (OCC) were carried out under a hood to avoid inhalation of solvent vapor by the analyst.

Extraction with acetone using a mortar and pestle

About 2 g of the homogenous representative sample of fresh tissue or about 2-5 g of flour samples was weighed (W) in dry and clean beaker. Sample weight varied according to the expected beta-carotene content in solution; the larger analytical sample, the sample weighed and subjected to extraction, was for samples expected to have small beta-carotene content. Besides, flour samples were rehydrated in deionized water and allowed to stand for 20 minutes at room temperature in order to allow efficient penetration of the extraction of solvent into the tissues; it is difficult to extract from dry flour. Acetone was the chemical used in this extraction method; it is inexpensive and readily available, and penetrates food tissues well. The weighed sample was then transferred in to a mortar to be blended and macerated with 50 ml cold acetone (acetone refrigerated for about 2 hours) with a pestle. The extract was filtered through a filter paper (whatman 42,125mm) in to 100ml conical flask. After filtration the residue was returned in to the mortar and was macerated again with fresh cold acetone and filtered as before, this process was repeated (three to four times) until the extract was devoid of color. During the extraction process, the mortar, the pestle, the filter paper and the residue were washed with small amounts of acetone. All the washings (rinsing) and extracts were collected and received in the same conical flask through the filter paper. Hence, extraction and filtration were repeated until the residue was colorless.

Partition to petroleum ether (PE)

The combined acetone extracts of the samples were poured in to a 500ml-separatory funnel with Teflon stop-cock where 40 ml of petroleum ether (PE) with boiling point of 40-60°c (to avoid prolonged heating), had been added. The volume of the petroleum ether was marked on the separatory funnel. 300 ml of distilled water was added, without shaking, by letting it flow along the walls of the funnel to avoid formation of an emulsion. The mixtures were left for some time until the two phases, the acetone and the petroleum ether phase, were separated and the lower aqueous phase was discarded; colored materials left the acetone phase and join the petroleum ether phase. Then the mixture was washed with distilled water to remove residual acetone in the mixture. This step was done repeatedly (3-4 times) with 200 ml distilled water during each time until the lower phase was completely discarded without discarding any of the upper phases as much as possible. The upper PE phase was collected in 50 ml volumetric flask and then separatory funnel was washed with PE followed by collecting the washings in the same volumetric flask by passing through the funnel.

Concentration of Petroleum ether extract or evaporation of solvent

The collected petroleum ether phase was transferred to drying flask and was evaporated to dryness on a rotary evaporator at 40^0C and pressure of 1 atmosphere; during rotation, the petroleum ether extract was concentrated. Then, the residue in the round bottom drying flask was dissolved with small petroleum ether (1ml) to introduce the concentrated PE extract in to petroleum ether rinsed silica gel open chromatographic column (OCC) Using Pasteur pipette.

Open column chromatographic separation

The introduced residue in to the open column chromatography (OCC) was eluted with petroleum ether and the β-carotene which passed through column as a yellow pigment was collected in a measuring cylinder and the volume was recorded (V) until no color is eluted through the chromatogram. The residue that passed the OCC was mixed well and put in cuvettes.

Quantification of β-carotene Spectrophotometrically

Absorbance (A) of the residue in the cuvette was read using UV-visible spectrophotometer at 450 nm (total absorption). The absorbance was between 0.2 and 0.8. Therefore, dilution of beta-carotene concentration was not necessary.

Calculate the β-carotene content using the subsequent formula:

$$\text{Amount of } \beta\text{-carotene content } (\mu g/g) = \frac{A*V*10^4}{A_{1cm}^{1\%} *W}$$

Where,
A = Absorbance
V= Volume (in ml) of β-carotene which goes as yellow pigment through column.
$A_{1cm}^{1\%}$ = Absorbance at a given wavelength of a 1% solution in 1 cm light-path Spectrophotometer cuvette
Absorption coefficient of β-carotene in PE is 2592.
W = Weight (in g) of sample

Conversion of β-carotene in to RAE

According to Haskell and others (2004), 1 retinol activity equivalent (RAE) = 1 μg of retinol= 13 μg of β-carotene from OFSP.

3.6.5.4 Mineral analysis

Calcium, Phosphorous and Iron content of flours and cookies were determined by Spectrophotometric technique from the ash obtained through dry ashing.

Calcium and Iron analysis

About 1g of ash was treated with 5 ml of 6N HCl to wet it completely and carefully dried on a low temperature hot plate to dryness. Seven point five ml of 3N HCl was added on each crucible and placed on the hot plate until the solution just boiled. The solution was cooled to room temperature and filtered through a filter paper (whatman 42,125mm) into 50ml volumetric flask. Again 5ml of 3N HCl was added to the crucible to dissolve the residue and it was heated until the solution just boiled. Then the solution was cooled and filtered into the previous 50ml volumetric flask. The crucible was washed with de ionized water three times and filtered and combined the washings into the 50ml volumetric flask. Then 2.5ml of lanthanum chloride solution was added in 50ml volumetric flask which contained the filtrate and the flask was filled with de ionized water up to the mark. Blank was prepared in parallel with the sample dissolving process using the same amount of reagents which were used for the sample. The sample solutions were transferred to polyethylene bottle.

Standard solutions: Five series of working standard for Fe and Ca were prepared from 1000ppm metals stock solutions with de ionized water containing 2.4 ml 3N HCl and 0.5ml lanthanum chloride in 10 ml volumetric flask used to make the sample and standard matrix similar. Iron, Zinc and Calcium Hallo cathode lamps were used as a radiation source for each elements and Air acetylene gas mixture was used as source of flame. After Flame Atomic Absorption Spectrophotometer (Varian SpectrAA-20 Plus, Varian Australia Pty., Ltd., Australia) turned on and optimized for each elements one after the other, maximum absorbance was obtained by adjusting the hallo Cathode lamps at specific slit width and wave lengths. Before the analysis of sample calibration graph (concentration versus absorbance) for each element using the standard solutions was prepared, then first Sample blank solution was run followed by the sample solutions. After each solution of standard or sample run the system was rinsed by aspirating de ionized water to avoid contamination. The measured samples blank concentration was deducted from that of the sample concentration. Duplicates of each sample were measured, and the concentrations were calculated using the average of each value.

Calculation:

Metal content (mg/100g) = [(A-B) x V]/ (10 *W)

Where,

W= Weight of sample in gram

V=Extract volume (50ml)

A = Concentration (μg/ml) of sample solution;

B = Concentration (μg/ml) of blank solution

Phosphorus determination

The sample solutions prepared for mineral determination was used for phosphorous determination. One ml of the clear extract (sample solution prepared for mineral determination) was diluted into 100 ml with deionized water. Five ml of the sample solution was added into test tubes. Exactly 0.5 ml of molybdate and 0.2 ml aminonaphtholsulphonic acid were added into the test tubes (sample solution) and mixed thoroughly step by step. The solution was allowed to stand for 10 minutes. Six series of working standard phosphorous solutions (0- 1µg/ml) were prepared by appropriate dilution of the phosphorous stock solution (1000 µg P/ml of KH2PO4) with deionized water using 10 ml volumetric flask. After manipulating the instrument operation procedure, the absorbance (A) the sample solution was measured at 660 nm against distilled water using UV-VIS spectrophotometer. The standard and sample blank solution was run with the sample. Calibration graph (concentration versus absorbance) prepared and used to quantify the amount of phosphorous in the sample.

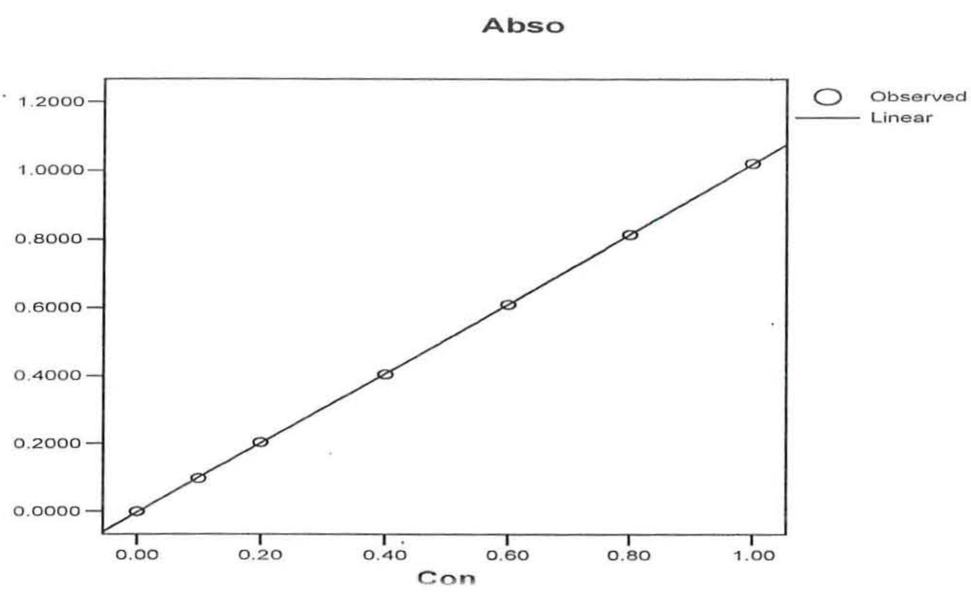
$$\text{Phosphorus in mg/100gm} = \frac{(A_S - A_B) \times \text{dilution factor} \times \text{extracted volume} \times 100}{\text{Slope} \times \text{weight of sample} \times 1000}$$

Where,

A_S= absorbance of sample

A_B= absorbance of blank

Slope= obtained from the calibration curve



Slope=1.0217, Intercept = -0.0022 and R^2=0.9999

Fig 3.6 A typical standard curve for phosphorous determination

3.7 Experimental Design and Data Analysis

3.7.1 Experimental Design

Blending ratio (Br) (SWF:OFSPF)	Cultivar (C)	
	C_1 (Tulla-OFSP)	C_2(Kulfo-OFSP)
Br_1 (SWF 100:0 **OFSPF)**	Br_1 C_1	Br_1 C_2
Br_2 (SWF 87.5:12.5 **OFSPF)**	Br_2 C_1	Br_2 C_2
Br_3 (SWF 75:25 **OFSPF)**	Br_3 C_1	Br_3 C_2
Br_4 (SWF 62.5:37.5 **OFSPF)**	Br_4 C_1	Br_4 C_2
Br_5 (SWF 50:50 **OFSPF)**	Br_5 C_1	Br_5 C_2

Table 3.7 Experimental design on the effect of blending ratio and cultivar on the Quality parameters of cookies developed from soft wheat flour (SWF) and Two cultivars of orange-fleshed sweetpotato (OFSP)

3.7.2 Data analysis

The results obtained for sensory analysis of cookie samples in the form of descriptive categories were converted to numerical scores from 1 to 9. The numerical scores obtained were subjected to statistical analysis and it was found that panelist had non-significant effect on the mean value of the results. The numerical scores obtained for all analysis of cookie samples were subjected to statistical analysis using statistical package SPSS, version 17. Since there were two factors; namely cultivar (C) and blending ratio (BR), their effects on the quality of cookies were analyzed with two-way ANOVA. When p values were significant ($p < 0.05$), the means of each parameter were compared using the Duncan's multiple range test (DMRt). Finally, the suggested models for each selected response were applied to predict the optimum formulation of the composite flour.

3.8 Optimization of composite flour formulation

A constrained, simplex lattice design was used to optimize the formulation of composite flour using Design expert soft ware (version 8.0.5). The design comprised 5 runs and the mean values of the runs were subjected for analysis. The selected responses were cookies' spread, energy value, beta-carotene content and overall acceptability by desiring cookies that fulfill at least 50% of RDA of pre-school children with attaining maximum overall acceptability, maximum energy and maximum spread.

4. Results and Discussion

4.1 Functional properties of composite flours

Functionality of the composite flour is the combination of functional properties of flours in regard to their proportion. Functional properties of flours depend on the type of raw materials and methods of processing which ultimately determines product quality and process effectiveness (Brennan, 2006). Thus, incorporation of OFSP flour in to soft wheat flour is expected to produce effect in the functional properties of the blended samples. The functional properties determine the application and use of food materials for various food products (Adeleke and Odedeji 2010). Hence, some of the functional properties of the flours such as moisture content, bulk density, water absorption capacity, oil absorption capacity, and dispersibility of the flours were analyzed and the average results are presented in Table 4.1.

Table 4.1 Functional properties of soft wheat flour and OFSP flours

Flours	Moisture (g/100g)	WAC (g/g)	OAC (ml/g)	BD (g/ml)	Dispersibility
SWF	14.628±0.05[c]	0.666±0.00[a]	0.961±0.00[b]	0.752±0.00[a]	21.000±0.00[a]
Tulla-**OFSPF**	6.054±0.04[a]	4.939±0.00[b]	0.954±0.00[b]	1.001±0.01[b]	88.000±0.00[b]
Kulfo-**OFSPF**	6.782±0.01[b]	5.598±0.00[c]	0.782±0.00[a]	1.094±0.00[c]	100.00±0.00[c]

All values are the means of duplicates ± standard deviation

For each column, mean values with the same lowercase superscript are not significantly different at the 0.05 level

4.1.1 Moisture

Flour moisture is typically an indication of flour quality where storage is concerned, since water can accelerate chemical or microbiological deterioration and has an impact on functionality in specific products (Van Hall, 2000). The moisture content of food products goes a long way in suggesting the shelf life of the product. The moisture content of SWF, *Tulla*-OFSPF and *Kulfo*-OFSPF were 14.628%, 6.054% and 6.782% respectively. The mean values obtained for the moisture contents of OFSP cultivars, 6.05% for *Tulla* and 6.78% for *Kulfo*, were in the range of moisture content of sweetpotato flour reviewed by Van Hall (2000) which is between 4.4 and 13.2%. Even though, there was a significant difference in moisture content among the three flours (Table 4.1), all the mean values of moisture content of the flours and the blended sample fall within the acceptable limit of dry products that is less than or equal to 15% (Adeleke and Odedeji 2010). The significant difference in moisture content between the two cultivars OFSP flours could be directly related to the initial moisture content of fresh-roots (Van Hall 2000); which was 75.86 and 78.01% for *Tulla* and *Kulfo* respectively.

