EFFECT OF FERMENTATION AND AMYLASE RICH FLOUR ON PHYSICO-CHEMICAL AND SENSORY PROPERTIES OF COMPLEMENTARY FOOD FROM OAT, BARLEY AND TEFF

M.Sc. Thesis

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April, 2016
Jimma, Ethiopia.

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M.Sc. Thesis

Submitted to the School of Graduate Studies Jimma University College of Agriculture and Veterinary Medicine

In Partial Fulfillment of the Requirements for the Degree of Master Science in Post Harvest Management (Durable crops)

By:

Alemgena Ayana

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I have completed my thesis research work as per the approved proposal and it has been evaluated and accepted by my advisers. Hence, I hereby kindly request the Department to allow me to present the findings of my work and submit the thesis.

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We, the thesis advisers have evaluated the contents of this thesis and found to be satisfactory, executed according to the approved proposal, written according to the standards and format of the University and ready to be submitted. Hence, we recommend the thesis to be submitted.

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DEDICATION

This thesis manuscript is dedicated to my beloved parents and my brother

STATEMENT OF THE AUTHOR

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BIOGRAPHICAL SKETCH

The author was born on 28 October 1990 in Jimma Arjo Woreda, East Wollega zone. He began his primary school at Chafe Arjo School in 1998 and attended junior and secondary high school at Mekonnen Demisew School from 2004 to 2005 at Arjo, Ethiopia. He continued his secondary and preparatory education in Arjo secondary and preparatory school from 2006 to 2009.At the same school he took Ethiopian School Leaving Examination in 2009 and successfully passed the examination and qualified to join Ethiopian Universities for higher studies. In 2010 he joined Jimma University graduated with Bachelor of Science degree (B.Sc) in post harvest management in July 2012. He also re-joined again Jimma University College of Agriculture and Veterinary Medicine in September 2014 to pursue post graduate study in Post Harvest Management specializing in Durable crops.

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ABBREVIATIONS AND ACRONYMS

Ab	Absorbance
ANOVA	Analysis of variance
AOAC	Association of Official Analytical Chemists
ARF	Amylase rich flour
BD	Bulk density
CIDA	Canada International Development Agency
CF	Complementary Food
cP	Centipoises
CSA	Central Statistics Authority
FAO	Food and Agriculture Organization
JUCAVM	Jima university College of Agriculture and Veterinary Medicine
LAB	Lactic Acid Bacteria
PEM	Protein Energy Malnutrition
RPM	Revolution per Minute
SSA	Sub Saharan Africa
SNNPR	Southern Nations Nationalities and People’s Region
TA	Titratable Acidity
WAC	Water Absorption Capacity
WFP	World Food Program
WHO	World health organization

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EFFECT OF FERMENTATION AND AMYLASE RICH FLOUR ON PHYSICO-CHEMICAL AND SENSORY PROPERTIES OF COMPLEMENTARY FOOD FROM OAT BARLEY AND TEFF

ABSTRACT

Inadequate complementary food is a major cause for the high incidence of child malnutrition, morbidity and mortality in many developing countries. In developing countries like Ethiopia, CF are usually made from starchy staples (cereals) with high viscosity, bulk density and anti-nutrients content. Literature indicates that there exist several modification techniques that increase nutrient density of CF. The purpose of this study was to evaluate the effects of fermentation and amylase rich flour (ARF) on physicochemical and sensory properties of complementary foods from oat, barley and teff. Experimental materials namely (teff, oat, malted barley and barley) used in this study were collected from Debrezeit and Sinana Agricultural research center, Asella malt factory and Holota local market respectively and prepared in to flour at laboratory.A3x3x3 factorial experiment (fermentation time, cereal type and malt concentration) in three blocks was set up. 2 and 5 % of ARF were added into oats, barley and teff flours and were fermented at room temperature (22-25^{o}C) for 24 and 48 hours at a concentration of 1:3 dilutions (w/v) and samples were withdrawn at 24h interval. Titratable acidity and pH were determined immediately at the end of each fermentation period and the samples were dried at 70°C for 36h. The samples were analyzed for proximate composition, anti-nutritional factors, and functional properties. The data was analyzed using analysis of variance (ANOVA) by using SAS version *9.2. Results showed that interaction effect of fermentation time, cereal type and ARF concentration had highly significantly ($p<0.01$) decreasing effect on pH, crude fiber, crude fat, total carbohydrate, phytic acid, tannin, bulk density, water absorption capacity and viscosity. In contrast TA, crude protein, calorific value were significantly ($P<0.01$) increased. The pH value was ranged between (3.89-6.40) and TA in (%) (0.09-0.80).The range mean values of proximate compositions in(%) were moisture(3.67–6.17),ash(1.75-2.71),protein(8.12–16.82),fat(1.63–4.55)fiber(1.58 5.96),CHO(71.20-78.18)and Energy in (kcal) (359.33–380.26). The results of anti-nutritional factors in (mg/100g) were phytate (18.63-175.07), tannin (0.84-42.89).The range mean values of functional properties were, bulk density (0.66g/ml-0.99g/ml), WAC (61.33%-143.12%) and viscosity (235cP-1016.33cP).The panelists ranked that appearance, aroma, taste, mouth feel and overall acceptability of the gruel prepared from oat, barley and teff flour that was subjected to 48h fermentation differ from others and least acceptable. In contrast gruels prepared from oat, teff and barley flour that was subjected to 48h fermentation period were acceptable with higher mean score 4.38, 4.16 and 4.02, for consistency. According to the results of this study, combined effect of fermentation and ARF appeared to be promising food processing method to significantly improve the nutrient and energy density and decrease the dietary bulkiness and viscosity values of CFs. Further research on mineral determination and shelf life study of oat, barley and teff based fermented CFs needs to be conducted in Ethiopia.*

***Key Words**: Fermentation, Amylase rich flour, physicochemical properties, Complementary food*

1. INTRODUCTION

Complementary feeding period is the most critical period in a child's life as an infant's transfer from nutritious and uncontaminated breast milk to the regular family diet with chance of vulnerable to malnutrition and disease (Yewelesew *et al.*, 2006).According to the WHO definition, a complementary food is normally a semi-solid food that is used in addition to breast milk and not to replace it and started at the age of six months. When a baby reaches 6-24 months old, breast milk alone is no longer sufficient to meet his nutritional requirement (Adenuga, 2010). Calories and other nutrients from complementary foods are needed to supplement breast-milk until the child is ready to eat the family diet (Nandutu and Howell, 2009).

The traditional complementary foods given to young children in most developing countries are based on local staples, usually cereals. During the cooking/reconstitution process, the starch granules swell and bind a large volume of water, resulting in gruels of high viscosity. Gruel of suitable feeding consistency contains a great deal of water and large volume relative to its contents of solid matter (Victor and Joseph, 2005).

In developing countries the interaction of poverty, poor health and poor complementary feeding practices has a multiplier effect on the general welfare of the children population. It also contributes significantly towards growth retardation, poor cognitive development, illness and death of children which leads to retarded national socioeconomic development (FAO, 2001; WHO, 2002; Anigo *et al*., 2007). Poor nutrition during infancy also leads to poor academic achievement, low incomes in adulthood and inadequate care for the children of subsequent generations (Grantham-McGregor *et al*., 2007).

