

**NUTRITIONAL AND SENSORY QUALITY OF EXTRUDED OAT,
SOYBEAN AND LINSEED COMPLEMENTARY INSTANT FOOD**

M.Sc. THESIS

HAILE TESFAYE

**JUNE, 2016
JIMMA, ETHIOPIA**

**NUTRITIONAL AND SENSORY QUALITY OF EXTRUDED OAT,
SOYBEAN AND LINSEED COMPLEMENTARY INSTANT FOOD**

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By

Haile Tesfaye

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APPROVAL SHEET

Jimma University College of Agriculture and Veterinary Medicine, Department of Post
Harvest Management

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Name of Student: Haile Tesfaye ID No. M.Sc. 06049/06

Program of Study: M.Sc. In Postharvest Management

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Complementary Instant Food.

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Major Advisor: Prof. Tefera Belachew (MD, M.Sc., PhD) signature _____ date _____

Co-Advisor: Sirawdink Fikreyesus (PhD Scholar) signature _____ date _____

Decision/suggestion of department graduate council (DGC)

Chairperson of DGC Name

Signature

Date

DEDICATION

This thesis manuscript is dedicated to my beloved father and brother who separated from me without seeing my fruits, to those who passed away from this world due to malnutrition in Ethiopia and finally I want to dedicate this thesis to those who scarify their life for the freedom of human being.

STATEMENT OF THE AUTHOR

I, the undersigned, declare that this Thesis is my work and is not submitted to any institution elsewhere for the award of any academic degree, diploma or certificate and all sources of materials used for this Thesis have been duly acknowledged. This Thesis has been submitted in partial fulfillment of the requirements for M.Sc. degree at Jimma University, College of Agriculture and Veterinary Medicine and is deposited at the University Library to be made available to borrowers under the rules of the library.

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Name: Haile Tesfaye

Place: Jimma University, Jimma

Date of submission: _____

Signature _____

BIOGRAPHICAL SKETCH

The author Haile Tesfaye was born October 1990 to his mother Mrs. Workitu Yadessa and Father Mr. Tesfaye Duguma in Muger, West Showa Oromia Regional State. He attended his elementary education at Muger Mekoda School from 1998-2006, upon completing his primary school education, he joined his secondary and preparatory education at Muger community high and preparatory school from 2006 to 2010. The author started his university education at Jimma University in September 2010 and graduated with Bachelor of Science degree (B.Sc.) in post harvest management in June 2013. After his graduation he directly joined Jimma University College of Agriculture and Veterinary Medicine in September 2013 again to pursue his graduate study in Post Harvest Management specializing in perishable crops.

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LIST OF ABBREVIATIONS

ALA	Alpha linolenic acid
ANF	Anti Nutritional Factor
AOAC	Association of Official Analytical Chemists
BD	Bulk Density
CSA	Central Statistics Agency
EPHI	Ethiopian Public Health Institute
FAO	Food and Agricultural Organization
HDL	High Density Lipoprotein
HTST	High Temperature, Short-Time
LDL	Low-Density Lipoprotein
LDPE	Low Density Poly ethylene
MOFED	Ministry of Finance and Economic Development,
MUFA	Mono Unsaturated Fatty Acid
PEM	Protein Energy Malnutrition
RDA	Required Daily Allowance
RTE	Ready to Eat
SNNP	South Nation and Nationality's of People
TVET	Technical and Vocational Education and Training
USDA	United States Development Agency
WAC	Water Absorption Capacity

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ABSTRACT

In Ethiopia child malnutrition is enormous challenge. Besides preparation and processing method, one of the major problems associated with macro and micronutrient deficiency in traditional complementary food is that, it is only from cereal source. Using blends of cereal, pulse and oilseed extruded composite flour(ECF) can solve these problems by improving nutritional quality; reducing macro and micro-nutrient deficiencies and producing safe and instant complementary food that needs minimum cooking time to reduce loss of heat liable nutrients. The blend of oat, soybean, linseed flour and premix (sugar, salt, moringa & fenugreek) composite flour was developed. Thirteen formulations of the composite flour were generated using D-optimal constrained mixture design with a range of oat 55-65%, soybean 11-23%, linseed 6-11% and 15%. A premix was added in equal proportion to all treatments to improve taste, flavor, mineral and vitamin. The blend was extrusion cooked at 130°C barrel temperature(BT), 150 rpm screw speed, (with the feeder delivering a feed rate of 5.1g/min; feed moisture content in the extruder barrel was 17% (170 g/kg). Nutritional and sensory qualities of the ECF were investigated using standard methods. The major responses of nutritional composition (protein, fat, carbohydrate, energy, β -carotene, iron, zinc and calcium, anti-nutritional factors (phytate and tannin), functional proprieties (bulk density, water absorption capacity) and sensory quality were analyzed. The results showed a significant ($P < 0.05$) difference in fat, protein, carbohydrate, ash, fiber, calcium, phytate, tannin, water absorption capacity (WAC) and bulk density of the ECF. The protein, fat, energy, tannin, phytate and mineral contents of the ECF increased with increasing ratio of soybean and linseed. However sensory acceptability and β - carotene content was decreased with increasing proportion of soybean in the blend. There was significant ($P < 0.05$) difference for aroma, taste, and consistency and over all acceptability of the gruel but appearance and mouth feel showed no difference for gruels. The blend ratios at 58.4%, 18.4% and 8.2% oat, soybean and linseed respectively were selected as overall optimum blend ratio with desirable nutritional composition which varied between 19.0-20.64%protein, 8-9.9% fat, carbohydrate 60.92- 63.7, 394.98-408.2kcal energy, 110- 124.4 mg/100 g calcium, 7-7.8 mg/100 g iron, 2.96-3.0 mg/100 g zinc, 1600- 1900.3 μ g/100 g, β - carotene and sensory attributes 3.75-4 overall acceptability (5 point hedonic scale).Extrusion cooked oat, soybean linseed blend enriched with premix showed significant improvement in the nutritional quality and sensory attributes. The result revealed that, the blend had good potential of improving protein, fat, energy, zinc, iron, & β -carotene of complementary food. Hence, using this optimum amount of formulation can enrich the nutritional value of CF.

Keywords: *Complementary Food, Extrusion Cooking, Instant Flour, Optimization, premix*

1. INTRODUCTION

1.1. Background

Malnutrition among children is the major health challenges in developing countries; it is evident that high prevalence of deaths each year among children aged less than five years old in the developing world is associated with malnutrition (Ijarotimi and Keshinro, 2012). This nutrition problem is ascribed to the inappropriate complementary feeding practices, low nutritional quality of traditional complementary foods and high cost of quality protein-based complementary foods (Eka *et al.*, 2010).

Globally, under nutrition in children is highly prevalent and remains a big challenge. According to United Nations FAO estimates, 11.11% of world populations were suffering from chronic undernourishment in 2012-2014 (FAO, 2014). Beyond all, children are the most visible victims of under nutrition. According to United Nations Children's Fund report, 25% and 8% of under-five year old children were estimated to be stunted and wasted respectively (UNICEF, 2014b). Similarly malnutrition child is an enormous challenge in Ethiopia.

In Ethiopia About 33.6 percent of the Ethiopian population are living below poverty line and cannot meet their daily minimum nutritional requirement of 2200 calories (MOFED, 2013). Based on recent Ethiopian demographic and health survey mini report, nationally 40%, 25% and 9% of children under age five were stunted, underweight and wasted respectively (CSA, 2014).

To overcome malnutrition problem a variety of complementary foods are available with high nutritive value, which are directly used for instant preparation of gruels. However these products are beyond the economic means of most families. So mothers use traditional cereal based gruel, as complementary foods for infants. These gruels usually have low energy density and poor protein, vitamin and mineral contents (Njongmeta *et al.*, 2003).

Complementary food could be improved by combining locally available food that complement each other in such a way that the new pattern of amino acids created by this combination is similar to that recommended for infants (Mensa-Wilmot *et al.*, 2001).

Thus; cereals and pulses play a predominant role in diets of developing countries. The use of cereal, pulse based food has long been advocated as alternative protein and energy source for children food products. Protein quality is a critically important problem in many developing countries, where human diets consist mainly of cereals (Serna-Saldivar and Rooney, 1995). The use of legume and oil seed in addition to cereal has been identified as a means of solving the problem of malnutrition associated with the consumption of cereal products (Ikujenlola and Fashakin, 2005).

Oat belongs to the Poaceae family, its unique among the cereals; with one of the rich sources of dietary fibers among cereals (Butt *et al.*, 2008). Oat grain when compared to wheat or rye grains is characterized by unusual nutritional composition (Butt *et al.*, 2008; Angioloni and Collar, 2012), because it contains a beneficial combination of nutritional compounds. Lipid content in oat grain is 2- 3 times higher than in other cereals (Butt *et al.*, 2008; Angioloni and Collar, 2012). Moreover it is characterized by high protein content (Butt *et al.*, 2008; Gambús *et al.*, 2011). Unique nutritional composition of oat grain contributes to growing worldwide interest in this cereal in aspect of human nutrition (Klava *et al.*, 2007).

Grain legumes or pulses (including soybeans) are rich and low-cost sources of dietary proteins and nutrients for a large part of the world's population (Egounlety and Aworh, 2003). Soybeans is an excellent source of protein (about 35-40%), hence the seed is the richest in food value of all plant foods consumed in the world. It is a very rich source of vegetable protein for all including growing children (Dandago and Igwe, 2006) and it has been identified as a suitable protein rich crop that could improve the nutritional and economic status of the general population in developing countries (Babajide *et al.*, 2003). Soybeans have great potential in overcoming protein-calorie malnutrition.

Linseed has been used as a precious nutritional product and as a traditional medicine from ancient times. It is richest source of alpha-linolenic acid, lignans and other nutritional components. Flaxseed has an amino acid profile comparable to that of soybean flour and contains no gluten (Hongzhi *et al.*, 2004).

Composition of flaxseed makes it more promising for its utilization in different food products. Flaxseed is one of the richest vegetarian sources of α -linolenic acid (omega 3 fatty acid) and soluble mucilage. In present era, consumer's trend towards functional food has increased significantly as health awareness rose.

Fenugreek is locally used as a pulse, spice and medicinal plant, and has a long history in Ethiopia (Gall and Zerihun 2009). Fenugreek seed can be utilized for value addition of cereals based food products to attain multiple benefits. It is used as a condiment and as a supplement to wheat and maize flour for bread making and as a constituent of the daily diet of general population.

Moringa is a "miracle plant" that has almost all the minerals and vitamins that the body needs for good health. The leaves, pods and flowers of this plant which are used as vegetable in many parts of the world have great nutritional value (Fuglie, 2001). Moringa leaves and pods can be an extremely valuable source of nutrients for people of all ages. The leaves can be dried, made into powder and stored for use when needed (Olusola, 2006).

Various processing technique have been used to prepare complementary food, fermentation, sprouting/germination, extrusion and less often toasting (Obizoba and Ati, 1991). Extrusion is a powerful food processing operation, which utilizes high temperature and high shear force to produce a product with unique physical and chemical characteristics (Pansawat *et al.*, 2008). Extrusion is able to break the covalent bonds in biopolymers, and the intense structural disruption and mixing which facilitate the modification of functional properties of food ingredients (Singh *et al.*, 2007).

In addition, the hot extrusion process denatures undesirable enzymes; inactivates some anti-nutritional factors (trypsin inhibitors, tannins and phytates); sterilizes the finished product; and retains natural colors and flavors of foods (Singh *et al.*, 2007).

The combination of high throughput rates, energy efficiency and versatility results in the potential for improved cost effectiveness and process rationalization over traditional production methods. In Africa, due to deforestation by utilization of wood for fuel, there is a great need for precooked foods (Peleme *et al.*, 2002).

Various technologies are being used to develop “Instant foods” and extrusion is an important technology for processing grain-based products and adds immense value to raw material. High-Temperature, Short-Time (HTST) extrusion cooking could be used to produce foods of high nutritional quality and in a ready-to-eat form. A simple, convenient, easy and quick to prepare food product besides being hygienic, free from microbial contamination and also convenient to eat is ‘Instant food’ (Tamlurkar, 2006). Low cost and nutritious instant foods can be developed using major crops in the country which may be blended or supplemented with each other to provide complete nutrition. Instant foods are prepared from almost all available food grains, either alone or supplemented with fruits, vegetables, legumes, oilseeds, flavoring agents, coloring agents, preservatives, etc.

Generally in addition to nutritional value extrudates are microbiologically safe, can be stored for long periods because of low moisture without need for refrigeration and requires less labor for handling and less packaging materials and storage space (Filli and Nkama, 2007).

1.2. Statement of the Problem

Adoption of recommended breastfeeding and complementary feeding practices and access to the appropriate quality and quantity of foods are essential conditions for fulfilling optimal nutrition for infants and young children (PAG, 1972). Nutritionally, it has been proven that breast milk is a complete and perfect food for the infant during the first six months of life (Lutter and Rivera, 2003). After 6 months breast milk alone can no longer be sufficient both in terms of quantity and quality to meet the nutritional requirements of infants, hence, appropriate complementary foods should be introduced (UNICEF, 2009).

In developing countries, including Ethiopia many families cannot afford commercial complementary foods to wean their infants due to high level of poverty, and thereby engage in weaning the children on cereals gruels (Ikpeme-Emmanuel *et al.*, 2012).

Scientific findings have shown that cereal gruels are the common complementary foods in developing countries, which is characterized by low energy density and protein due to large volume of water relative to its solid matter contents during preparation (Inyang and Zakari, 2008; Igyor *et al.*, 2011). To increase the energy density of the gruel, more of the solid matter are needed, which will makes the gruel too thick and viscous for infant to eat and too bulky for their stomach. As a result, infants are unable to fulfill their energy and other essential nutrients requirements (Ikujenlola & Fashakin, 2005), hence, there is increased in protein-energy malnutrition among weaning children.

In Ethiopia poor feeding practices, methods of complementary food processing and shortfall in food intake are the most important direct factors responsible for malnutrition and illness among children. The presence of non-nutrient constituents (anti-nutritional factors) in plant-based foods has been shown to also negatively influence the bioavailability of nutrients. In addition, a combination of nutritionally inferior diets, improper processing and improper feeding practices are also major contributing factors to the development of childhood malnutrition (Jacobs and Rubery 1988. Poor processing methods and hygiene have also been identified as other factors responsible for low nutrient density in local complementary foods due to knowledge gap about simple processing techniques to produce nutritious food.

Therefore, there is the need to develop nutritious, storage stable and affordable complementary food from locally available raw materials by application household or small to medium scale production technologies had been strongly recommended as a viable and sustainable approach to address the problem of malnutrition .In addition, developing ready to eat infant food that need minimal processing and safe to eat is valuable way of tackling child malnutrition.

1.3. Research Questions

The study tried to answer the following questions:

1. Does the extruded composite flour meet nutrient requirements of infant?
2. Are the extruded composite flour's gruel sensory attribute are acceptable?

1.4. Objectives

1.4.1. General objectives

- ✚ To develop extrusion cooked composite flour for better nutritional composition, sensory attributes and less anti nutritional factor

1.4.2. Specific objectives

- ✚ To develop instant composite flour from locally available raw materials
- ✚ To evaluate nutritional composition of extrusion cooked composite flour
- ✚ To evaluate functional properties and anti nutritional factors of extruded composite flour
- ✚ To determine sensory acceptance of gruel prepared from extruded composite flour
- ✚ To determine the optimum blend of oat, soybean and linseed flour with suitable nutritional quality and sensory acceptability.

1.5. Significance of the Study

Children are the country's hope of tomorrow. For intelligent and brilliant child to be developed early complementary feeding play a determinant role. Adequate nutrition during infancy and early childhood is fundamental to the development of each child's full human potential. To improve nutritional requirements of infant, the status of lactating mother in feeding children, complementary food given and processing techniques of complementary food is crucial, therefore this study was carried out with the intent of developing nutritious instant composite flour which can meet minimum nutrient requirements of infants. Developing instant nutritious infant flour is one way of reducing malnutrition, it minimizes nutrients loss due to high cooking temperature and it saves time of cooking, in addition it creates job opportunities for small scale micro enterprise who want to produce instant composite flour from locally available materials. This will have contribution to the solution of the pervasively high level of stunting among children that sets in during the complementary feeding Period.

2. LITERATURE REVIEW

2.1. Child Malnutrition

Child malnutrition is the most devastating problem currently facing the majority of the world's poor. The level of under nutrition among children remains unacceptable throughout the world, with large number of children living in developing world (WHO, 2008). The major risk factors associated with infant and early childhood mortality and morbidity are poor infant feeding practices as well as childhood and maternal under-nutrition (WHO, 2009).

Prevalence of Protein Energy Malnutrition (PEM) in infants after six months of age is high in Africa (Lalude and Fashakin, 2006). This is because infants at this stage of development require higher energy and proteins in their diet so as to meet the increasing demand for metabolism. Protein energy malnutrition is an important nutritional deficiency condition that often occurs during the critical transitional phase of complementary food infants, complementary crippling their physical and mental growth. This condition can be prevented to a large extent by introducing complementary foods of good quality and quantity at right proportion and at right stage.

Complementary feeding period is the time when malnutrition starts in many infants, contributing significantly to the high prevalence of malnutrition in children less than 5 years of age worldwide (Daelmans and Saadeh, 2003). Poor feeding practices as well as lack of suitable complementary foods are responsible for under nutrition with poverty exacerbating the whole issue. In developing countries, children are mostly weaned on starchy, bulky gruels which are characterized by both low energy and low nutrient density.

Malnutrition remains one of the most common causes of morbidity and mortality among children throughout the world (WHO, 2008). Particularly stunting, is still a severe public health problem in Sub Saharan Africa. For example, about 35 % of preschoolers are stunted; while 29 % are underweight in Sub-Saharan Africa (Leenstra *et al.*, 2005). UNICEF (2009) reported that more than 4.6 million annual deaths of children occur in Africa due to malnutrition.

Ethiopia is one of the countries in the Sub-Saharan Africa with the highest rates of malnutrition in children (Taylor, 2012). Under nutrition is currently the most wide spread and serious health problem of children. In Ethiopia, 40 and 9% of below five years children are stunted and wasted, respectively (CSA, 2014). A higher rate of child under five years mortality due to protein-energy malnutrition is also reported in Ethiopia (Government of the Federal Democratic of Ethiopia, 2008) and Sudan (Asma *et al.*, 2006). Furthermore, pocket studies on micronutrient deficiencies indicate higher prevalence of iron and zinc deficiency especially in young children.

In general, rural children and children of uneducated mothers are more likely to be stunted, wasted, or underweight than other children (CSA and Macro, 2006). Regional variation in nutritional status of children is substantial. Stunting levels are above the national average in Amhara and SNNP. Wasting is higher than the national average in Somali, Benishangul-Gumuz, Amhara, Tigray and Dire Dawa. The percentage of underweight children is above the national average in Somali, Amhara, Tigray and Benishangul-Gumuz (CSA and Macro, 2006).

2.2. Complementary Foods and Problems during Complementing

Complementary foods are foods other than breast milk that is given to a breastfeeding child (Agostoni *et al.*, 2008). WHO (2001) also states that any nutrient containing foods or liquids other than breast milk given to young children during the period of complementary feeding are defined as complementary foods and the period during which other foods or liquids are provided along with breast milk is called the complementary feeding period. The transition from milk to solid or adult food is a critical period in the life of a child as the complementary feeding practices by the mother profoundly determines the child's growth and development (Olorunfemi *et al.*, 2006). This period which could start from four to six months of age, varies from one socioeconomic status to the other.

The ability of breast milk to meet the requirements for macronutrients and micronutrients becomes limited with increasing age of infant. The use of local foods formulated in the home and guided by the following principles: high nutritional value to supplement breastfeeding, acceptability, low price, and use of local food items is one of the major strategy to overcome

child malnutrition and death (Dewey and Brown, 2003; Pelto *et al.*, 2003). In order to solve these nutritional problems, several efforts have been made to formulate complementary foods from local food materials; however, it is evident recently that there is heavy reliance on most of these food materials thereby leading to inaccessibility by many mothers as a result of the cost of the materials and mode of production processes (Ijarotimi, 2008). Thus most families have to depend on the local material and technology for the preparation of complementary foods.

Infant foods in many parts of Ethiopia are cereal or root crop-based because these are major components of the family's diet. Timely introduction of complementary foods during infancy is necessary for both nutritional and developmental reasons, and to enable the transition from milk feeding to family foods. It also has effect on growth and neurodevelopment. Complementary food should not be introduced in any infant before 17 weeks and all infants should start complimentary feeding by 26 weeks.

One of the major nutritional problems faced by young children in African during the complementary feeding period is that complementary foods (which are commonly prepared in the form of porridges) made from the major staple foods (mainly cereals and roots and tubers) have low nutrient density, mainly because they are high in unmodified starch and low in fat. A young child must consume a large volume of this porridge if its energy and other nutrient requirements are to be met during the period when breast milk alone is insufficient. Many children are unable to eat such quantities mainly by virtue of their small stomachs, resulting in insufficient intakes of energy, protein, and other nutrients (Badau *et al.*, 2006).

Traditional complementary foods are typically watery gruels of low energy density and protein content. Often they are not consumed immediately after preparation. Unhygienic conditions of preparation and storage may lead to infection with enteropathogenic bacteria. Infants in the complementary feeding period are potentially at risk in developing countries, and many nutritional problems arise with the introduction of solids.

The preservation process, poor hygiene sanitation and inadequate knowledge of complementary food preservation introduce the risks of gastrointestinal and parasitic infection because of the heavy contamination of foodstuffs with infecting organisms.

In addition, too early introduction of complementary foods may lead to diarrhoea through the ingestion of thin, contaminated feed with insufficient calorie and protein. Too late introduction may lead to under nutrition owing to insufficient milk intake.

2.3. Recommended Dietary Allowance for Infants and Children

Good nutrition is the key to ensuring that growth and development proceeds optimally. Infants require all the essential vitamins and minerals and also need adequate amounts of calories and protein. The Recommended Dietary Allowance (RDA) is an estimate of the minimum daily average dietary intake level that meets the nutrient requirements of nearly all 97 to 98 percent healthy individuals in a particular life stage and gender group. The RDA is intended to be used as a goal for daily intake by individuals as this value estimates an intake level that has a high probability of meeting the requirement of individual about 97.5 % (Food and Nutrition Board, 2005).

Table 1: Recommended dietary allowance for infants and children per day

Nutrient	0-6 Months	7-12 Months	1-3 Years
Energy (calories)	520-570	676-743	992-1046
Protein (g)	9.1	13.5	13
Fat (g)	31	30	Nd
Carbohydrate (g)	65	90	130
Vitamin A (μg RE)	400	500	300
Calcium (mg)	200	260	700
Iron (mg)	.27	11	7
Zinc (mg)	2	3	3
Iodine (μg)	110	130	90

Source; Food and Nutrition Board, Institute of Medicine, 2005

2.4. Nutritional Composition of Ingredients Used in Extruded Composite Flour

Cereals and legumes are the two most important flowering plants used in agriculture. Most cereal grains are higher proportionally in the sulfur bearing amino acids. When soy is blended with cereals, the resulting balance of sulfur bearing amino acids with the other essential amino acids is substantially improved. Popelka *et al.* (2004) reported that lysine which is very high in soy is low in most cereal grains. Thus, the final amino acid balance of mixture of soy with other products generally results in an improved protein nutritional value of soy and a far superior nutritional value of the other products with soy.