The mean moisture content of SWF, 14.63%, was above the basis moisture content of soft wheat flour described by Sumnu and Sahin (2008) which is specified as 14%. The 1.05% increase in moisture content of the SWF might be due to the initial moisture content of the wheat kernel or storage conditions of the flour before laboratory analysis. The flours with low moisture content could have long shelf life (Van Hall, 2000). Hence, both cultivars of OFSP flours tend to have long shelf life compared to soft wheat flour.

4.1.2 Water absorption capacity (WAC)

Water absorption capacity is an important functional property required in food formulations especially those involving dough handling, since an increase in water absorption lead to the weakened dough and decrease dough development and extendibility (Ojinnaka 2009). Water absorption capacity is also strongly correlated to cookie spread; the higher water absorption capacities could have contributed to the lower spread ratio because rapid partitioning of free water to hydrophilic sites during mixing increased dough viscosity, thereby limiting cookie spread (Manoela et al., 2006). The mean values for water absorption capacity of OFSP flours were 4.939 g/g for Tulla cultivar and 5.598 g/g for Kulfo cultivar while that of soft wheat flour was 0.666 g/g (Table 4.1) indicating that both cultivars of OFSP flours have higher water absorption capacity than SWF by 648.5% (*Tulla*) and 748.5% (*Kulfo*). Therefore, OFSP flours have higher affinity for water which was informed by their lower moisture content, 6.04% and 6.78% for *Tulla* and *Kulfo* respectively. This high percentage of water absorption could be because of more fiber content in the OFSP flour than wheat flour. The differences in water absorption is mainly caused by the greater number of hydroxyl group which exist in the fiber structure and allow more water interaction through hydrogen bonding as described by Nasser et al., 2008. Besides, processing like blanching could have effect on WAC as observed by Emmanuel et al., 2010. According to their study taro flour from blanched samples had high WAC value than the unblanched samples and the given reason was protein subunits dissociates on heating and the dissociating of the protein subunits during the blanching regimen increase the number of hydrophilic groups which are the primary sites of water binding of protein. Similarly, the OFSP slices were blanched during processing which could have effect on water absorption capacity. Therefore, the high water absorption capacity of OFSP flour could be due to the method of processing (blanching) or/and the high fiber content of OFSP flour than wheat flour (Table 4.4.1). Generally, the mean values for WAC of the flours were significantly different; suggests that addition of OFSP flour to wheat flour could affect the water absorption capacity of the composite flour. The water absorption capacity of the blended samples was on increase as more and more OFSP flour was added to SWF (Figure 4.1). The WAC values for the blends ranged from 0.66 to 2.80 g/g (*Tulla* cultivar) and from 0.66 to 3.13 g/g (*Kulfo* cultivar) and there was a significant difference ($p < 0.05$) in the WAC of the blended samples as a function of blending ratio, cultivar and their interaction (Appendix II).

The blend samples from *Kulfo*-OFSPF and SWF had higher WAC compared to the blend samples from *Tulla*-OFSPF and SWF. Composite flour that contains 50% OFSP flour in the composite flour had the highest score for WAC value. This could be due to high fiber content of OFSP flour, particularly the Kulfo cultivar. A similar trend was also observed by Tesfaye et al., 2011. In their study of partial substitution of wheat flour with carrot and QPM flour, as the proportion of carrot flour in the blend increased, the water absorption capacity was increased and this was related to the higher fiber content of the carrot flour compared to wheat and QPM flour.

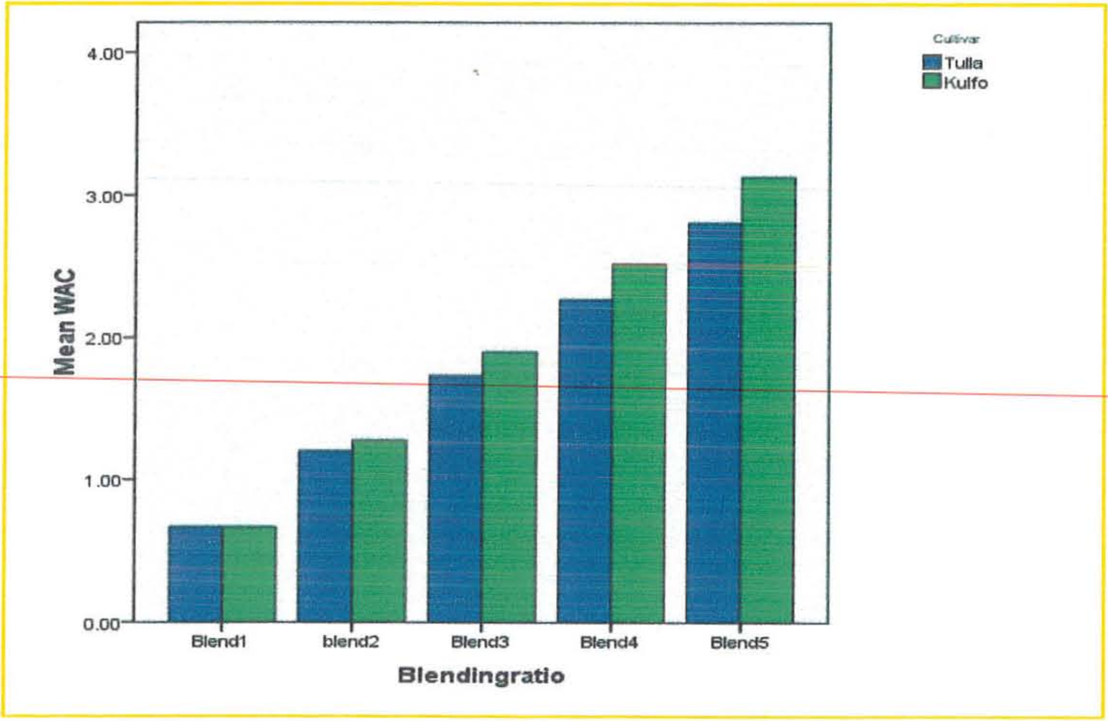


Fig 4.1 Water absorption capacity of SWF-OFSP composite flours

4.1.3 Oil Absorption Capacity (OAC)

Oil absorption capacity or fat absorption capacity is the ability of the flour protein to physically bind fat by capillary attraction and it is of great importance, since fat acts as flavor retainer and also increases the mouth feel of foods, especially bread and other baked foods such as cookies (Emmanuel et al., 2010). The oil absorption capacity of flours was 0.961, 0.954 and 0.782 ml/g for SWF, *Tulla*-OFSPF and *Kulfo*-OFSPF respectively. The mean values for OAC of SWF was not significantly different from *Tulla*-OFSPF but from *Kulfo*-OFSPF. The variations in the presence of non-polar side chains of flour proteins, which might bind the hydrocarbon side chains of oil among the flour, explain differences in the oil binding capacity of the flour. The result justifies that *Tulla*-OFSPF and SWF could be a better flavor retainer than *Kulfo*-OFSPF.

4.1.4 Bulk Density (BD)

The bulk density (BD) is a reflection of the load that samples can carry if allowed to rest directly on one another and it is generally affected by the particle size and density of the flour. Therefore, it gives an indication of the relative volume of packaging material required; high bulk density is a good physical attribute as described by Emmanuel et al., (2010). The mean values obtained for bulk density was 0.75, 1.00 and 1.10 g/ml for SWF, *Tulla*-OFSPF and *Kulfo*-OFSPF respectively. The mean bulk density values were generally higher for the two cultivars of OFSP flours (100%) than that of 100% SWF (Table 4.1); all the mean values for BD were significantly different ($p < 0.05$). This might be due to the influence of molecular structure of OFSP starch. Higher bulk density is desirable for the greater ease of dispersibility, mixing, packaging and reduction of paste thickness (Udensi and Okoronkwo 2006). Therefore, OFSPF is very fine and better in terms of mixing and requires less packaging material compared to SWF.

4.1.5 Dispersibility

Dispersibility indicates the ability of flours to reconstitute by adding water. The dispersibility values for the three flours were 21%, 88% and 100% for SWF, *Tulla*-OFSPF and *Kulfo*-OFSPF respectively. The mean values obtained were significantly different; the highest dispersibility score was 100% for *Kulfo*-OFSPF and the lowest was that of 21% for SWF. With this respect, OFSP flours seem to have better dispersibility and reduced pasting thickness.

4.2 Physical properties of cookies

Physical parameters of cookies are dependent on gluten strength of flour and shortening used. Physical analysis of cookies is an important matter from consumer as well as bakers point of view. OFSP flour supplemented cookies along with control were analyzed for physical characteristics including diameter, thickness and spread ratio on the day of baking after 30 minutes of cooling. Varying levels of supplementation exhibited significant differences on the physical parameters of cookies. Mean values for aforementioned traits as a function of blending ratio and cultivar are presented in Table 4.2A and 4.2B.

Table 4.2.1 Effect of blending ratio on physical properties of cookies

Blending Ratio	Diameter (mm)	Thickness(mm)	Spread ratio
Br 1	37.667 ± 0.00^{d}	4.333 ± 0.00^{a}	8.693 ± 0.00^{b}
Br 2	37.584 ± 0.09^{c}	4.333 ± 0.00^{a}	8.683 ± 0.02^{b}
Br 3	37.000 ± 0.54^{b}	4.333 ± 0.00^{a}	8.538 ± 0.09^{a}
Br 4	36.250 ± 0.55^{a}	4.292 ± 0.06^{a}	8.428 ± 0.12^{a}
Br 5	35.333 ± 0.47^{a}	4.250 ± 0.08^{a}	8.421 ± 0.18^{a}

All values are the means of duplicates ± standard deviation
For each column, mean values with the same lowercase superscript are not significantly different at the 0.05 level

Table 4.2.2 Effect of cultivar on physical properties of cookies

Cultivar type	Diameter(mm)	Thickness(mm)	Spread ratio
Tulla-**OFSPF**	37.000±0.85[b]	4.317±0.06[a]	8.606±0.12[b]
Kulfo-**OFSPF**	36.533±1.07[a]	4.300±0.05[a]	8.499±0.19[a]

All values are the means of duplicates ± standard deviation
For each column, mean values with the same lowercase superscript are not significantly different at the 0.05 level

4.2.1 Diameter (D)/ Width (W)

The principal criterion for good cookie-making quality is the diameter increase during the baking process and a large cookie diameter is considered as superior (Sumnu and Sahin 2008). Diameter of cookies was significantly influenced ($p < 0.05$) by blending ratio and cultivar but not by their interaction (Appendix III). The mean values obtained for the cookies diameter supplemented with various level of OFSP flour were ranged from 37.667 of cookie from control (100% SWF) to 35.333 mm of cookie from BR5 (50% OFSP flour) which showed decreasing trend for diameter with increasing the proportion of OFSP flour in the composite flour (Table 4.2.1). Cultivar also had effect on the diameter of cookies; the highest score for diameter, 37.000 mm, was from *Tulla*-OFSP and decreased for *Kulfo*-OFSP, 36.533mm as shown in Table 4.4.2. This could be due to high WAC value of OFSP flour which in turn depends on high fiber content; OFSP had higher fiber content 7.823 g(Tulla) and 8.252 g (Kulfo) per 100 g than 2.180 g per 100 g of wheat (Table 4.2.2). A similar decreased in diameter was also reported for cookie prepared with Wheat and sweet potato composite flour (Singh et al., 2008), and wheat and residue from king palm processing (Manoela et al., 2006) in which both sweet potato flour and residue from king palm processing contains a higher fiber content than wheat, and as their proportion increased in the formulation a gradual decease in diameter was found. The other reason could be the reduction of gluten as result of SWF reduction in the composite flour, since gluten affects the extensibility of cookie dough while baking; the less SWF, the less gluten. Therefore, the decrease in cookie diameter could be attributed to increase of water absorption capacity of the composite flours which in turn depends on their fiber content; when WAC increases, the cookie dough become more viscous that limits the increase in diameter of the cookie when baked and/or due to reduction of SWF as OFSPF increased in the formulation, since the formulation of composite flour is non-orthogonal.

4.2.2 Thickness (T)/ Height (H)

The mean values for thickness of OFSP supplemented cookies showed non momentous decrease when the proportion of OFSP increase in the composite flour; ranged from 4.33 mm of control (Br_1) to 4.25 of Br_5 (Table 4.2.1) and cultivar had also insignificant effect on this parameter of cookies as shown in Table 4.2.2. Both blending ratio and cultivar had no significant effect ($p > 0.05$) on the thickness of cookie, so their interaction too (appendix III). The result was in agreement with Tesfaye et al., (2010) in which a similar non-significant decrease in the average thickness of the cookies was observed as the proportion of carrot and quality protein maize flour was increased in the formulation.