In Ethiopia, children in rural area are especially prone to nutrient deficiencies as they eat from the family dish, which is predominantly plant-based (Melaku *et al*., 2005). For this reason, infant malnutrition due to nutritionally inadequate diets is one of the major concerns in Ethiopia. Where the most serious nutritional deficiency in infants and young children are protein energy malnutrition (PEM); which contributes to more than 50% of childhood mortality in developing countries (Temesgen, 2013)

Nationally under five age Ethiopian children 40% stunted, 9% wasted 27% underweight. Rural children are more likely to be underweight (27 percent) than urban children (15 percent). The proportion of underweight children varies by region. Addis Ababa has the lowest proportion of underweight children, at 7 percent, while Affar has the highest prevalence of underweight children, at 44 percent (CSA, 2014).

The other problem in protein energy malnutrition is the presence of anti-nutritional factors, which binds minerals and reduces availability of nutrients on consumption (Ijarotimi and Keshinro, 2012). Many processing methods are investigated to reduce the level of anti-nutritional factors in the food material. Fermentation, germination, soaking (Rasha *et al*., 2011), dehulling (Oseni, 2011), heat treatment (Soetan and Oyewole, 2009; Oladele *et al*., 2009; Hernandez *et al*., 2011).

Cereal fermentation is one of the oldest biotechnological techniques of processing raw materials used in worldwide. Fermented foods and drinks have been included in the diet of various cultures for centuries (Marko *et al.,* 2014). Animal and plant tissues subjected to the action of microorganisms and/or enzymes to give desirable biochemical changes and significant modification of food quality are referred to as fermented food (Sahana & Fauzia, 2003). Natural fermentation is applied in traditional African food preparation is an effective method of improving the protein and complex carbohydrate digestibility of cooked cereals (Chavan *et al*., 2006). Fermented cereal porridges (and gels) are important staple food items for people of the West African sub region and are also important complementary foods for infants (Taiwo, 2009). Fermented foods having acidic pH are microbiologically safe and can be stored for a long time (Khetarpaul, 2007).

The incorporation of amylase-rich flour (ARF) has been reported as an effective means of producing liquefied foods from their solid base, and can be processed using malted cereals such as maize, rice, millet or sorghum to develop amylotic enzymes(Barragan-Delgado and Serna-Saldivar 2000). Malting is a traditional practice in developing countries primarily used in the production of alcohol and non-alcoholic beverages. During malting of grains, α-amylase and β-amylase activity is developed. These enzymes effectively degrade starch granules, reducing their water-binding capacity (Yusuf *et al*., 2008).

1.1. Statement of the Problem

One of the major nutritional problems faced by young children in the eastern and southern African region during the complementary feeding period is that CFs (which are commonly prepared in the form of porridges) made from the major staple foods (mainly cereals and roots and tubers) found in the region have a low nutrient density, mainly because they are high in unmodified starch. 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).

The high rates of malnutrition reported in cereal consuming areas of Africa reveal the bulkiness and high viscosity of the predominant cereal-based complementary food. As a result, mothers commonly dilute the porridge with water to reduce its viscosity (John *et a*l., 2008).Since young children have small gastric capacities, they are unable to consume enough of the diluted porridge to meet their energy requirements and consequently may become malnourished (Ramakrishna *et al.,* 2006). This problem of high viscosity, low energy density, or both in complementary food is often referred to as "dietary bulk" (Sucan, 1987). The availability of ready-made snacks and baby foods is limited in local markets and whatever items are available, they are expensive. Moreover, they are not available in remote areas (Ali *et al*., 2006).

The addition of a small quantity of amylase rich flour (ARF), to thick cereal porridges is known to improve the digestibility of storage proteins and starch, and to dramatically reduce the viscosity and bulk density of such porridges, thus allowing for the preparation of a more nutrient-dense food that is sufficiently liquid for a young child to consume. The bulky nature of the cereals based weaning food is the other discouraging factor for many infants from consuming it (Gernah *et al*., 2011). Therefore, there is need to develop nutritious food, nutrient dense, low dietary bulk low viscosity and affordable complementary food from locally produced cereals by simple technologies like fermentation process combined with ARF has been strongly recommended as sustainable approach to address the problem of infant malnutrition.

1.2. Research Questions

i. What is the effect of fermentation and ARF on physico-chemical and sensory properties of products from oat ,barley and teff?
ii. Is there any difference in nutritional content and functional properties change before and after fermentation process and addition of ARF?
iii. Is fermentation process can reduce the anti- nutritional factors present on selected starchy staples?
iv. Is there change in pH and titratable acidity of the cereals flour's before and after fermentation process?

1.3. Study Objectives

1.3.1. General objective

To investigate the effect of fermentation and Amylase rich flour on nutritional composition and sensory properties of complementary food from oat, barley and teff

1.3.2. Specific objectives

- ❖ To determine effect of fermentation and ARF on changes in pH and total titratable acidity of oat ,barley and teff based complementary food.
- ❖ To evaluate effect of fermentation and addition of ARF on nutritional composition and functional properties of complementary food from oat ,barley and teff .
- ❖ To analyze effect of fermentation and ARF process on some anti-nutritional factors (phytic acid and tannin) contents of oat ,barley and teff based complementary food.
- ❖ To study effect of fermentation and ARF on consumer acceptance of the gruel prepared from oat ,barley and teff.

1.4. Significance of the Study

The present level of nutritional insecurity facing developing countries, especially African countries including Ethiopia , requires that maximum research and development efforts should be made to exploit and promote the food uses of all available food crops that can easily be digestible and low viscosity food especially for under two years age children . The cereals crops are used as staple food in Ethiopia but despite its nutritional bioavaliabity of the potential starchy staple cereals crop still remains underutilized.

Development of complementary food from oat, barley and teff by fermentation process provides a means of enhancing and upgrading it's nutritional quality and starch digestibility. This will lead to increased utilization of the crop and for improved food and nutrition security as well as more income to families engaged in the production of the cereals crops in response to increase in demand. Enhancing the utilization of the starch staples crops through product development by low cost technology is essential in promoting nutritional security and meeting nutritional needs. The results obtained from this study enhance the understanding and knowledge about starchy staples crops value addition through low cost processing method by using fermentation and addition of Amylase Rich flour to get maximum nutrient and energy density and low viscosity product from oat, barley and teff crops.

2. LITERATURE REVIEW

2.1. Malnutrition Problem in the World

Malnutrition is globally the most important risk factor for illnesses and death, affecting especially hundreds of millions of pregnant women and young children. It is currently the leading cause of global burden of disease (Ezzati *et al.*, 2002). Malnutrition among infants in low-income countries is an important public health introduced after the breastfeeding period (Faruque *et al*., 2008). According to Onabanjo (2008) malnutrition, when it is severe, can cause premature death, permanent disability and fragility in face of many deadly diseases. Among different types of malnutrition, micronutrient malnutrition affects more than one-half of the world s population, especially women and preschool children (FAO, 2004).

Globally malnutrition is the most important risk factor for illnesses and death, affecting especially hundreds of millions of pregnant women and young children. However evidence has shown that child death and malnutrition are not similar problems throughout the world. They cluster in sub-Saharan Africa and south Asia, and in poor communities within these regions (de Onis and Blossner, 2003). Studies have also shown that malnutrition is the most common cause of immunodeficiency worldwide (Ezzati *et al.*, 2002).

According to USAID (2011) malnutrition has become one of the major world health problems facing developing countries. More than one third of children in the developing world are undernourished. Throughout the developing world, malnutrition affects almost 200 million children worldwide and contributes to more than 3.5 million child deaths each year. It affects physical growth, morbidity, mortality, cognitive development; reproduction, and physical work capacity (Mahgoub *et al*., 2006).The World Health Organization defines malnutrition as "the cellular imbalance between supply of nutrients and energy and the body's demand for them to ensure growth, maintenance, and specific functions. Severe malnutrition, typified by wasting, oedema or both, occurs almost exclusively in children (Brabin and Coulter, 2003).