According to Omima *et al.* (2010), legume seed grain proteins are the natural supplement to cereal grain protein in producing and overall essential amino acid balance. The protein quality cereals can be improved by combining it with other rich sources of protein (Popelka *et al.*, 2004). Oilseeds are leading suppliers of superior quality and specialty vegetable oils to nutritional products, natural food and premium snack food worldwide.

2.4.1. Cereals

Cereals are widely utilized as food in African countries than in the developed world (Makinde and Ladipo, 2012). Compositionally cereals consist of carbohydrate and less in amount of protein and essential minerals. Cereal grains are considered to be one of the most important sources of dietary proteins, carbohydrates, vitamins, minerals and fiber for people in developing countries. However, the nutritional quality of cereals and sensorial properties of their products are sometimes inferior or poor in comparison with milk and milk products. This is because cereal is deficient in certain essential amino acids (*i.e.*, lysine and tryptophan), and additionally is characterized by low starch availability, presence of anti-nutrients (phytic acid, tannins and polyphenols) and the coarse nature of the grains (Vasal, 2001).

Oat

Oat is belonging to the *Poacea* family (Butt *et al.*, 2008). Oats have been used in the Nordic diet for almost two thousand years. However, they have been replaced more and more by other cereals since the beginning of the 19th century. Nowadays oats have regained a new interest due to its health-promoting effects (Duss and Nyberg, 2004).

Oats are a difficult raw material for bread making due to the absence of gluten compared for example to wheat. Therefore, oats are often used in porridge, breakfast cereals or in baking blended with wheat flour (Butt *et al.*, 2008). Oat products are consumed as ingredients in baked foods or in porridge for many years due to their high percentage of nutritious protein (Rzedzicki *et al.*, 2000).

In comparison to other cereals, these are characterized constitute large amount of total protein, carbohydrate (primary starch content), crude fat, dietary fiber (non-starch), unique antioxidants and considerable vitamins and mineral content. Oats are the source of low cost protein with a protein content of 15-20% (dry matter basis) in dehulled oat grain. Moreover, people suffer from coeliac disease can tolerate oat protein due to the absence of gluten (Ahokas *et al.*, 2005). Nowadays the interest of using oats in designing food products is increasing due to the high amounts of soluble fiber, i.e., β -glucan (Mohamed *et al.*, 2009).

Oat β -glucans are able to influence lipid mechanism in human organisms and lower the total level of cholesterol in blood. Several studies have showed that food products containing oat β -glucan can have a cholesterol-lowering effect (Wood *et al.*, 2007). Oat β -glucans not only decrease the total blood cholesterol, but also decrease the low-density lipoprotein (LDL) cholesterol and increase high-density lipoprotein (HDL) cholesterol. LDL is often termed as bad cholesterol, while HDL is regarded as good cholesterol. Due to such health promoting properties, oat products have gained popularity to become as a part of human diet either as whole or as a fractionated form mostly as ready-to-eat (RTE) breakfast cereals, snack foods or bread (Mirmoghatadaie *et al.*, 2009). Snack foods have become a part of the lifestyle especially in the Western countries and cereal grains are generally used as their raw materials. Oats are generally considered healthy, being touted commercially as nutritious which has led to wider appreciation of oats as human food.

A good taste and an activity of stimulating metabolic changes in the body make nutritive value of oats high for both human and animals (Peltonen-Sainio *et al.*, 2004). Total carbohydrate content (including cellulose and non-starch polysaccharides) may reach 75-80% of the dry matter. The amylase content of oat starch ranges 16-18% to 28.5–28.7% (Mirmoghtadaie *et al.*, 2009).

Oats contain a high percentage of protein and balanced composition of amino acids which have proved them highly nutritive in comparison to other cereals (Petkov *et al.*, 2001). Highest protein content occurs in oat grout (12.4 to 24.5%) among cereals. Relatively high protein levels occur in the bran (about 20%) and lower in hull (less than 2%).

Table 2: Nutritional composition of whole grain oat and oat bran per 100gm

Nutrients	Whole grain oat	Oat bran
Protein	15 - 17	15 - 18
Starch and sugars	59 - 70	10 - 50
Fat	4.5	6.5
Total dietary fiber	12	14-15
Ash	3.5	2.4
β-Glucan	2 - 6	5 - 20
Cellulose	14	2.5
Lignin	2.4	4.5

Source: (Usman *et al.*, 2010)

A high lipid content of oats makes them different from other cereals and is a good source of essential unsaturated fatty acid. The percentage varies from 3.1 to 10.9% but in some high oil varieties it may be as high as 15% (Peterson and Wood, 1997). Triglycerides constitute the main component of lipids and phospholipids, glycolipids, sterols are also present in considerable quantities.

Dietary fibers (non-starch polysaccharides) also known as roughage or bulk are the edible parts of the plants and essential constituents of human nutrition. Although they are not a nutrient, they are nevertheless an important component of our diets. Ingested dietary fiber moves along into the large intestine where it is partially or completely fermented by gut bacteria.

Oats comprises a very balanced profile of both soluble and insoluble dietary fibers. A high intake of dietary fiber is positively related to several preventive medical and nutritional effects. Dietary fiber complex with its antioxidants and other phytochemicals is most effective against cardiovascular disease and some types of cancer, lowering lipid levels (Slavin *et al.*, 2000). Mineral content which is 2-3% in oat include phosphorus, potassium, magnesium, and calcium as main components as in other cereal.

Table 3: Mineral composition of oats in (100 gm)

mineral	amount (mg)
Calcium	53.85
Iron	4.78
Magnesium	179.22
Phosphorus	529.87
Potassium	434.42
Sodium	1.95
Zinc	4.02
Manganese	4.98

Source: (USDA, 2005)

2.4.2. Legumes

On a worldwide basis, legumes contribute about one-third to human direct protein intake, while also serving as an important source of fodder and forage for animals and of edible and industrial oils. Grain legumes or pulses (including soybeans) are rich and low-cost sources of dietary proteins and nutrients for a large part of the world's population. Legumes play an important role in human nutrition as they are rich source of protein, calories, certain minerals and vitamins (Baloch and Zubair, 2010).

It is well documented that cereal proteins are deficient in certain essential amino acids, particularly in lysine (Anjum *et al.*, 2005) whereas legumes contained adequate amount of lysine (Sai-Ut *et al.*, 2009). It is advisable to enhance the protein content of the diet through easily available and accessible plant protein sources especially legumes to improve the nutritional status of the low-income groups of population (Khattab and Arntfield, 2009).

Soybean

The soybean (*Glycine max*) is one of the most important food plants of the world, and seems to be growing in importance. It is an annual crop, fairly easy to grow, that produces more protein and oil. It is a versatile food plant that, used in its various forms, is capable of supplying most nutrients. Soybean protein quality has been the subject of intense investigation for several decades due to soybean's increasing importance as human food resource.

Soybean is often called the “miracle bean” because of its chemical composition and diverse applications for food, feed, and non-food uses. It has an exceptionally high content of both protein and fat (Kim *et al.*, 2008). The oil and protein content together account for about 60 % of the weight of dry soybean flour; protein 37 % and oil 22 % (Kim *et al.*, 2008). The protein and oil component of soybean are not only high in terms of quantity but also in quality. Soybeans is an excellent source of protein (about 35-40%), hence the seed is the richest in food value of all plant foods consumed in the world, which provides all the essential amino acids in the amounts needed for human health (Kure *et al.*, 1998).

Most of the essential amino acid present in soybean is available in an amount that is close to those required by animals and humans. The protein – digestibility – correlated amino acid score is close to 1, a rating that is the same for animal proteins such as an egg white and casein. Soybean is a very rich source of vegetable protein for all including growing children (Dandago and Igwe, 2006); and it has been identified as a suitable protein rich crop that could improve the nutritional and economic status of the general population in developing countries (Babajide *et al.*, 2003).

The amino acid profile of soy protein complements that of cereals (wheat, rice, or corn). Cereals, the main food staple consumed globally contain insufficient quantities of the amino acids lysine, tryptophan, and threonine. Soy protein is particularly valuable because its amino acid composition complements that of cereals. Soybeans are limiting in the sulfur containing amino acids, cystein and methionine, but contain sufficient lysine to overcome the lysine deficiency of cereals (Bair and Snyder, 1980). Soybeans have great potential in overcoming protein-calorie malnutrition. Although soybean is not indigenous to Africa, it has received tremendous popularity as a cheap protein source in developing countries (Nwabueze, 2007).

In addition to excellent nutritional properties, soy protein provides superior “functional “properties for processing. Soy proteins can be processed to become excellent water binders and fat emulsifiers. These properties are especially important when soy protein products such as textured soy protein, soy concentrates, and soy isolates are added to extend or improve the quality of meat products.

Soybeans have been processed in a number of ways, but a more recent processing method is extrusion cooking (Iwe and Ngoddy, 2000). It is a cost-effective processing method that is now widely used to improve the nutritive value of legumes, primarily as a means of reducing the levels of heat-labile, non-nutritive compounds. Baraga *et al.* (1985) indicated high extrusion temperature can affect the molecular structure of tannins and polyphenols. This chemical modification may alter solubility of tannin or chemical reactivity. Extrusion cooking at 150°C and combined effect of other variables on ANFs were reported (Anuonye *et al.*, 2010).

2.4.3. Oilseeds

Oilseeds are leading suppliers of superior quality and specialty vegetable oils to nutritional products, natural food and premium snack food worldwide. The oil content of small grains for example, wheat is only 1 to 2%, but that of oilseeds ranges from about 40% for sunflower and rapeseeds like canola. Seed oils from flax (linseed) and castor bean are used for industrial purposes. They do not contain an appreciable amount of carbohydrate, but, contain high level of B vitamins.

Oilseeds are energy dense foods; although oilseeds contain protein (14– 32 g/100 g) and carbohydrate (ranging from less than 1 g/100 g to more than 34 g/100 g), most of the food energy they provide is as fat (which provides 9 kcal or 37 kJ/g). Oilseeds vary widely in their fatty acid composition but tend to be rich in MUFA. Generally, oilseeds are a source of fiber, phosphorus, iron and magnesium; many oilseeds are also a source of vitamin E (an antioxidant), niacin and folate.

Flaxseed (linseed)

Flaxseed, or Linseed (*Linum Usitatissimum*), is a blue flowering crop and a member of family Linaceae. Globally, Flaxseed is grown as either oil crop or a fiber crop with fiber linen derived from the stem of fiber varieties and oil from the seed of linseed varieties (Vaisey-Genser *et al.*, 2003). The plant is native to west Asia and the Mediterranean. As the source of linen fiber flax has been cultivated since at least 5000 BC, today it is mainly grown for its oil.

Beyond its oilseed crop ability, proximate composition of flaxseed makes it more promising for its utilization in different food products. Flaxseed is one of the richest vegetarian sources of α -linolenic acid (omega 3 fatty acid) and soluble mucilage. Alpha linolenic acid cannot be synthesized by the human body from any other substance therefore it is considered as an essential fatty acid. The essential fatty acids requirements for the human body can be fulfilled by intake of flaxseed products (Morris, 2004).

In present era, consumer's trend towards functional food has increased significantly as health awareness rose. Flaxseed can be one stop for novel high quality source of nutrition. In the World's food supply, flax is making its mark as a functional food. Flax confers its health benefits due to the presence of α -linolenic acid (ALA), the essential ω -3 fatty acid, and phytochemicals such as lignans (Morris, 2004). Flaxseed oil and canola oil have the lowest levels of the nutritionally undesirable saturated fatty acids. The level of the desirable monounsaturated in flax oil is modest.

The major nutritional components of flaxseed include oil, viscous lignan-rich fibres (mucilage), protein and minerals. A100g portion of flaxseed provides 534Kcal energy and contains approximately 7% carbohydrates, 10% protein, 53% total fat and 21% dietary fat. Flaxseed is high in most of the B vitamins, magnesium (Mg) and manganese (Mn) (Dolson, 2010). About 73% of the fatty acids present in flaxseeds are polyunsaturated fatty acids (Madhusudhan, 2009). Flaxseed is a source of good-quality protein and albumins and globulins are the storage proteins of flaxseed with globulins forming the highest portion (58-66% of the total seed protein) (Chung *et al.*, 2005).

2.4.4. Premix

Premixes are combination of different ingredients added in order to meet recommendations for vitamin and mineral contents and confer suitable organoleptic characteristics to the flours. The formula calculations were made with data from food composition tables (Mouquet *et al.*, 2003).

2.4.4.1. Moringa (*Moringa stenopetala*)

Moringa is a tropical plant belonging to the family *Moringaceae* that grows throughout the tropics. Moringa is a multipurpose tree of significant economic importance, as it has vital nutritional, industrial, and medicinal applications. Moringa was domesticated in the east African lowlands and is indigenous to southern Ethiopia. Many different ecotypes and varieties of moringa are found in Ethiopia. It is native to Ethiopia. Moringa is an important indigenous vegetable in south western Ethiopia where it is cultivated as a food crop. The Gofa, Konso, Burji, and Gamo tribes consume its leaves as a vegetable, especially during the dry season (Abuye *et al.*, 2003).

Moringa is native to Ethiopia, (“Haleko” in Gofa areas, “Shelagda” in the Konso language, and “*Shiferaw*”) in Amharic (Seifu, 2015). Moringa is particularly important as human food because the leaves have high nutritional value (high amounts of essential amino acids and vitamins A and C) (Abuye *et al.*, 2003).

Moringa is a favorite and main component of the daily meal of the Konso, Gamo, and Gofa people in southern Ethiopia (Endeshaw, 2003; Personal observation). In the Konso area, moringa leaves are eaten almost every day like spinach together with cereal balls. It was reported that about 50% of the people in the Konso district of southern Ethiopia get their food from moringa (Endeshaw, 2003).

Moringa tree has both nutritional and medicinal values. The leaves, flowers, and green pods of Moringa are eaten as a staple vegetable and are rich in proteins, Ca, Fe, and P. Moringa leaves are rich in protein source, which can be used to solve worldwide malnutrition or under nutrition problems (Thurber and Fahey, 2009). Moringa leaves contain seven times the vitamin C of oranges, four times the vitamin A of carrots, four times the calcium of milk, three times the potassium of bananas, and two times the protein of yoghurt (Mathur, 2005).

Moreover, moringa leaves contain all the essential amino acids (Mathur, 2005; Melesse *et al.*, 2009) and vitamins A and C among others (Mathur, 2005). Hundred grams of dried leaves contain 27.1 g protein, 16.3 mg vitamin A, 17.3 mg vitamin C, 2.0 g calcium, 1.3 g potassium

and 28.2 mg iron, in addition to 19.2 g dietary fiber and several other nutrients.(Fuglie, 2001).

Vitamins are present at nutritionally significant levels averaging 28 mg/100 g of vitamin C and 16 mg/100 g of beta carotene (Abuye *et al.*, 2003). Among the wide range of green leafy vegetables, Moringa is the richest source of beta-carotene (vitamin A) and provides other important micronutrients (Mathur, 2005). Minerals such as K, Fe, Zn, P, and Ca also exist in significant concentrations with average values of 3.08 mg/100 g iron and 792.8 mg/100 g Ca (Abuye *et al.*, 2003). Reports indicated that high vitamin content of moringa leaves could be used to reduce child and maternal mortality rates by 30-50% (Seifu, 2015).

2.4.4.2. Fenugreek (*Trigonella foenum-graecum* L.)

Fenugreek belonging to the family *Leguminosae* is an annual legume mainly used as a spice crop in many parts of the world. Fenugreek is an old medicinal plant and has been commonly used as a traditional food and medicine. It is locally used as a pulse, spice and medicinal plant, and has a long history in Ethiopia (Gall and Zerihun 2009).

Fenugreek seed can be utilized for value addition of cereals based food products to attain multiple benefits. It is used as a condiment and as a supplement to wheat and maize flour for bread making and as a constituent of the daily diet (Uhl, 2000). Adding fenugreek fiber to refined flours helps to fortify with a balance of soluble and insoluble fiber.

Flour fortified with 8–10% fenugreek fiber has been used to prepare bakery foods like pizza, bread, muffins, and cakes with acceptable sensory properties (Srinivasan, 2005). Commercially, fenugreek has been utilized in the food industry as stabilizer, emulsifier, thickening agent, spice, supplement of wheat and rice flour, beverage, coffee substitute, and tea.

In Ethiopia, it is known as “*Abish*” (in Amharic) and has been considered as medicinal plant to lower blood glucose level, treat gastric disease and abdominal discomfort, boost milk production in lactating mothers, and to increase body weight. It is also used, for softening and preserving of “*Ingera*”, infant food preparation, food blend, as beverage, spice.

The health beneficial effects of fenugreek are attributed to its nutritional composition (3–5% moisture, 25–30% protein, 20–30% galactomannan, 20–25% insoluble fiber, 7–9% lipids, 5–7% saponins and 3–4% ash) that include mucilaginous fiber, lysine-rich protein, free amino acids, saponins, flavonoids, and volatile oils (Raju and Bird, 2006). About 20% of fenugreek seed is gel-forming soluble fiber, similar to guar gum, oat bran and psyllium husk. The insoluble fiber, which constitutes 30% of fenugreek seed, is bulk-forming like wheat bran (Mathur and Chaudhary, 2009; Srinivasan, 2005).

The seeds of fenugreek contain lysine and L- tryptophan rich proteins, mucilaginous fiber and other rare chemical constituents such as saponins, coumarine, saponins and trigonelline, which are thought to account for many of its presumed therapeutic effects, may inhibit cholesterol absorption and thought to help lower sugar levels (Bukhari *et al.*, 2008). Fenugreek is reported to have restorative and nutritive properties and to stimulate digestive processes (Khosla *et al.*, 1995).

Fenugreek seeds have also been reported to exhibit pharmacological properties such as anti-tumor, anti-viral and antioxidant activity. However, the seeds are bitter in taste due to presence of saponins which limit their acceptability in foods. It has been possible to debitter fenugreek seeds by employing various processing methods such as soaking, germination, roasting etc.

Traditional preparation methods may affect reduction in the bitterness of the seeds and make possible its incorporation in various recipes. Earlier studies reported that sprouting or overnight soaking; washing of fenugreek seeds in running water and roasting removes the bitterness to a certain extent and makes possible its use in increased quantities for incorporation into various preparations which are commonly consumed. The beneficial effects of processing of fenugreek seeds may be attributed to an increase in low methoxy salts of calcium and magnesium as well as proto-pectin.

2.4.4.3. Salt and sugar

The presence of salt (sodium chloride) primarily contributes to the improvement of flavor and iodine contents of the product. Added salt to food improves the sensory properties of virtually

every food that humans consume, and it is cheap. There are many reasons for adding salt to foods. The main reason is that, in many cases, added salt enhances the positive sensory attributes of foods, even some otherwise unpalatable foods; it makes them “taste” better (Fu, 2008).

2.5. Extrusion

Extrusion is defined as "shaping by force through a specially designed opening often after previous heating of the material" (Harper, 1981). Extrusion technology has created a huge impact in the food industries towards shaping and deriving ready to eat products (Fellows 2009). The use of extrusion in the food processing has increased its popularity due to its versatility, cost-effectiveness, environmental friendliness and better product output (Guy 2001a).

Extrusion cooking has been used in a large number of food applications as it has some unique positive features compared with other heat processes. Nowadays, the food extruder is considered as a high-temperature short-time bioreactor that transforms raw ingredients into a variety of modified intermediate and finished products. During the extrusion process, the material is treated not only by heating, but also by intense mechanical shearing, compression and torque, which are able to break the covalent bonds in biopolymers (Singh *et al.*, 2007). Thus, the functional properties of the food ingredients are rapidly modified due to the combined influence of temperature, pressure, shear and time (Carvalho and Mitchell, 2000). Food extrusion also permits inactivate the undesirable enzymes that can affect the quality and eliminate several anti-nutritional factors, such as trypsin inhibitors, haemagglutinins, tannins and phytates (Singh *et al.*, 2007).

The general differences between the can be single or twin screw extruders. The single screw extruders are classified into, low shear forming extruder, low shear cooking, medium shear cooking and high shear cooking single screw extruders. The size and shape of the extrudates and efficiency of the extruder performance are interdependent on the operational parameters like temperature, pressure and screw speed (Fellows, 2009).

The advantage of using twin screw extruders is versatility to process wide range of products like tortillas, cereal snacks, extruded corn snacks, and multigrain snacks. However due to high capital and maintenance costs, single screw extruders are considered to be cost-effective when compared to twin screw extruders.

2.5.1. Processing parameters affecting extruded products

The extrusion process involves the loading of raw materials in the feeding hopper where the screw conveys through the raw materials. When the raw materials pass down the barrel, the volume is reduced and thereby the food is compressed under pressure into a semi-solid, plasticized mass. At that stage, the dough must have attained the ideal form to create a structure in the extruded product. This transformation of the native raw materials has an optimum range for most product types. For some products, only small changes in native biopolymers are required, whereas for others a very high degree of change must be achieved. For each individual product type, the process must be maintained in a balance between the independent input variables to obtain the ideal conditions within the barrel to achieve this level of transformation (Guy, 2004).

In the confined space of the barrel, the dough is compressed and heated to high temperatures, while subjected to high pressures, before being extruded through the dies into the atmosphere (Guy, 2004). The selection of right extruder for the production of ready to eat (RTE) or cereal snacks depends on the nature of raw materials used, bulk density and type of product to be produced (Fellows, 2009).

The residence time in the extrusion plays an important role in the performance of the product which can be controlled by screw speed. The twin screw extruders, they are classified based on degree of co-rotation and the degree of interconnection between the two screws (Fellows, 2009). The twin screw extruders are also classified into; Co-rotating intermeshing, Co-rotating non-intermeshing, Counter-rotating intermeshing and 4 Counter-rotating non-intermeshing twin-screw extruders.

The processing parameters play important role in determining the quality output of the extruded products. The process controlling of the product depends on various primary and

secondary extrusion process parameters. The primary process parameters include feed rate, screw speed, barrel temperature, moisture content, feed formulation and die configuration. The secondary process parameters include die temperature, pressure and torque (Chessari and Sellahewa 2001).

The pre-conditioning treatment of the raw materials with the help of hot water or steam for about 4-5 minutes helps in gelatinization of starch and protein denaturation of the raw materials during the extrusion processing (Bailey *et al.*, 1995). With respect to the extrusion parameters, processing temperature, water content in the feed and shearing rate plays an important role in the expansion of extrudates during extrusion processing (Guy 2001b). Due to pronounced starch-protein interactions and the cross linking of disulphide bonds in proteins upon high-moisture heat conditions (Mahasukhnothachai *et al.*, 2010), it can be concluded that conventional cooking may not improve nutritional quality of sorghum.