Singh et al., (2010) also reported the non-significant decreasing trend of cookie diameter while the proportion of sweet potato increased in composite flour. The non significant decrease could be due to the thin thickness of the cookies during sheeting; dough was sheeted to a thickness of 4mm, which is very limited surface area, unlike diameter of cookie (35mm) to get significant change of the cookies thickness while measuring.

4.2.3 Spread factor (SF)

Cookie spreads (flows) during baking until the point at which the viscosity suddenly increases. However, the degree of spread depends on the viscous property of the dough which is in turn influenced by the recipe, ingredients, procedures and conditions used in cookie production (Pareyt and Delcour, 2008). Spread factor is the ratio which depends upon the values of width (diameter) and thickness (height) and is one of the main factors in achieving a good quality product (Dogan, 2006). The spread was significantly affected ($p < 0.05$) by blending ratio, cultivar and their interaction (Appendix III). The spread was greater for cookies made from control (Br_1), 8.693, and decreased with increasing the proportion of OFSP flour, 8.421, for Br_5 as shown in Table 4.2.1. However, there were no significant differences in spread of cookies made from Br_1 and Br_2 and similarly among cookies made from Br_3, Br_4 and Br_5.

The data analysis has also shown that cookies made from *Tulla*-OFSP and SWF composite flour had the highest score for spread ratio, 8.606, than 8.499 of cookies made from *Kulfo*-OFSP and SWF composite flour. There are several views on the mechanisms by which the spread ratio of cookies is reduced when wheat flour is supplemented with non-wheat flours. It has been established that cookie spread is strongly correlated to the water absorption capacities of the flour (Vieira et al., 2007); the more WAC of the composite flours, the less spread of the cookies. The mean values obtained for spread factor of the cookies were in accordance with the above mentioned factor. As it can be seen from Table 4.1, the WAC of the OFSP flours, specifically Kulfo cultivar was higher than soft wheat flour. Therefore, addition of OFSP flour to wheat flour cookie production could affect the spread. But, the significant decreases in spread of cookies depend on the type of cultivar of OFSP and their ratio in the composite flour.

4.3 Sensory characteristics of Cookies

To the consumer, the most important quality attributes of a food are its sensory characteristics. These determine an individual's preference for specific food products, and a small difference between similar products can have a substantial influence on acceptability (Fellows 2000). Therefore, during new product development, conducting sensory evaluation is an important matter to measure, analyze and interpret reactions of humans to those characteristics of food products as they are perceived by the senses of sight, smell, taste, touch and hearing, ultimately to understand the acceptability of food products. Hence, the sensory characteristics such as color, crispiness, taste, flavor and overall acceptability of cookies made from OFSPF and SWF blend were evaluated and their mean values are listed in the Table 4.3.1.

Table 4.3.1 Effect of blending ratio on sensory characteristics of cookies

Blending ratio	Color	Crispiness	Taste	Flavor	Overall Acceptability
BR1	6.813±1.36[b]	8.500±0.51[e]	7.750±0.84[d]	8.000+0.51[d]	7.688+0.59[c]
BR2	7.686±0.93[bc]	7.844±0.45[d]	7.719±0.46[d]	7.844+0.52[d]	7.656+0.55[c]
BR3	8.031±0.57[d]	7.594±0.50[c]	7.375±0.66[c]	7.344+0.55[c]	7.500+0.67[c]
BR4	7.500±1.19[c]	6.813±0.59[b]	6.500±0.76[b]	6.375+0.98[b]	6.406+1.01[b]
BR5	6.375±1.44[a]	6.031±0.47[a]	5.250±0.76[a]	5.250+0.88[a]	5.219+0.83[a]

All values are the means of sixteen replication ± standard deviation
For each column, mean values with the same lowercase superscript are not significantly different

Table 4.3.2 Effect of Cultivar on sensory characteristics of cookies

Cultivar	Color	Crispiness	Taste	Flavor	Overall Acceptability
Tulla-**OFSP**	7.325±1.05[b]	7.438 ±0.94[b]	7.175 ±1.03[b]	7.250±1.03[b]	7.138±1.03[b]
Kulfo-**OFSP**	6.850 ±1.51[a]	7.275 ±1.04[a]	6.663 ±1.27[a]	6.675±1.39[a]	6.650±1.34[a]

All values are the means of sixteen replication ± standard deviation
For each column, mean values with the same lowercase superscript are not significantly different

4.3.1 Color

Color, perceived by the sense of sight, is an important parameter among various sensory attributes. It is the first of the organoleptic senses that a consumer experiences in any food. Color was found to be significantly affected ($p<0.05$) by blending ratio, cultivar and their interaction (Appendix IV). The acceptance for the color of cookie from BR3 was found superior scoring 8.031, while cookie from BR5 was the least in terms of color's acceptance scoring 6.375 than the rest of the cookies as shown in Table 4.3.1. It was also observed that color acceptance was significantly higher for cookies made from *Tulla*-OFSP and SWF composite flour scoring 7.325 than cookies made from *Kulfo*-OFSP and SWF composite flour scoring 6.850 (Table 4.3.2). There was an increasing trend in the acceptability of the cookies' color with an increase in the amount of OFSP flour in the blend until 25% but a decreasing trend after this point. This might be due to the beautiful light yellow color of the cookie imparted by OFSP flour apparently at low level in the composite flour and the decrease could be due to orange color and burned edge of the cookies. The result justify that replacement of wheat flour with OFSP flour up to 25% would improve the acceptability of the cookies with respect to color; OFSP flour could add natural color to baked products such as cookies with significantly improved acceptability.

4.3.2 Crispiness

Crispiness is the textural properties of a food product perceptible by mechanical, tactile and where appropriate, visual and auditory receptors. The quality score in response to crispness of the cookies exhibited significant changes ($P<0.05$) in this trait due to blending proportion and cultivar but not ($P>0.05$) by their interaction (Appendix IV). It was observed that cookies supplemented with different levels of OFSP behaved differently; the more OFSP flour the less crispiness. At every level of blending of OFSP flour, the score obtained for crispiness was significantly different; ranged from 8.500 to 6.031 (Table 4.3.1). The lowest score for crispiness was found for cookie made from BR5 while the highest score was for cookie made from control (100% soft wheat flour). Cookies made from *Tulla*-OFSP and SWF composite flour were significantly crisp, scoring 7.438, from that of Cookies made from *Kulfo*-OFSP and SWF composite flour which scored 7.275 as shown in Table 4.3.2. The decreasing trend in quality scores for crispness of cookies was certainly due to increase in moisture content that has an inverse correlation with the crispness (Mahmood, 2008). Similarly, Fellows (2000) stated the texture of foods is mostly determined by the moisture; loss of moisture from the interior is required to produce the desired crisp texture. With this respect, it can be concluded that the reason why the cookies have showed a decreased in their crispiness score with increased OFSP flour in the blend, is due to a parallel increase in the moisture content of cookies which could be the result of high fiber content.

4.3.3 Taste

Taste, one of the parameters related to the acceptability of any product, is a sensation perceived by the taste buds and influenced by the texture, flavor and composition of the foods. Taste attributes consist of saltiness, sweetness, bitterness and sourness which are detected by the taste buds at the tip, sides, and back of the tongue. Taste was found to be significantly influenced ($p<0.05$) by cultivar, blending ratio and their interaction (Appendix IV). According to Table 4.3.1, cookie from control and Br_2 were not significantly different and the cookie from the control had the highest score in this trait. Despite the fact that sweet potato has apparently high sucrose compared to wheat flour, the score of the cookies for taste decreased, ranged from 7.750 of Br1 to 5.250 Br_5, when the ratio of OFSP flour increase in the blend. This could be due to the effect of other sensory attributes, probably flavor because most panelists have claimed about the distinct flavor of the cookies where the ratio of OFSPF was high. The acceptability for test of cookies was also significantly affected by cultivar where cookies made from *Tulla*-OFSP and SWF composite flour had sensory score of 7.175 but 6.663 for cookies made from *Kulfo*-OFSP and SWF composite flour as shown in table 4.3.2.

4.3.4 Flavor

Flavor, the main criterion that makes the product to be liked or not, is one of the sensory qualities of a food product and the perception is synthesis of taste and smell impressions, along with texture and also influenced by appearance. It was found that blending ratio, cultivar and their interaction ($P<0.05$) had a significant influence on flavor of the cookie (appendix IV). The mean values obtained for acceptability of flavor of cookies decreased as the ratio of OFSP flour in the blend increased. The highest score for this trait, 8.000, was for cookies made from 100% SWF (BR1) and the lowest score, 5.250, was for BR5 (Table 4.3.1). With respect to cultivar, cookies made from *Tulla*-OFSP and SWF composite flour had significantly higher score (7.250) than cookies made from *Kulfo*-OFSP and SWF composite flour which was 6.675 and (Table 4.3.2). This could be due to some volatile compounds produced by OFSP, strongly from *Kulfo*-OFSP which are perceived by olfactory tissue of the nasal cavity.

4.3.5 Overall Acceptability

Overall acceptability, the last sensory attribute analyzed by panelists, was significantly affected ($P<0.05$) by blending ratio, cultivar and by their interaction (appendix IV). Generally, there was declining trend in the overall acceptability of cookies during increase of OFSP flour in the composite flour. Maximum score was obtained by 100% SWF cookies (7.688) that decreased to 5.21 of Br_5 (Table 4.3.1). The *Tulla*-OFSP was significantly better in this respect than *Kulfo*-OFSP, which was 7.138 and 6.65 respectively (Table 4.3.2). The decrease in score for overall acceptability as the OFSPF increased in the formulation was probably due to the decreased acceptability for crispiness, taste and flavor as more OFSPF was incorporated in SWF, especially for *Kulfo*-OFSP. Besides, flour from *Kulfo*-OFSP had lower fat absorption capacity than all other flours which would have a negative impact on retention of flavor and mouth feel of the cookies. However, cookies from Br_1, Br_2 and Br_3 were not significantly different for overall acceptability, probably due to the effect of yellow color imparted by OFSPF. The result justifies that irrespective of cultivar, cookies developed from 25% OFSP flour and 75% SWF had similar overall sensory acceptability with cookies from 100% SWF.

4.4 Nutritional value

4.4.1 β-carotene and dry matter content of fresh OFSP roots

Table 4.4.1 β-carotene and dry matter content of fresh OFSP cultivars

Cultivar	β-carotene (μg/g)	Dry matter (%)
Tulla-**OFSP roots**	206.54±2.94^{a}	24.21±0.02^{b}
Kulfo-**OFSP roots**	312.86 ±1.26^{b}	22.00±0.52^{a}

All values are the means of duplicates ± standard deviation
For each column, mean values with the same lowercase superscript are not significantly different at the 0.05 level

Dry matter determination was necessary in order to calculate the beta-carotene content on a dry weight basis (dwb). According to Table 4.4.1, the mean values for beta-carotene content of 1 gram of fresh OFSP roots from cultivar *Kulfo* was significantly higher ($p<0.05$) than from cultivar *Tulla*. The beta-carotene content of fresh *Kulfo*-OFSP roots which had moisture content of 78% was 312.86 μg/g (dwb) whereas the beta-carotene content of fresh *Tulla*-OFSP roots which had moisture content of 75.8%, was 206.54 μg/g (dwb). Therefore, both cultivars of OFSP roots demonstrated a potential for a significant contribution to vitamin A in the diet.

4.4.2 Nutritional Value of flours

4.4.2.1 β-carotene content of flours

Table 4.4.2.1 β-carotene content of flours

Flours	β-carotene (μg/g)
SWF	2.668±0.04^{a}
Tulla-**OFSPF**	184.900±1.70^{b}
Kulfo-**OFSPF**	234.486±3.09^{c}

All values are the means of duplicates ± standard deviation
For each column, mean values with the same lowercase superscript are not significantly different at the 0.05 level

The amounts of beta-carotene in flours from the two cultivars OFSP were significantly different ($p<0.05$). β-carotene content in *Tulla*-OFSP flour was 184.9 μg/g (dwb) which is obtained from fresh roots that had β-carotene content of 206.54 μg/g (dwb) while β-carotene content in *Kulfo*-OFSP flour was 234.486 μg/g (dwb) which is obtained from fresh roots that had initial β-carotene content of 312.86 μg/g (dwb); the losses of pro-vitamin A was 10.48% for *Tulla*-cultivar and 33.42% for *Kulfo*- cultivar. The result obtained was in agreement with Bechoff (2009), Sweet potato cultivar had a significant effect ($p<0.05$) on carotenoid losses in processing like drying and cultivars with higher initial moisture and beta-carotene content occurred to be related to higher level of carotene loss in processing. However, Flours made from the two cultivars of OFSP had large amount of beta-carotene content compared to wheat flour. Therefore, OFSP flour could be a potential source of provitamin A and contribute for beta-carotene in formulation of composite flour.