There are two (2) elements of malnutrition: protein–energy malnutrition and micronutrient deficiencies. Apart from marasmus and kwashiorkor (the two forms of protein– energy malnutrition), deficiencies in iron, iodine, vitamin A and zinc are the main manifestations of malnutrition (Muller and Krawinkel, 2005). The degree and distribution of protein–energy malnutrition and micronutrient deficiencies in a given population depends on many factors: the political and economic situation, the level of education and sanitation, the season and climate conditions, food production (Brabin and Coulter, 2003), cultural and religious food customs, breast-feeding habits, prevalence of infectious diseases, the existence and effectiveness of nutrition programs and the availability and quality of health services (F.A.O., 2004).

2.1.1. Protein energy malnutrition

Protein-energy malnutrition (PEM) may be present at any time during the life cycle, but it is more common in the extreme ages that is, during infancy/childhood and in the elderly (Castaneda *et al.*, 1995). The present review will be restricted mostly to the condition present during infancy and childhood. Protein energy malnutrition in children (PEM) is a pathologic depletion of the body's lean tissues caused by starvation, or a combination of starvation and catabolic stress (Castaneda *et al.*, 1995). The underlying mechanisms include decreased food intake because of anorexia, decreased nutrient absorption, increased metabolic requirements and direct nutrient losses (Gonzalez-Barranco and Rios-Torres, 2004).

Patients that lose 10–20 percent of their body weight may have moderate PEM. Losing 20 percent of body weight or more is generally classified as severe PEM (Gonzalez-Barranco and Rios-Torres, 2004; Hamer *et al.*, 2004). Primary PEM results from a diet that lacks sufficient sources of protein and/or energy. Secondary PEM usually occurs as a complication of chronic diseases such as AIDS, cancer, chronic kidney failure, inflammatory bowel disease, and other illnesses that impair the body's ability to absorb or use nutrients or to compensate for nutrient losses (Hamer *et al.*, 2004).

Marasmus and kwashiorkor are the two (2) forms of the protein– energy malnutrition. Protein-energy malnutrition (PEM) is a problem in many developing countries, most commonly affecting children between the ages of 6 months and 5 years. The condition may result from lack of food or from infections that cause loss of appetite while increasing the body's nutrient requirements and losses. Children between 12 and 36 months old are especially at risk since they are the most vulnerable to infections such as gastroenteritis and measles (WHO, 2000).

2.1.1.1. Kwashiorkor

Kwashiorkor, also called wet protein-energy malnutrition, is a form of PEM characterized primarily by protein deficiency. This condition usually appears at the age of about 12 months when breastfeeding is discontinued, but it can develop at any time during a child's formative years (Manary *et al.*, 1998). Kwashiorkor usually manifests with fluid retention (oedema) usually starting in the legs and feet and spreading, in more advanced cases, to the hands and face. Oedema may be detected by the production of a definite pit as a result of moderate pressure for 3 seconds with the thumb over the lower end of the tibia and the dorsum of foot. Because of oedema, children with kwashiorkor may look "fat" so that their parents regard them as well fed (Manary *et al.*, 1998).

2.1.1.2. Marasmus

Early marasmus occurs usually in the first year of life in children who have been weaned from breast milk or who suffer from weakening conditions like chronic diarrhoea. It is frequently associated with contaminated bottle-feeding in urban areas (Pinstrup-Andersen *et al.*, 1993). Primarily marasmus is caused by energy deficiency from prolonged starvation. It may also result from chronic or recurring infections with marginal food intake (de Onis *et al.*, 1993). Marasmus is characterized by stunted growth and wasting of muscle and tissue (Hoffer *et al.,* 1999*)*. Wasting indicates recent weight loss, whereas stunting usually results from chronic weight loss. The major nutritional indicators studied are: stunting (low height-for-age); underweight (low weight-for-age); and wasting (low weight-for-height). Of the three (3), wasting is the most dangerous and signifies acute malnutrition (Muller and Krawinkel, 2005).

2.2. Malnutrition Problems in Africa and Ethiopian Children

Malnutrition is a major public health problem throughout the developing world, particularly in southern Asia and sub-Saharan Africa (Schofield and Ashworth, 1996); (WHO and UNICEF, 2004) Foods eaten in developing countries contain high levels of carbohydrate with very low or no proteins due to the high cost of protein rich foods and highly viscous and hence low in nutrient content and energy density (Olusola *et.al,* 2009). In most Sub-Saharan African (SSA) countries, millions of people depend on maize for their daily food and especially plays an important role in infant feeding. For many, it is the largest source of protein.

In Nigeria rural community children are traditionally complement on cereal- or tuber-based gruels that have been found to be highly viscous and hence low in nutrient content and energy density. For example, akamu (also known as pap) as consumed contains only 0.5% protein and 1% fat, as compared with the 9% protein and 4% fat contents of the original corn. Of the protein content, 98% of the original tryptophan in maize is lost during processing of akamu (Peter *et.al*, 2002).

In Ethiopia, particularly in rural communities, children are traditionally complement on cereal like maize, sorghum, barley and root crops like potato, sweet potato based gruels that have been found to be highly viscous and hence low in nutrient content and energy density(Zewditu *et al*., 2001). Ethiopia is one of the countries in the Sub-Saharan Africa with the highest rates of malnutrition in children. Malnutrition results from the interaction between poor diet and disease and leads to most of the anthropometric deficits observed among children in Ethiopia (Zewditu *et al*., 2001). Complementary foods are not introduced in a timely fashion for many children. At 6-8 months, only one in two children is receiving complementary foods. The use of a bottle with a nipple is not widespread in Ethiopia (CSA and Macro, 2006). More than half of Ethiopian children ages 6- 59 months are classified as anaemic, with 21 percent mildly anaemic, 28 percent moderately anaemic, and 4 percent severely anaemic (CSA and Macro, 2006).

The level of malnutrition is significant with nearly one in two (47 percent) Ethiopian children under five years of age stunted (short for their age), 11 percent wasted (thin for their height), and 38 percent underweight. 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).

2.3. Complimentary Foods

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. Infant foods in many parts of Ethiopia are cereal or root crop-based because these are the major components of the family's diet. Timely introduction of complimentary weaning 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. Complimentary weaning food should not be introduced in any infant before 17 weeks and all infants should start complimentary feeding by 26 weeks. The ability of breast milk to meet the requirements for macronutrients and micronutrients becomes limited with increasing age of infant. Furthermore as infants develop they show desire of foods other than milk (Agostoni *et al*., 2008).

A complementary food is normally a semi-solid food that is used as additional to the breast milk and not to replace it. Complementary foods are mostly prepared in the form of thin porridges. In most developing countries they are introduced directly to the regular household diet made of cereal or starchy root crops. Inadequate complementary food is a major cause for the high incidence of child malnutrition, morbidity and mortality in many developing countries and the weaning period is the most critical period in the child's life (Ijarotimi, 2008).

In most African countries, traditional complementary foods are based on local starchy staples usually fermented, ground cereals that are processed into porridges such as Ogi. Sorghum has been widely used to produce Ogi which is a popular complementary food in Nigeria (Olorunfemi *et al*., 2006).

Poor child feeding practices of low-income families is responsible for the high prevalence of Protein energy malnutrition among children in developing countries (Ijarotimi, 2008). In order to solve these nutritional problems, several efforts have been made to formulate weaning 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.