2.5.2. Physical and chemical changes during extrusion process

The changes occurring during the extrusion cooking plays a major role in determining the shape and crispness of the extrudates which is an important characteristic for cereal based snacks (Guy 2001b). One of the important phenomena during the process of extrusion is the process of gelatinization. The process of starch gelatinization helps in gas-holding capacities that result in expansion of extrudates (Guy 2001b).

2.5.2.1. Physical change during extrusion process

During the process of starch gelatinization, breakage of intermolecular hydrogen bonding results in the increase in the absorption of water resulting in swelling of starch granules (Fellows 2009). As the temperature gradually increases, starch molecules are gelatinized which results in the formation of viscous fluid melt. The fluid melt forms the outer coating for the foam bubbles that contain superheated water vapor. During the exit of materials from the extruder die, there is sudden drop in pressure which results in expansion of bubbles by loss of moisture by the process of evaporation. These physical changes during the extrusion process increases the viscosity of material followed by formation of glassy state depending on the degree of vaporization of water in the extrudates structure (Guy 2001a).

The expansion of the extrudates greatly depends on the content of amylose and amylopectin present in the starch granules (Guy 2001b). Higher content of amylose in the starch results in low viscous fluid melt thereby resulting in greater expansion of foods during the extrusion processing. Other process techniques such as germination and fermentation were also found to improve protein digestibility (Correia, *et al.*, 2010). On the other hand, extrusion cooking is an established technique gaining importance in the production of several snack and other ready-to-eat foods.

Advantages of extrusion cooking include high throughput, low energy consumption, absence of process effluents and high efficiency when it comes to raw material selection, their texture and shapes (Harper, 1981).

Key functions such as agglomeration, degassing, dehydration, expansion, homogenization, mixing, pasteurization, protein-denaturation, shaping, shearing, texture alteration, thermal cooking and unitizing are few conditions that are generated by a food extruder for use in a variety of food uses that include food, feed and industry applications.

2.5.2.2. Effect of extrusion on the nutritional properties of extrudates

The bioavailability of nutrients during the processing of foods is always considered important when obtaining a nutritional product. The advantages of extrusion cooking with respect to the nutritional content of the final product are the inactivation of antinutrients, destruction of aflatoxins and increasing the digestibility of fiber (Singh *et al.*, 2007; Saalia and Phillips 2011).

The denaturation of proteins during the extrusion processing caused inactivation of antinutrients such as lectin and antitrypsin inhibitors resulting in the increase of protein digestibility (Kitabatake and Doi 1992).

During the process, disulphide bonds break and reunite, while the high molecular proteins dissociate into smaller subunits (Guy 2001a). During high extrusion processing with lower water content of the feed initiates non-enzymatic browning reaction termed as Maillard reaction. The nutritional effect of protein and amino acid availability was negatively affected by browning and caramelization involving proteins and sugars (Singh *et al.*, 2007).

The addition of antioxidants (e.g. phenolics) also reduced the effect of lipid oxidation in the extrudates and thereby resulting in the better retention of nutritional properties (Camire *et al.*, 2005). The process of extrusion cooking at higher extrusion temperatures also enhanced the process lipid oxidation in extruded corn based snacks (Zadernowski *et al.*, 1997). There are also certain effects of extrusion in relation to dietary fiber content. Increase in the total dietary fiber content of the extruded barley flours was determined with respect to content of soluble dietary fiber. Effect of extrusion on the dietary fiber content led to the transformation of insoluble dietary fiber to the soluble dietary form in addition to the formation of resistant starch and enzyme resistant glucans through the process of transglycosidation (Vasanthan *et al.*, 2002).

Vitamin losses were also reported in the foods that were produced through extrusion. α tocopherol content in the extruded peas decreased with an increase in the extrusion temperature (Grela *et al.*, 1999). Milder extrusion temperatures (150 °C) and short residence time resulted in better retention of heat-sensitive vitamins (vitamin B1, B2). Singh *et al.* (2007) summarized that the heat-sensitive vitamins were lost during extrusion. Athar *et al.* (2006) reported that there was 44-62% retention of B vitamins in snacks during the extrusion processing of cereals and resulted in higher stability of riboflavin (vitamin B2) and niacin (vitamin B3). Absorption of minerals can be enhanced by the process of extrusion (Alonso *et al.*, 2001).

2.5.2.3. Effect of extrusion on the antinutritional properties of extruded products

Traditional artisanal technologies such as decortication, soaking, germination and fermentation of cereal-based foods reduce the levels of tannins and phytates, increase bioavailability of amino acids and mineral elements and improve protein and starch digestibility (Lorri and Svanberg, 1993). But these technologies are limited by their laborious and time demanding nature.

Extrusion of legumes at higher temperature (180 °C) and water content (22%) helped in the complete elimination of trypsin inhibitors in the extrudates (El-Hady and Habiba 2003). Soaking of beans and peas for a period of 16 hours followed by extrusion processing resulted in better elimination of antinutrients in the extrudates (El-Hady and Habiba 2003).

The extrusion of wheat, rice and barley at 140 °C and water content (20%) resulted in more than 50% reduction in the content of phytates, trypsin inhibitors and oxalates in the extruded cereal snacks (Kaur *et al.*, 2013). Elimination of protease inhibitors can be successfully achieved by the process of extrusion at higher temperatures while the complete inhibition of gossypol can be achieved by increasing the water content of the feed during the extrusion processing.

Table 4: Effect of extrusion on antinutrients contents of different crops

Material	Extrusion conditions	Phytates (mg/g m)	Trypsin inhibitors (U /mg d.m.)	Tannins (mg /100 g d.m.)	Oxalates (%)
Raw Faba beans		6.1	1.85	485	n.d.
Extruded Faba beans	T' 180°C, Wc - 22%	4.8	*	362	n.d.
Raw Peas		8.5	13.7	2	n.d.
Extruded Peas	T' 180°C, Wc - 22%	7.6	*	200	n.d.
Raw Wheat		35.9	46.7	n.d.	0.4
Extruded Wheat	T' 140°C, Wc - 20%	16.2	13.4	n.d.	0.2
Raw Rice		36.8	46.3	n.d.	0.4
Extruded rice	T' 140°C, Wc - 20%	16.3	12.5	n.d.	0.2
Raw Barley		34.7	43.3	n.d.	0.3
Extruded Barley	T' 140°C, Wc - 20%	13.3	17.7	n.d.	0.2

* - negligible amounts, T' - temperature, Wc -water content

N.d.-not determined

Source: (Kaur *et al.*, 2013, El-Hady and Habiba 2003)

2.6. Instant Flour

The global lifestyle, which is characterized by limited free time and increased working hours, have turned consumers to the consumption of ready-to-eat products. In addition, children worldwide are attracted to several snack products which are particularly tasty and easy to be consumed. With a growth rate of 12-15 % per year, food market is gaining general acceptance throughout the globe (Prescha *et al.*, 2012). Therefore, food industries have increased the production of ready-to-eat products using several processes. This has obviously presented a huge opportunity for the food industry to develop and market convenience food products (Barbet, 2012).

The Food Standards Agency (FSA, UK) defines ready-to-eat products as: ‘any food for consumption without further heating or processing’. This definition covers both open and pre-wrapped ready-to eat products and is intended to apply whether the ready-to-eat food may be consumed hot or cold. The expression ‘further heating or processing’ is not intended to include food preparation activities such as light washing, slicing, chopping, portioning, marinating or preservation carried out by the consumer by way of preference to an otherwise ready to eat food item.

Various technologies are being used to develop “Instant foods” and extrusion is an important technology for processing grain-based products and adds immense value to raw material ranging from corn, wheat and rice to sorghum, oats and soybeans.

Low cost and nutritious instant foods may be developed using major crops, which may be blended or supplemented with each other to provide complete nutrition. Processing of blends of cereals with legumes and oilseeds into forms which combine the advantage of nutritive value and convenience of use stands a better chance of success. Ready-to-eat (RTE) foods are increasingly popular with the consumer predominantly due to their convenience of consumption and ease of preparation and storage and consumer appeal factors such as convenience, value, attractive appearance and texture (Harper, 1981).

3. MATERIALS AND METHODS

3.1. Description of the Study Site

The study was conducted at Jimma University College of Agriculture and Veterinary Medicine (JUCAVM), Post-harvest management laboratory. JUCAVM is located in Jimma zone, Oromia regional state in southwest Ethiopia. The experiment on the extrusion process was conducted in the food processing laboratory of the School of Chemical and Food Engineering, Faculty of Engineering, Bahir Dar University. Protein, fat, fiber, and phytate analysis were done at Ethiopian Public Health Institute (EPHI) Addis Ababa and mineral analysis was conducted at Debre Zeit Agricultural Research Center. Other parameters were measured in JUCAVM's post-harvest management and soil laboratories.

3.2. Experimental Materials

About 35kg of unhusked oat grain *sinana one* variety was collected from Sinana Agricultural, Research Center, 10kg of soybean *crack 63K* variety was collected from Jimma agricultural research Center, 5kg of linseed, *Kulumsa one* variety was brought from Kulumsa Agriculture Research Center, *chala variety* of fenugreek and *moringa stenopetila* was brought from Debira Zeit Agricultural Research Center and Alagae agricultural technical and vocational education and training (TVET) college respectively to Jimma university. The selection of variety was based on its production and availability in the society. The criteria for selection of each ingredient are presented in (Table 5).

The materials used for this study were known variety of oat, soybean and linseed.

3.2.1 Oat

Oat was selected based on social acceptability as a complementary food (Tsegaye, 2015) and its nutrition composition. Oats are unique among the cereals; one of the rich sources of dietary fibers among cereals belongs to the Poaceae family (Butt *et al.*, 2008). Oat grain is characterized by unusual chemical composition (Butt *et al.*, 2008; Angioloni and Collar, 2012), because it contains a beneficial combination of nutritional compounds. Lipid content in oat grain is 2-3 times higher than in other cereals (Butt *et al.*, 2008; Angioloni and Collar, 2012). Moreover it is characterized by high protein content, which is a good source of aminoacids (Butt *et al.*, 2008; Gambuś *et al.*, 2011).

3.2.2. Soybean

Soybean was selected based on its nutritional composition and its production in the area. Soybean is an important source of high quality, inexpensive protein and oil. Soybean has the highest protein content of all food crops and is second only to peanut in terms of oil content (18%) among food legumes. Compared to other protein-rich foods such as meat, fish, and eggs, soybeans is by far the cheapest. It also has a superior amino-acid profile compared to other sources of plant protein (Saskia and Martin, 2008). Soybean is often called the “miracle bean” because of its chemical composition and diverse applications for food, feed, and non-food uses. It has an exceptionally high content of both protein and fat (Kim *et al.*, 2008).

3.2.3. Linseed

Linseed was selected depending on its dominant production in the area south west Ethiopia (availability of the ingredient in the area), their acceptance as complementary food of child in the area (Nejat un published data) and its nutritional profile. Linseed presents a nutritional and pharmacological value as a source of biologically active phytochemicals such as linolenic acid and lignans and is thus widely used as a dietary supplement (Attoumbéré *et al.*, 2011). The linseed contains approximately 40% oil, 30% dietary fiber, 20% protein, 6% moisture and 4% ash (Oomah Mazza, 2001).

3.2.4. Premix

Premix (sugar 9.9% & salt 0.6%) was selected based on literature (Mouquet *et al.*, 2003) and 1.5% fenugreek and 3% moringa were selected based on nutritional composition to fulfill vitamin A, mineral requirements and to improve flavor of composite flour. Premix was added to confer suitable organoleptic characteristics to the flour.

3.2.4.1. Moringa

Moringa was selected because of its nutritional composition, hundred grams of dried leaves of moringa contain 27.1 g protein, 16.3 mg vitamin A, 17.3 mg vitamin C, 2.0 g calcium, 1.3 g potassium and 28.2 mg iron, in addition to 19.2 g dietary fiber and several other nutrients (Fuglie, 2001). Some articles and research studies have reported that the dry leaves of moringa contain 7 times more vitamin C than orange, 10 times vitamin A than carrot, 17 times calcium than milk, 15 times potassium than bananas, 25 times iron than spinach and 9 times proteins than yogurt (Fuglie, 1999)

3.2.4.2. Fenugreek

Fenugreek is locally used as a pulse, spice and medicinal plant, and has a long history in Ethiopia (Gall and Zerihun 2009). Fenugreek seed can be utilized for value addition of cereals based food products to attain multiple benefits. It is used as a condiment and as a supplement to wheat and maize flour for bread making and as a constituent of the daily diet of general population. The fenugreek seed is very bitter but does have interesting proximate composition. Protein content ranges between 23 and 43% of the seed, carbohydrate represents up to 58%, moisture make up about 10 - 13% of the seed, lipid represent 5-6 % and minerals make up less than 1% (Elnasri, 2006).

3.2.4.3. Sugar and salt

Sugar was used to improve the taste of the product. Salt and sugar can be also used for preservative purpose of instant infant flour. Salt (sodium chloride) was added primarily to the improvement of flavor and iodine contents of the product. Adding salt to food improves the sensory properties of virtually every food that humans consume.

Table 5: Selection criteria of ingredients used for formulation

Ingredients	Reason of selection	Reference
Oat	Carbohydrate source; Socially accepted by the community for infant feeding, Good nutritional profile	Baseline survey Butt <i>et al.</i> , 2008
Soybean	Protein source ; used as ingredient of complementary food Soybean is a very rich source of vegetable protein for all including growing children	Baseline survey Dandago and Igwe, 2006
linseed	Fat source; Dominantly produced among oil seeds, sometimes used as infant complementary food A rich source of nutrients it finds its use in various food product formulations	Baseline survey Mantri <i>et al.</i> , 2012
	Sugar ; To increase acceptability of taste	Mouquet <i>et al.</i> , 2003
Premixes	salt ; Improve iodine contents of the formulation Enhances the positive sensory attributes of foods, it makes them “taste” better Moringa; Nutritional profile;	Mouquet <i>et al.</i> , 2003, Fu, 2008 Fuglie <i>et al.</i> , 2001
	Fenugreek; commonly used for seasoning purposes, in Ethiopia the seeds are prepared for infant feeding	Baseline survey, (Fazli and Hardman, 1968)

3.3. Sample Preparation

The procedures for samples preparation are presented below

3.3.1. Oat flour preparation

Oat flour was prepared according to the procedure of Hooda and Jood (2003). The oat was cleaned of dirt, stone and other impurities. The cleaned oat was dehusked by de husker (Hangyuhp20, Germany).

After the husk is removed it was sorted manually to obtain uniform size and to remove broken kernels and shrunken ones. The cleaned oat was milled into fine flour using grinding mill (Karl Kolb D-6072 Drrich. West Germany) following by sieving (0.5mm) to get uniform sized flour. The flour was then stored in polyethylene bags at 4°C for extrusion and further analysis.

3.3.2. Soybean flour preparation

Soybean flour was prepared according to Famurewa and Raji (2011). The soybean grains were hand sorted and cleaned to get similar grains and to discard the defective grains including other contaminants. The cleaned soybean was poured in to boiling water and boiled for 30 minutes. The hot water was drained out, and the boiled soybean was placed in basin of fresh water and agitated between fingers to remove the hulls. The dehulled soybean was oven dried at of 60°C for 13 hours. Dehulled dry soybeans were milled (Karl Kolb D-6072 Drrich. West Germany) followed by sieving (0.5mm) to produce fine flour. The flour was then stored in polyethylene bags at 4°C for further analysis.

3.3.3. Flaxseed (linseed) flour preparation

The linseed flour was prepared according to Chetana *et al.* (2010). Linseed was cleaned manually to remove dust particles, damaged seeds, seeds of other crops and other impurities such as weeds. The cleaned linseed was lightly roasted at 100°C for 15 minutes. It was continuously stirred with ladle for proper and uniform roasting until a nutty flavor was developed. The roasted linseed was milled (Karl Kolb D-6072 Drrich. West Germany) followed by sieving (0.5mm) to get full fat flaxseed flour. The resulting flour was stored in airtight polyethylene bag at 4°C for further analysis.

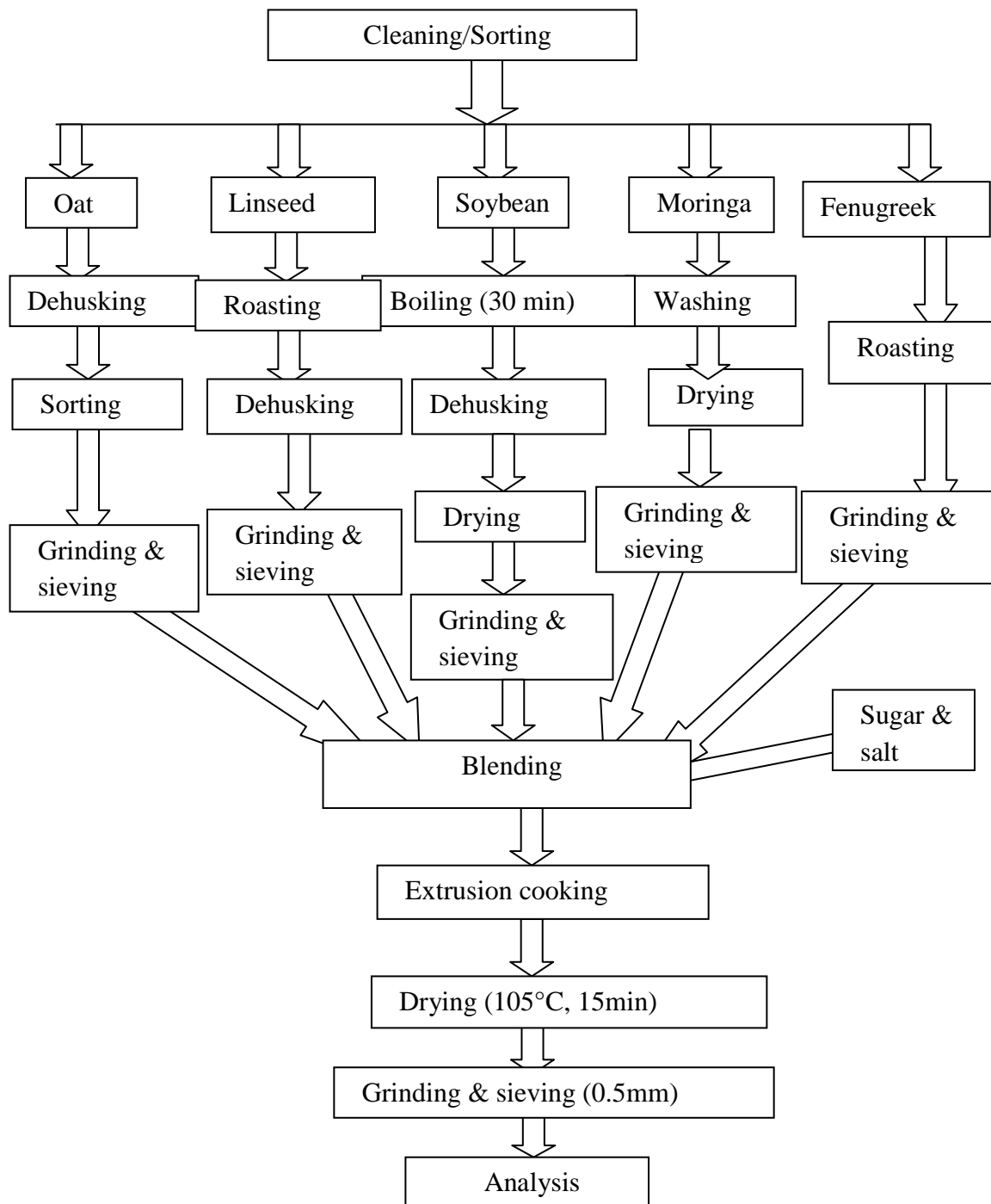
3.3.4. Fenugreek flour preparation

Fenugreek flour was prepared according to Hemlata and Pratima (2015). Fenugreek seeds was cleaned of any foreign material and defectives and roasted in an open pan at 103 ± 5 °C for 7 minutes to impart aroma generating compound. It was continuously stirred with ladle for proper and uniform roasting until it became slight brown and left a peculiar aroma.

The seeds were ground in grinder (Karl Kolb D-6072 Drrich. West Germany) and passed through sieve (0.5mm) to get uniform sized flour. The resulting flour was stored in airtight polyethylene bag at 4°C for further analysis.

3.3.5. Moringa leaf powder preparation

Moringa leaves were prepared according to methods of Gebretsadikan *et al.* (2015). Fresh mature moringa leaves were carefully harvested and the damaged and diseased leaves were discarded. The leaves were washed with clean water at 1:5 leaves to water ratio to remove dusts and pests, and subsequently drained on the draining table for about one hour before taking them to the drying room at ambient temperature and relative humidity. Dried leaves were milled using the grinding mill (Karl Kolb D-6072 Drrich. West Germany) followed by sieving (0.5mm) to produce fine flour. The powder was stored in airtight LDPE packages which also protect the flour from ultra-violet light and kept in dark place.



Source; (Hooda and Jood, 2003, Hemlata and Pratima, 2015, Famurewa and Raji 2011, Gebretsadikan *et al.*, 2015)

Figure 1: Flow diagram of sample preparation and processing

3.4. Experimental Design and Treatment Combinations

The experimental was designed design was with the aid of Minitab software version 16, D-optimal mixture design was used to generate the treatment combination; accordingly 13 runs were computer generated. The treatment combinations are presented in (Table 6).

Table 6: Treatment combination of oat, soybean, linseed and premix of composite flours

Run order	Oat	Soy bean	Linseed	Premix	100%
1	57.92	19.92	7.17	15.0	100.01
2	57.42	17.92	9.67	15.0	100.01
3	56.00	23.00	6.00	15.0	100.00
4	65.00	11.00	9.00	15.0	100.00
5	62.42	15.42	7.17	15.0	100.01
6	55.00	19.00	11.0	15.0	100.00
7	62.42	15.42	7.17	15.0	100.01
8	57.42	19.92	7.67	15.0	100.01
9	63.00	11.00	11.0	15.0	100.00
10	59.83	16.83	8.34	15.0	100.00
11	55.00	23.00	7.00	15.0	100.00
12	61.42	13.92	9.67	15.0	100.01
13	65.00	14.00	6.00	15.0	100.00

Premix (9.9% sugar, 3% moringa, 1.5% fenugreek and 0.6% salt)

Upper and lower constraints for each ingredient (Table 7) were determined based on data from food composition tables (Souci *et al.*, 2008) and literature. Premix was added to meet recommendations for vitamin and mineral and confer suitable organoleptic characteristics to the flours (Mouquet *et al.*, 2003). According to WHO/ FAO (2004), composite instant infant formula are expected to contain 10-15(g/100g) fat, greater than15 (g/100g) protein and 400-425 kcal) energy.