4.4.2.2 Proximate composition of flours

Table 4.4.2.1 proximate composition of flours

Flours	Moisture content	Ash (g)	Protein (g)	Fat (g)	Fiber (g)	CHO (g)
SWF	14.628±0.05^c	1.315±0.02^a	10.661±0.00^c	1.570±0.02^c	2.180±0.08^a	69.644±0.13^a
Tulla-**OFSPF**	6.054±0.04^a	3.071±0.00^c	5.091±0.49^b	0.972±0.00^a	7.823±0.00^b	76.629±0.49^b
Kulfo-**OFSPF**	6.782±0.01^b	2.632±0.03^b	4.030±0.00^a	1.443±0.00^b	8.252±0.00^c	76.861±0.45^b

All values are the means of duplicates ± standard deviation
For each column, mean values with the same lowercase superscript are not significantly different at the 0.05 level

The mean values for proximate composition of flours are expressed in terms of 100 g of flours and the results obtained are presented in table 4.4.2.1. OFSPF, compared to SWF, contains appreciable amounts of minerals, as can be inferred from their mean ash content which was 3.071% for *Tulla*-OFSPF and 2.632% for *Kulfo*-OFSPF compared to that of 1.315% SWF. The level of crude protein which was 5.091 % for *Tulla*-OFSPF and 4.030 % for *Kulfo*-OFSPF was lower than that of SWF, 10.661 %. This result justifies that there is compositional difference between OFSPF and SWF in their levels of protein in which SWF had significantly different protein than OFSP flours which might contribute to the higher protein content of cookies. The fat content of all flours were the least component of their proximate composition which is in agreement with Woolfe (1992) and Pareyt and Delcour (2008) in which fat represents the least percent of sweet potato composition and lipids are minor soft wheat flour constituents (typically 2%) respectively. However, the Fat content was significantly higher for SWF (1.57%) than *Tulla*-OFSPF (0.972 %) and *Kulfo*-OFSPF (1.443 %). A crude fiber level of OFSPF was analyzed to be 7.823% (*Tulla*) and 8.252% (*Kulfo*) which was significantly higher ($p<0.05$) compared to SWF (2.18 %). OFSP is a tuber plant this could be the reason why it contains higher fiber content than wheat. The carbohydrate content of OFSPF was significantly higher ($p<0.05$) than wheat flour and this could be due to high starch content of OFSP as described by Van Hall (2000). Therefore, blending of OFSPF to SWF will contribute to increase nutrients which were significantly lower ($p<0.05$) in one of the component of composite flours.

4.4.2.3 Energy value of flours

Table 4.4.2.3 Energy value of flours

Flours	Energy (kcal)
SWF	335.358±0.39[a]
Tulla-**OFSPF**	337.072±0.21[b]
Kulfo-**OFSPF**	336.553±0.15[b]

All values are the means of duplicates ± standard deviation
For each column, mean values with the same lowercase superscript are not significantly different at the 0.05 level

The energy values of both cultivars of OFSPF (*Tulla*- and *Kulfo*-) were significantly higher ($p<0.05$) than energy values of SWF. The energy values were 337.072 Kcal, 336.553 Kcal and 335.358 Kcal per 100 g of *Tulla*-OFSPF, *Kulfo*-OFSPF and SWF respectively. This could be due to high carbohydrate content of OFSPF, since carbohydrate is one of the sources of energy.

4.4.2.4 Mineral content of flours

Table 4.4.2.4 Mineral content of flours

Flours	Phosphorus (mg)	Calcium (mg)	Iron (mg)
SWF	17.564±1.13[b]	26.085±0.15[a]	3.887±0.20[b]
Tulla-**OFSPF**	18.670±0.15[b]	77.688±1.49[b]	0.757±0.00[a]
Kulfo-**OFSPF**	13.935±0.56[a]	92.500±2.19[c]	0.810±0.01[a]

All values are the means of duplicates ± standard deviation
For each column, mean values with the same lowercase superscript are not significantly different at the 0.05 level

Based on the above Table, the highest score for Calcium, phosphorous and Iron contents were 92.5mg for *Kulfo*-OFSPF, 18.67mg for *Tulla*-OFSPF and 3.887mg for SWF respectively. OFSPF are also good sources of minerals like Calcium and phosphorous but Iron compared to SWF. Therefore, supplementation of OFSPF to SWF for cookie making will contribute for increasing contents of minerals like calcium and Phosphorous but reduce iron content.

4.4.3 Nutritional value of cookies

4.4.3.1 Proximate composition of cookies

Cookies developed from composite flours of SWF and the two cultivars of OFSP flours, *Tulla and Kulfo*, at different blending ratio were analyzed for moisture, ash, crude protein, crude fat, crude fiber, total carbohydrate content. The mean values for proximate composition are expressed in terms of 100 g of cookie and the results obtained are presented in the following tables as a function of blending ratio and cultivar.

Table 4.4.3.1.1 Effect of blending ratio on proximate composition of cookies

Blending ratio	Moisture (%)	Ash (g)	Protein (g)	Fat (g)	Fiber (g)	CHO (g)
Br1	5.110±0.00[a]	1.351±0.00[a]	6.103±0.01[e]	18.516±0.28[b]	1.250±0.22[a]	67.671±0.04[e]
Br2	5.212±0.02[b]	1.494±0.05[b]	5.705±0.05[d]	18.502±0.26[ab]	1.726±0.03[b]	67.361±0.07[d]
Br3	5.269±0.02[c]	1.612±0.04[c]	5.298±0.13[c]	18.483±0.44[ab]	2.200±0.04[c]	67.139±0.10[c]
Br4	5.341±0.02[d]	1.727±0.07[d]	4.894±0.16[b]	18.468±0.67[ab]	2.670±0.06[d]	66.908±0.10[b]
Br5	5.403±0.03[e]	1.876±0.08[e]	4.486±0.20[a]	18.439±0.10[a]	3.152±0.06[e]	66.662±0.09[a]

All values are the means of duplicates ± standard deviation

For each column, mean values with the same lowercase superscript are not significantly different at the 0.05 level

Table 4.4.3.1.2 Effect of cultivar on proximate composition of cookies

Cultivar	Moisture (%)	Ash (g)	Protein (g)	Fat (g)	Fiber (g)	CHO (g)
Tulla	5.253±0.99[a]	1.651+0.22[b]	5.383+0.54[b]	18.433+0.64[a]	2.171+0.69[a]	66.109+0.40[a]
Kulfo	5.281±0.12[b]	1.573+0.17[a]	5.211+0.67[a]	18.520+0.17[b]	2.228+0.73[b]	66.187+0.35[a]

All values are the means of duplicates ± standard deviation

For each column, mean values with the same lowercase superscript are not significantly different at the 0.05 level

4.4.3.1.1 Moisture

The moisture content of a food is of great significance for many scientific, technical and economic reasons. Lesser the moisture content of the cookies, better its storage stability (Mahmood 2008). The moisture content of cookies was significantly affected ($P<0.05$) by blending ratio, cultivar and their interaction (Appendix V). The mean values for moisture content among cookies with different blending ratio (Table 4.4.3.1.1) elucidated statistically significant variations; ranged from 5.111 to 5.401. Moisture content of cookies was also affected by cultivar in which the highest score of was 5.253 for cookies made from *Tulla*-OFSP and SWF composite flour while the lowest score was 5.281 for cookies made from *Kulfo*-OFSP and SWF composite flour. It was observed that cookie from control had the least moisture content and a progressive increase in moisture content occurred when more OFSP was incorporated.

This increasing in moisture could be attributed to the higher water absorption capacity of OFSP flour than wheat flour (Table 4.1). The phenomenon of moisture increase in cookies during supplementing of non-wheat flour, which has high WAC value, to wheat flour is also supported by Tesfaye et al., (2010) who reported the increase of moisture content of cookies with increased amount of carrot flour supplementation in the composite flour and this was due to the high water absorption capacity of carrot flour. Similarly, Tekle et al., (2009) also reported that with an increase in the taro flour proportion in the cookies, there was an increase in the moisture content from average value 1.746% of the control to 2.153% of the 100% taro flour cookie due to the same reason; high WAC value of taro flour than wheat flour. Therefore, the increased moisture content of cookies with increased ratio of OFSPF in the composite could be due to high fiber content of OFSPF, especially for cultivar *Kulfo*.

4.4.3.1.2 Ash

Ash content in a food substance indicates inorganic remains after the organic matter has been burnt away and provides an estimate of the total mineral content of a food. The ash content of the cookies was found significantly influenced ($P<0.05$) by blending ratio, cultivar and their interaction (Appendix V). As listed on the table 4.4.3.1.1, ash content varied significantly from 1.351% of Br_1 (control) to 1.876% of Br_5 as proportion of OFSP flour was increased. This could be due to higher ash content of OFSP flour than wheat flour (Table 4.4.1); the control cookie was found significantly lower than all of the other blend proportions. This entails that incorporation of OFSP flour to wheat flour for cookie making could enhance the mineral intake, as ash is indicative of the amount of minerals contained in any food sample (Olaoye et al., 2007). A similar increase in ash content was also observed by Tesfaye et al., (2010) in their study of blending wheat flour with carrot and QPM flour; as the proportion of carrot flour was increased, in which carrot flour had the highest ash content (5.4%) than wheat (0.69%) and QPM (0.46%), the ash content of cookies were also increased.

4.4.3.1.3 Protein

The crude protein content of the cookies was significantly influenced ($P<0.05$) by blending ratio, type of cultivar and their interaction (Appendix V). The means for protein content of cookies illustrated significant decrease with increased OFSP flour in the composite flour; ranged from 6.103 of the control cookie to 4.486 of Br_5 cookie (Table 4.4.3.1.1) in which both cultivars of OFSP flour have a lower amount of protein content than wheat flour. However, cookies made from *Tulla*-OFSP and SWF composite flour had significantly higher ($p<0.05$) crude protein content than cookies made from *Kulfo*-OFSP and SWF composite flour, which were 5.383% and 5.211% respectively. The decrease in protein content of cookies as the ratio of OFSP increased in the blend, therefore, could be due to the less protein content of OFSP flours.

4.4.3.1.4 Fat

Dietary fat is the most concentrated source of energy; supplying 9 kcal/g (Yeung and Laquatra 2003). The fat content of the cookies was observed to be significantly affected ($p<0.05$) by blend proportion, cultivar and their interaction (Appendix V). As the amounts of OFSP flour in the composite flour increased, the amount of fat in the cookies was decreased. This is may be due to incorporation of OFSP flour that have smaller amount of fat content than wheat flour (Table 4.4.1). The means for fat content of supplemented cookies (Table 4.4.3.1.1) explicated non significant differences with varying levels of OFSP flour until Br_4 ranged from 18.516 of control to 18.439. Cookies made from *Kulfo*-OFSP and SWF composite flour had significantly higher fat content than cookie made from *Tulla*-OFSP and SWF composite flour, 18.520% and 18.433% respectively.

4.4.3.1.5 Crude fiber

Dietary fiber has been postulated to have beneficial effects on diabetes, atherosclerosis, cancer, and appendicitis, prevention of duodenal ulcer formation and varicose veins (Yeung and Laquatra 2003). The crude fiber content of the cookies was found significantly affected ($p< 0.05$) by blending ratio, cultivar and their interaction. (Appendix V). An increasing in crude fiber content of the cookies was observed as the proportion of OFSP flour was increased in the formulation (Table 4.4.3.1.1) In this respect, the control cookies were found inferior. As it can be seen from the raw material proximate composition (Table 4.4.1) the fiber content of the OFSP flour was found 8.252% and 7.823% but 2.18% for wheat flour. This result indicated that OFSP flour contain larger amount of crud fiber than wheat flour. This could be the reason that an increasing in fiber content was observed as the proportion of OFSP flour was increased. Similar result was also observed by Manoela et al., (2006) in their study of blending wheat flour with residue from king palm processing which contains higher fiber content than wheat flour and the control cookie was found significantly lower than all of the other blend proportion.

4.4.3.1.6 Carbohydrate

Carbohydrate content of cookies was expected to be higher since cookies have high sugar content and the raw materials have higher percentage of carbohydrate content than the other proximate components; 69.644 %, 76.629 % and 76.861 % for SWF, Tulla and Kulfo respectively (Table 4.4.1). The carbohydrate content of the cookie was found significantly influenced ($p< 0.05$) by blending ratio only (Appendix V). The decreasing in carbohydrate content was observed as the blending ratio of OFSP flour was increased. The control cookie was found significantly larger in carbohydrate content than all of the other blend proportion. The decreasing in carbohydrate content could be due to increase in moisture, ash and fiber content of cookies as the proportion of OFSP flour in the formulation was increased which leads to a reduction in carbohydrate content since carbohydrate was calculated by difference. A similar reduction in carbohydrate content was also reported by Manoela et al., (2006) in their study of blending wheat flour with residue from king palm processing which contained a higher fiber, ash and fat content than wheat flour.

4.4.3.2 Energy value of cookies

Table 4.4.3.2.1 Effect of blending ratio on energy values of cookies

Blending ratio	Energy(Kcal)
Br_1	466.732±0.07[e]
Br_2	463.778±0.25[d]
Br_3	461.092±0.20[c]
Br_4	458.418±0.29[b]
Br_5	455.540±0.46[a]

All values are the means of duplicates ± standard deviation
For column, mean values with the same lowercase superscript are not significantly different at the 0.05 level

Table 4.4.3.2.1 Effect of cultivar on energy values of cookies

Cultivar	Energy (Kcal)
Tulla	460.950+4.24[a]
Kulfo	461.274+4.034[b]

All values are the means of duplicates ± standard deviation
For column, mean values with the same lowercase superscript are not significantly different at the 0.05 level

The energy values of cookies from OFSP-wheat composite flour were ranged from 466.732 to 455.540 Kcal/100g. The energy value of cookies was found significantly influenced ($p< 0.05$) by both blending ratio and cultivar only (Appendix VI). A decreasing in energy values of cookies was observed as the blending OFSP was increased. This is associated with a reduction in protein, fat and carbohydrate content and an increasing in ash, crude fiber and moisture content. A similar reduction in energy value of cookies was also found by Manoela et al., (2006) in their study of blending wheat flour with residue from king palm processing; the control cookies were significantly larger in energy value than all of the other blend proportion, like the results obtained in this paper.