During the weaning process, children are particularly vulnerable to malnutrition. The weaning period is a very critical period in promoting child nutrition and survival. The period would have been less problematic if only mothers know how to feed their children with locally available food materials. Studies indicated that the vast majority of children in developing countries are not only improperly fed but are also underfed (Akeredolu *et al*., 2005). 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.

Supplementation of cereals with locally available legumes rich in protein and lysine would increases the protein content of cereal-legume blends and their protein quality through mutual complementation of their individual amino acids (Abebe *et al*., 2006). 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).

Furthermore, the bulkiness of most traditional Complementary foods and high concentrations of fiber and inhibitors are a major factor in reducing the nutritional benefits. Studies have shown that growth faltering in children coincides with the introduction of low nutrient density weaning foods, improper feeding practices and gastrointestinal infections (Mbithi-Mwikya *et al*., 2002). Mothers need education on the importance of complementary foods and about hygienic processing, preparation and storage of these foods (WHO, 2001).

2.4. Complementary Food Development in Ethiopia

Complementary food can be generally produced following traditional technologies such as malting, popping and fermentation and modern food processing technologies such as roller drying and extrusion cooking (Sachdev and Choudhury 2004). Mostly weaning prepared in the form of tin porridge or gruels are used for ready consumption (Lutter and Rivera , 2003) The choices of complementary foods are affected by factors like family dietary pattern, influence of elders, cultures, customs, beliefs of food taboos, previous experience of feeding patterns, agriculture, inadequate nutritional knowledge, geography and climates (Lutter and Rivera , 2003). Development of complementary food is guided by high nutritional value to supplement breast feeding, acceptability, low price and use of local food items.

The concept of improved feeding of infant and young children is not well understood by most families in Ethiopia. The point at which infants begin the actual weaning process i.e. the introduction of grain based solid foods is not the same throughout the country. It varies considerably with the ethnic make-up of the population the degree of urbanization and the socio economic status of the families (Suhasini and Malleshi , 2003). In general infants in the rural areas starts very late from 8 to 12 months of age whereas urban infants begin at about 5 months (Melaku *et al*,.2005).

When mothers introduce solid foods to their infants, they traditionally give gruel made from a variety of cereals. As the infants grow, porridge is given in addition to gruel and both foods are given together until about the end of the 2nd year. When the child is about 2 years of age “fetfet “ketta” and “dabo” are given; soon afterwards the child is introduced to an adult diet consisting mainly of thin leavened bread (*Injera*) with hot sauce *(wot*) made from legume (split or ground and spiced) (Melaku *et al,* .2005).

Generally, weaning foods in Ethiopia are made from cereals or starchy tubers such as maize, sorghum, millet, rice and yam, potato, barley and yam. Traditional complementary food gruels or porridge are based on starchy staple foods, such as wheat, rice, maize, barley, oat, teff, millet or sorghum but in some areas also forms starchy roots or tubers that produce viscous porridges that are difficult for children to consume (Ramakrishna *et al,*. 2006). As a result, mothers commonly dilute the porridge with water to reduce its viscosity. Such dilution, however, also reduces the energy density of the mixture. Since young children have small gastric capacities, they are unable to consume enough of the diluted porridge to meet their energy requirements and consequently may become malnourished. This problem of high viscosity, low energy density, or both in complementary food is often referred to as "dietary bulk" (Sucan, 1987).

Children consuming these foods grow poorly and have higher mortality rates. Increasing the nutrient density of complementary foods is a strategy commonly recommended for improving child nutrition It is possible to achieve an adequate nutrient intake by increasing the daily intake of such low nutrient-dense foods, but the volume of the food to be consumed may be too large to allow the child to ingest all the food necessary to cover nutrient needs. For instance, an infant aged 4–6 months would need about 62 g of corn gruel to meet daily need of energy of 740 (kcal), and protein need of 13 g (Sucan ,1987) . This is an impossible target considering the size of an infant's stomach.

2.5. Functional Properties of Complimentary Food

The functional properties of flours play an important role in manufacturing of products (Akubor *et al.,* 2013). Kinsella (1976) have been defined the functional properties are refer to those physical and chemical properties; that influence the behavior of proteins in food systems during processing, storage, cooking and consumption. The functional properties such as water and oil absorption capacities, particle size distribution, bulk density and gelatin of flour are important to determine; either the flour would be useful in development of food, those functional properties affect processing applications, formulations, food quality, storage thereby altering the texture, and sensory qualities of food product beyond satisfying the basic nutritional requirements (Mahajan and Dua, 2002; Wu *et al.*, 2009).

2.5.1. Bulk Density

Bulk density is a function of particle size, while particle size being inversely proportional to bulk density (Onimawo and Akubor, 2012). The differences in the particle size may be the cause of variations in bulk density of flours. Bulk density is an indication of the porosity of a product which influences package design and could be used in determining the type of packaging material required, material handling and application in wet processing in the food industry (Kinsella, 1987). According to Peleg and Bagley (1983), bulk density depends on the combined effects of interrelated factors such as the intensity of attractive inter-particle forces, particle size, and number of contact points. Bulk density is also important in infant feeding where less bulk is desirable. The low bulk density of flour would an advantage in the use of the flour for preparation of complementary foods (Akubor *et al.,* 2013)

2.5.2. Water Absorption Capacity

Water absorption of flour is dependent mainly on the amount and nature of the hydrophilic constituents and to some extent on pH and nature of the protein (Gordon, 1993). Water absorption characteristic represents ability of the product to associate with water under conditions when water is limiting such as dough's and pastes (Akubor *et al.,* 2013). Water absorption capacity of flour may depend on the higher polar amino acid residues of proteins that have an affinity for water molecules (Yusuf *et al*., 2008). The major chemical components that enhance the WAC of flours are proteins and carbohydrates. These constituents contain hydrophilic parts, such as polar or charged side chains (Lawal and Adebowale, 2004).

Water absorption is important as it may change flow regimes in batters or flour suspensions and it affects final product quality (Barrera, *et al*., 2013). Damaged starch granules have modified rheological and structural properties, and are able to absorb water. Glutenins and gliadins also have a significant role in water absorption capacity. Flour water absorption capacity may also be attributed to weax cross linking reactions (oxidative gelation) (Kweon, Slade, & Levine, 2011). It is correlated with increases in batter viscosity caused by increased concentrations of free radicals in the flour. Free radicals initiate oxidative gelation, increasing cross linking between ferulic acid residues. The cross-links form a polymeric network that is capable of sequestering water and increasing flour water absorption capacity (Bettge & Morris, 2007).

2.5.3. Viscosity

During infancy, viscosity of a complementary food is the most important determinant of energy density. The viscosity of gruels in turn depends in large part on the degree of starch gelatinization (Kabeir *et al*., 2004).An infant needs a gradual transition from fluid to solid foods which allow the infant to develop feeding skills. Early in weaning, the infant will reject very viscous foods by spitting them out. During weaning inadequate intake of energy and protein occurs due to traditionally prepared complementary foods, which are bulky in nature (have high viscosity but low energy density) and limit the infant's ability to eat enough (Agostoni *et al*., 2008) .

Infant can easily consume low viscosity complementary food as much a possible of the food and more solid can be added to the mixture; this will no doubt increase the nutrient density of the gruels which is highly beneficial to the infants. The enzyme, α- amylases, breaks down the starch molecules of the cereals grain to dextrin eventually to maltose and glucose. These sugars always have low viscosity. Thus, starch degradation reduces the viscosity of thick cereal porridges without dilution with water while simultaneously enhancing their energy and nutrient densities (Almeida-Dominguez *et al*.1993).