Table 7: lower and upper limits of ingredients in mixture design

Ingredient	Lower	Upper
Oat	55	65
Soybean	11	23
Linseed	6	11
Premix	15	15

The proportion of oat, soybean and linseed is converted to 85% in order to fulfill the requirements of protein and fat and the remains 15% premix. The experimental region and experimental points are presented in below in figure 2

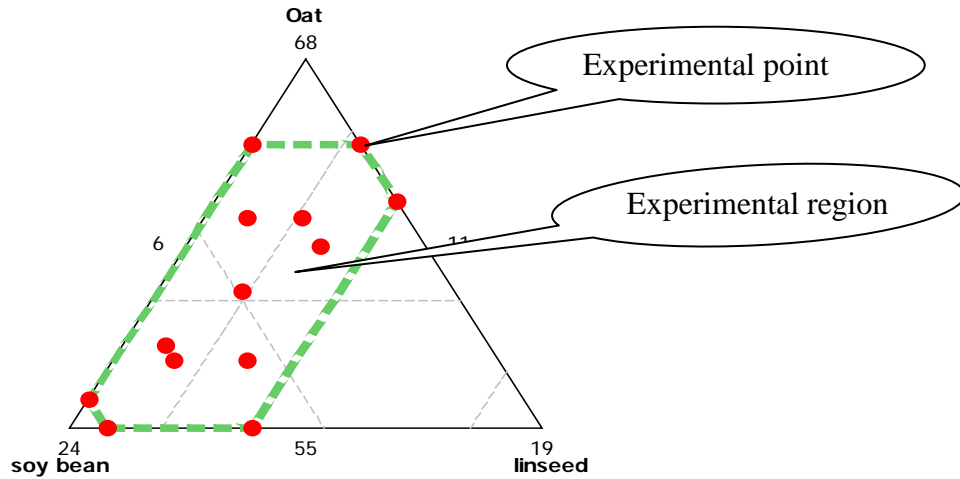


Figure 2: Simplex design plots for the 3-component mixture formulations

3. 5. Extrusion Processing

Before extrusion processing, the flours were mixed and homogenized for 5 minutes in a planetary cake mixer (H.LB20/B, Hungary). Extrusion process was conducted using a pilot scale co-rotating twin screw food extrusion cooker (model Clextal, BC-21 N0 194, Firminy, France). The Length to Diameter (L/D) ratio for extruder was 20:1. The diameter of the hole in the die was 6 mm with a die length of 27 mm.

Necessary calibration and adjustment of the material feed rate and water flow rate were performed prior to the main extrusion cooking process. The barrel temperature of third zone, which was located just before the die was allowed to operate at different temperatures starting of 100 ° C to 140 ° C, was adjusted based on literature.

By looking at the characteristics of the products from the extrusion, the barrel temperature which varied from 120°C to 130°C was selected for the experiment based on preliminary work. The moisture content of the material was adjusted by varying the water injection rate of the pump. The pump was adjusted to give moisture content of 17 and 20% using hydration

equation (Equation 1). Screw speed was set at 150, 200 and 250 rpm based on literature. The selected moisture and screw speed levels were also chosen by looking the product characteristics at different moisture and screw speed at pre-test experiments.

Finally the blend was extruded using a twin screw co-rotating extruder. The extruder was consisting of three independent zones of controlled temperature in the barrel, 70, 100 and 130°C, respectively. The extruder was operated at barrel temperature of 130°C, screw speed of 150 rpm with the feeder delivering feed rate of 5.1g/min. Feed moisture content in the extruder barrel was 17% (170 g/kg). The extrudates were collected and dried in a dry oven at 105° C for 15 minutes (Carine *et al.*, 2010), allowed to cool at room temperature. The dried extrudates were milled and sieved (0.5mm) finally, the flour was stored in polyethylene bags at 4°C for further analysis.

$$W_a = S_w \left(\frac{m - m_a}{100 - m} \right) \dots \dots \dots \text{Equation 1}$$

Where:

W_a= weight of water added (g)

m_a=original flour moisture content (% weight base)

m= Required dough moisture level (% weight base)

S_w = sample flour weight (g)

3.6. Preparation of Gruel

In total, 15 gruels samples were prepared; 13 gruel samples from composite instant flour and 2, gruel samples from ‘mittin’ (blends of soybean, barley, lentil, wheat, peanut, fenugreek, linseed, faba bean, sorghum and oat used as complementary food) and barley gruel as a control. Fifty gram of instant flour was added to 150ml of pure tape water warmed at 75°C and stirred with wooden ladle until it attains desirable paste consistency similar to the traditional gruel consumed (Thathola and Srivastava, 2002) for extruded composite flour for control (‘mittin’ and barley flour) gruel sample preparation time was 90°C for 30 minutes. After preparation, gruels were allowed to cool to 45°C before sensory evaluation.

3.7. Analysis Methods

3.7.1 Proximate analysis

Proximate compositions of extruded composite flour were determined using standard methods. All analyses were performed in duplicate.

3.7.1.1. Moisture content determination

Moisture content of the composite flour, samples were determined by dry air oven method according to (AOAC, 2005) method, 925.10. The petridish was dried at $130 \pm 3^\circ\text{C}$ for 1 hour and placed in desiccator and weighed after cooling. About 3g of flour were weighed by analytical balance and put on dry dish. Dry dish and its contents put in the oven (LEICESTER LE67 5FT, England) maintained at $130^\circ\text{C} \pm 3^\circ\text{C}$ for 1hour. The sample containing dish was transferred to a desiccator and weighed soon after reaching room temperature. Then, the moisture content was estimated by the formula:-

$$\text{Moisture (\%)} = \frac{W_2 - W_3}{(W_2 - W_1)} \dots\dots\dots \text{Equation 2}$$

Where:

W1 = Weight of the drying petridish,

W2= Mass of the drying petridish and the sample before drying,

W3 = Mass of the drying petridish and the sample after drying

3.7.1.2. Ash determination

Ash was determined by the method of the Association of Official Analytical Chemists (AOAC, 2000), using the official method 923.03. Clean porcelain crucible, dried at 120°C in an oven was cooled in desiccators and weighed (M1). Then about 2.0 g samples were weighed into a previously dried and weighed (M2) porcelain crucible. The crucible with the contents were placed in a Muffle furnace (Gallenkamp, Model FSL 340-0100, U.K.) set at 550°C for three hours to ignite until ashing was complete. After this period the crucible with its content was removed and cooled in desiccators. The crucible with the residue was weighed (M3). The weights of the ash were expressed as a percentage of the initial weight of the samples. The total ash was expressed as percentages on dry matter basis as follows

$$\text{Ash content (\%)} = (M3 - M1)/(M2 - M1) \times 100 \dots\dots\dots\text{Equation 3}$$

Where:

(M2-M1) is sample mass in g on dry base and

(M3-M1) mass of ash in gm

3.7.1.3. Crude fat determination

Crude fat content of composite flour was determined by Soxhlet extraction according to (AOAC, 2005) method, 2003.06. About 2 g of sample was weighed and put into a thimble. The thimble and its contents were placed in to extraction flask. The thimble and its extraction flask were transferred in to extraction unit. The sample contained in the thimble was extracted with the solvent diethyl ether in a Soxhlet extraction apparatus (Foss Tecator, Sweden) for 6 hours. After the extraction completed, the extraction thimble was dried in the oven (Memmert 854 schwabach, West Germen) for 30 minutes at 102°C ± 2°C to remove moisture. Then it was removed from the oven and cooled in desiccators. The cup and its contents were weighed. The crude fat was determined by the following formula:-

$$\text{Crude fat (\%)} = \frac{W2-W1}{\text{weight of sample}} \dots\dots\dots\text{Equation 4}$$

Where:

W1= Weight of extraction flask before extraction

W2 = Weight of extraction flask after extraction

3.7.1.4. Protein determination

The crude protein content of composite flour was analyzed by Kjeldahl method of nitrogen analysis according to AOAC (2005) Method, 978.04. About 0.3 gm of sample, 1 g of catalyst mixture of K₂SO₄ and CuSO₄ and 5 ml of sulfuric acid were added to digestion flask (Kjeldahl flask KF250, Technical Glass Products, Inc., USA) at about 370°C digested until the solution becomes clear. Then, distillation took place by adding 25 ml of 40% NaOH and using 25 ml of boric acid with 10 drops of indicator solution. As the distillation was going on, the pink color solution of the receiver flask turned green indicating the presence of ammonia. The green color solution was then titrated with 0.1N HCl solutions. At the end point, the green color turned to red pink color, which indicated that, all the nitrogen trapped as ammonium

borate have been removed as ammonium chloride. The distillate was titrated with standardized 0.1N sulfuric acid to a reddish color. Finally the percentage of nitrogen content was estimated using the following formula:

$$\text{Total nitrogen (\%)} = \frac{(T-B) \times N \times 14.007 \times 100}{\text{weight of sample}} \dots \text{Equation 5}$$

Where:

Crude protein = 6.25 * total nitrogen

T - Volume in ml of the standard acid solution used in the titration for the test material

B - Volume in ml of the standard acid solution used in the titration for the blank determination

N - Normality of standard sulphuric acid

3.7.1.5. Crude fiber determination

The crude fiber was determined by the non-enzymatic gravimetric method of (AOAC, 2005) method, 920.168. About 1.5 g food sample was placed into 600mL beaker. A 200 ml of 1.25% H₂SO₄ solution was added to each beaker and allowed to boil for 30 min by rotating and stirring sometimes. During boiling, the level was kept constant by addition of hot distilled water. After 30 min, 20 ml of 28% potassium hydroxide solution was added in to each beaker and again allowed to boil for another 30 min. The level was still kept constant by addition of hot distilled water. The solution in each beaker was then filtered through crucibles containing sand filter 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% H₂SO₄ solution, hot distilled water, 1% NaOH solution, 1% H₂SO₄, hot distilled water and finally with acetone. Each of the crucibles with their contents was dried for 2 hours at 130°C in the oven (Mettler 854 Schwabach, West Germany), cooled in desiccators and weighed (M1). Then again they were ashed for 30 min at 550°C in furnace (Stuart 2416, UK) and were cooled in desiccators. Finally the mass of each crucible was weighed (M2) to subtract ash from fiber. The crude fiber was calculated from the equation

$$\text{Crud fiber (\%)} = \frac{M1-M2}{\text{weight of sample}} \dots \text{Equation 6}$$

Where:

M1 = mass of crucible and wet residue

M2 = mass of crucible and ash residue

3.7.1.6. Carbohydrate Determination by difference

The carbohydrate contents was determined by subtracting the summed up percentage compositions of moisture, protein, lipid, fiber, and ash contents from 100 g of the sample (Otitoju, 2009).

$$\text{Carbohydrate (\%)} = 100 \% - (\text{Moisture\%} + \text{Protein \%} + \text{Fat \%} + \text{Ash \%} + \text{Fiber\%}) \dots \dots \dots \text{Equation 7}$$

3.7.1.7. Gross Energy

Energy was determined by calculation from fat, carbohydrate and protein content using the Atwater's conversion factor (Golden, 2009).

$$\text{Energy (kcal)} = \text{kcal Protein\%} + \text{kcal fat\%} + \text{kcal carbohydrate\%} \dots \dots \dots \text{Equation 8}$$

3.7.2. Mineral Analyses

Calcium, zinc and iron were determined by Flame Atomic Absorption Spectrophotometer (AAS) (Auto sampler AA 6800, Japan) method as per the (AOAC, 2005) method, 985.35. One g of composite flour samples were ashed and weighed flask used for filter the digested ash was washed with 10% HNO₃ (nitric acid) and Ashes were obtained from dry ashing was wetted completely with 5ml of 6N HCl and dried on hot plate by low temperature. A 7ml of 3N HCl was added to the dried ash and heated on the hot plate until the solution just boils. The ash solution was cooled to room temperature at open air in a hood and filtered through a filter paper (Whatman 42, 125 mm) into a 50 ml graduated flask and 5ml of 3N HCl was added into each crucible dishes and re-heated until the solution just boiled, then cooled and filtered into the flask. The crucible dishes were again washed three times with de-ionized water; the washings were filtered into the flask. A 2.5ml of 10% Lanthanum chloride solution was added into each graduated flask to suppress interferences during calcium reading. Then the solution was cooled and diluted to the mark (50ml) with de-ionized water. A blank was

prepared by taking the same procedure as the sample. Then the solution was used to determine Ca, Zn, and Fe. The sample and standard were atomized by using reducing air-acetylene for Ca and oxidizing air-acetylene for zinc and iron as a source of energy for atomization.

For iron content determination absorbance was measured at 248.4 nm. For zinc concentration determination, absorbance was measured at 213.9 nm. Calcium content determination, absorbance was measured at 422.7 nm. Mineral content were calculated using the following formula: -

$$\text{Mineral content (mg/100g)} = \frac{[(A-B)*V]}{10W} \dots\dots\dots \text{Equation 9}$$

Where:

W= Weight (g) of samples;

V= Volume (V) of extract

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

B = Concentration (µg/ml) of blank solution

3.7.3. β-carotene determination

Extraction of total β-carotene content was followed the method of Sadler and others (1990), with minor modifications. Briefly, 1 g of A sample flour was mixed with of 1gram CaCl₂.2H₂O and 50 ml extraction solvent (50% hexane, 25% acetone, and 25% ethanol, containing 0.1% BHT) and shaken 30 min at ambient temperature. After adding 15 ml of distilled water, the solution was frequently shaken for a further 15 min. The organic phase, containing the β-carotene was separated from the water phase, using a separation funnel, and filtered using Whatman filter paper No.1. The extraction procedure was carried out under subdued light to avoid degradation of carotenoids. β-carotene was estimated from a standard curve of beta carotene standard product of Sigma Aldrich dissolved in the same solvent combination. To draw the calibration curve, beta carotene standard stock solution was prepared by accurately weigh 0.01g beta-carotene standard and dissolved in 20 ml solvent which was similar to extraction solvent used to extract samples (50 % hexane, 25 % acetone, and 25 % ethanol) and made the volume to 100 ml using the same solvent. From the stock

solution 0, 2, 3, 4 and 5ml were added in to 100ml flask and diluted to give 0, 0.1, 0.2, 0.4, and 0.8 mg/L of beta carotene standard. Extracted beta carotenes were determined using double beam UV-Vis spectrophotometer (T80, china) at 450nm wavelength. Finally, the β -carotene of each sample was calculated using the standard calibration curve equation

3.7.4. Anti nutrient determinations

3.7.4.1. Phytate content determination

The method described by Vaintraub and Lapteva (1988) was used for phaytate determination. Five g of dried sample was weighed and extracted with 10ml of 0.2N HCl for 1 hour at an ambient temperature and centrifuge at (3000rpm/30minut). The clear supernatant was used for the phytate estimation. 2 ml of wade reagent was added to 3ml of the supernatant sample solution then homogenize and centrifuged the solution (3000rpm/10minut). The absorbance at 500nm was measured using UV-Vis spectrophotometer. The phytate concentration was calculated from the difference between the absorbance of the blank (3ml of 0.2N HCl +2ml of wade reagent) and that of assayed sample. The amount of phytic acid was calculated using phytic acid standard curve and result was expressed as phytic acid in $\mu\text{g/g}$ fresh weight.

$$\text{Phytate } ((\mu\text{g})/\text{g}) = \frac{\text{absorbance} - \text{intercept}}{\text{density} * \text{slpoe} * \text{wieght}} \dots\dots\dots \text{Equation 10}$$

3.7.4.2. Tannin content determination

Tannin content was determined by the modified vanillin-HCl methods Latta and Eskin (1980). Two grams sample was extracted with 50 ml 99.9% methanol for 20 minutes at room temperature with constant agitation. After centrifugation for 10 min. at 653 rpm, 5 ml of vanillin-HCl (2% Vanilli and 1% HCl) reagent was added to 1 ml aliquots, and the color developed after 20 min. at room temperature was read at 500 nm. Correction for interference light natural pigments in the sample was achieved by subjecting the extract to the conditions of the reaction, but without vanillin reagent. A standard curve was prepared using Catechin (Sigma Chemical, St. Louis, MO) after correcting for blank, and tannin concentration will be expressed in g/100 g.

$$\text{Tannin } (\mu\text{g/g}) = \frac{(A-B) - \text{intercept}}{\text{wieght}} * \text{slpope} * d \dots\dots\dots \text{Equation 11}$$

Where:

A =sample absorbance

B= blank absorbance

d=density of solution (0.791g/ml)

3.7.5. Functional Properties Determinations

3.7.5.1. Water Absorption Capacity

Water Absorption Capacity (WAC) was determined using the method of Adebowale *et al.* (2005). Ten ml of distilled water was added to 1gram of the sample in a beaker. The suspension was stirred using magnetic stirrer for 3 minutes. The suspension obtained was thereafter centrifuged at 3500 rpm for 30 minutes, and the supernatant was measured into a 10 ml graduated cylinder. The water absorbed by the flour was calculated as the difference between the initial volume of the sample and the volume of the supernatant.

$$WAC = \frac{W_3 - W_2}{W_1} \dots\dots\dots \text{Equation 12}$$

Where:

W1= weight of flour,

W3= weight of flour plus tube after centrifuge

W2= weight of tube

3.7.5.2. Bulk density determination

The bulk density of the composite flour was analyzed according to the method stated by Oladele and Aina (2007) in which a mass of 50 g of the sample was put in to a 100 ml measuring cylinder. The cylinder was tapped continuously until a constant volume was obtained. The bulk density was then calculated as weight of the grounded flour in (g) divided by its volume (ml).

$$\text{Bulk density (g/cm}^3\text{)} = \frac{\text{weight of sample}}{\text{volume of sample after tapping}} \dots\dots\dots \text{Equation 13}$$

4. RESULTS AND DISCUSSIONS

In this chapter, data collected on proximate compositions (moisture, ash content, protein, crude fiber, fat, and carbohydrate), energy, minerals (Fe, Ca and Zn), total carotenoids, anti-nutritional factors (phytate and tannin), functional properties (water absorption capacity and bulk density) and sensory attributes (appearance, aroma, taste, mouth feel, consistency and overall acceptability) are described, presented in different tables/figures and discussed.

4.1 Proximate Compositions of Instant Complementary Flour

Mean values of moisture content, crude protein, crude fat, total ash, crude fiber, carbohydrate and energy density contents of extruded complementary flour are presented in Table 8. The minimum and maximum values for moisture, protein, fat, fiber, ash, carbohydrate and energy density of extruded composite instant flours were 3.2 - 5.6%, 18.0 - 20.6%, 6.8 - 9.9%, 3.8 - 4.3%, 2.3 - 3.1%, 59.8 - 63.7% and 384.5 - 408.2kcal/100g respectively.

Table 8: Proximate composition and energy contents (kcal/100g) of the instant complementary flour

Mixture composition (%)				Proximate composition (%)						Energy
Oat	soybean	linseed	Premix*	MC	PC	FC	FiC	AC	CC	
57.9	19.9	7.2	15.0	4.1	19.5	8.5	3.9	2.7	61.4	399.8
57.4	17.9	9.7	15.0	4.0	19.4	8.7	3.9	2.8	61.1	400.8
56.0	23.0	6.0	15.0	3.2	20.5	8.7	3.9	2.9	60.9	403.7
65.0	11.0	9.0	15.0	5.2	18.2	7.1	4.1	2.6	62.7	387.2
62.4	15.4	7.2	15.0	4.7	18.7	7.6	4.0	2.7	62.3	392.5
55.0	19.0	11.0	15.0	3.2	19.6	9.9	4.0	3.1	60.2	408.2
62.4	13.9	8.7	15.0	5.0	18.8	7.5	4.1	2.8	61.9	390.3
57.4	19.9	7.7	15.0	4.0	19.9	8.7	3.8	2.9	60.6	400.8
63.0	11.0	11.0	15.0	4.6	18.1	7.0	4.3	2.3	63.7	390.1
59.8	16.8	8.3	15.0	4.5	19.0	8.5	3.9	2.8	61.4	397.9
55.0	23.0	7.0	15.0	3.4	20.6	9.3	3.8	3.1	59.8	405.0
61.4	13.9	9.7	15.0	4.3	18.7	7.8	4.1	2.6	62.5	395.0
65.0	14.0	6.0	15.0	5.6	18.0	6.8	4.2	2.5	62.9	384.5

Premix (9.9% sugar, 3% moringa, 1.5% fenugreek and 0.6% salt)

MC=moisture content; PC=Protein content; FC=crude fat content; FiC= crude fiber content; AC=ash content; CC= carbohydrate content,

Analysis of variance outputs of proximate composition of instant complementary flour are summarized in Appendix table 1. P-values for quadratic models, interaction among components and R squared values were indicated. ANOVA p-values of all the proximate compositions (except for moisture content and energy) indicate that quadratic models and interaction among the mixture components are significantly ($p < 0.05$) different. Coefficient of determination (R^2 values) for proximate composition indicated that models can sufficiently predict the responses.

4.1.1. Moisture content

The moisture contents of the extruded composite flours varied from 3.2% to 5.6%. ANOVA has shown that the model for predicting moisture content is not satisfactory. The Coefficient of determination in indicates that the model developed in predicting moisture contents could explain 96.11% of the variations (Appendix table 1). The regression model for moisture content was presented as equation in (Appendix table 2)

At higher proportion of oat in the blend, the moisture contents of the infant flours was also high (5.6%), and at high proportion of soybean and linseed the moisture content was low (3.2%). Increasing proportion of oat in the formulation increased the moisture content of formulations. On the other hand increasing proportions of soybean and linseed decreased the moisture content of formulations (Figure 3).

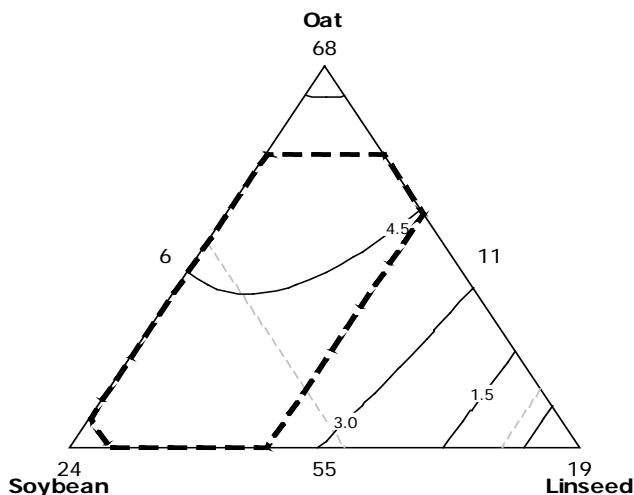


Figure 3: Contour plot of a moisture content (%) of instant complementary flour

The moisture levels were less than 7% similar to values reported by Filli *et al.* (2010) in extruded cereals. The increasing of moisture contents of the instant infant flour with increasing of oat may be due to high moisture contents of oat than soybean and linseed. The result of this study is in agreement with the findings of Iwe and Ngoddy, (1998) who reported that addition of soybean flour reduced the moisture and the carbohydrate contents. Ojinnaka *et al.* (2013) also reported that moisture level of the samples decreased with increased inclusion of soybean flour. The decrease in moisture level with increase in level of substitution with soybean flour might serve as an indication of increased storage stability (Adebayo-Oyetoro *et al.*, 2010).