4.4.3.3 β-carotene content of cookies

Table 4.4.3.3.1 Effect of blending ratio on β-carotene content of cookies

Blending ratio	β-carotene (μg/g)	β-carotene (μg/ 100g)	RAE (per 200g)
Br 1	1.496+0.01[a]	149.6	23.016
Br 2	16.636+1.78[b]	1663.6	256.000
Br 3	29.037+3.41[c]	2903.7	446.724
Br 4	36.884+2.14[d]	3688.4	567.446
Br 5	41.047+4.33[e]	4104.7	631.492

All values are the means of duplicates ± standard deviation
For the second column, mean values with the same lowercase superscript are not significantly different at the 0.05 level

Table 4.4.3.3.2 Effect of cultivars on β-carotene content of cookies

Cultivar	β-carotene (μg /g)	β-carotene (μg/ 100g)	RAE (per 200g)
Tulla-**OFSP**	23.010+14.02[a]	2301.0	354.000
Kulfo-**OFSP**	27.031+16.38[b]	2703.1	415.862

All values are the means of duplicates ± standard deviation
For the second column, mean values with the same lowercase superscript are not significantly different at the 0.05 level

The content of cookies' Beta-carotene, which is a precursor of vitamin A, was found to be significantly influenced ($P < 0.05$) by blending ratio, cultivar and their interaction (Appendix VII). As it can be seen from table 4.4.3.3.1, the beta-carotene content of the control cookie (100% wheat) was found to be the lowest as compared to the blend proportion, since the initial beta-carotene content of the soft wheat flour was apparently low, only 2.668 μg/g (dwb); as the proportion of the OFSP flour increased in the formulation, the beta carotene content was also increased. It is also observed that the beta-carotene content was significantly high for cookies made from *Kulfo*-OFSPF and SWF blend than *Tulla*-OFSPF in the blend (Table 4.4.3.3.2). This might be due to high beta-carotene content of *Kulfo*-OFSP flour (234.486 μg/g (dwb)) than that of flour from Tulla-OFSP (184.900 μg/g (dwb)).

Generally, after baking of cookies at temperature of 160°c for 8 minutes, the percent loss in beta-carotene content was 5.12%, 13.75%, 21.18% and 35.01% in Br2, Br3, Br4, Br5 when there was *Tulla*-OFSPF in the blend while 7.39%, 15.88%, 31.55% and 41.55% when there was *Tulla*-OFSPF in the blend of Br2, Br3, Br4 and Br5. Therefore, the loss was high for high beta-carotene. The loss for the control was only 1.9%. Irrespective of cultivars, 200 g of cookies developed from Br_3, which had the same overall acceptability with the control, provide 446.724 RAE. This means 200 g cookies from Br_3 can provide more than 100% of mean requirements for all age-class-age groups except for lactating women which is 99.27%. But the same cookies provide 99.27 and more than 100% pre-school children and 55.84% for pregnant women and 52.56 % for lactating women, which are the most VAD affected segment of population, of their recommended dietary allowance (RDA). Therefore, OFSP flour can be supplemented to wheat flour for cookie making with the objective of increasing vitamin A precursor.

4.4.3.4 Mineral content of cookies

Table 4.4.3.4.1 Effect of blending ratio on mineral content of cookies

Blending ratio	P (mg)	Ca (mg)	Fe (mg)
Br1	10.088±0.14[a]	14.995±0.50[a]	2.150±0.03[e]
Br2	10.120±0.24[a]	19.699±0.92[b]	2.030±0.03[d]
Br3	10.120±0.43[a]	24.262±1.18[c]	1.773±0.02[c]
Br4	10.123±0.66[a]	29.500±1.90[d]	1.578±0.04[b]
Br5	10.218±0.94[a]	33.833±2.88[e]	1.365±0.02[a]

All values are the means of duplicates ± standard deviation
For each column, mean values with the same lowercase superscript are not significantly different at the 0.05 level

Table 4.4.3.4.2 Effect of cultivar on mineral content of cookies

Cultivar	P (mg)	Ca (mg)	Fe (mg)
Tulla	10.524±0.35[b]	23.300±6.22[a]	1.778±0.31[a]
Kulfo	9.743a±0.28[a]	25.615±7.96[b]	1.780±0.30[a]

All values are the means of duplicates ± standard deviation
For each column, mean values with the same lowercase superscript are not significantly different at the 0.05 level

Cookies from different blending ratio of SWF and the two cultivars of OFSP flours were analyzed for mineral contents such as Phosphorous, Calcium and Iron. The phosphorous content of cookies was found significantly influenced ($P < 0.05$) by cultivar and interaction of cultivar and blending ratio but not by blending ratio (Appendix VIII). The calcium content of cookies was found significantly influenced ($P < 0.05$) by cultivar, blending ratio and their interaction (Appendix VIII). However, the Iron content of cookies was found significantly influenced ($P < 0.05$) by blending ratio only (Appendix VIII). The decreasing in Iron content and increasing in calcium content was observed as the blending ratio of OFSP flour increase in the blend (Table 4.4.3.4.1). These could be due to high content of Fe in SWF than wheat flour and high content of Ca in OFSP flour than wheat flour. Cultivar had also significant effect on the mineral content (Ca and P but not Fe) of cookies. Ca content was significantly higher for cookies made from SWF and *Kulfo*-OFSPF blend but the P content was significantly higher for cookies made from SWF and *Kulfo*-OFSPF blend (Table 4.4.3.4.2). This could be due to higher P content of *Tulla*-OFSPF and Higher Ca content of *Kulfo*-OFSPF. Therefore, supplementing of OFSPF to wheat flour for cookie making could also contribute for minerals like Ca and P.

4.5 Optimum formulation of composite flour

Table 4.5.1 Summery of simplex lattice mixture design for 2 components and 4 responses

Component 1 SWF (%)	Component 2 *Tulla*-OFSPF(%)	Response 1 Cookies' spread	Response 2 Cookies' overall acceptability	Response3 Cookies' energy (Kcal)	Response 4 Cookies'beta-carotene content (μg/100g)
87.500	12.500	8.693	7.750	458.663	1509.700
62.500	37.500	8.462	7.000	453.169	3505.700
100.000	0.000	8.693	7.688	461.731	149.700
50.000	50.000	8.392	5.500	450.262	3730.900
75.000	25.000	8.612	7.750	455.925	2609.100

According to the results obtained above in section 4.2 and 4.3, cookies made from soft wheat flour and *Tulla*-OFSP composite flours had better physical quality (larger spread) and sensory acceptability in terms of color, crisspiness, taste, flavor and overall acceptability than cookies made from soft wheat and *Kulfo*-OFSP composite flours; that means the primary qualities of cookies, before nutritional improvements, were found better when the OFSP flour in the composite flour is from *Tulla* than *Kulfo* cultivar. However, the nutritional quality in regard with β-carotene content, was less in *Tulla*-cultivar. There was no expectetion of parallel increase or decrease for all important qualities parameters of cookies during incorporation of non-gluten flour to wheat flour. Rather, the improvement of one quality caused the decrease of the other parameter. For instance, during serial addition of OFSP flour to wheat flour for cookie development, the more OFSPF in the composite flour, the high β-carotene content (nutritional improvement) but negatively affected the physical and sensory quality of cookies required by consumers. This indicates, to obtain high quality cookies, compromisation of all the quality indeces is very important. Therfore, the formulation of the composite flour from *Tulla*-OFSP and SWF for development of acceptable and β-carotene rich cookies was optimized by giving equal degree of importance for all responses using numerical Optimization technique.

Table 4.5.2 Goals of responses for optimization

Name	Goal	Lower limit	Upper limit
SWF	In range	50	100
OFSPF	In range	0	50
Spread	Maximize	8.392	8.693
Acceptability	Maximize	5.5	7.75
Energy	Maximize	450.262	461.731
Beta-carotene	Target **(2925)**	149.7	3730.9

Criteria were set to achieve cookies that have high β-carotene content and the desired quality characterstics; high energy value, large spread and high acceptability. Beta-carotene content was targeted to fulfil 100% of RDA for pre-school children from 200 g of cookies and the other characteristics were maximized as much as possible.

Table 4.5.3 Suggested models for each responses

Response	Name of response	Minimum Value	Maximum value	Suggested model
Y1	Spread	8.392	8.693	Linear
Y2	Acceptability	5.5	7.75	Quadratic
Y3	Energy	450.262	461.731	Linear
Y4	Beta-carotene	149.7	3730	Quadratic

- **Response 1: Cookies' spread**

The suggested model for spread was linear model and the final equation in terms of actual components was expressed as;

Spread = 0.08737***SWF** + 0.080706**OFSPF** with prediction value of 8.54418

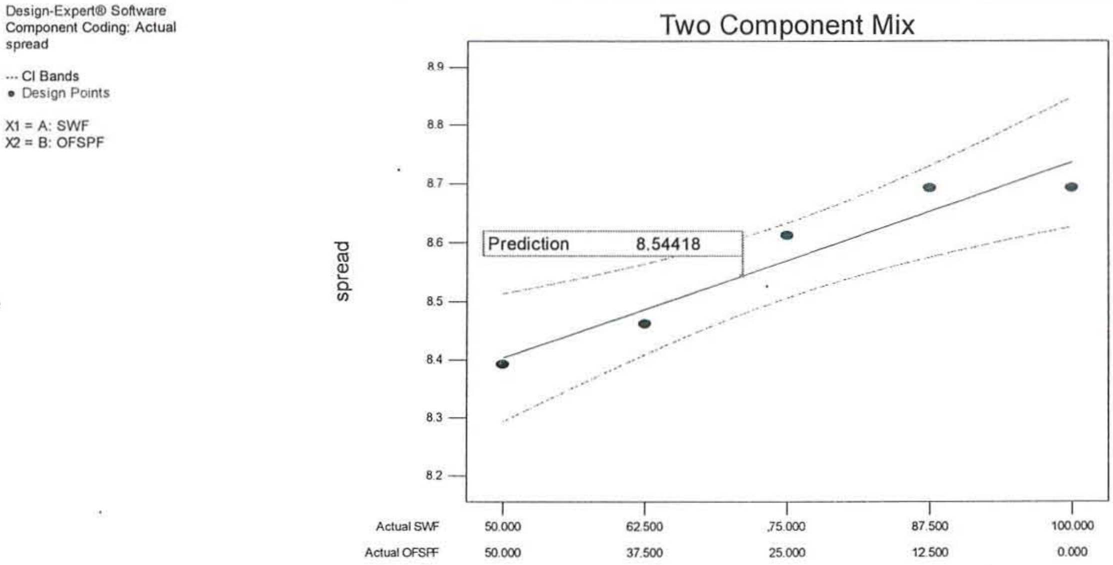


Figure 4.5.1 Cookies' spread against blending ratio

❖ **Response 2: Cookies' overall acceptability**

The suggested model for overall acceptability was quadratic model and the final equation in terms of actual components was expressed as:

Overall Acceptability = 0.076094* **SWF** - 0.05346* **OFSPF** + 0.001771***SWF*OFSPF** with prediction value of 7.50227

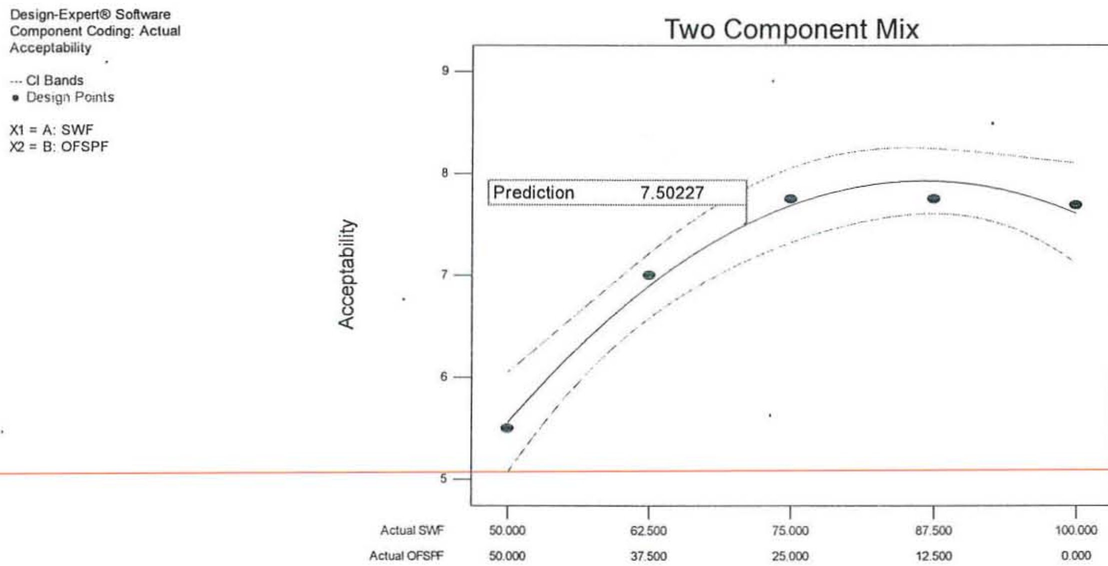


Figure 4.5.2 Cookies' overall acceptability against blending ratio

❖ **Response 3: Cookies' energy value**

The suggested model for energy value was linear model and the final equation in terms of actual components was expressed as:

Energy value = 4.616364***SWF** + 4.388908***OFSPF** with prediction value of 455.055