2.6. Nutritional importance of Oat, Barley and Teff for Complementary Food Development

2.6.1. Oats (*Avena sativa*)

Oats have numerous uses in food. Most commonly, they are rolled or crushed into oatmeal, or ground into fine oat flour. Oat widely used for the production of ready to eat breakfast cereals, bakery goods, snacked food, infant food, and other products. Oat posses a number of unique features it can be used as whole groat products that impart a characteristic nutty flavor, of a comparatively high nutritional value. However, oat lack gluten and it is thus not possible to produce high quality leavened bread from oat without the addition of wheat (Anderson B *et al*,. 1991). Oats are generally considered "healthy", or a health food, being considered commercially as nutritious. The discovery of the healthy cholesterol-lowering properties has led to wider appreciation of oats as human food. Oat bran is the outer casing of the oat. Its consumption is believed to lower LDL ("bad") cholesterol, and possibly to reduce the risk of heart disease (Sandberg and Andlid, 2002).

Oats have higher oil content and thus a higher energy density. Oat protein has relatively high amino acid balance and oats also contains significant amounts of dietary fiber which is high in soluble fiber. This soluble fiber, which is composed mainly of β-glucan, is also known as oat gum. Oats also contribute significant amounts of dietary minerals (Mg, P, Fe, Cu and Zn) and vitamins (thiamin, vitamin E, folate, niacin)(Svanberg ,1987).The calcium content is higher than all the food grains and best quality protein with the presence of essential amino acids, some pro-vitamin A, vitamin B and phosphorus (Frias *et al*,..2005). Oat is the only cereal containing a globulin or legume-like protein. Globulins are characterized by dilute salt solution and water solubility.

Oat protein is nearly equivalent in quality to soy protein, which has been shown by the World Health Organization to be equal to meat, milk, and egg protein. The protein content of the hull-less oat kernel (groat) ranges from 12–24%, the highest among cereals. Oats is a well adapted fodder crop grown for a long period of time in the highlands of Ethiopia (Sandberg and Andlid , 2002).

It is produced by some peri-urban dairy cattle producers and by smallholder farmers who own crossbred dairy cows (Svanberg, 1987). Oats being an annual forage crop is highly useful for integration into the prevailing mixed crop- livestock farming systems of the highlands on accounts of its short-term yielding characteristics, use in overcoming seasonal feed shortages, convenience in crop rotations and its fodder conservation characteristics. Moreover, farmers can easily grow it because its husbandry is similar to that of other cereals such as barley and wheat. In view of its good nutritional profile, low allergy bearing, flavor compatibility, and cost, oats are a useful component of infant foods oat flour is often the ingredient of choice, and specifications include low levels of free fatty acids and absence of hull when flour is used for infant nutrition (Svanberg, 1987).This shows that blending oat in complementary food formulation is good sources of essential nutrients.

2.6.2. Barley (*Hordeum vulgare*)

Barley is one of the most important staple food crops in the high lands of Ethiopia. Currently, barley is widely consumed as a food grain with desirable nutritional contents consumed as a foods and snacks are increasingly available, driven by recent research findings, which show that barley fibers contains beta–glucans and tocotrineols, chemical agents known to lower serum cholesterol level (Lee *et al*,. 2007). In Ethiopia barley is the fifth most important cereal crop next to maize, tef, sorghum and wheat. It is the staple food grain for Ethiopian high lands who mange the crop with indigenous technologies and utilized different parts of the plants for various purposes. Barley is used as flour, as semolina, and as whole-dehulled grain. A large variety of dishes, including soups, bread, and couscous are made from barley products. Preparations include both product from fully mature grains and grains harvested at physiological maturity (*Azenbou*) some recipes, such as *Besso* (fine flour of well-roasted barley grain moistened with water, butter or oil), Zurbegonie (same type of flour used for *Besso* suspension in cold water with sugar) (Lee *et al.*, 2007).

Chiko (besso molded with butter and spice), which have long shelf life, can only be prepared from barley grain. Other recipes, such as *Genfo* (thick porridge), *Kolo* (de-hulled and roasted barley grain served as snack), and *Kinche* (thick grits) are most popular when made from barley grain (Temesgen, 2013)

Barley is the one preferred grain, after tef, for making the traditional bread called *Injera*, which can be used either solely or in combination with tef flour or other cereal flours. Other recipes, such as Dabbo (bread), Kitta (thin, unleavened, dry bread) and *Atmit* (gruel) can be prepared with barley or blended with other cereal flours. Among local beverages Tella ,shameta and korafe are prominent, and best made from barley grain. Barley spikes both unripe at milk or dough stage and ripe and dry are also roasted over flame and the grain is consumed as snack called *Eshete or Wotelo* if the spikes are unripe, or *Enkuto* if the roasted barley spikes are dry (Anderson B *et al*,.. 1991). Barley is also traditionally used in the preparation of gruel utilized as complementary food. Barley is mainly used as a source of carbohydrates, although the protein content is also important (Anderson B et *al*,.. 1991). Barley protein is composed of 19 amino acids, but low in lysine and methionine. This might be the reason why most traditional barley recipes are prepared or consumed along with legumes or animal products to supplement the deficient amino acids.

2.6.3. Teff (*Eragrostisis teff (*Zucc) Trotter*)*

Teff is one of the major and indigenous cereal crops in Ethiopia, where it is believed to have originated and has the largest share of area under cereal crop production. According to Central Statistics Authority teff accounts about 34,340,420.63 quintal of production in an area of 2,722,739.42 ha with grain yield 12.61 qt/ha (Firmin *et al*.., 1986). It provides over two thirds of the human nutrition in the country. This cereal is considered high in nutritional quality, but limited information. there are available about its usefulness in complementary food blends. Teff flour is primarily used to make a fermented, sour dough type, flat bread called *Injera*. The crop is also an excellent source of fiber and iron, and has high amount of calcium, potassium and other essential minerals found in an equal amount than in other cereal grains. While the reported high iron content of teff seed has been refuted by some works, the lack of anemia in Ethiopia is considered due to the available iron from *Injera* (Gahlawat and Sehgal , 1992). Teff flour blending in all complementary food will enrich the products with iron and other essential minerals.

2.7. Effect of Fermentation and Malting on Nutritional Quality of Complementary Food.

2.7.1. Fermentation

All over the world, fermented foods provide an important part of human diet. Fermented foods and beverages provide about 20-40% of human food supply. Traditional food fermentation is capable of improving nutrients of the food, preserve it by generating acidic condition, detoxify and reduce cooking time of the food (Fagbemi *et al*., 2005).

The majority of traditional cereal-based foods consumed in Africa are processed by natural fermentation. Fermented cereals are particularly important as complementary foods for infants and as dietary staples for adults (FAO, 1999). Combining fermentation with cooking, either fermenting then cooking or cooking then fermenting improve the nutrient quality and drastically reduce the anti-nutritional factors to safe levels much greater than any of the other processing methods tested (Chavan *et al*., 2006). Lactic acid fermentation of cereals generally improves extractability of minerals, probably because of the decreased content of phytic acid in the fermented cereal product (Eltayeb, 2007; Khetarpaul and Chauhan, 2007).

Over the centuries, fermentation has evolved and been refined and diversified. Today, a variety of food products are derived from this technology in households, small-scale food industries as well as in large enterprises. Furthermore, fermentation is an affordable food preservation technology and of economic importance to developing countries (Motarjemi, 2002). It enhances the nutritional quality of foods and contributes to food safety particularly under conditions where refrigeration or other foods processing facilities are not available (Motarjemi, 2002).