In contrary to these results, Asare *et al.* (2012) reported that a blend of rice and legumes had relatively high moisture content as compared to rice extruded alone, which suggests that the incorporation of the legumes (groundnut and cowpea) to the system might have resulted in the observed higher moisture values.

Moisture is one of the important quality parameter that determine the shelf life of food, as increasing of moisture decrease shelf life of products by facilitating growth of microorganisms. Low moisture content of flours prevents microbial activity and extends the shelf life of products (Tizazu *et al.*, 2010; Kikafunda *et al.*, 2006). The low moisture values of the products indicated that, the product is shelf stable.

4.1.2. Crude fiber content

The crude fiber contents of the composite instant infant flours varied from 3.8% to 4.3%. ANOVA has shown that highly significant ($p < 0.01$) difference quadratic model and interaction among the components except interaction between oat and soybean which no significance ($p > 0.05$) difference. The model developed in predicting product fiber contents could explain 98.86% of the variation (Appendix table 1). The regression model for crude fiber content was represented as equation in (Appendix table 2) as indicated with three variables.

The minimum crude fiber contents of the formulation was recorded at highest proportions of soybean in the blend of oat, soybean and linseed (55%: 23%: 7%) respectively and the maximum value of fiber contents of composite instant infant flour was obtained at high proportions of oat flour in the blend (65%: 14%: 6%) as indicated in Table 8. Increasing proportions of oat and linseed flour increases the crude fiber contents of the formulation on other hand decreasing the proportion of oat and linseed flour and increasing proportion of soybean flour decreased the crude fiber contents of the formulation as showed in (Figure 4) below.

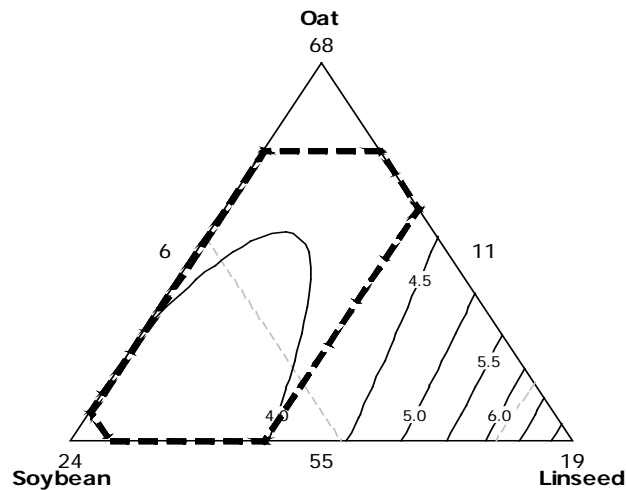


Figure 4: Contour plot of crude fiber content (%) of instant complementary flour

Increased fiber contents of formulation as oat and linseed flour increases might be related to high fiber contents of oat and linseed. Similar result was reported by Ganorkar and Jain (2014), increasing proportion of linseed flour in wheat based cookies increased crude fiber content, this could be accounted by the fact that linseed is far higher in crude fiber content compared with wheat flour. The fiber contents of soybean might be reduced due to husk removal during sample preparation. Dehulling removes the hulls, which contain much fiber (Kikafunda *et al.*, 2006). The result of the finding is in line with the report of Ghasemzadeh and Ghavide (2011), incorporation of de-hulled legumes instead of whole legume grains, reported to decrease fiber contents.

The result of present finding of crude fiber content was less than the maximum fiber content of 5% of the Protein Advisory Group Recommendations (1972). According to Codex Alimentarius Commission (FAO/WHO, 2013) recommendation, fiber intake for children should not exceed 5 g per 100 g of food on dry weight basis. Therefore the result of crude fiber contents of extruded composite flour is within the range of this recommendation.

Although fiber content in food is believed to prevent or alleviate maladies such as cardiovascular diseases, diabetes, and colon cancer (Azizah and Zainon, 1997), because it consists of cellulose and lignin, its estimation affords an index for evaluation of dietary fiber (Eddy *et al.*, 2007). High fiber content reduce energy intake through a suppressing effect on appetite, and may increase fecal losses of energy due to reduced absorption of fat and carbohydrate.

4.1.3. Protein content

The minimum and maximum values of protein contents of the formulation were 18.0 and 20.6% respectively. Analysis of variance (ANOVA) has shown highly significant difference ($P < 0.01$) in protein content of extruded composite flour samples in quadratic model, interaction between soybean and linseed but, the interaction of soybean with linseed showed significant ($p < 0.05$) effect on protein contents of extruded composite flour samples. The value of coefficient of determination indicates that model developed for predicting product protein contents could explain 98.94% of the variation (Appendix table 1). The regression model for protein was shown by equation in Appendix table 2

The minimum values of protein content was recorded at highest proportion of oat flour in the formulation (65%:14%:6%) of oat, soybean and linseed respectively. The mean maximum values of protein contents of the formulation was recorded at highest soybean and linseed flour content in the formulation (55%: 23%: 7%) of oat, soybean and linseed respectively. Figure 6 indicates that the protein content of extruded composite flour increased with an increasing proportion of soybean and linseed in the blend on the other hand increasing proportion of oat flour in the blend decreased the protein contents of the formulation.

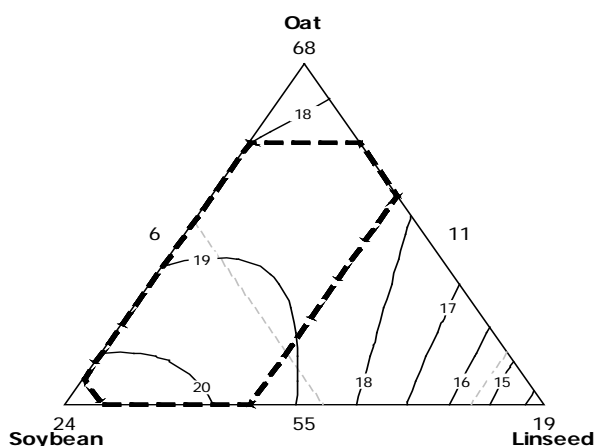


Figure 5: Contour plot of protein content (%) of instant complementary flour

The highest protein content at highest percentage of soybean in oat, soybean and linseed blend might be due to high protein contents of soybean and linseed. Similar results were reported by different researchers, the increase in the protein content and energy values of foods that have been enriched with soybean has been reported by many investigators (Bangoura and Zhou, 2007; Martin *et al.*, 2010). Omwamba and Mahungu (2014) also reported that the inclusion of a soy protein source in cereal flour increases the protein content and improves the amino acid balance in the extruded products which ultimately improves the nutritional value of the products.

According to Hotz and Gibson (2001), the blending of cereal based foods with legumes and their processing methods improves the protein content of the flour. Onwulata *et al.* (2001) reported incorporation of protein rich materials into extruded products will not only increase the utilization of products but also improve the products nutritive value by increasing the protein content.

The recommended dietary allowance (RDA) for the protein content in the complementary foods was recommended to be $\geq 15\%$ (FAO, 2004) and the result of present study can satisfy the protein requirements of children under the age of two years. Therefore, fortifying cereals with pulses and linseed can satisfy the protein requirements of young children in developing countries like Ethiopia and can reduce protein malnutrition.

4.1.4. Fat content

The mean fat contents of the formulations were showed in Table 8. The mean minimum and maximum values of fat contents of the extruded composite flour varies from 6.8% to 9.9%. ANOVA showed significant difference ($p < 0.05$) in fat content of the extruded composite flours at quadratic model and interactions of soybean with linseed. The regression models, developed to predict the fat content of the formulation was explained 98.39% of the variation (Appendix table 1). The regression model for crude fat was shown as equation in Appendix table 2

Minimum fat content was recorded at highest proportion of oat flour in the formulation (65% oat, 14% soybean and 6% linseed). The maximum fat content was recorded at high proportion of soybean and linseed in the formulation (55% oat, 19% soybean and 11% linseed). Increasing the proportion of soybean and linseed flour in the blend increased the fat content of the formulation, however increasing the proportion of oat flour decreased the fat content of the formulation as indicated by Figure 6.

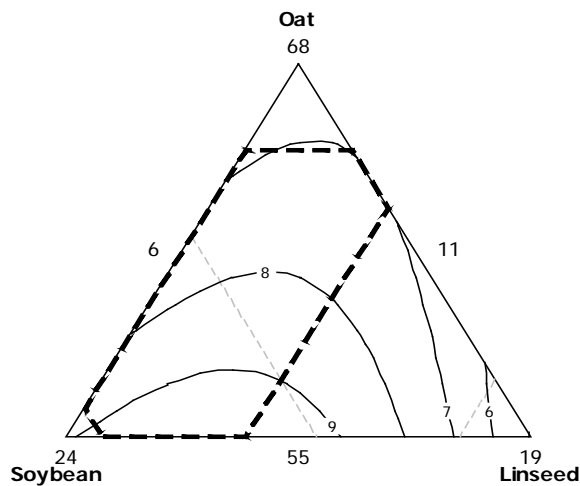


Figure 6: Contour plot of fat content (%) of instant complementary flour

The fat content of the extrudates increased consistently with increasing soybean and linseed proportions for all extrudates of the oat-soybean-linseed blends and decreased with increased proportion of oat flour in the blend, this may be because soybean and linseed is known to contain considerably high amounts of fat.

This result is in agreement with results of Joel *et al.* (2011) who reported that fat content of wheat flour increased with the addition of soybean flour. A research by Okoye *et al.* (2008) revealed that fat content increased from 0.82 to 18.4% as the amount of soybean increase from 0% to 100% in the final product. A study conducted by Aleem *et al.* (2012) on blending of soybean flour in composite flour showed that the fat content increased with increasing soybean.

Plant fat is a good source of energy and helps in absorption of most fat soluble vitamins and minerals. Dietary lipids provide essential fatty acids and facilitate the absorption of fat-soluble vitamins (Uauy *et al.*, 2000). In the current study, soybean and linseed were used as the main fat suppliers. Soybean oil can provide the recommended amounts of fat (WHO, 1998). Vegetable oils can be included in foods meant for infants and children, which will not only increase the energy density, but also be a carrier for fat soluble vitamins and provide essential fatty acids. Soybean and cereals contain unsaturated fats (Wikramanayake, 1996) which does not increase the cholesterol in the blood hence can be recommended for children.

Therefore, enrichment of cereals crops with legumes and oilseed improves the fat content of complementary food of infant and young children with very high fat requirement to compensate glucose deficiency due to high body activity at this particular developmental period. Protein Advisory group (1972) recommended fat content of a complementary feeding should be not more than 10% due to oxidative deterioration. The result of the study can satisfy the Protein Advisory group's recommendation.

4.1.5. Ash content

The mean ash contents of the formulations were indicated in Table 8. The minimum and maximum ash content of the extruded composite flour ranges from 2.3 to 3.1% respectively. ANOVA showed a highly significant ($p < 0.01$) effect of proportion of oat, soybean and linseed and quadratic model on ash content of the blend. The regression models, developed to predict the ash content, explained 94.50% of the variations (Appendix table 1). The regression model for ash was shown as equation in (Appendix table 2) indicating quadratic model with three variables

The minimum ash content was recorded at higher proportions of oat flour in oat, soybean and linseed blend (63%:11%:11%) respectively. The maximum ash contents of the formulation was recorded at highest proportion of soybean flour in oat, soybean and linseed blend (55%:23%:7) respectively. The ash contents of the formulations increased with increasing the proportion of soybean and decreased with increasing the proportion of oat and linseed flour in the formulation as shown in Figure 7

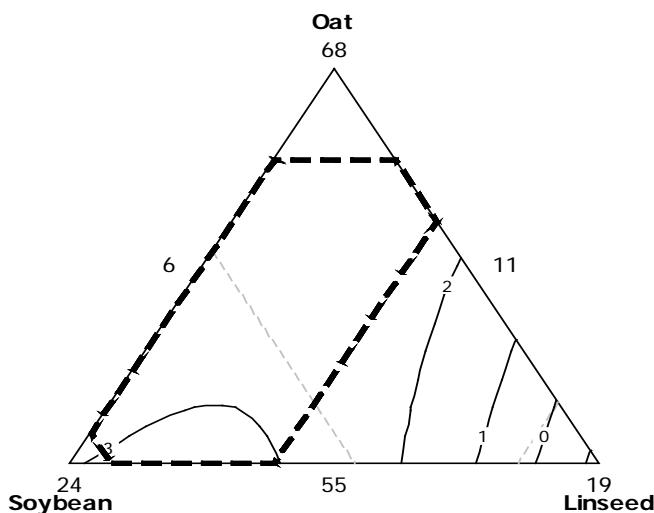


Figure 7: Contour plot of ash content (%) of instant complementary flour

Ash content of the blends increased with increasing content of soybean flour in the blend. This might be because of high percentage of inorganic contents in soybean than oat and linseed. The finding is in line with the report of Okoye *et al.* (2008) who stated that ash contents of the blended products increased as the level of soybean flour inclusion increase. The increased noted in the ash content of the malted sorghum-soy flour may be attributed to the higher mineral content of soybean, legumes have been reported to be good sources of ash Bolarinwa *et al.* (2015)

Acceptable ash content of complementary food, as given by the Protein Advisory Group recommendations (1972), should not exceed 5%. The extruded instant flour studied in the current study was within this limit as well.

4.1.6. Carbohydrate content

The mean carbohydrate contents of the formulations were indicated in Table 8. The minimum and maximum value of carbohydrate content of the extruded composite flour varies from 59.8 to 63.7% respectively. ANOVA has shown there was highly significant difference ($p < 0.01$) in quadratic model and interaction among the components on carbohydrate contents of extruded composite flour.

The coefficient of determination for carbohydrate indicates that models developed to predict the carbohydrate content was explained 97.86% of the variations (Appendix table 1). The regression model for carbohydrate was shown as Equation in (Appendix table 2) indicating quadratic model with three variables.

The minimum carbohydrate content was obtained at highest soybean and lowest oat and linseed blend (55%: 23%: 7%), maximum carbohydrate content was obtained at high oat and flaxseed proportion in the blend and lowest soybean (63%:11%: 11%). The increasing of oat and linseed flour in the formulation increased carbohydrate contents of the formulation on the other hand increasing the proportion of soybean decreased the carbohydrate contents as indicated by Figure 8.

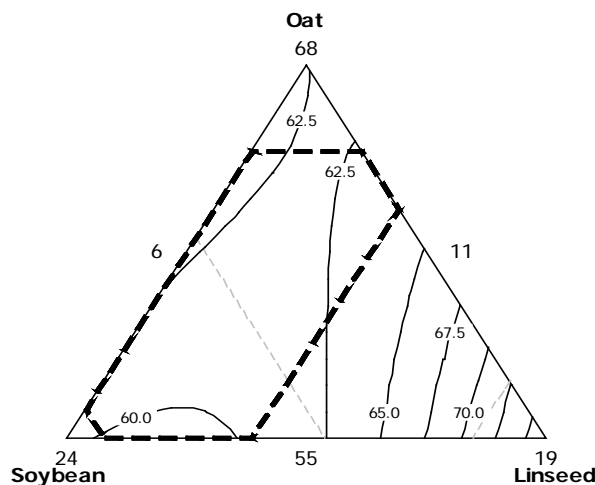


Figure 8: Contour plot of carbohydrate content (%) of instant complementary flour

Carbohydrate is an important source of energy in diet. It includes sugars or polymers of sugar such as starch that can be hydrolyzed to simple sugars. Carbohydrate containing food is important vehicle for protein, vitamins, minerals and other food components such as phytochemicals and antioxidants (Bowman and Russel, 2001).

All products containing high proportions of oat had higher carbohydrate content compared to those that had lower proportions of oat. This could be because cereals like oat have high carbohydrate contents than soybean. The results obtained in this study is similar to the findings of Okoye *et al.* (2008) who reported a decrease in carbohydrate content of wheat-soybean flour blend with increasing soy flour substitution. Sefa-Dedeh *et al.* (2001) also reported that addition of legumes decreases the carbohydrate content of cereal-based traditional foods. In addition, Bolarinwa *et al.* (2015) concluded that carbohydrate content of the malted sorghum soybean flour decreased with increase in the proportion of the soybean flour in the mixes, which indicate that soybeans are not good sources of carbohydrate.

4.1.7. Gross Energy

The mean energy contents of the formulations were presented in (Table 8). The minimum and maximum energy content of the formulation varies from 384.5 to 408.2kcal/100g respectively. ANOVA has shown no significant ($p > 0.05$) difference statistically at both quadratic and interaction among the components. The coefficient of determination regression models, developed to predict the energy content explained 98.14% of the variations (Appendix table 1). The regression model for energy was shown as equation in (Appendix table 2) indicating quadratic model with three variables.

The minimum energy density of the blend was recorded at highest proportions of oat in the blend (65%: 14%: 6 %.) oat, soybean and linseed respectively. The maximum energy density was recorded at high proportion of soybean and linseed (55%:19%:11%) oat, soybean and linseed respectively in the blend. The energy content of the formulations increased with increasing the proportion of soybean and linseed flour and decreased with increasing proportion of oat flour in the blend as shown by Figure 9.

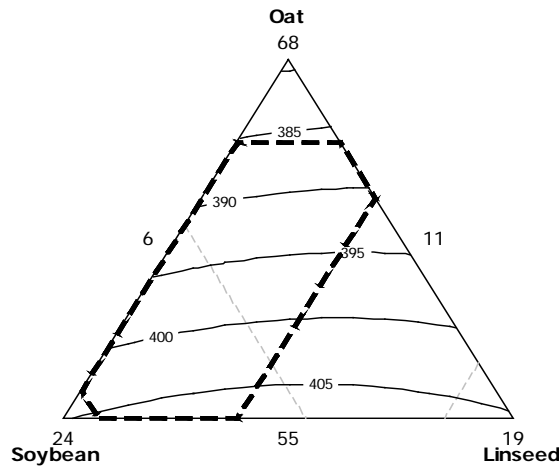


Figure 9: Contour plot of energy content (kcal) of instant complementary flour

Increasing proportion of soybean and linseed flour in the blend increased the energy density of the composite instant flour, it may be due to high fat content in soybean and linseed which contributes to high energy contents, as fat provides more than double of carbohydrate and protein energy.

The result of this thesis is supported by Michaelsen *et al.* (2009) who reported that, the most important factor influencing energy density is the fat content, as the energy density of fat (9 kcal per g) is more than double that of protein and carbohydrate (4 kcal/g). Induja *et al.* (2012) also reported that fortified high protein and high calorie supplementation of biscuits revealed that maximum amount of calorie was found in the biscuits containing the highest amount of soybean flour. Onabanjo *et al.* (2009) in his study on characteristics of complementary food observed that the high fat content of legume and oil crops flour further increased the energy density of the products developed from different formulas.

High energy density in foods has been pointed out as a major cause of fast growth and well nourishment among undernourished children (Dewey and Brown, 2003). Enrichment of oat based formulations with soybeans and linseed improved their energy densities. Energy density is one of the most important qualities of foods for wasted children. If the energy density is too low and food is bulky, then the child would not be able to get enough calories (Michaelsen *et*

al., 2009). Energy density is most important for children, as they have an increased energy need for catch-up growth.

The energy content of instant composite flour was above the specified minimum value of 380kcal for fortified blended foods by technical specifications of FAO (2004). Generally, the formulations with high proportions of soybean had higher energy density. This was because soybean is rich in fat and protein which contribute to higher energy density.

4.2. Mineral Content of Instant Complementary Flour

The mean mineral (calcium iron and zinc) content of composite instant flours is presented in the Table 10 below. The minimum and maximum calcium, iron and zinc contents in (mg/100gm) were, 101.3-124.4, 6.6-7.8 and 2.9-3.0 respectively.

Table 9: Mineral content of instant complementary flour

Mixture composition (%)				Mineral contents (mg/100gm)		
Oat	Soybean	Linseed	Premix*	Ca	Fe	Zn
57.9	19.9	7.2	15.0	116.9	7.5	3.0
57.4	17.9	9.7	15.0	117.6	7.4	3.0
56.0	23.0	6.0	15.0	121.2	7.8	3.0
65.0	11.0	9.0	15.0	101.3	6.6	2.9
62.4	15.4	7.2	15.0	107.8	7.0	3.0
55.0	19.0	11.0	15.0	122.4	7.4	3.0
62.4	13.9	8.7	15.0	107.2	6.9	2.9
57.4	19.9	7.7	15.0	118.7	7.5	3.0
63.0	11.0	11.0	15.0	104.9	6.6	2.9
59.8	16.8	8.3	15.0	113.1	7.2	3.0
55.0	23.0	7.0	15.0	124.4	7.8	3.0
61.4	13.9	9.7	15.0	110.0	6.9	2.9
65.0	14.0	6.0	15.0	102.5	6.8	2.9

Premix (9.9% sugar, 3% moringa, 1.5% fenugreek and 0.6% salt)

Analysis of variance outputs for mineral (calcium iron and zinc) content of composite instant flours are summarized in Appendix table 3. ANOVA has shown that minerals (Ca, Fe and Zn) contents were not significantly ($P > 0.05$) different statistically but there increment of minerals with increasing of soybean in the blend.

4.2.1. Calcium

The mean calcium contents of the formulations are presented in Table 10. The minimum and maximum calcium content of composite instant flour ranges from 101.3-124.4mg/100gm respectively. ANOVA has shown that the model for predicting calcium content is satisfactory. The model developed for predicting product calcium contents could explain 99.73% of the variations (Appendix table 3) and the regression model for predicting calcium content is presented in Equation (Appendix table 4)

Minimum calcium contents was recorded at the highest proportion of oat flour (65%:14%:6%) in the blend and maximum calcium content of the formulation was recorded at high proportion of soybean and linseed flour (55%:19%:11%). The increasing of soybean and linseed flour increased the calcium contents of the formulation, on contrary the increasing of oat flour decreased the calcium contents of the formulation as indicated by Figure 10

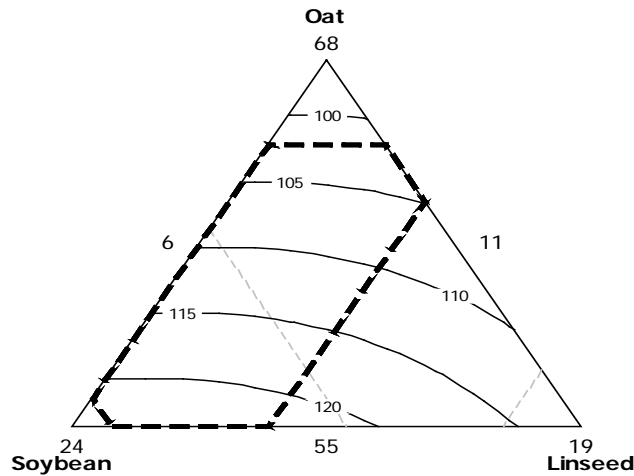


Figure 10: Contour plots of Calcium contents (mg/100gm) of instant complementary flour

The calcium contents of the composite instant flour increased with increasing of soybean and linseed, which indicates that soybean and linseed contain higher amounts of calcium. The result of the current research in agreement with a research done by Bolarinwa *et al.* (2015) who reported that, calcium content of the composite flour increased as the proportion of soy flour increases in the composite flour mixes. A similar result has been reported by Rawat *et al.* (1994), soybean fortified chapattis contained higher calcium than wheat flour chapattis.