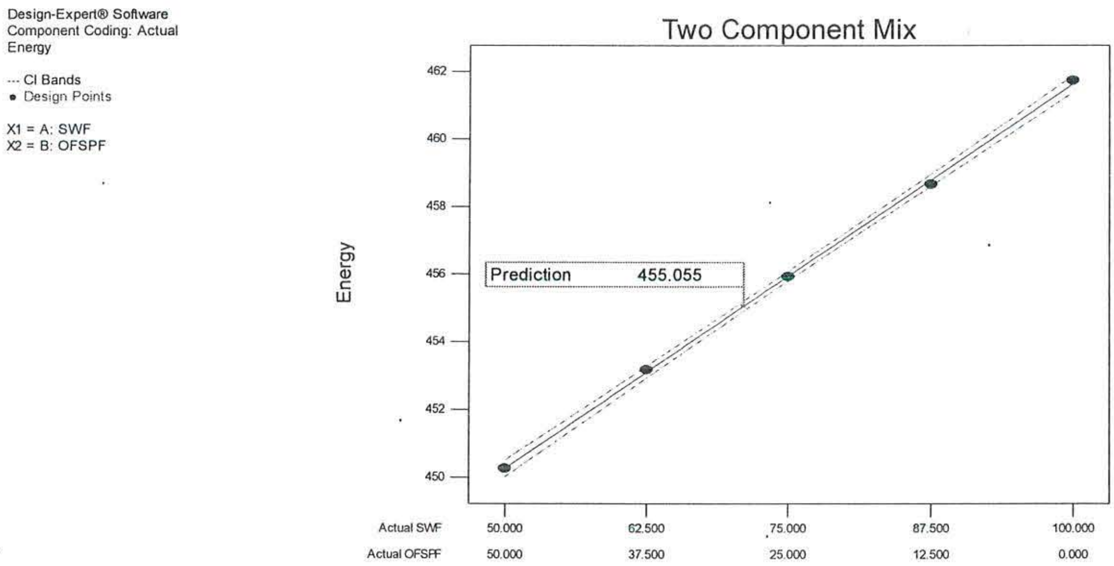


Figure 4.5.3 Cookies' energy against blending ratio

- **Response 4: Cookies' beta-carotene content**

The suggested model was quadratic and the final equation in terms of actual components was expressed as:

Beta-carotene= 1.1614***SWF** + 17.9166***OFSPF** +1.13024***SWF*OFSPF** with prediction value of 2925

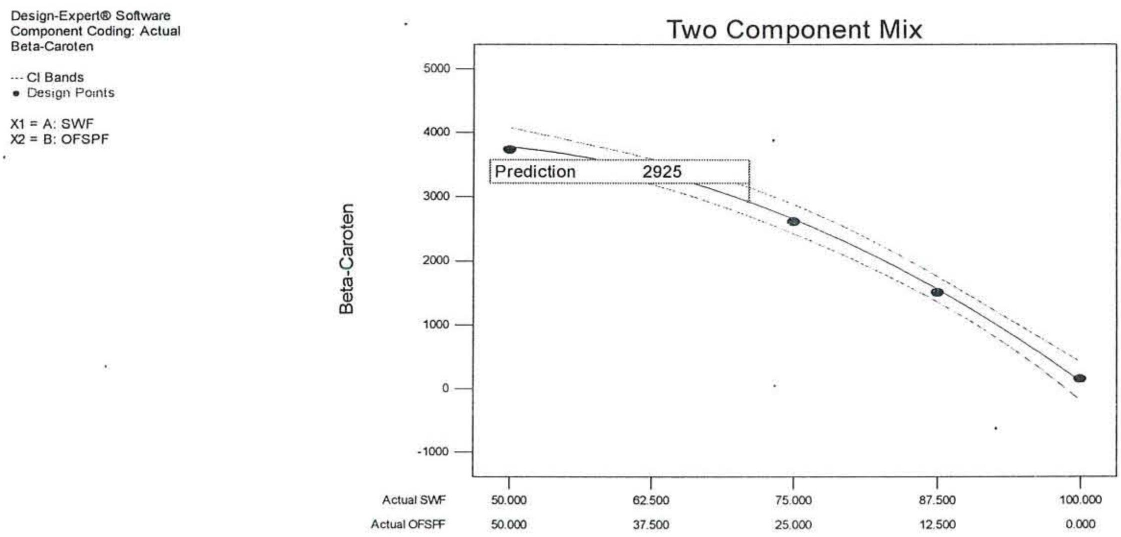


Figure 4.5.4 Cookies' beta-carotene content against blending ratio

❖ **Desirability**

Table 4.5.4 Desirability (overlay of the desired responses)

Component 1 SWF (%)	**Component 2 *Tulla*-OFSPF(%)**	**Response 1 Cookie' spread**	**Response 2 Cookie' overall acceptability**	**Response 3 Cookie' energy (Kcal)**	**Response 4 Cookie' beta-carotene content**
71.065	28.935	8.54418	7.50227	455.055	2925

The primarily selected formulation of *Tulla*-OFSP and soft wheat composite flour for development of beta-carotene rich cookies to satisfy 100 % RDA of pre-school children from 200 g of cookies with the desired quality characteristics of cookies was 71.065**SWF: OFSPF** 28.935 with prediction value of 0.714

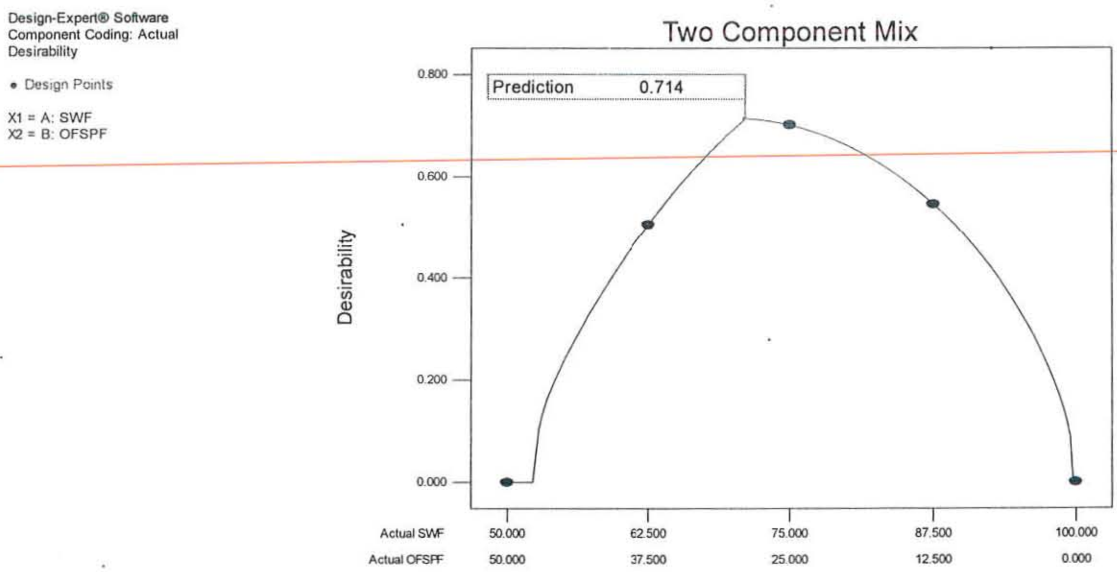


Figure 4.5.5 Desirability against blending ratio

5. Conclusion and Recommendations

5.1 Conclusion

In order to tackle vitamin A deficiency by increased consumption of OFSP, development of OFSP-based finished product and evaluation of the product in terms of it's quality parameters is an important step before recommending the product to consumers. This paper tried to investigate the possibility of incorporating OFSP flour to wheat flour for producing β-carotene rich cookies with their desired quality characteristics. For this effect, OFSP flour (from two cultivars namely *Tulla* and *Kulfo*) was supplemented to wheat flour at different blending ratio based on blending points obtained from the simplex lattice design. Cookies were then developed and their physical, sensory, and nutritional qualities were evaluated as a function of blending ratio and cultivar. Moreover, functional properties of the composite flours were also analyzed. The research output demonstrated that supplementation of OFSP flour to wheat flour for cookie making can contribute for increased β-carotene content and other nutrients with their required physical quality and sensory acceptability.

The study indicated that increasing the ratio of OFSP flour in the OFSPF-SWF blend generally increased cookies' ash, fiber, moisture, beta-carotene and Calcium content. This was largely due to the high content of fiber, ash, beta-carotene and calcium in OFSPF compared to wheat flour but decreased the protein, fat, carbohydrate and Iron content of cookies and energy value of cookies which could be due to low content of protein, fat and Iron in OFSPF and high content of fiber and ash in OFSPF respectively than wheat flour. Similarly, the other parameters, spread and sensory acceptability in terms of color, crispiness, taste, flavor and overall acceptability were also significantly affected ($p<0.05$) due to blending ratio. The highest scores for sensory property, except color, and physical property were for cookies made from 100% SWF (control). However, cookies from 25% OFSP and 75% SWF blend had similar overall acceptability with cookie from 100% SWF.

The study also showed that cookies made from *Tulla*-Cultivar OFSPF and SWF composite flour was significantly better ($p<0.05$) than cookies made from *Kulfo*-Cultivar OFSPF and SWF composite flour in terms of spread ratio, sensory acceptability (color, crispiness, taste, flavor, and overall acceptability) and nutrients like protein and phosphorous but significantly lower for nutrients like beta-carotene, calcium and fiber. The other nutrients like Iron and carbohydrate content of cookies were not significantly influenced ($p>0.05$) due to cultivar type. Generally, OFSP flour can be supplemented to wheat flour for development of sensory acceptable, beta-carotene rich and nutritious cookies. Hence, the ratio of flours in the composite flour to produce beta-carotene rich cookies with their desired quality characteristics (large spread, high acceptability and high energy) was optimized and found that cookies developed from 71.0655% SWF and 28.935% *Tulla*-OFSPF blend could be the best to attain all the desired properties optimally with prediction value of 0.714 for desirability. In general, the result of this study indicated that supplementation of orange-fleshed sweet potato flour to wheat flour is possible and very promising to develop β-carotene rich cookies.

which mix was found with higher beta-carotene? and is it also liked better than the others?

5.2 Recommendations

Based on the study result, supplementation of orange-fleshed sweet potato flour to soft wheat flour is very promising to develop β-carotene rich cookies. Therefore, the following recommendations are made.

- In order for communities benefit from high β-carotene content of OFSP and to combat VAD through finished products, trials and efforts for development of various OFSP-based value added products should be encouraged.
- Agricultural sectors should play role to adopt and cultivate various cultivars of OFSP roots to increase the availability, accessibility and consumption of OFSP throughout the country.
- All stakeholders who involve in combating VAD in the country should involve and take part to promote the potential benefit of OFSP by developing various OFSP-based finished food products and advertising them through mass media.
- Further studies on bioefficacy of cookies made from the OFSP-SWF composite flour on pre-school children is needed to be studied.
- Studies on level of Anti-nutrients of OFSP at different processing stages also need to be studied.

References

AACC. **(2000)**. Approved methods of American association of cereal chemists. *Arlington,10th edition.*

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.* Vol (2) pp. 087-091

Adu-Afarwuah, S., Lartey, A., Brown K.H., Zlotkin, S., Briend, A. and Dewey K.G. **(2008).** Home fortification of complementary foods with micronutrient supplements is well Accepted and has positive effects on infant iron status in Ghana. *Am. J. Clin. Nutr.* 87(4):929-938.

AOAC. **(2000).** Association of Official Analytical Chemists. *Official Method of Analysis, 16th Edn. Washington, DC.*

Bechoff, A., Dufour, D., Dhuique-Mayer, C., Marouzé, C., Reynes, M. and Westby, A. **(2009).** Effect of hot air, solar and sun drying treatments on provitamin A retention of OFSP. *Journal of Food Engineering* 92, 164-171.

Bechoff, A., Westby, A., Owori, C., Menya, G., Dhuique-Mayer, C., Dufour D. & Tomlins K. **(2010a).** Effect of drying and storage on the degradation of total carotenoids in orange-fleshed sweetpotato cultivars. *Jour of the Science of Food and Agriculture, 90*, 622-629.

Bechoff, A., Tomlins, K.I., Dhuique-Mayer, C., Dove, R. & Westby, A. **(2010b).** On farm Evaluation of the impact of drying and subsequent storage on the carotenoid content of Orange-fleshed sweet potato. *International Journal of Food Science & Technology.*

Bengtsson, A., Namutebi, A., Larsson Alminger, M. and Svanberg, U. **(2008).** Effects of Various traditional processing methods on the all-trans-b-carotene content of orange-Fleshed sweetpotato. *Journal of Food Composition and Analysis* 21, 134-143.

Berdanier, C.D., Feldman, E. B., & Dwyer, J. **(2008).** *Hand Book of Nutrition and Food, 2nd edition. CRC Press, Tylor and series group, USA.*

Bradley, D.G. and Min, D.B. **(1992).** Singlet oxygen oxidation of foods. *Critical Reviews in Food Science and Nutrition* 31, 211-236.

Brennan, G.J. **(2006)** Food Processing Handbook. WILEY-VCH Verlag GmbH & Co. KGaA, Weinheim, Germany.

Britton, G., Liaanen-Jensen, S. and Pfander, H. **(2008).** Carotenoids, Volume 4: Natural Functions. Birkhaüser: Basel, Switzerland.

Castillon Leon, J., Alpeeva, I.S., Chubar, T.A., Galaev, I.Y., Csoregi, E. and Sakharov, I.Y. (2002). Purification and substrate specificity of peroxidase from sweet potato tubers. *Plant Science* 163, 1011-1019.