The increase in insoluble protein digestibility during fermentation suggests that fermentation causes structural changes in the cereal storage proteins (prolamins and glutelins), making them more accessible to pepsin attack. Thus natural fermentation, as applied in traditional African food preparation is an effective method of improving the protein digestibility of cooked cereals (Goyal and Khetarpaul, 1994; Taylor and John, 2002; Chavan *et al*., 2006).

Fermented cereal porridges (and gels) are important staple food items for people of the West African sub-region and are also important complementary foods for infants (Taiwo, 2009). Fermented foods having acidic pH are microbiologically safe and can be stored for a long time (Khetarpaul and Chauhan, 1989).

In Africa, the majorities of cereal-based foods are consumed in the form of porridges and naturally fermented products (Kabeir *et al*., 2004). Many desirable changes occur during the fermentation process of cereal grains due to the breakdown of complex compounds into simple forms and the transformation into essential constituents (Kabeir *et al*., 2004).

The microbial groups involved in spontaneous fermentation of cereal flour include *Lactobacillus spp*, *Acetobacter spp*. and *Saccharomyces cervisiae* (Kabeir *et al*., 2004; Tsaousi *et al*., 2008). Recently, instead of the traditional lactic acid bacteria, the use of probiotics bacteria such as *Bifidobacteria* to improve the therapeutic quality of food has gained considerable interest. *Bifidobacteria* are beneficial for human beings of all ages, as they are the predominant member of the endogenous intestinal flora, capable of improving the balance of the intestinal microflora by preventing colonization of pathogens, activating the immune system and increasing protein digestion (Kabeir *et al*., 2004).

Table 1: Effect of fermentation on nutritional quality of food and potential health benefits

Effect on food	Potential health benefit
Break down of starch by amylases	Reduces bulk and increases energy density
Reduction of phytic acid	Improved absorption of minerals and protein
Decrease in pH	Improved food safety
Reduction in lactose content (only milk products)	Better tolerance in individuals with lactase deficiency
Increase in lactic acid bacteria	Better food safety potential probiotic effects
Synthesis of B vitamins	Better vitamin B status

Source: Stanbury *et al*., (2003)

Fermentation causes changes in food quality including texture, flavor, appearance, nutrition and safety. The benefit of fermentations may include improvement in palatability and acceptability by developing improved flavors and textures (Sahana and Fauzia, 2003). The changes occurring during the fermentation process are mainly due to enzymatic activity exerted by the microorganisms and/or the indigenous enzymes in the grain (Ulf and Wilbald, 1998).

Fermentation also leads to a general improvement in the shelf life, taste and aroma of the final product. During cereal fermentations several volatile compounds are formed, which contribute to a complex blend of flavors in the products. The presence of aroma representatives; diacetyl, acetic acid and butyric acid make fermented cereal based products more appetizing (Katongole, 2008).

2.7.2. Malting

Malting is the germination of cereal grains in moist air under controlled conditions. The primary objective being to promote the development of hydrolytic enzymes, which are not present (or are present in limited amounts) in un-germinated grains. The main enzymes produced during germination that intervene in the hydrolysis of starch are α- and ß-amylases (Badau *et al.,* 2005). The α-amylases are liquefying enzymes that convert starch into soluble sugars (Traore *et al.*, 2004), while β amylases are saccarfying enzymes that release soluble sugars.

Alpha amylase activity has been observed to increase during germination of cereals, especially sorghum and millet. This enzyme hydrolyses amylase and amylopectin to dextrins and maltose, thus reducing the viscosity of thick cereal porridges without dilution with water while simultaneously enhancing their energy and nutrient densities (Gibson and Ferguson, 1998). Malting of cereals is a processing procedure traditionally used in many African countries for the manufacture of alcoholic drinks (Dewar *et al.*, 1997; Taylor and Dewar, 2001) like opaque beers; complementary foods, and other traditional dishes (Serna-Saldivar and Rooney 1995). The malting process can be divided into three physically distinct operations, i.e. steeping, germination (sprouting) and drying (Dewar *et al.*, 1997).

Malting causes up to 30 % dry matter loss (Chavan and Kadam, 1989); decreased levels of prolamine, fat, tannins and starch; and increased levels of free amino acids, albumins, lysine, reducing sugars, and most vitamins including synthesis of vitamin B-12 and C (Almeida-Dominguez *et al.*, 1993). The activation of intrinsic amylases, proteases, phytases, and fibre degrading enzymes disrupts protein bodies. Pelembe (2001) observed that malting (germination) significantly reduced the mousy odour, characteristic of ground pearl millet meals when stored.

Malting induces the synthesis of hydrolytic enzymes such as starch degrading enzymes (α-Amylases) and proteases. Malting has been claimed to improve the nutritive quality of Cereals. Due to enzymatic breakdown of starch to sugars (dextrin and maltose), reducing their water-binding capacity and consequently lowering the viscosity and bulk density of porridge and gruel made from cereals. Germination also improves protein quality and digestibility and the content of riboflavin, niacin, and vitamin C is increased and the content of anti-nutrients is reduced (Mbithi-Mwikya *et al*., 2000 and Hotz and Gibson, 2007). The terms 'sprouting', malting', and 'germination' are used interchangeably to refer to the process of soaking grains in water until saturated and germinating them under controlled conditions. The term malting is more commonly used when grains, especially barley, are soaked and germinated for brewing purposes; however, sprouting has been reported to improve the nutritional quality of seeds by increasing the contents and availability of essential nutrients and lowering the levels of anti-nutrients (Mbithi-Mwikya *et al*., 2000).

Different traditional processes used in cereal malting were characterized and some biochemical modifications occurring in seeds were studied to examine the possibility of using malted cereal flours to reduce the viscosity of gruels (Traore *et al*. 2004). During the malting of a variety of cereal grains, a significant increase in sucrose, glucose and fructose content. Flours from malted cereals are useful characteristics (α-amylase activity and nutrient contents) for incorporation into infant complementary flours to improve the energy and nutrient density of gruels (Badau *et al.*,2005).

2.8 Sensory Properties of Complementary Foods

Sensory evaluation is a scientific method used to evoke measure, analyze and interpret responses to products as perceived through the senses of sight, smell, touch, taste, and hearing. It is an irreplaceable tool in food industry while interacting with the key sectors in food production (Meilgaard., *et al* 1991).When consumers buy a food product, they can buy nutrition, convenience and image. In sensory evaluation, judges are asked to score the products for appearance, color, flavor, taste and overall acceptability using a scorecard of Hedonic Rating Scale. This test relies on people's ability to communicate their feelings of like or dislike. Hedonic testing is popular because it may be used with untrained people as well as with experienced panel members. A minimum amount of verbal ability is necessary for reliable results (sadana and chabra, 2004).

Sensory quality should be considered as key factors in food acceptance since consumers seek food with certain sensory characteristics. The acceptance of a food will depend on whether it responds to consumer needs and on the degree of satisfaction that it is able to provide (Heldman, 2004). The process by which man accepts or rejects food is of a multi-dimensional nature. Its structure is both dynamic and variable, not only among different individuals within a group but also within the same individual in different contexts and periods of time. Acceptance of a food is basically the result of the interaction between food and man at a certain moment (Shepherd, 1989).