Jan *et al.* (2000) also reported that oilseeds flour contained appreciable quantity of mineral which resulted in increase in mineral contents of composite flours. In addition to soybean and linseed flour the presence of moringa contributes for high calcium contents of the formulation. Calcium was observed to be higher in moringa compared with other plant sources (Nkafamiya *et al.*, 2010).

The calcium contents of the extruded composite flour varies from 101.3 to 124.4mg/100gm, the RDA for 6 to 12 months and 13 to 36 months infant was 270 mg and 500 mg per day respectively (Food and Nutrition Board, 2005). The current blended product can meet only 37.62 to 46.06% and 20 to 24.88 % of the RDA for infant and children respectively. The blended products have better calcium content but still it needs calcium fortification or enrichment with calcium rich agricultural products to meet the RDA for infants.

4.2.2. Iron

The minimum and maximum iron contents of extruded composite flour were 6.55- 7.82 mg/100g respectively. The coefficient of determination models, developed to predict the iron content, explained 99.87% of the variations (Appendix table 3) and the regression model for predicting iron content is presented in Equation (Appendix table 4). ANOVA has shown that iron content did not show significant difference ($P > 0.05$) statistically however, there was an increment in the iron content from 6.6 to 7.8mg/100gm with an increase in proportion of soybean flour. The iron contents of the flour were increased with increasing of soybean flour, but decreased with increasing of oat flour as indicated by contour plot (Figure11); this might be due to high mineral contents of soybean as compared to oat and linseed.

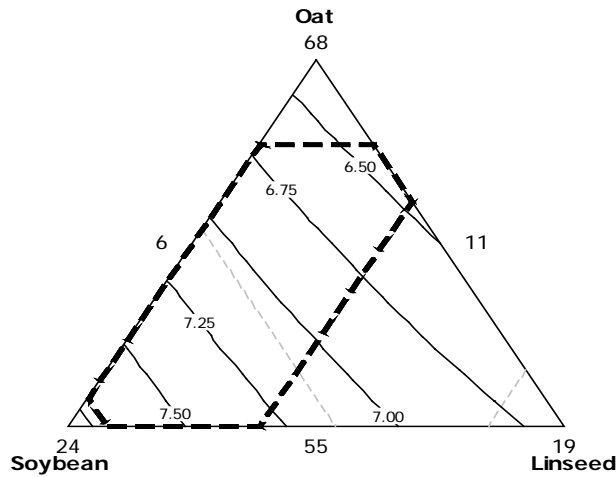


Figure 11: Contour plots of iron contents (mg/100gm) of instant complementary flour

The result of the current research was similar to a research done by, Bolarinwa *et al.* (2015) who reported that, iron content of composite flour increased as the proportion of soybean flour increases in the composite flour mixes. Gebretsadikan *et al.* (2015) also reported that the composition of iron increased as the proportion of soybean and moringa flour increased in formulations.

According to Food and Nutrition Board (2005) the RDA of iron for infant of 6 to 12 month was 11 mg/ day and 7 mg/day for children of 13 to 36 months. The iron content of the current formulations ranged from 6.6 - 7.8 mg/100gm, which can meets 59.54 to 71 % of the RDA for 6 to 12 month life stage infants and 93.57 to 100% for 13 to 36 months life stage children in lower and higher value respectively. The formulation of this study can fulfill iron requirements of infants 70-100%.

4.2.3. Zinc

The mean zinc contents of the formulation were showed in Table 10. The minimum and maximum zinc content varies from 2.9 to 3.0mg/100gm respectively in extruded composite flour. ANOVA has shown that the model for predicting zinc content is satisfactory. The regression models, developed to predict the zinc content, of the formulation was explained 98.97% of the variations (Appendix table 3) and the regression model for predicting zinc content is presented as equation (Appendix table 4).

The minimum zinc content of the formulation was obtained at highest proportion of oat and linseed flour and the maximum zinc contents of the formulation were obtained at highest proportion of soybean flour. The zinc contents of the formulation were increased with increasing the proportion of soybean flour in the blend Figure 12.

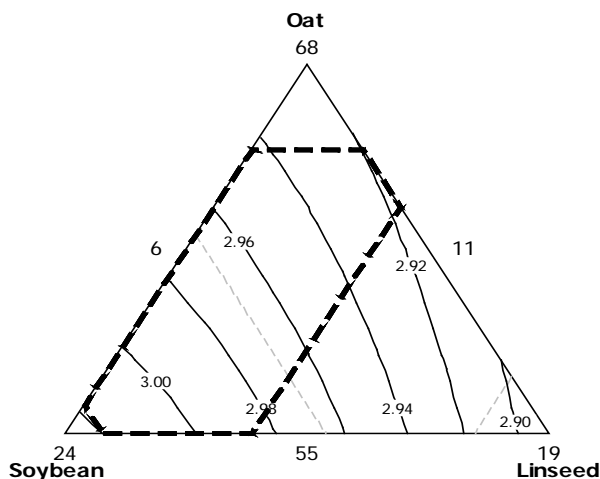


Figure 12: Contour plots of zinc contents (mg/100gm) of instant complementary flour

The result of the current research was similar to a research done by, Bolarinwa *et al.* (2015) who reported that, zinc content of the composite flour increased as the proportion of soybean flour increases in the composite flour mixes. Isingoma *et al.* (2015) reported that, moringa fortified porridges had the best nutrient composition and were able to meet zinc recommendations of the CODEX requirements.

The RDA of zinc for infant under age of two years is 3mg/100gm (Food and Nutrition Board, 2005). The current developed formulation can provide minimum and maximum of 2.9 and 3.0mg/100gm zinc respectively, which can fulfill 97 to 100% zinc RDA of infants under two years. The presence of moringa in the extruded composite flours boosted the mineral contents of the product.

Some scholars have shown the potential of moringa in improving child nutrition (Thierry *et al.*, 2013, Odinakachukwu *et al.*, 2014). Adequate processing and careful blending of the locally available foods has been encouraged as a measure of improving intake of nutrients among children in areas with limited resources (Lombor *et al.*, 2009).

4.3. β -carotene content of Instant Complementary Flour

The mean total β -carotene content of extruded composite flour is presented in the Table 10. The β -carotene content of composite flours varied between 1308.78 to 1900.33 $\mu\text{g}/100\text{g}$ as represented in Table 10.

Table 10: β -carotene content of the instant complementary flour

Mixture composition (%)				β - carotene $\mu\text{g}/100\text{g}$
Oat	Soybean	Linseed	Premix*	
57.9	19.9	7.2	15.0	1532.8
57.4	17.9	9.7	15.0	1731.3
56.0	23.0	6.0	15.0	1308.8
65.0	11.0	9.0	15.0	1900.3
62.4	15.4	7.2	15.0	1782.5
55.0	19.0	11.0	15.0	1552.5
62.4	13.9	8.7	15.0	1828.0
57.4	19.9	7.7	15.0	1688.2
63.0	11.0	11.0	15.0	1480.0
59.8	16.8	8.3	15.0	1715.1
55.0	23.0	7.0	15.0	1496.5
61.4	13.9	9.7	15.0	1716.0
65.0	14.0	6.0	15.0	1672.8

Premix (9.9% sugar, 3% moringa, 1.5% fenugreek and 0.6% salt)

The mean total β - carotene content of composite instant flours varied from 1308.8 to 1900.3 $\mu\text{g}/100\text{g}$. ANOVA has shown highly significant ($P < 0.01$) difference in β -carotene content of composite instant flours samples in quadratic model and interaction among the ingredients (Appendix table 3) The regression models developed to predict the β - carotene content of composite instant flours, was explained 96.69% of the variations (Appendix table 3) and the regression model predicting beta carotene contents was indicated as Equation (Appendix table 4).

The highest beta carotene was recorded at higher proportions of oat and linseed in the blend and the lowest was recorded at highest soybean content in the blend of oat-soy- linseed as showed in Table 10. β - Carotene contents of the formulation was decreased with increasing the proportion of soybean flour in the formulation and increased with decreasing the proportion of soybean flour in the formulation.

The high beta carotene content of the extruded infant composite flour may be due to the presence of moringa in the formulation. This is supported by Abraham *et al.* (2013), who found that beta-carotene content increased significantly with increase in moringa leaf powder which is due to the much higher level of β -carotene in moringa leaf powder as compared to wheat flour. Moringa has demonstrated the potential of improving vitamin A (Thurber and Fahey, 2009). The result of this study is also in agreement with findings of Gebretsadikan *et al.* (2015) who reported that an increase in total carotenoid content was observed for porridges prepared from higher proportions of orange fleshed sweet potato and moringa.

4.4. Anti-Nutritional Factors of Instant Complementary Flour

The mean values of anti nutritional (phytate and tannin) contents obtained from extruded composite flour are shown in Table 11. The minimum and maximum values of anti nutritional factor contents of extruded composite flour were 158.93-191.33mg/100gm and 8.4-22.89mg/100gm for phytate and tannin respectively.

Table 11: Anti nutritional factor contents of instant complementary flour

Mixture composition (%)				Anti nutritional contents (mg/100gm)	
Oat	Soybean	Linseed	Premix*	Phytate	Tannin
57.9	19.9	7.2	15.0	180.60	13.69
57.4	17.9	9.7	15.0	181.57	14.64
56.0	23.0	6.0	15.0	178.43	17.27
65.0	11.0	9.0	15.0	170.82	10.41
62.4	15.4	7.2	15.0	174.80	10.82
55.0	19.0	11	15.0	190.56	22.89
62.4	13.9	8.7	15.0	170.02	11.29
57.4	19.9	7.7	15.0	187.85	15.63
63.0	11.0	11.	15.0	158.93	16.52
59.8	16.8	8.3	15.0	175.48	14.22
55.0	23.0	7.0	15.0	191.33	21.13
61.4	13.9	9.7	15.0	177.39	12.03
65.0	14.0	6.0	15.0	180.57	8.40

Premix (9.9% sugar, 3% moringa, 1.5% fenugreek and 0.6% salt)

Analysis of variance (ANOVA) for models, interaction of components and coefficient of determination (R^2 value) are summarized in Appendix table 5. ANOVA indicates that there was significant ($P < 0.05$) difference in anti-nutritional (phytate and tannin) contents of

extruded composite flour. Coefficient of determination (R^2 values) of antinutritional factor contents of extruded composite flour indicated that the models can sufficiently predict the responses.

4.5.1. Phytate

The minimum and maximum mean value of phytate content of the composite instant flour varies from 158.93-191.33mg/100gm respectively. ANOVA table showed that there was significant ($p < 0.05$) difference at only quadratic model. The regression models developed to predict the phytate content, was explained 89.86% of the variations (Appendix table 5). The regression model for phytate was shown by Equation in (Appendix table 6) in the quadratic model with three variables.

Phytate present in raw materials and foods of plant origin are suggested to be a major factor responsible for lowering the availability of minerals and some proteins (Shimelis and Rakshit, 2006).

The lowest phytate (158.93 mg/100gm) was recorded at minimum proportion of soybean and high proportion of oat and linseed in oat-soybean-linseed blend (63%-11%-11%). The highest phytate content (191.33mg/100gm) was recorded at maximum proportion of soybean in oat-soybean-linseed blend (55%-23%-7%). Contour plot (Figure 13) indicates that, phytate content of the extruded composite flour was increased as the proportion of soybean flour increased in the blend. In contrary to this phytate content decreased with increasing proportion of oat flour in the blend. This may be due to phytic acid contents of soybean that contribute for high phytate content at high proportion of soybean in the blend.

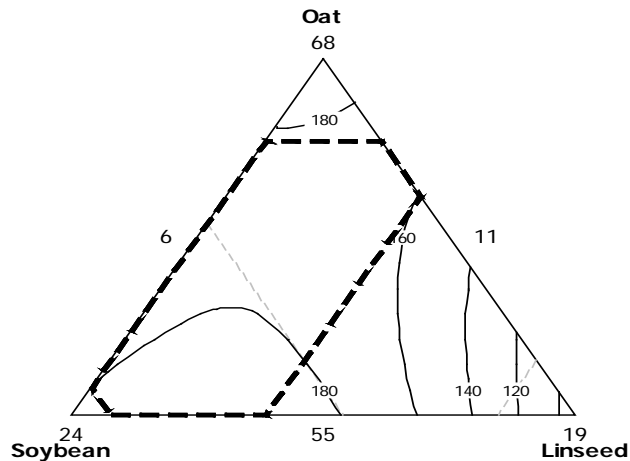


Figure 13: Contour plot of phytate content (mg/100gm) of instant complementary flour

The result of the finding is in agreement with other findings. Liener (2000) found that phytate content of a bread increased as the amount of soybean flour increased in the bread. This is because of the high amount of phytate found in soybeans. Tajoddin *et al.* (2011) also reported that phytate content is high in legumes and decrease the bioavailability of essential mineral and bioavailability of protein by forming insoluble phytate mineral and phytate protein complex.

4.5.2. Tannin

The minimum and maximum tannin content of extruded composite flour varies from 8.40 to 22.89 mg/100gm. Tannin content of the extruded composite flour showed a significant ($p < 0.05$) effect at quadratic model, however the interaction effect was not significant ($p > 0.05$). The regression models, developed to predict the tannin content, explained 89.86% of the variations (Appendix table 5). The regression model for tannin was shown by equation (Appendix table 6) in the quadratic model with three variables.

Tannins form insoluble complexes with proteins thereby decreasing the digestibility of proteins (Uzoehina, 2007). Tannins also decrease palatability, cause damage to intestinal tract, and enhance carcinogenesis (Makkar and Becker, 1996).

The maximum tannin content was recorded at high proportion of soybean and linseed in oat-soybean-linseed blend (55%:19%:11%) and minimum tannin content was recorded at highest oat proportion in the blend (65%:14%:6%). Tannin content of the formulations increased with increasing the proportion of linseed and soybean flour in the formulation. On the other hand increasing the proportion of oat flour decreased the tannin contents of the formulation as indicated in Figure 14.

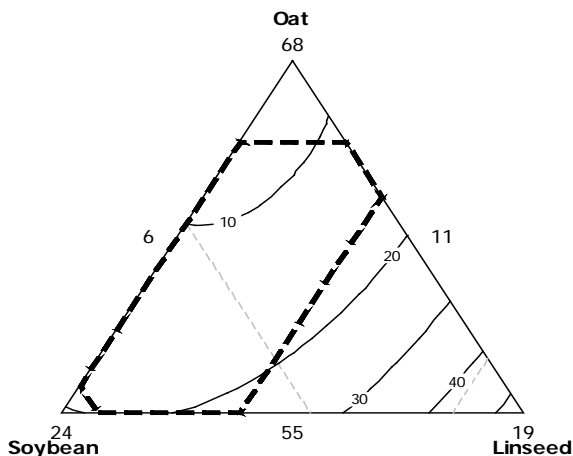


Figure 14: Contour plot of tannin content (mg/100gm) of instant complementary flour

The tannin contents of the flour increased as the proportion of soybean and linseed increased. This might be due to high anti nutritional (tannin) contents of soybean and linseed. The finding of the present study is in agreement with that of Folake *et al.* (2012) who concluded that amount of tannin in bread increased as the amount of soybean flour increased.

4.5. Functional Properties of Complementary instant Flour

The mean functional properties (water absorption capacities and bulk density) of composite instant flour were presented in Table 12. The bulk density ranges from 0.93 to 0.98 gm/ml for extruded composite instant flour. The Water absorption capacity of extruded instant flour varies from 3.06 to 3.41ml.

Table 12: Functional properties of instant complementary flour

Mixture composition (%)				Functional properties	
Oat	Soybean	Linseed	Premix*	BD (gm/ml)	WAC (ml)
57.9	19.9	7.2	15.0	0.94	3.19
57.4	17.9	9.7	15.0	0.95	3.28
56.0	23.0	6.0	15.0	0.93	3.31
65.0	11.0	9.0	15.0	0.97	3.10
62.4	15.4	7.2	15.0	0.96	3.07
55.0	19.0	11.0	15.0	0.96	3.41
62.4	13.9	8.7	15.0	0.96	3.14
57.4	19.9	7.7	15.0	0.94	3.24
63.0	11.0	11.0	15.0	0.94	3.24
59.8	16.8	8.3	15.0	0.95	3.20
55.0	23.0	7.0	15.0	0.94	3.30
61.4	13.9	9.7	15.0	0.95	3.15
65.0	14.0	6.0	15.0	0.98	3.06

Premix (9.9% sugar, 3% moringa, 1.5% fenugreek and 0.6% salt)

The analyses of variance (ANOVA) of functional properties of composite instant infant flour were summarized in Appendix table 5. ANOVA has shown significance ($p < 0.05$) difference.. High coefficient of determination (R^2 value) indicates that the model can sufficiently predict the responses.

4.4.1. Bulk density

The mean values of bulk density of composite instant flour ranges from 0.93 to 0.98gm/ml. ANOVA has shown a highly significant ($p < 0.01$) difference in quadratic model, interaction of oat with soybean and soybean with linseed. The regression models, developed to predict the bulk density of the extruded composite flour explained 97.97% of the variations (Appendix table 5).

The bulk density of the flour is high in blend at which the proportion of oat is highest and low at highest proportion of soybean and linseed in the blend, as described in the (Table 12). This may relate to high fiber contents in oat which contributes for increasing bulk density of the flour. Increasing the proportion of oat flour in the formulation increased the fiber contents of extruded composite flours. In opposite to this, increased proportion of soybean decreased fiber contents of the extruded composite flour as indicated in contour plot (Figure 15).

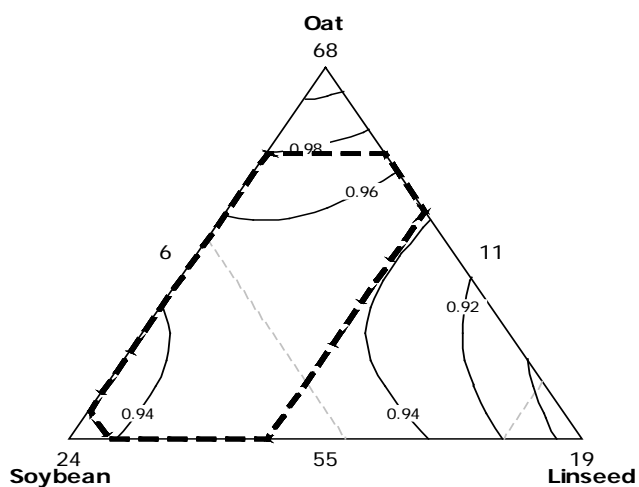


Figure 15: Contour plot of bulk density (g/ml) of instant complementary flour

Bulk density is a measure of heaviness of flour (Oladele and Aina, 2007). The value of bulk density of this finding is within the range of 0.96- 0.98gm/ml reported by Kavitha and Parimalavalli (2014) for extruded complementary foods.

The finding of this thesis is in agreement with the finding of Veronica *et al.* (2006) who reported that as fiber and protein-rich materials are added to starchy materials, the density of expanded product is increased. Bolarinwa *et al.* (2015) reported that the bulk density of the composite flour decreased with increasing level of soybean flour substitution in the mixes. Ryu, (2004) reported that bulk density of the product increases with increasing fiber content. Siddharth, (2014) also reported that the higher bulk density may be due to the presence of more crude fiber in the composite flour sample.

4.4.2. Water Absorption Capacity

The mean value of water absorption capacity (WAC) of extruded flour varies from 3.06- 3.41ml. ANOVA has shown highly significant ($p < 0.01$) effects in quadratic model and interaction between oat with linseed, soybean with linseed, but the interaction of oat and soybean was not significant ($p > 0.05$). Coefficient of determination model, developed to predict the WAC of the extruded composite flour explained 96.75% of the variations (Appendix table 5). The regression model for WAC of extruded composite flour was

represented in Equation (Appendix table 6) as indicated in quadratic model with three variables.

The minimum water absorption capacity of composite instant flour was recorded at highest proportion of oat flour in the formulation and maximum values of water absorption capacities were recorded at high proportion of soybean and linseed flour in the formulation as indicated in contour plot (Figure 16) for extruded composite flour.

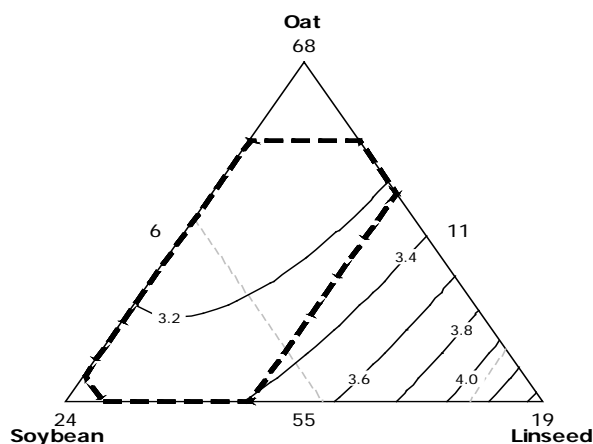


Figure 16: Contour plot of WAC in (ml) of instant complementary flour

The water absorption capacity gives an indication of the amount of water available for gelatinization (Kulkarni, *et al*, 1990). Water absorption capacity depends on the availability of the hydrophilic groups to bind water molecules and the gel-forming ability of the macromolecules. Indications are that the damaged starch granules absorb considerable amounts of water at room temperature and swells resulting in increased viscosity (Colonna *et al*. 2004).

The water absorption capacity of the blend was increased as the proportion of soybean and linseed increased in the blend. This may be, because of high water absorption capacity of soybean as compared to oat; the protein denatured during extrusion cooking made the soybean flour to absorb more water.

The result is in agreement with the observation of Igyor *et al*. (2011) who suggested that protein functions in binding water and fat while retaining them. Thus, the availability of

soybean protein has increased its ability to absorb water. Amarjeet *et al.* (1995), who reported that increased water absorption with increased soybean flour fortification. Ojinnaka *et al.* (2013) also reported that water absorption capacity increasing with increased inclusion of soybean flour to the samples

4.6. Sensory Quality of Gruel Made from Instant Complementary Flour

The mean values of sensory attributes (appearance, aroma, taste, mouth feel consistency and over all acceptability) are presented in Table 13. The minimum and maximum values of appearance, aroma, taste, mouth feel, and consistency and over all acceptability of instant composite flour were 2.96-3.72, 3.56-3.77, 3.45-3.80, 3.40-3.80, 3.60-3.96, and 3.61-4.0 respectively. The mean score of sensory attributes of the gruel prepared from barley flour and ‘mittin’ were also indicated (Table 13). The mean scores of sensory evaluation showed that all the gruel prepared from extruded composite flours were within the acceptable range (scores > 3) from 5 point hedonic scale.