CIP. **(2006).** International Potato Center (CIP) Report. McKnight Foundation CCRP East African Regional Sweetpotato Project Annual Report (April 2005–March 2006). International Potato Center, Nairobi, Kenya.

Coutsoudis, A. **(2000).** The relationship between vitamin A deficiency and HIV infection: review of scientific studies. Report prepared for the WHO Consultation on vitamin A in the first six months of life. WHO, Geneva.

Demissie, T., Ali, A., Mekonnen, Y., Haider, J., and Umeta, M. **(2009).** Demographic and Health-related Risk Factors of Subclinical Vitamin A Deficiency in Ethiopia. *Journal of Health and Nutrition.* 5:666-673

Demissie, T. **(2006).** Ethiopian national vitamin A deficiency survey report: socio-demographic, clinical, health, food access and consumption aspects. *Ethiopian Health and Nutrition Research Institute (EHNRI), Addis Ababa.*

Dogan, I.S. **(2006).** Effect of Oven Types on the Characteristics of Biscuits Made from refrigerated and Frozen Dough. *Journal of Food Technology and Biotechnology,* 44(1), 117–122

Ellie, W. and Sharon, R., R. **(2008).** *Understanding Nutrition. 7th edition.* 11:368-377

Emmanuel C. A. I., Osuchukwu N. C. and Oshiele L. **(2010).** Functional and sensory properties of wheat (*Aestium triticium*) and taro flour (*Colocasia esculenta*)composite bread. *African Journal of Food Science* Vol. 4(5), pp. 248-253

Ethiopian Health and Nutrition Research Institute. **(2009).** Nutrition baseline survey report for the national nutrition program of Ethiopia.

Ethiopian Health and Nutrition Research Institute. **(2006).** National Micronutrient Survey on Vitamin A Deficiency.

FAO/WHO. **(1988).** Requirements of vitamin A, Iron, Folate, and Vitamin B 12. *Report of a joint FAO/WHO Expert consultation Group, FAO Food and Nutrition series No. 23, FAO/WHO. Rome, Italy.*

FAO. **(1997).** Preventing micronutrient malnutrition: *a guide to food based dietary approaches. Washington: FAO & ILSI.*

FAO/WHO. **(2002).** Vitamin A, in Human Vitamin and Mineral Requirements. *Report of joint FAO/WHO expert consultation Bangkok, Thailand.* pp 87-107.

Food and Agricultural organization of the United Nations. **(2008).**Ethiopia Nutrition Profile Nutrition and Consumer Protection Division, FAO.

Gayathri G.N., Platel K., Prakash J. and Srinivasan K. **(2004).** Influence of antioxidant spices on the retention of b-carotene in vegetables during domestic cooking processes. *Food Chemistry* **84**, 35-43.

Gibson, R.S. **(1990).** Principles of nutritional assessment. Oxford Uni. Press, New York, USA. p.129-141.

Hagenimana, V., Carey, E., Gichuki, S.T., Oyunga, M.A. and Imungi, J.K. **(1999).** Carotenoid contents in fresh, dried and processed sweetpotato products. *Ecology in Food and Nutrition* **37**, 455-473.

Haskell MJ, Jamil KM, Hassan F,.Peerson JM, Hossain MI, Fuchs GJ, Brown KH. **(2004).** Daily consumption of Indian spinach (*Basella alba*) or sweet potatoes has a positive effect on total-body vitamin A stores in Bangladeshi men. *Am J Clin Nutr* 80:705–14.

Hodges, R.E., H.E. Sauberlich, J.E. Canham, D.L. Wallace, R.B. Rucker, L.A. Mejia and M. Mohanram. **(1978).** Hematopoietic studies in vitamin A deficiency. *Am. J. Clin. Nutr.* 31:876–885.

Hugo, L.F., Rooney, L.W. and Taylor, J.R.N. **(2000).** Malted sorghum as a functional ingredient in composite bread. *Cereal Science* 79(4): 428-432.

ILSI. **(2004).** International Life Sciences Institute. Nutritional and Safety Assessments of Foods and Feeds Nutritionally Improved through Biotechnology. *Comprehensive Reviews in Food Science and Food Safety*. 7: 81-91.

Johns, T., Eyzaguirre, P.B. **(2007).** Biofortification, biodiversity and diet: A search for complementary applications against poverty and malnutrition. *Food Policy Journal* 32, 1–24.

Kassaye T. Olivier R. Timothy J and Becklake M R. **(2001).** Prevalence of vitamin A deficiency in children aged 6–9 years in Wukro, northern Ethiopia. *Bulletin of the World Health Organization* 79: 415–422.

Klemm R. D. W., West K.P., Palmer A.C., Johnson Q., Randall P., Ranum P., and Northrop-Clewes C. **(2010).** Vitamin A fortification of wheat flour: Considerations and current recommendations. *Food and Nutrition Bulletin*, vol. 31, no. 1.

Kosambo, L., Carey, E. E., Misra, A. K., Wilkes, J., & Hagenimana, V. (1998). Influence of age, farming site and boiling on pro-vitamin A content in sweet potato (Ipomoea batatas Lam.). *Journal of Food Composition and Analysis*, 11, 305–321.

Kosambo L. **(2004).** Effect of storage and processing on all-trans-b-carotene content in fresh sweetpotato (*Ipomoea batatas* (L.) Lam) roots and its products. *CIP Funded Research Project: Annual Report (July 2003 – June 2004). Kenya Industrial Research and Development Institute. Nairobi. 11pp.*

Lazic Z. R. **(2004).** *Design of Experiments in Chemical Engineering, A Practical Guide. WILEY-VCH Verlag GmbH & Co. KGaA, Weinheim, USA.*

Low, J.W., 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 Community and International Nutrition* 137, 1320-27.

Low, J., Kapinga, R., Cole, D., Loechl, C., Lynam, J., and Andrade, M. **(2009).** Nutritional Impact with Orange-fleshed Sweetpotato, CIP-Social sciences working paper, 1. 74p.

Mahmood, S., M.S. Butt, F.M. Anjum and H. Nawaz. **(2008).** Baking and storage stability of retinyl acetate (vitamin A) fortified cookies. Pak. J. Nutr.7(4):586-589.

Manley, D., **(1998)**. Baking and cooling of biscuits. Wood head Publishing Ltd, England.

Manoela, A.V., Karina, C.T., Rossana, P., Sandra, R. P., & Edna, R. A., **(2006)**. Physicochemical and sensory characteristics of cookies containing residue from king palm (Archontophoenix alexandrae) processing. International Journal of Food Science and Technology 43, 1534–1540

McLaren, D.S., and Frigg, M .**(2001).** Sight and Life guidebook on vitamin A in health and disease. Basel, Switzerland, Task Force Sight and Life.

Mdziniso, P., Hinds, M.J., Bellmer, D.D., Brown, B. and Payton, M. **(2006).** Physical Quality and carotene content of colar-dried green leafy and yellow succulent vegetables. Plant Foods for Human Nutrition **61**, 13-21.

Mepba, H.D., Eboh, L., and Nwaojigwa, S.U. **(2007).** Chemical Composition, Functional and Baking Properties of Wheat-Plantain Composite Flours. African Journal of Food, Agriculture, Nutrition and Development. 7(1)

Nassar, A.G., AbdEl-Hamied, A.A., and El-Naggar E.A., **(2008)**. Effect of Citrus by-Products Flour Incorporation on Chemical, Rheological and Organolepic Characteristics of Biscuits. World Journal of Agricultural Sciences 4 (5), 612-616

Ojinnaka, M.C., Akobundu, E.N.T., and Iwe, M.O. **(2009).** Cocoyam Starch Modification Effects on Functional, Sensory and Cookies Qualities. Pakistan Journal of Nutrition 8 (5), 558-567.

Oladele, A. K., and Aina, J. O. **(2007).** Chemical Composition and Functional Properties of Flour Produced from Two Varieties of Tigernut (Cyperus esculentus). African Journal of Biotechnology, 6 (21) 2473-2476.

Olaoye, O.A., Onilude, A.A., and Oladoye, C.O. **(2007)**. Breadfruit Flour in Biscuit Making: Effects on Product Quality. *African Journal of Food Science*, 020-023.

Owori, C. B., Lemaga, R.O.M., Mwanga, A., Namutebi and Kapinga, R. **(2007).** Sweetpotato recipe book: sweetpotato processed products from Eastern and Central Africa. *African Crop Science Society, Kampala, Uganda.* 93pp.

Pareyt, B. and Delcour, J.A .**(2008).** The Role of Wheat Flour Constituents, Sugar, and Fat in Low Moisture Cereal Based Products: A Review on Sugar-Snap Cookies. *Journal of Critical Review in Food Science and Nutrition*, 48(9), 824 – 839

Pedersen, L., Kaack, K., Bergsoe, M.N., and Adler-Nissen J .**(2004).** Rheological properties of biscuit dough from different cultivars, and relationship to baking characteristics. *Journal of Cereal Science* 39: 37–46.

Rodriguez-Amaya D.B. **(1996).** Assessment of the pro-vitamin A contents of foods-the Brazilian experience. *Journal of Food Composition and Analysis* 9, 196-230.

Rodriguez-Amaya DB. **(1997).** Carotenoids and food preparation: the retention of provitamin A carotenoids in prepared, processed, and stored foods. *Arlington, VA, John Snow and Opportunities for Micronutrient Interventions Project*

Rodriguez Amaya D.B. **(2001).** *A Guide to Carotenoid Analysis in Food.* ILSI Press, Washington DC. 64pp.

Rodriguez Amaya D.B. and Kimura M. **(2004)** *HarvestPlus Handbook for Carotenoid Analysis.* Copyright HarvestPlus, Washington DC. 63pp.

Roomi MW, Ivanov V, Kalinovsky T, Niedzwiecki A, Rath M. **(2005).** In vivo antitumor Effect of ascorbic acid, lysine, proline and green tea extract on human colon cancer cell HCT 116 xenografts in nude mice: evaluation of tumor growth and immunohistochemistry. *On col Rep 13:421–5.*

Rucker, B., R. **(2001).** *Handbook of Vitamins.* Marcel Dekker, USA.

Shrestha, A.K., and Noomhorm, A. **(2002)**. Comparison of Physico-Chemical Properties of Biscuits Supplemented with Soy and Kinema Flour. *International journal of food science and nutrition,* 37, 361-368

Shittu, T.A., Raji, A. O. and Sanni, A.O. **(2007)**. Effect of baking time and temperature on some physicalo properties of bread loaf. *Food Research International* 40(2): 280-290.

Siems, W. , Wiswedel, I., Salerno, C., Crifo, C., Augustin, W., Schild, L., Langhans, C.D. and Sommerburg, O. **(2005).** Carotene breakdown products may impair mitochondrial functions_potential side effects of high-dose b-carotene supplementation. *Journal of Nutritional Biochemistry* **16**, 385–397.

Singh, S, Riar, C. S., and Saxena, D. C. **(2008).** Effect of Incorporating Sweet Potato flour to Wheat Flour on the Quality Characteristics of Cookies. *African Journal of Food Science*, 2, 065-072

Sommer A. **(1994)**. Vitamin A deficiency and its consequences: a field guide to detection and control, 3rd ed. Geneva, World Health Organization,

Sommer A. **(1998).** Xerophtalmia and vitamin A status. In: Progress in Retinal and Eye Research. Great Britain: Elsevier Science Ltd 17, 9-31.

Sumnu, S., And Sahin, S. **(2008).** *Food Engineering Aspects of Baking Sweet Goods*, CRC Press Taylor & Francis Group.

Taha, M. R., Majdi, A. A., and Khalil, I. E. **(2006).** Effect of Chickpea, Broad Bean, or Isolated Soy Protein Additions on the Physicochemical and Sensory Properties of Biscuits. *Jordan Univ. of Science and Technology, Jordan.*

Tekle, A., Worku S. and Retta N. **(2009).** The effect of blend proportion and baking condition on the quality of cookie made from Taro and wheat flour blend (unpublished).

Tesfaye B., Zegeye, A. and Admassu, S. **(2010).** Development of cookies from wheat, Quality protein maize and carrot composite flour. Lambert publishing academy.

Tofu, A., Anshebo, T., Tsegaye, E., and Tadesse, T. **(2007).** Summary of progress on Orange-fleshed sweetpotato research and development in Ethiopia. *Proceedings of the 13th ISTRC Symposium. Arusha, Tanzania.* pp 728-731.

Udensi, E. A., and Okoronkwo, K. A., **(2006)**. Effects of Fermentation and Germination on the Physico-Chemical Properties of *Mucuna cochinchinensis* Protein Isolate. *African Journal of Biotechnology,* 5 (10), 896-900

Van Hal, M. **(2000).** Quality of sweetpotato flour during Processing and storage, *Food Reviews International, 16: 1, 1 — 37*

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

Vieira, M.A, Tramonte, K.C., Podesta, R., Avancini, S.R.P., Amboni, R. D. de M. C. and Amante E.R. **(2007)**. Physicochemical and Sensory Characteristics of Cookies Containing Residue from King Palm (Archontophoenix alexandrae) Processing. *International Journal of Food Science and Technology,* 43, 1534–1540

Wade, P. **(1998).** *Biscuits, cookies and crackers:* The principle of the craft 1st ed. Elsevier applied Sciences, London.