Food characteristics (chemical and nutritional composition, physical structure and properties), consumer characteristics (genetic, age group, gender, physiological and psychological state) and those of the consumer's environment (family and cultural habits, religion, education, fashion, price or convenience) the influence of consumers´ decision to accept or reject a food (Shepherd, 1989; Shepherd and Sparks, 1994). Apart from the characteristics of the food itself and the sensations consumers experience when ingesting it, a consumer's purchase choice and even the degree of pleasure when consuming it can be influenced by their attitude and opinion about the nutritional characteristics (Bruhn *et al*., 1992), safety (Resurreccion and Galvez, 1999; Hashim *et al.*, 1996, Wilcock *et al*. 2004). For today's consumers, the primary consideration for selecting and eating a food commodity is the product's palatability or eating quality, and other quality parameters, such as nutrition and wholesomeness are secondary(Meiselman & MacFie, 1996; Lawless & Heymann, 1998). In order for players in the food and beverage industry, to have a market edge/success, they should ensure that the quality of food is appealing and appetising or more specifically that the eating quality attributes of; aroma, taste, aftertaste, tactual properties and appearance is acceptable to the consumer so that they crave for more. Thus if we accept that food quality is that "which the consumer likes best" and that the grades of quality are understood more by the degree of desirable attributes and absence of undesirable characteristics which are primarily detected by the consumer's sensory organs, then a good method of deciding quality of a food is through sensory evaluation (Bruhn *et al*., 1992),.

Sensory analysis can be considered to be an interdisciplinary science that uses human panelists sensory perception related to thresholds of determination of attributes, the variance in individual sensory response experimental design to measure the sensory characteristics and the acceptability of food products, as well as many other materials. Since there is no one instrument that can replicate or replace the human psychological and emotional response, the sensory evaluation component of any food study is essential and the importance of good experimental design cannot be over emphasized in sensory experiments (Lawless & Klein, 1989; Meiselman, Mastroianni, Buller, & Edwards, 1999).Sensory analysis is applicable to a variety of areas such as; inspection of raw materials, product development, product improvement, cost reduction, quality control, selection of packaging material, shelf life/storage studies, establishing analytical/instrument/sensory relationship and process development (Wilcock *et al*. 2004).

3. MATERIALS AND METHODS

3.1. Description of the Study Area

The study was conducted in Jimma zone, Oromia regional state in southwestern Ethiopia. Jimma town is located at 346 km South-West of Addis Ababa. The laboratory Analysis was conducted at Jimma University College of Agriculture and Veterinary Medicine (JUCAVM), Postharvest laboratory and animal nutrition laboratory, in 2015. JUCAVM is located at south western part of Ethiopia at 346km from Addis Ababa and is 7º.33''N latitude and 36.57ºE latitudes with elevations of 1710 m.a.s.l.

3.2. Raw Material Collection and Sample Preparation

3.2.1. Raw material collection

The experimental materials used for laboratory analysis were oats(15kg), barley (15kg), Teff (15kg) and malted barley (10kg).Oat Sinana (01) variety was collected from Sinana Agricultural Research center, barley BH-1307 variety was collected from Holota Agricultural Research center , teff Kuncho variety was collected from Debrezeit Agricultural Research center and malted barley holker variety was collected from Asella malt factory.

Figure 1: Raw materials used in the experiment.

3.2.2. Sample preparation

3.2.2.1. Preparation of oats flour

Preparation of oats flour was done according to the method developed by Shewarega (2012) with some modification. The oats grains were hand sorted and cleaned to get similar grains and to discard the defective grains. Cleaning was done by winnowing and hand sorting, and then cleaned grains were debranned using grain mill (HANGYU HP 20, Germany). The bran and grits were separated by winnowing. Finally, the grits were milled by using grain mill (KARLKOLB D-6072, Dreich, West Germany) to pass through 0.5mm sieve. The milled sample was packed in airtight polyethylene bags and stored at room temperature, until next processing step.

3.2.2.2. Preparation of barley flour

Preparation of barley flour was done according to the method developed Abraha (2013) with some modification. Barley grain was cleaned, and sorted, by winnowing and hand sorting. Cleaned grains were lightly roasted at 140°C for 5 min on stainless steel iron pan using electric heating to facilitate debranning process. After debranning, the bran and grits were separated by winnowing. Finally, the grits were milled by using grain mill (KARLKOLB D-6072, Dreich, West Germany) to pass through 0.5mm sieve. The milled sample was packed in airtight polyethylene bags and stored at room temperature, until next processing step.

3.2.2.3. Preparation of Teff flour

Preparation of teff flour was done according to the method developed by (Mariam Idreis,2004).The Teff grains were sieved to remove stones and extraneous matter by laboratory test sieve (Endecotts Ltd., London England) then milled by using grain mill (KARLKOLB D-6072, Dreich, West Germany) to pass through 0.5mm sieve. The milled sample was packed in airtight polyethylene bags and stored at room temperature, until used.

3.2.2.4. Preparation of Amylase rich flour (ARF)

Preparation of ARF flour was done according to the method developed by Afoakwa *et al.*, (2010) with some modification. Dry malted barley grains were cleaned by winnowing and hand sorting. Then milled by using grain mill (KARLKOLB D-6072, Dreich, West Germany) to pass through 0.5mm sieve. The milled sample was packed in air tight polyethylene bags and stored at room temperature, until used.

3.2.2.5. Fermentation process

Fermentation process was done by the method developed by Mihiret (2009) and Afoakwa *et al*., (2010) with some modification. Different levels of ARF concentration were added to each of the three selected starchy staples flours namely (oats, barley and teff) separately according to the levels indicated in the experimental design. Then mixed uniformly by homogenizer and added in distilled water in plastic buckets at a concentration of 1:3 dilutions (w/v). The flour slurry was allowed to ferment naturally with only the microorganisms borne on or inside the seeds (endogenous micro flora on the seeds) at room temperature for 24 and 48h in plastic containers. The fermentation water was decanted and the fermented samples were withdrawn and transferred to aluminum dishes after each fermentation time and dried in a hot air oven-drier (LEICESTER LE67 5FT, England) at 70 °C for 36h. Dried samples were ground with a miller (KARLKOLB D-6072, Dreich, West Germany) to pass a 0.5 mm sieve and packed in airtight polyethylene plastic bags and stored at room temperature until it was needed for chemical analysis. All samples were analyzed for pH, titratable acidity, moisture content, ash, and crude fiber, fat, crude protein, tannins, phytate, viscosity, bulk density, and water absorption capacity.