Table 13: Mean score of sensory result of gruel evaluated by panelist.

Mixture composition (%)				Sensory score (5 point hedonic scale)					
Oat	Soy bean	Linseed	Premix*	Appearance	Aroma	Taste	Mouth feel	consistency	OA
57.92	19.92	7.17	15	3.39	3.60	3.48	3.65	3.67	3.62
57.42	17.92	9.67	15	3.36	3.72	3.55	3.72	3.68	3.80
56.00	23.00	6.00	15	3.32	3.64	3.56	3.60	3.76	3.72
65.00	11.00	9.00	15	3.20	3.68	3.80	3.48	3.96	4.00
62.42	15.42	7.17	15	3.46	3.64	3.63	3.54	3.80	3.64
55.00	19.00	11.0	15	3.12	3.77	3.45	3.68	3.68	3.61
62.42	13.92	8.67	15	3.36	3.62	3.66	3.60	3.84	3.86
57.42	19.92	7.67	15	3.24	3.60	3.52	3.68	3.60	3.96
63.00	11.00	11.0	15	2.96	3.68	3.52	3.72	3.93	3.80
59.83	16.83	8.33	15	3.48	3.56	3.64	3.63	3.64	3.76
55.00	23.00	7.00	15	3.44	3.74	3.60	3.80	3.80	3.84
61.42	13.92	9.67	15	3.28	3.60	3.56	3.60	3.88	3.75
65.00	14.00	6.00	15	3.72	3.76	3.60	3.40	3.80	3.76
Barley				3.56	3.60	3.72	3.68	3.84	3.96
‘Mitin’				3.72	3.52	3.32	3.64	3.84	3.76

Premix (9.9% sugar, 3% moringa, 1.5% fenugreek and 0.6% salt), OA= Overall acceptability, Mitin

Sensory qualities are the main criterion that makes a product to be liked or disliked (Falola *et al.*, 2011). Sensory analysis or sensory evaluation is a scientific discipline that is done by the use of human senses for the purposes of evaluating the suitability and preference of the consumer to the products. Organoleptic properties are the aspects of food or other substances as experienced by the senses, including taste, sight, smell, and touch (Delwiche, 2004)

Mean comparisons of gruel from composite instant flour with gruel from ‘*mitin*’ and barley flour was made to see the sensory acceptance of the currently developed formulation with what the community is already using. Accordingly, in all sensory attributes, the gruel made from extruded composite flour was preferred. The maximum appearance score for ‘*mitin*’ and the developed formulation was equal as indicated in Table 13 but for others the currently developed product was preferred.

Analysis of variance, for quadratic, interaction among the ingredients and coefficient of determination in % for sensory attributes (appearance, aroma taste, mouth feel, consistency and overall acceptability) are summarized in Appendix table 7. ANOVA showed that all the sensory attributes (except for appearance and mouth feel) are significantly ($p < 0.05$) different. The values of Coefficient of determination (R^2 values) of sensory attributes indicated that the models can sufficiently predict the responses (Table 13)

4.6.1. Appearance

The appearance of the gruel sensory score ranges between 2.96 and 3.72. ANOVA has showed that the model developed for predicting appearance is satisfactory. The coefficient of determination models, developed to predict the appearance could explained 82.90% of the variations (Appendix table 7) and the regression model for predicting appearance is presented as equation (Appendix table 8).

The minimum score of appearance was recorded at the highest linseed and high soybean flour in the formulation. The maximum score for appearance was obtained at the highest proportion of oat flour in the gruel. The sensory acceptance of appearance of the gruel increased as the proportion of oat flour increased in the formulation, but the sensory score of appearance was decreased with increasing proportion of soybean and linseed flour in the gruel (Figure 17).

It may be due to white color of oat flour that makes the appearance of the gruel more acceptable at high proportion of oat flour.

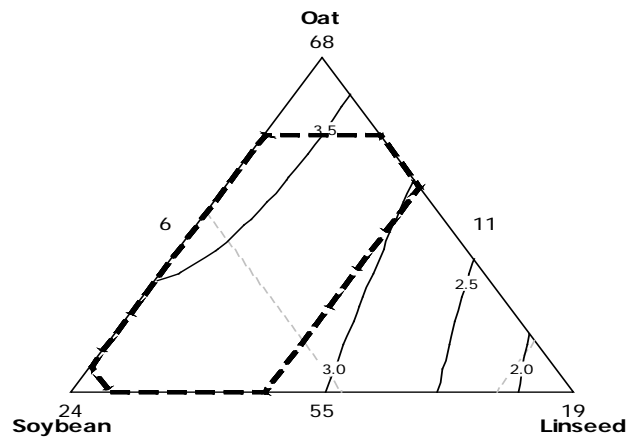


Figure 17: Contour plot of appearance (5 point hedonic scales) of the gruel

Appearance is one of organoleptic attributes that evokes the initial response of panelist. Vision plays a major role in sensory analysis and the appearance of food can have a major effect on its acceptability (Tizazu *et al.*, 2010). The finding of present study is in agreement with United States Department of Agriculture, (2007) report; the probable reason for the low score of appearance at high percentage of linseed could be brown color of linseed which became dark brown at high baking temperature. Maillard reaction may have also contributed to the darker color of bakery products due to the high protein content of flaxseed and soy bean (Borrelli *et al.*, 2003).

Additionally the presence of moringa affects the appearance of the gruel due to its green color (chlorophyll contents of moringa leaves). This finding is in line with the result of Gebretsadikan *et al.* (2015) who reported that the presence of moringa influences the degree of acceptance of a product.

Even though the added moringa reduce the appearance of the gruel, the gruel prepared from extrusion cooked composite has the same maximum sensory score for appearance with the reference gruel made from ‘mittin’ but its better preferred than barley flour gruel.

4.6.2. Aroma

The mean aroma scores of the gruels vary from 3.56 to 3.77. The aroma of the gruel showed highly significant ($p < 0.01$) difference in quadratic model and interaction between oat and soybean and showed significant ($p < 0.05$) difference in interaction between oat and linseed. The interaction of soybean and linseed showed no significant difference. The regression models, developed to predict the aroma explained 81.89% of the variations (Appendix table 1) and the regression model for predicting aroma is presented as equation (Appendix table 8).

The lowest aroma score was recorded at high proportion of oat in the blend (59.83%, 16.83% and 8.33%) in oat-soybean and linseed blend respectively and the highest score for aroma was recorded at high proportion of soybean and linseed in the blend (55%, 19% & 11%) oat, soybean and linseed blend respectively. Contour plot Figure 18 indicated that aroma of the gruel increases with increasing of oat flour until certain point, but increasing of linseed flour proportion increases aroma score of the gruel consistently.

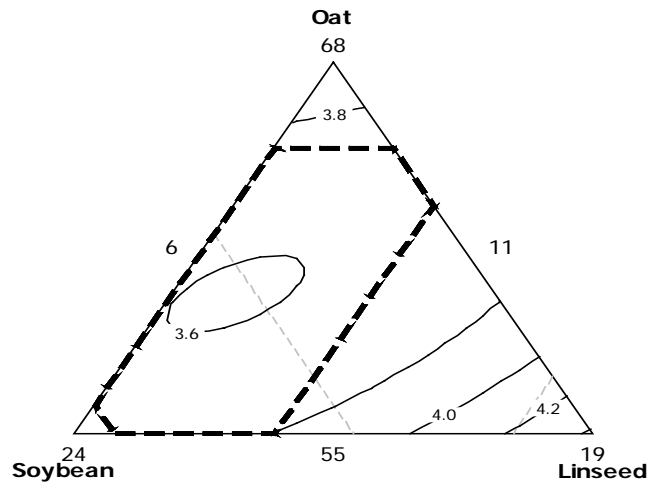


Figure 18: Contour plot of aroma (5 point hedonic scales) of the gruel

The high preference of aroma attribute at high soybean and linseed may be due to flavor imparted by soybean and linseed up on heating during extrusion cooking. This result is in agreement with Ojinnaka *et al.* (2013) who reported that the increase in aroma ratings could be due to the presence of flavor imparted by the oils in soybean.

The extrusion process enhances the acceptability of gruel prepared from extruded soybean flour because of dextrinization and starch breakdown (Mensah and Tomkins, 2003).

The gruel prepared from extruded composite flour scored high sensory values as compared to the reference gruel prepared from 'mittin' and barley flour, this might be because of ingredients used and the extrusion cooking that enhance the acceptability of the gruel.

Aroma is an integral part of taste and general acceptance of the food before it is put in the mouth (Muhimbula *et al.*, 2011). Aroma enhances acceptability of complementary foods. The sensations of taste and smell are functions of flavor which is a complex of sensations (Iwe, 2007). It is the flavor of a food that ultimately determines its acceptance or rejection, even though its appearance evokes the initial response.

4.6.3. Taste

The mean sensory score of taste for the gruels varies from 3.45 to 3.8. ANOVA has showed that the model for predicting taste was significant ($p < 0.05$) difference in quadratic and interaction with the ingredients. The regression models, developed to predict the taste explained 82.60% of the variations (Appendix table 7) and the regression model for predicting taste is presented as equation (Appendix table 8)

The minimum taste score (3.45) of the gruel was recorded at high proportion of soybean and linseed (55%: 19%: 11%) oat, soybean and linseed blend respectively. The maximum taste sensory score (3.80) of the gruel was recorded at high proportion of oat in the formulation (65%: 11%: 9%). The taste of the gruel increased with increasing proportion of oat where as decreased with increasing proportion of linseed in the blend as indicated by contour plot (Figure 19). This may be due to development of taste during extrusion cooking in which oat starch is degraded in to its simplest form.

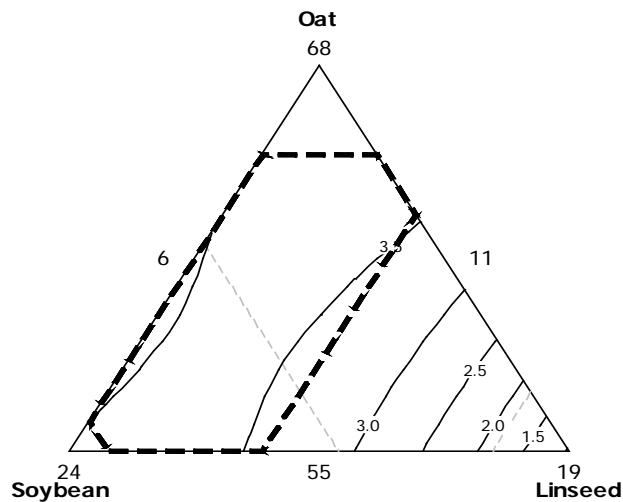


Figure 19: Contour plot of taste (5 point hedonic scales) of the gruel

The sensory acceptance taste of the gruel was increased with increasing proportion of oat. This may be due to beany taste of soybean and unacceptable high nutty flavor imparted by linseed. The finding of this thesis is in agreement with the result of Tortoe *et al.* (2014) who suggests that higher proportions of legume flour in the blend reduced taste for the porridge. The observation is likely to have resulted from coarseness and beany flavor introduced by the flour from legumes, which affected taste of the porridge. Beany flavor is related to lipoxygenases and is undesirable and greatly reduces the acceptability of products containing cowpea and soybean (Bott and Chambers, 2006).

Other studies also showed a reduction in acceptability of cowpea-fortified complementary foods as a result of the high cowpea inclusion levels because of the coarseness and beany flavor imparted by the cowpea (Olapade *et al.*, 2012). Similar studies indicated that acceptability of bread fortified with soybean flour also reduced with increase in proportion of soybean (Natal *et al.*, 2013).

The sensory score for the reference ('mittin' and barley) is very low as compared to extrusion cooked gruel. This may be related to extrusion cooking of the formulation in which starch is degraded in to its simplest sugar form and nature of the ingredient used in the extruded composite flour gruel's.

4.6.4. Mouth feel

The mean mouth feel score varies from 3.40 to 3.80. ANOVA has showed no significant difference ($p > 0.05$) among the samples and quadratic model in terms of the mouth feel; even though the mean value varies from 3.40 to 3.80. The coefficient of determination regression models, developed to predict the mouth feel explained 82.07% of the variations (Appendix table 7) and the regression model for predicting taste is presented by equation (Appendix table 7).

The least mouth feel value was scored at maximum proportion of oat in the blend (65%:14%:6%) and maximum sensory value of mouth feel was recorded at highest proportion of soybean and linseed in the blend (55%:19%:11%) oat, soybean and linseed blend respectively as indicated in Figure 20.

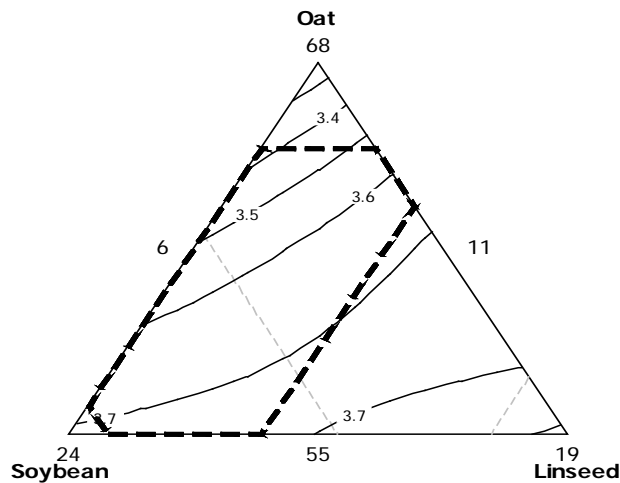


Figure 20: Contour plot of mouth feel (5 point hedonic scales) of the gruel

The mouth feel is very important in a complementary food as it will determine the amount of food an infant would consume since they can only swallow a smooth gruel not a coarse one. The reference gruel sample prepared from 'mittin' and barley flour had relatively low sensory score for mouth feel as compared to extruded composite flour gruel samples. The probable reasons for high mouth feel score for extruded composite flour may be the extruded composite flour was grinded in to fine flour. Similar result was reported by Swai (2013)

grinding the extrudates after extrusion resulted in very fine textured flour that made cooked gruel to be very smooth.

4.6.5. Consistency

The mean value for the consistency scores of the samples by the panelists varies from 3.60-3.96. ANOVA has shown that the model for predicting consistency is significant ($p < 0.05$) difference at quadratic model and interaction between oat and soybean. The coefficient of determination indicates that models, developed to predict the consistency explained 84.47% of the variations (Appendix table 7) and the regression model for predicting consistency is presented as equation(Appendix table 8).

The minimum mean values of sensory score for consistency was recorded at higher proportion of soybean flour where as the maximum mean sensory score of consistency was recorded at high proportion of oat and linseed in formulation. The sensory acceptance of the gruel for consistency was increased with increasing proportion of oat and linseed where as increasing the proportion of soybean flour decreased the sensory acceptance for consistency of the as shown in gruel figure 21.

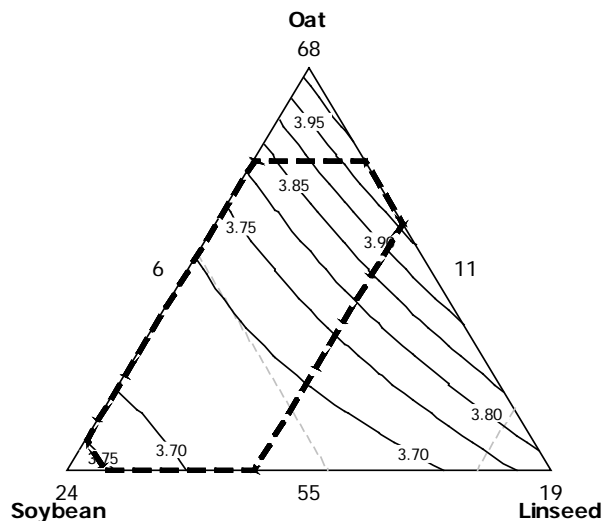


Figure 21: Contour plot of consistency (5 point hedonic scales) of the gruel

Increasing of oat flour proportion in the blend increased the consistency score of the gruel; this may be due to starchy properties of oat and linseed that makes the gruel viscous.

The sensory score of the reference gruel is less as compared to extrusion cooked flour gruel; the probable reason may be the nature of the ingredients and processing methods. A very thick consistency would need increased effort to swallow, and therefore may limit the food intake in young children have not fully developed their ability in this aspect (King and Ashworth, 1987).

4.6.6. Overall acceptability

The sensory score of overall acceptability of the complementary gruel ranged from 3.61 to 4. The ANOVA has shown that the model for predicting overall acceptability is satisfactory. The coefficient of determination models, developed to predict the overall acceptability of the gruel explained 61.23% of the variations (Appendix table 7) and the regression model for predicting taste is presented as Equation (Appendix table 8).

The minimum overall acceptability was scored at high proportion of soybean, linseed and low proportion of oat flour in the blends (55%-19%-11%). The highest mean sensory acceptance score was recorded at the highest proportion of oat and lowest soybean flour in oat, soybean and linseed (65%-11%-9%) respectively. Figure 22 indicates that over all acceptability of the gruel increased with increasing proportion of oat where as decreased with increasing the proportion of soybean flour in the formulation.

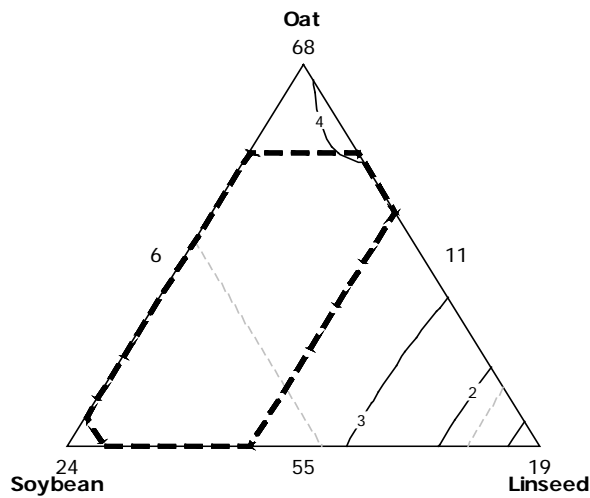


Figure 22: Contour plot of overall acceptability (5 point hedonic scales) of the gruel

Even though the presence of soybean and linseed improved the nutritional quality of the gruel, at higher contents of these ingredients the overall acceptability of the gruel is lower. At high percentages of soybean the appearance and taste of the gruel was low, which may contribute to the lower preference of gruel at high proportion of soybean.

The result of current study is in line with, Tortoe *et al.* (2014) who suggested that higher proportions of the legume flour in the blend reduced general preference for the porridge. The observation is likely to have resulted from a combination of reasons such as coarseness and beany flavor introduced by the flour from legumes, which affected taste of the porridge.

Singh *et al.* (1996); also suggested that the decrease in over all acceptability scores of the chapattis prepared from blended wheat flour with increasing the level of soybean flour incorporation may be attributed due to the typical undesirable taste soybean flour had.

The overall acceptability score for gruel made from extrusion cooked flour is greater than the reference gruel made from barely and ‘Mittin’ flour. This might be because of ingredient contents (used) in extruded composite flour that gives better preference for overall acceptability score of the gruel.

4.7. Optimal Mixture

4.7.1. Optimization based on nutritional composition

The main objective of using mixture design is to determine optimum formulation that can fulfill minimum nutritional requirements of child under two years. The regions of interest for nutritional composition of; protein, fat, carbohydrate, calories, calcium, Iron, and zinc varied between 19.0-20.6%, 8-9.9%, 61.09-63.7%, 394.98-408.2kcal/100g, 110-124.4mg/100g 7.0-7.8mg/100g and 2.97-3.0 mg/100g respectively. The ranges were selected as higher values of each response are within the range of recommended nutrient requirements for children under age of two years.

The white region in figures 23 indicates that any point within this region represents an optimum combination of oat, soybean and linseed, with desirable nutritional composition. The overall optimum nutritional composition were found in the blend of 58.3%, 19.1% and 7.6% oat, soybean and linseed respectively from range of oat 55-65%, soybean 11-23% and linseed 6-11%.

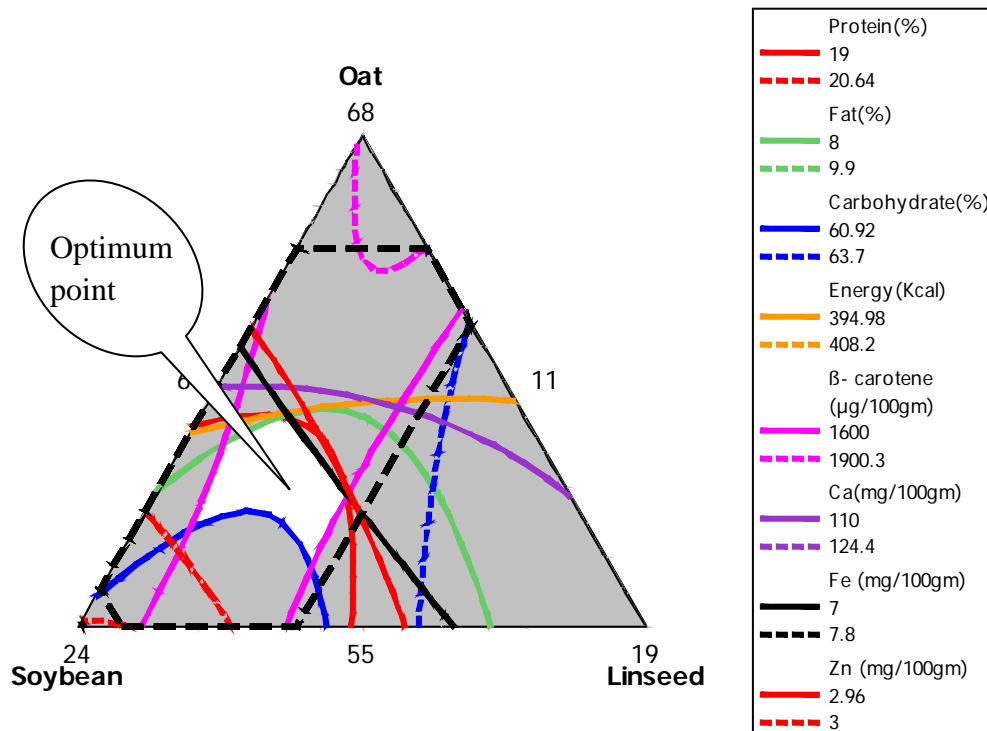


Figure 23: Overlaid contour plots of instant complementary flour nutritional composition

4.7.2. Optimization based on sensory evaluation

To obtain the optimum region for appearance, aroma, taste, mouth feel, consistency and overall acceptability for gruel the ingredients were mixed at different ratio. The optimum formulation should maximize consumer acceptance, it is possible to develop a product with all sensory qualities that would satisfy consumer's acceptability of the product. Sensory optimization was done based on each sensory attributes as received hedonic rating >3.36 from 5 point hedonic scale.