Watts B.M, Ylimaki G.L., JefferyL.E., Elias L.G. **(1990).** *Basic Sensory Methods for Food Evaluation.* Ottawa, Ont., IDRC,. IDRC-277e, pp 43-66

West, C.E., Sijtsna, S.R., Kouwenhoven, B., Rombout J.H.W.M. and Zijpp, A.J.V. **(1992).** Epithelia damaging virus infections affect vitamin A status of Chicken. *J. Nutr.* p.333–339

West, K.P. **(2002).** Extent of Vitamin A Deficiency among Preschool Children and Women of Reproductive Age. *Journal of Nutrition* **132,** 2857–2866.

WHO. **(1995).** Global Prevalence of Vitamin A Deficiency. *Micronutrient Deficiency Information System Working Paper No. 2.* Geneva, Switzerland: World Health Organization.

WHO. **(1996).** Indicators for assessing vitamin A deficiency and their application in monitoring and evaluating intervention programmes. Geneva, Switzerland: World Health Organization.

WHO. **(2003).** Report nutrition micronutrient deficiencies: combating vitamin A eficiencies. Geneva, Switzerland: World Health Organization.

WHO. **(2004).** Vitamin A deficiency (Chap. 4) In: Comparative Quantification of Health Risks. *Global and Regional Burden of Disease Attributable to Selected Major Risk Factors* (V.1). World Health Organisation, Geneva.

Woolfe, J., A. **(1992).** *Sweetpotato: An Untapped Food Resource,* Cambridge University Press, Cambridge.

Yeung, D. L., and Laquatra, I. **(2003).** *Hand Book of Nutrition.* Heinz Corporate Research Center, Canada.

Zoulias, E.I., Oreopoulou, V., and Kounalaki, E. **(2002).** Effect of Fat and Sugar replacement on Cookie Properties. *Journal of the Science of Food and Agriculture,* 82, 1637–1644

Appendices

Appendix-I: Score sheet for sensory evaluation of cookies

Panelist code………. Sample code……………. Date…………………

Please evaluate each of the Samples of cookies provided on the white tray for the sensory attributes being evaluated in order from the left to the right as shown on the table and indicate how much you like or dislike the coded sample for each attributes by checking the appropriate phrase.

Category scale	Color	Crispiness	Taste	Flavor	Overall Acceptability
Like Extremely					
Like Very much					
Like Moderately					
Like Slightly					
Neither like nor dislike					
Dislike Slightly					
Dislike Moderately					
Dislike Very much					
Dislike Extremely					

Comments: -

Thank you

Appendix-II: Water absorption capacity (WAC) of composite flours

Tests of Between-Subjects Effects

Dependent Variable: WAC

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	13.451[a]	9	1.495	9.489E4	.000
Intercept	66.092	1	66.092	4.196E6	.000
Blending ratio	13.248	4	3.312	2.103E5	.000
Cultivar	.135	1	.135	8.570E3	.000
Blending ratio * Cultivar	.068	4	.017	1.076E3	.000
Error	.000	10	1.575E-5		
Total	79.543	20			
Corrected Total	13.451	19			

a. R Squared = 1.000 (Adjusted R Squared = 1.000)

Appendix-III: Physical properties of cookies

3.1 Diameter

Tests of Between-Subjects Effects

Dependent variable: Diameter

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	17.028[a]	9	1.892	21.328	.000
Intercept	27035.584	1	27035.584	3.048E5	.000
cultivar	1.088	1	1.088	12.266	.006
blend	15.419	4	3.855	43.451	.000
cultivar * blend	.521	4	.130	1.469	.283
Error	.887	10	.089		
Total	27053.499	20			
Corrected Total	17.915	19			

a. R Squared = .950 (Adjusted R Squared = .906)

3.2 Thickness

Tests of Between-Subjects Effects

Dependent variable: Thickness

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.029[a]	9	.003	1.291	.347
Intercept	371.255	1	371.255	1.473E5	.000
cultivar	.001	1	.001	.553	.474
blend	.022	4	.006	2.213	.141
cultivar * blend	.006	4	.001	.553	.701
Error	.025	10	.003		
Total	371.309	20			
Corrected Total	.054	19			

a. R Squared = .537 (Adjusted R Squared = .121)

3.3 Spread

Tests of Between-Subjects Effects

Dependent Variable: spread

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.440[a]	9	.049	8.516	.001
Intercept	1462.871	1	1462.871	2.547E5	.000
cultivar	.057	1	.057	10.005	.010
blend	.279	4	.070	12.147	.001
cultivar * blend	.104	4	.026	4.511	.024
Error	.057	10	.006		
Total	1463.369	20			
Corrected Total	.498	19			

a. R Squared = .885 (Adjusted R Squared = .781)

Appendix-IV: Sensory characteristics of cookies

4.1 Color

Tests of Between-Subjects Effects

Dependent Variable: Color

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	114.775[a]	9	12.753	11.808	.000
Intercept	8037.225	1	8037.225	7441.875	.000
Blending ratio	75.650	4	18.912	17.512	.000
Cultivar	9.025	1	9.025	8.356	.004
Blending ratio * Cultivar	30.100	4	7.525	6.968	.000
Error	162.000	150	1.080		
Total	8314.000	160			
Corrected Total	276.775	159			

a. R Squared = .415 (Adjusted R Squared = .380)

4.2 Crispiness

Tests of Between-Subjects Effects

Dependent Variable: crispiness

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	119.131[a]	9	13.237	52.859	.000
Intercept	8658.306	1	8658.306	34575.599	.000
Cultivar	1.056	1	1.056	4.218	.042
Blending ratio	116.912	4	29.228	116.718	.000
Cultivar * Blending ratio	1.163	4	.291	1.161	.331
Error	37.563	150	.250		
Total	8815.000	160			
Corrected Total	156.694	159			

a. R Squared = .760 (Adjusted R Squared = .746)

4.3 Taste

Tests of Between-Subjects Effects

Dependent Variable: Taste

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	160.006[a]	9	17.778	43.056	.000
Intercept	7659.056	1	7659.056	18548.673	.000
Cultivar	10.506	1	10.506	25.444	.000
Blending ratio	143.975	4	35.994	87.170	.000
Cultivar * Blending ratio	5.525	4	1.381	3.345	.012
Error	61.938	150	.413		
Total	7881.000	160			
Corrected Total	221.944	159			

a. R Squared = .721 (Adjusted R Squared = .704)

4.4 Flavor

Tests of Between-Subjects Effects

Dependent Variable: Flavor

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	190.400[a]	9	21.156	55.309	.000
Intercept	7756.225	1	7756.225	20277.712	.000
Cultivar	13.225	1	13.225	34.575	.000
Blending ratio	168.837	4	42.209	110.351	.000
Cultivar * Blending ratio	8.337	4	2.084	5.449	.000
Error	57.375	150	.382		
Total	8004.000	160			
Corrected Total	247.775	159			

a. R Squared = .768 (Adjusted R Squared = .755)

4.5 Overall Acceptability

Tests of Between-Subjects Effects

Dependent Variable: Overall Acceptability

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	164.006[a]	9	18.223	38.398	.000
Intercept	7603.806	1	7603.806	16022.068	.000
Blending ratio	147.912	4	36.978	77.917	.000
Cultivar	9.506	1	9.506	20.031	.000
Blending ratio * Cultivar	6.587	4	1.647	3.470	.010
Error	71.187	150	.475		
Total	7839.000	160			
Corrected Total	235.194	159			

a. R Squared = .697 (Adjusted R Squared = .679)

Appendix-V: Proximate composition of cookies

5.1 Moisture

Tests of Between-Subjects Effects

Dependent Variable: Moisture

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.212[a]	9	.024	881.239	.000
Intercept	554.910	1	554.910	2.078E7	.000
Cultivar	.004	1	.004	144.727	.000
blend	.206	4	.052	1.932E3	.000
Cultivar * blend	.002	4	.000	14.914	.000
Error	.000	10	2.670E-5		
Total	555.122	20			
Corrected Total	.212	19			

5.2 Ash

Tests of Between-Subjects Effects

Dependent Variable: Ash

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.700[a]	9	.078	161.964	.000
Intercept	51.961	1	51.961	1.082E5	.000
Cultivar	.030	1	.030	63.491	.000
blend	.658	4	.165	342.518	.000
Cultivar * blend	.012	4	.003	6.027	.010
Error	.005	10	.000		
Total	52.666	20			
Corrected Total	.705	19			

a. R Squared = .993 (Adjusted R Squared = .987)

5.3 Crude protein

Tests of Between-Subjects Effects

Dependent Variable: Protein

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	6.765[a]	9	.752	267.599	.000
Intercept	561.175	1	561.175	1.998E5	.000
Cultivar	.147	1	.147	52.232	.000
blend	6.543	4	1.636	582.292	.000
Cultivar * blend	.076	4	.019	6.747	.007
Error	.028	10	.003		
Total	567.968	20			
Corrected Total	6.793	19			

a. R Squared = .996 (Adjusted R Squared = .992)

5.4 Crude Fat

Tests of Between-Subjects Effects

Dependent Variable: Fat

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.061[a]	9	.007	7.570	.002
Intercept	6831.132	1	6831.132	7.681E6	.000
Cultivar	.030	1	.030	33.768	.000
blend	.014	4	.004	4.035	.033
Cultivar * blend	.016	4	.004	4.555	.024
Error	.009	10	.001		
Total	6831.202	20			
Corrected Total	.069	19			

a. R Squared = .872 (Adjusted R Squared = .757)

5.5 Crude fiber

Tests of Between-Subjects Effects

Dependent Variable: Fiber

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	9.044[a]	9	1.005	1.864E3	.000
Intercept	96.743	1	96.743	1.794E5	.000
Cultivar	.016	1	.016	29.395	.000
blend	9.019	4	2.255	4.182E3	.000
Cultivar * blend	.009	4	.002	4.079	.032
Error	.005	10	.001		
Total	105.792	20			
Corrected Total	9.049	19			

a. R Squared = .999 (Adjusted R Squared = .999)

5.6 Carbohydrate

Tests of Between-Subjects Effects

Dependent Variable: CHO

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	2.489[a]	9	.277	43.366	.000
Intercept	90177.347	1	90177.347	1.414E7	.000
Cultivar	.030	1	.030	4.747	.054
blend	2.448	4	.612	95.967	.000
Cultivar * blend	.011	4	.003	.419	.792
Error	.064	10	.006		
Total	90179.899	20			
Corrected Total	2.552	19			

a. R Squared = .975 (Adjusted R Squared = .953)

Appendix-VI: Energy value of cookies

Tests of Between-Subjects Effects

Dependent Variable: Energy

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	308.656[a]	9	34.295	718.876	.000
Intercept	4160760.394	1	4160760.39	8.722E7	.000
Cultivar	.523	1	.523	10.968	.008
blend	307.933	4	76.983	1.614E3	.000
Cultivar * blend	.199	4	.050	1.044	.432
Error	.477	10	.048		
Total	4161069.527	20			
Corrected Total	309.133	19			

a. R Squared = .998 (Adjusted R Squared = .997)

Appendix-VII: Beta-carotene content of cookies

Tests of Between-Subjects Effects

Dependent variable: Beta-carotene

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	4262.928[a]	9	473.659	5.636E3	.000
Intercept	12519.958	1	12519.958	1.490E5	.000
Cultivar	80.806	1	80.806	961.569	.000
blend	4149.495	4	1037.374	1.234E4	.000
Cultivar * blend	32.627	4	8.157	97.063	.000
Error	.840	10	.084		
Total	16783.727	20			
Corrected Total	4263.769	19			

a. R Squared = 1.000 (Adjusted R Squared = 1.000)

Appendix-VIII: Mineral content of cookies

8.1 Phosphorous (P)

Tests of Between-Subjects Effects

Dependent Variable: Phosphorous

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	4.675[a]	9	.519	38.926	.000
Intercept	2053.756	1	2053.756	1.539E5	.000
cultivar	3.050	1	3.050	228.535	.000
blend	.039	4	.010	.724	.595
cultivar * blend	1.587	4	.397	29.726	.000
Error	.133	10	.013		
Total	2058.565	20			
Corrected Total	4.809	19			

a. R Squared = .972 (Adjusted R Squared = .947)

8.2 Calcium (Ca)

Tests of Between-Subjects Effects

Dependent Variable: Calcium

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	942.843[a]	9	104.760	616.087	.000
Intercept	11963.533	1	11963.533	7.036E4	.000
cultivar	26.803	1	26.803	157.627	.000
blend	902.161	4	225.540	1.326E3	.000
cultivar * blend	13.879	4	3.470	20.405	.000
Error	1.700	10	.170		
Total	12908.076	20			
Corrected Total	944.544	19			

a. R Squared = .998 (Adjusted R Squared = .997)

8.3 Iron (Fe)

Tests of Between-Subjects Effects

Dependent Variable: Iron

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	1.651[a]	9	.183	138.955	.000
Intercept	63.297	1	63.297	4.795E4	.000
cultivar	2.000E-5	1	2.000E-5	.015	.904
blend	1.651	4	.413	312.638	.000
cultivar * blend	3.000E-5	4	7.500E-6	.006	1.000
Error	.013	10	.001		
Total	64.961	20			
Corrected Total	1.664	19			

a. R Squared = .992 (Adjusted R Squared = .985)

Declaration

I certify that this work has not been accepted in substance for any degree, and is not currently submitted for any degree other than that of Masters of Science in Addis Ababa University. I also declare that this work is the result of my own investigations except where otherwise stated.

Name: Adugna Melkie

Signature: ______________

Date: June 19, 2011

The thesis has been submitted with my approval as a supervisor.

Name: Melesse Abdisa (PhD)

Signature: ______________

Date: ______________

Name: Nigusse Reta (Professor)

Signature: ______________

Date: ______________