The white region in Figure 24 represents an optimum combination of oat, soybean and linseed, with desirable sensory acceptability. The optimum point prediction shows that 62.8% oat, 14% soybean and 8.2% linseed. The optimal point for appearance, aroma, taste, mouth feel, consistency and overall acceptability were found to be ranged from 3.38-3.72, 3.60-3.77, 3.6-3.8, 3.54-3.8, 3.67-3.96 and 3.75-4.0 respectively. The ranges were selected as higher score indicates better preference of the gruel and with suitable chemical composition also.

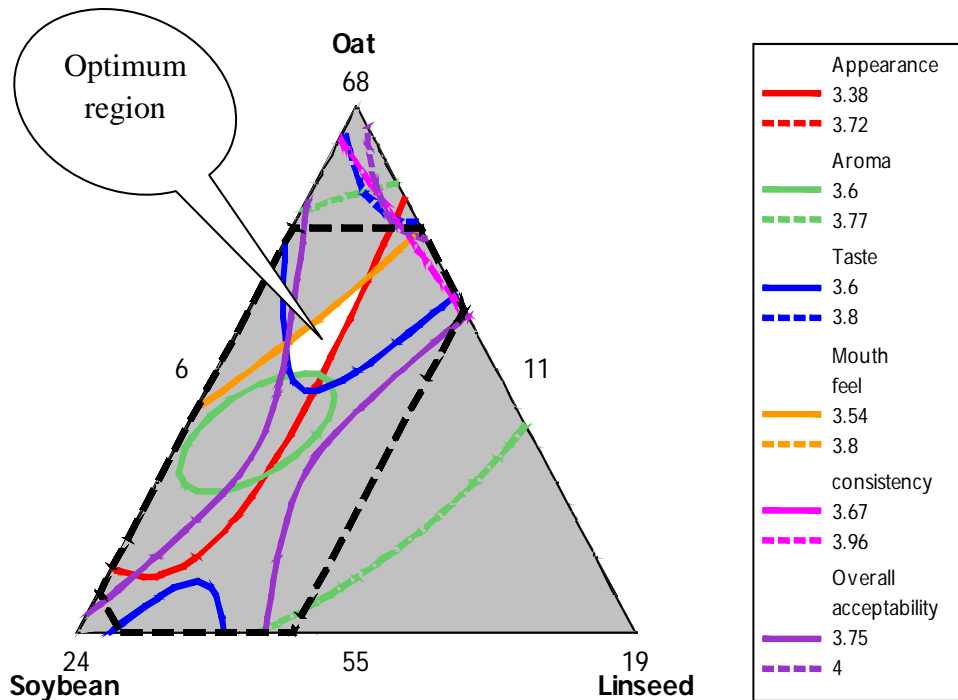


Figure 24: Overlaid contour plots of instant complementary flour gruel sensory attributes

4.7.3. Overall optimal mixture compositions

The study focused on determining the optimal blend ratio of individual food source (ingredients) that are suited to produce instant infant flour with desirable nutrient compositions and acceptable sensory attributes. In order to determine the optimum formulation, the regions of acceptability in the contour plot for protein, carbohydrate, fat, calorie, iron, calcium, zinc, beta carotene and overall sensory attribute were superimposed.

Superimposition of contour plot regions of interest protein (19.0-20.6%), fat (8-9.9%), carbohydrate (60.92-63.7%), energy (394.98-408.2kcal/100gm), calcium (110-124.4mg/100gm) iron (7-7.8mg/100g), zinc (2.96-3.0 mg/100g), beta carotene (1600-1900.3µg/100gm) and overall acceptance received hedonic ratings (3.75- 4.0) resulted in optimum regions for extruded composite flour. The white region in Figure 25 indicates that any point within this region represents an optimum combination of oat, soybean and linseed, with optimal nutritional composition and sensory attributes. The overall optimum blends were found at 58.4, 18.4 and 8.2 oat, soybean and linseed respectively in a range of 55-65% oat, 11-23% soybean and 6-11% linseed.

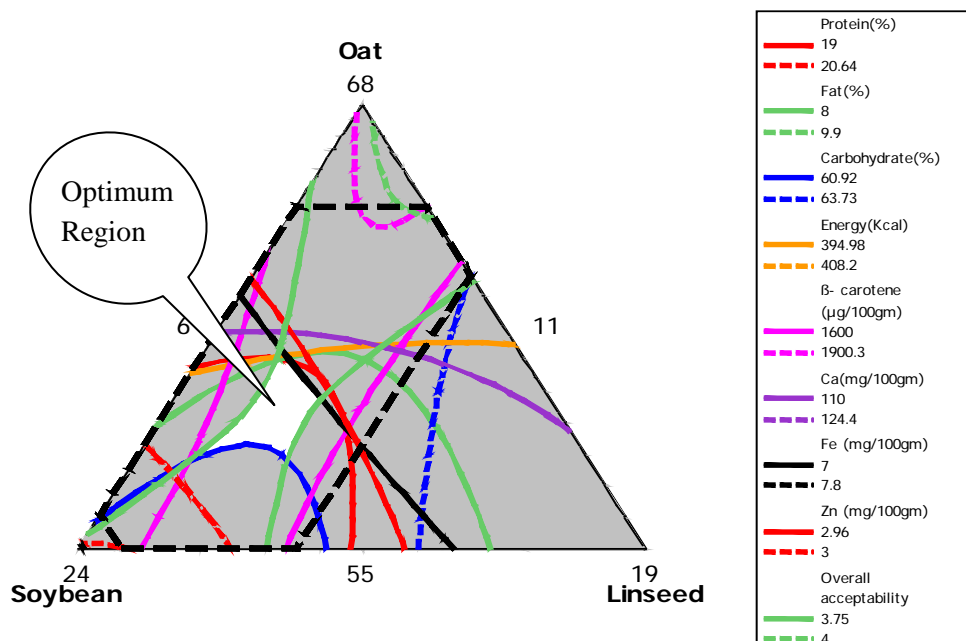


Figure 25: Overlaid contour plots of instant complementary flour nutritional composition and gruel sensory attributes'

5. SUMMARY AND CONCLUSIONS

5.1. Summary

The current study was aimed at developing new instant infant flour, which is enriched with protein, fat, iron, zinc and vitamin A (β -carotene) to be used as a complementary food for under two years old children. The product was developed from mixture of oat (55%-65%), soybean (11%-23%) linseed (6%-11%) and 15% premix (sugar, salt fenugreek and moringa).

The result of proximate composition in this study indicated that, the protein, fat and energy content were significantly improved with increasing the proportion of soybean and linseed flour in the composite flour. Whereas, moisture, fiber and carbohydrate contents of the extruded composite flour were increased with increasing proportion of oat flour in the blend. Moreover, besides the presence of moringa as a source of mineral and vitamin in the composite flour, calcium, iron, and zinc content of the product increased significantly with the increasing proportion of soybean and linseed flour. Anti- nutritional contents (phytate and tannin concentration) in the extruded composite flour were also increased as the proportion of soybean flour increased. The β -carotene content was decreased with increasing proportion of soybean flour.

In other way, the functional properties of the extruded composite flour showed significant ($p < 0.05$) difference. Thus, the water absorption capacity of the extruded composite flour was increased with increasing proportion of soybean and linseed flour in the formulation. While, bulk density was decreased with increasing proportion of soybean and increased with increasing proportion of oat in the blend.

The sensory acceptance of the gruel show little decrement with increasing proportion of soybean flour. whereas, the gruel prepared from highest oat content in the blend scored better value in terms of overall acceptability but, not nutritionally.

Finally, the optimization study indicated that, the optimal blend of composite flour containing 58.4% oat, 18.4% soybean and 8.4% linseed was selected as superior in terms of chemical composition (protein 19.0-20.6%, fat 8-9.9%, carbohydrate 60.92-63.7%, energy 394.98-408.2kcal/100g, calcium 110-124.4mg/100g, Iron 7-7.8mg/100g, zinc 2.96-3.0) and sensory qualities (3.75-4 overall acceptability) .

5.2. Conclusion

This result revealed that extrusion cooked oat, soybean, linseed and premix (sugar salt fenugreek and moringa) blend can be used to produce nutritious and ready-to-use composite flour. The extruded composite flour have good potential to improving vitamin A (β -carotene), fat, zinc, iron, protein and energy rich complementary foods. The blends were extruded to provide pre-cooked foods that could be reconstituted at 75°C to a porridge or gruel, eliminating prolonged cooking or degradation of heat labile nutrients. The use of these locally grown cereals, legumes and oilseed could make a great contribution to the fight against malnutrition in the study area.

The optimum mixture of oat, soybean and linseed flour with added premix (sugar, salt, moringa and fenugreek) induced significant improvement in the composite flour's nutritional quality and gruel's sensory attributes.

Generally, the study successfully produced a nutritious and energy-dense complementary food with acceptable sensory attributes from locally available raw materials. The product developed can be used as ready-to-use meal that needs minimum processing time. Consumption of these alternative food ingredients in a form of gruel can suppress nutrient deficiencies and can play a significant role in developing countries as a viable long-term food-based strategy to control nutrient deficiencies.

6. FUTURE LINE OF WORK

The extruded composite instant flour developed has a good nutritional content and sensory acceptability and can fulfill nutrient requirement of children under the age of two years for fat, protein carbohydrate, energy, and Fe, Zn and beta carotene. Therefore, the concerned bodies should give attention for the developed instant composite flour to make it available to rural area of our country, especially, southwest parts of Ethiopia, where malnutrition is the main problem of infants and children.

Developing instant infant flour at small scale creates great chance both in creating job opportunities and in reducing malnutrition. Therefore, all the concerned bodies should give attention to supporting small scale entrepreneurs in producing instant infant flour.

On the other hand, this study did not cover the digestibility and bioavailability of the macronutrients and amino acid of the developed product; therefore, it needs further investigation. In addition, starch characteristics as well as the shelf life of extruded composite flour needs further investigation.

Finally, the composite flour developed in this study did not meet all the recommended micronutrients especially calcium requirements for infants and children under two years. Therefore, fortification with appropriate micronutrients or micronutrient-dense foodstuffs will be necessary.

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8. APPENDICES

Appendix table 2: Analysis of variance (ANOVA) for proximate composition and energy

Source	MC	FiC	PC	FC	AC	CC	Energy
Linear	0.11	0.00	0.01	0.26	0.01	0.01	0.69
Quadratic	0.13	0.00	0.01	0.02	0.01	0.00	0.94
X1*X2	0.86	0.67	0.06	0.27	0.01	0.01	0.85
X1*X3	0.28	0.00	0.01	0.18	0.01	0.01	0.81
X2*X3	0.10	0.00	0.04	0.02	0.00	0.00	0.75
R ²	96.11	98.86	98.64	98.39	94.50	97.86	98.14

X1=oat; X2=soybean; X3=linseed; MC=moisture content; FiC= crude fiber content; PC=protein content; FC=crude fat content; AC=ash content & CC= carbohydrate content R²=Coefficient of determination in %

Appendix table 3: Estimated regression coefficients of proximate compositions and energy contents of instant complementary flour

Response	Estimated regression coefficients equations
Moisture	$0.61X_1 + 113.9X_2 - 32X_3 + 180.3X_1X_2 + 34.7X_1X_3 + 116.4X_2X_3$
Crude fiber	$6.7X_1 + 5.7X_2 + 175.6X_3 - 2.7X_1X_2 - 209.9X_1X_3 - 220.06X_2X_3$
Protein	$13.5X_1 + 76.6X_2 - 288.1X_3 - 77.0X_1X_2 + 389.9X_1X_3 - 306.2X_2X_3$
Crude fat	$6.3X_1 + 30.3X_2 - 183.3X_3 - 47.6X_1X_2 + 203.1X_1X_3 + 432.3X_2X_3$
Crude Ash	$5.3X_1 + 31.4X_2 - 184.3X_3 - 63.7X_1X_2 + 198.9X_1X_3 + 311.1X_2X_3$
Carbohydrate	$66X_1 + 36X_2 + 801X_3 + 75X_1X_2 - 84419X_1X_3 - 1173X_2X_3$
Energy	$373.4X_1 + 737.2X_2 + 405.2X_3 - 442.1X_1X_2 + 10.2X_1X_3 + 419.6X_2X_3$

X1=oat; X2=soybean; X3=linseed

Appendix table 4: Analysis of variance (ANOVA) for minerals and beta carotene contents

Source	Calcium	Iron	Zinc	β-carotene
Linear	0.059	0.001	0.341	0.000
Quadratic	0.642	0.763	0.935	0.000
X1*X2	0.945	0.643	0.754	0.098
X1*X3	0.314	0.866	0.700	0.000
X2*X3	0.276	0.568	0.590	0.000
R ²	99.73	99.87	98.97	96.69

X1=oat; X2=soybean; X3=linseed, R²=Coefficient of determination in %

Appendix table 5: Estimated regression coefficients of minerals and beta carotene contents of instant complementary flour

Response	Estimated regression coefficients equations
Calcium	$58.3 X_1 + 238.3 X_2 - 1.357 X_3 - 9.9 X_1 X_2 + 503.7 X_2 X_3$
Iron	$4.48 X_1 + 13.66 X_2 + 10.49 X_3 + 2.5 X_1 X_2 - 3.06 X_1 X_3 - 11.41 X_2 X_3$
Zinc	$2.8465 X_1 + 3.675 X_2 + 1.3579 X_3 - 0.378 X_1 X_2 + 1.5767 X_1 X_3 + 2.4192 X_2 X_3$
Beta-carotene	$2088X_1 + 1521X_2 - 242993X_3 - 18981X_1X_2 + 280258X_1X_3 + 366032X_2X_3$

X1=oat; X2=soybean; X3=linseed

Appendix table 6: Analysis of variance (ANOVA) for antinutritional factor and functional properties

Source	Phytate	Condensed tannin	Bulk Density	WAC
Linear	0.416	0.015	0.346	0.004
Quadratic	0.015	0.059	0.000	0.022
X1*X2	0.073	0.089	0.001	0.192
X1*X3	0.656	0.158	0.172	0.021
X2*X3	0.061	0.182	0.012	0.024
R ²	89.86	92.87	97.97	96.75

X1=oat; X2=soybean; X3=linseed, R²=Coefficient of determination in%

Appendix table 7: Estimated regression coefficients of anti-nutritional factors and functional properties of instant complementary flour

Response	Estimated regression coefficients equations
Phytate	$362X_1 + 921 X_2 - 2402X_3 - 1997X_2X_3 + 1493 X_1X_3 + 7821X_2X_3$
Tannin	$43X_1 + 508 X_2 + 1671X_3 - 745X_2X_3 - 2009 X_1X_3 - 2054 X_2X_3$
Bulk density	$1.4783X_1 + 1.889X_2 + 1.514X_3 - 3.514X_1X_2 - 4.054X_1X_3 + 7.972X_2X_3$
WAC	$1.4783X_1 + 1.889 X_2 + 1.514X_3 - 3.514X_1X_2 - 4.054X_1X_3 + 7.972 X_2X_3$

X1=oat; X2=soybean; X3=linseed

Appendix table 8: Analysis of variance (ANOVA) p-values for sensory acceptability value of the gruel

Source	Appearance	Aroma	Taste	Mouth feel	consistency	Overall acceptability
Linear	0.296	0.009	0.015	0.781	0.47	0.062
Quadratic	0.092	0.006	0.029	0.242	0.031	0.096
X1*X2	0.630	0.008	0.02	0.713	0.043	0.075
X1*X3	0.895	0.036	0.01	0.332	0.917	0.040
X2*X3	0.342	0.401	0.01	0.869	0.714	0.059
R ²	82.90	81.89	82.60	82.07	84.47	61.23

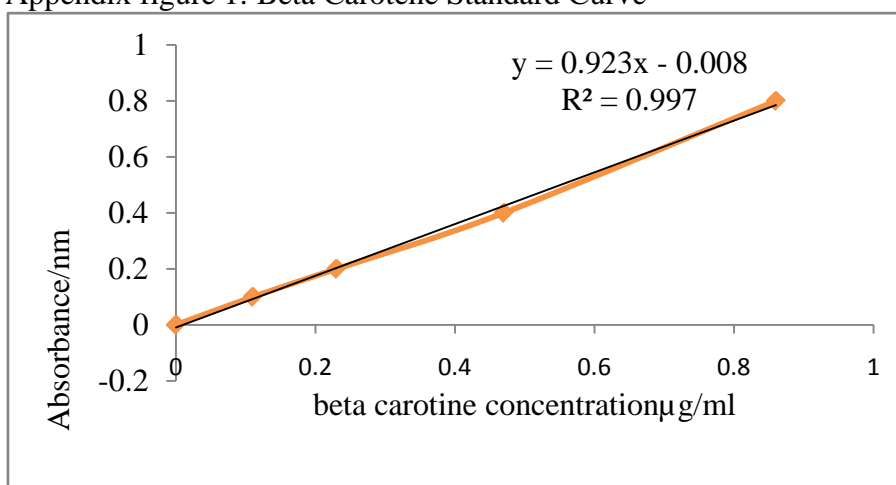
X1= oat; X2=soybean; X3=linseed, R²=Coefficient of determination in%

Appendix table 9: Estimated regression coefficients of sensory acceptability of instant complementary gruel

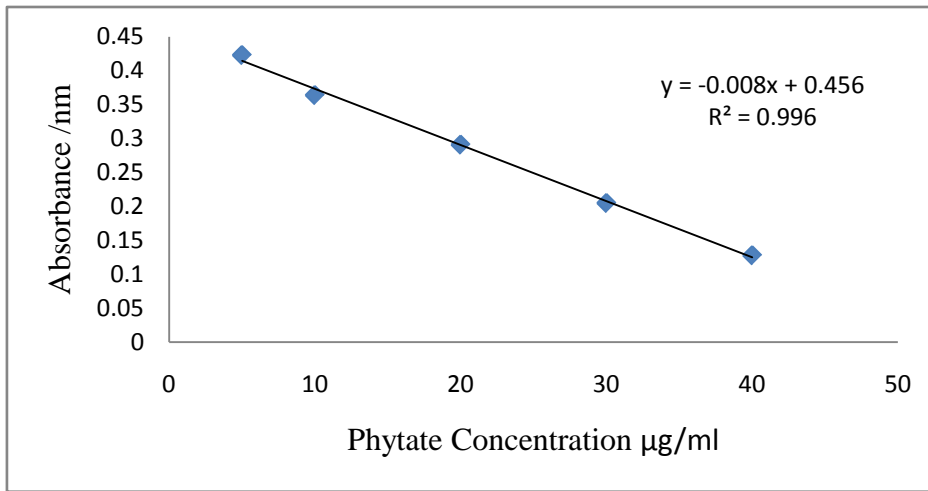
Response	Estimated regression coefficients equations
Appearance	$5.02X_1 - 10X_2 - 26.61X_3 + 13X_1X_2 + 11.97X_1X_3 + 97.1X_2X_3$
Aroma	$7.8X_1 + 20.19X_2 + 59.24X_3 - 35.18X_1X_2 - 85.25X_1X_3 - 32 X_2X_3$
Taste	$5X_1 + 53.5X_2 - 124.3X_3 - 37.2X_1X_2 + 146.6X_1X_3 + 162.1X_2X_3$
Moth feel	$1.19X_1 + 4.36 X_2 - 27.66X_3 + 5.75X_1X_2 + 53.02X_1X_3 + 9.48X_2X_3$
Consistence	$5.7X_1 + 25.25X_2 - 1.13X_3 - 37.11X_1X_2 + 5.44X_1X_3 - 20.84X_2X_3$
Overall acceptability	$4.7X_1 + 30.7X_2 - 172.8X_3 - 51.8X_1X_2 + 210.5X_1X_3 + 206.4X_2X_3$

X1= oat; X2=soybean; X3=linseed

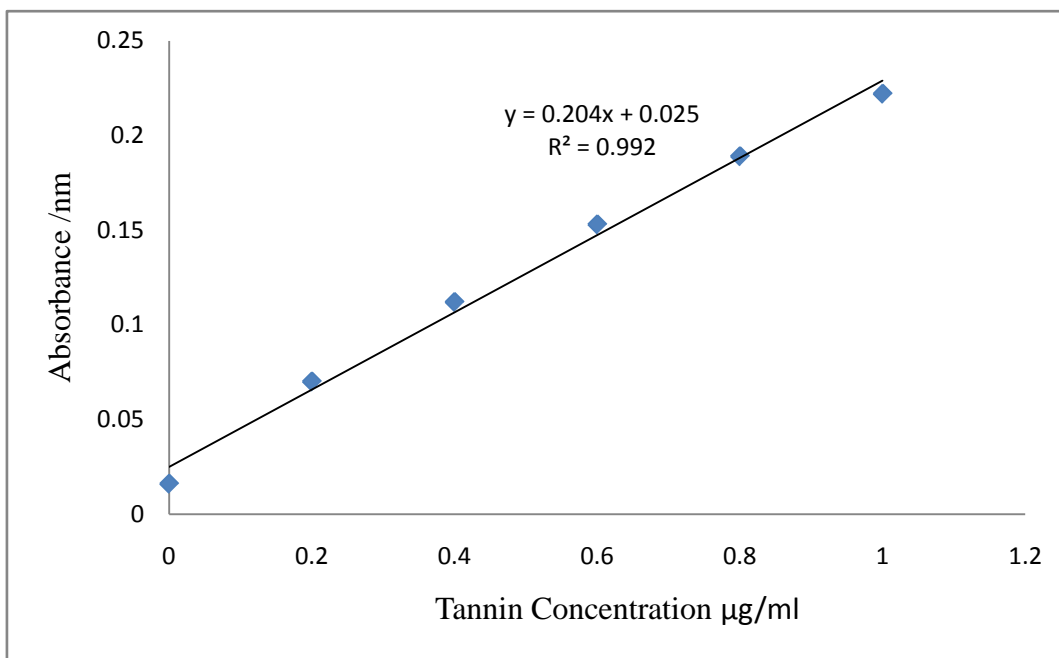
Appendix figure 1: Beta Carotene Standard Curve



Appendix figure 2: Phytate standard curve



Appendix figure 3: Tannin standard curve



Appendix table 10: Sensory evaluation questionnaire form

Please look at and taste each sample of gruel in order from left to right as shown on the ballot. Indicate how much you like or dislike each sample by checking the appropriate phrase of category which is listed below and mark your choice with the number that corresponds to your preference on each parameter.

- 1. Dislike Very Much
- 2. Dislike
- 3. Neither Like or Dislike
- 4. Like
- 5. Like Very Much

Sample code	Appearance	Aroma	Taste	Mouth feel	consistency	Overall acceptability
210						
211						
200						
201						
120						
122						
102						
100						
011						
012						
010						
022						
222						
111						
333						
123						

General Comments /suggestion about the product