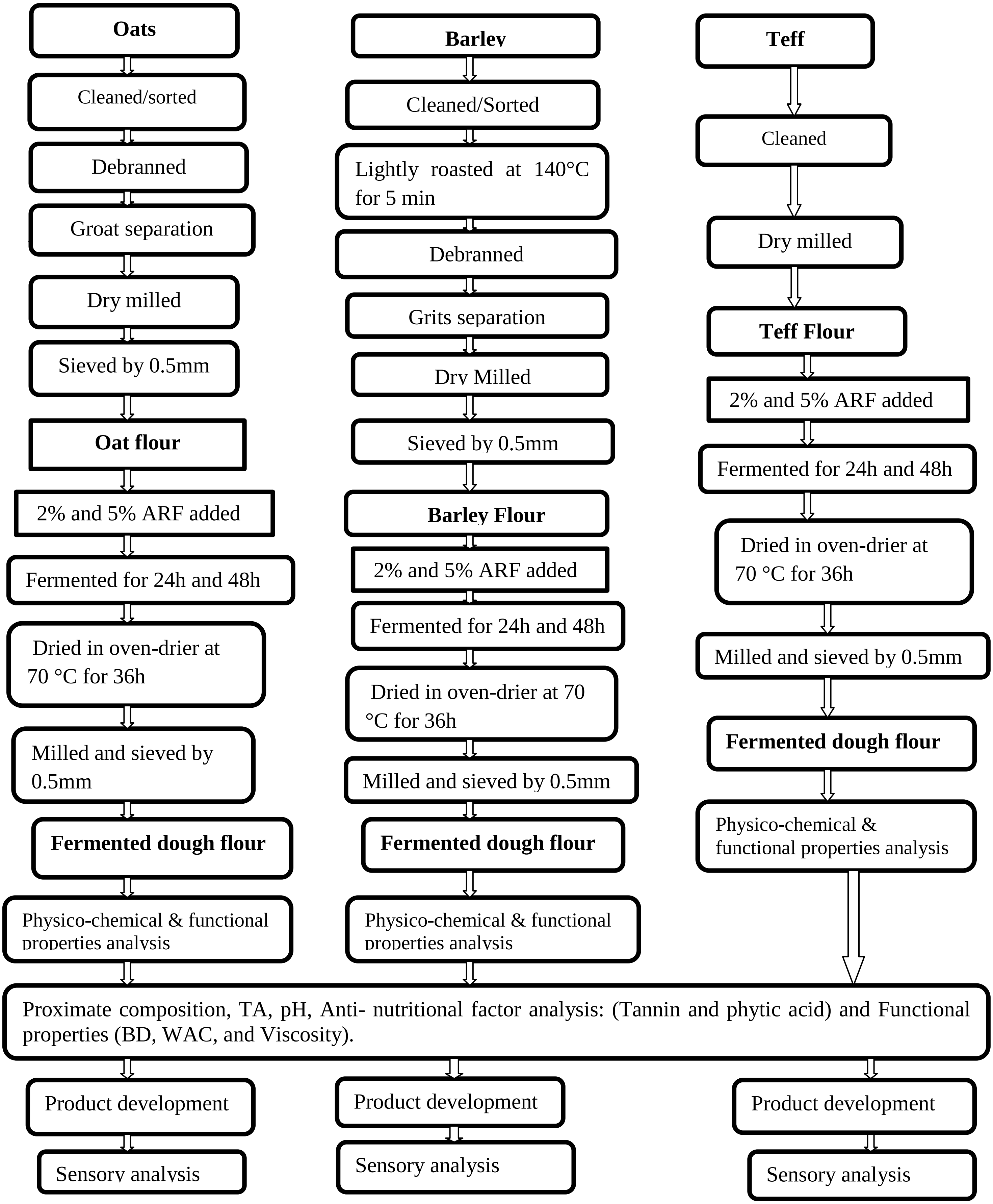

Figure 2: Flow diagram of experimental framework and preparation of flour sample

3.3. Experimental Factor and Design

The experiment had three factors namely Type of cereals, fermentation time and Amylase rich flour (ARF). Factor A=type of cereals (three levels), factor B=fermentation time (Three levels) factor C=amylase rich flour (three levels).

The treatments were arranged in 3x3x3 factorial combinations run in randomized complete blocks and replicated three times. The treatments consists of three type of cereals(oats, barley and teff),three fermentation time(0,24 and,48h)and three levels of ARF(0,2 and ,5%).

3.4. Laboratory Analysis

3.4.1. pH determination

The pH of the samples was determined according to the method of Pearson (1971). For the non fermented flour 10 g of the sample was added to 30 ml of distilled water and stirred for10 min.The pH meter was calibrated using pH 4.0 and 7.0 buffer. After calibration pH values of the samples were measured using a pH meter (SET/HO11, Mauritius).The pH of the fermented samples was determined by dipping the electrode of the pH meter in the homogenate fermented mixture slurries after the end of each fermentation period. Triplicate determinations were made in all cases.

3.4.2. Determination of titratable acidity

Total titratable acidity, expressed as percentage of lactic acid, was determined by titrating 30 ml of the homogenate samples used for pH determination against 0.1 N NaOH. First the distilled water (1L) used for titration was titrated with 0.1 N NaOH and the volume of 0.1 N NaOH consumed by water titration was considered as a blank. The volume of 0.1 N NaOH used for titration of the sample was noted after correcting the blank and triplicate determination was made (Pearson, 1971)

$$\%\text{Lactic Acid} = V \times 0.009008 \times 100 / W \quad \text{......} \quad \text{Eq.1}$$

Where: V=Volume of 0.1 N NaOH used for sample titration; 0.009008=Factor equivalent in which 1ml of 0.1N NaOH =0.009008g $C_3H_6O_5$; W=Weight in gram of sample in the mixture.

3.4.3. Determination of proximate composition

3.4.3.1. Determination of moisture content (%)

Moisture content of fermented and unfermented samples was determined by dry air oven method according to (AOAC, 2011) method 925.10. The petri dish was dried at 130 ± 3^{o}C for 1 hr and placed in desiccator and weighed after cooling for 30 minute. Two g of flour samples were weighed by analytical balance and put on dry dish. Dry dish and its contents put in the oven uncovered (LEICESTER LE67 5FT, England) maintained at 102^{o}C ± 3^{o}C for 1 hr. The sample containing dish covered was transferred to a desiccator and weighed soon after reaching room temperature and reweighed until the constant mass obtained then, the moisture content was estimated by using formulas.

$$\text{Moisture (\%)} = \frac{(M_{intial} - M_{dried})}{M_{intial}} x100\% \text{...Eq.2}$$

3.4.3.2. Determination of total ash (%)

A total ash content of unfermented and fermented flour sample was determined by (AOAC, 2005) method 923.03. The crucibles were cleaned and dried at 120^{O}C and ignited at 550^{o}C in furnace (Model SX-5-12, China) for 30 min. The crucibles were removed from the furnace and were placed in desiccators to cool down to room temperature. The mass of crucibles were measured by analytical balance and the reading was recorded as (M1). Three g of the flour samples were weighed in to crucibles (M2). And the sample was dried at 120^{O}C for 1 hr in drying oven (LEICESTER LE67 5FT, England). The sample was placed in furnace at 550^{O}C until free from carbon and the residues appear grayish white (for about 8 hrs). The sample was removed from the furnace and placed in the desiccator followed by weighing the mass and recorded as (M3):

$$\text{Total ash(\%)} = \left(\frac{M3 - M1}{M2 - M1}\right) x100 \text{...Eq.3}$$

Where; M2 = Weight of crucible + sample before ashing, M1 = Weight of empty crucible

M3 = Weight of crucible + sample after ashing.

3.4.3.3. Determination of crude protein (%)

The crude protein content of unfermented and fermented flour was analyzed by Kjeldahl (AOAC, 2005) method 988.05 of nitrogen analysis. About 0.3gm of sample was measured by analytical balance (model Model:ABJ220-4M, WB1151070, Australia) , 1gm of catalyst mixture of K_2SO_4 and $CuSO_4$ and 5ml of sulfuric acid added to each digestion flask (Kjeldahl flask KF250,German) which contain the mixture of sample and catalysts. The solution (0.3 gm of sample + 1gm of K_2SO_4 and copper sulfate + 5 ml of H_2SO_4) immediately placed in digestion flask at about 420^{o}C for 3-4 hrs, until the solution becomes clear. The digested sample was then transferred into the distillation apparatus and 25 ml of 40% (w/v). NaOH was continually added to the digested sample until the solution turned cloudy which indicated that the solution had become alkaline. The mixtures were then steam distilled and the liberated ammonia was collected into a 200 ml conical flask containing 25 ml of 4% boric acid plus mixed methyl red indicator solution. Next distillation was carried out into the boric acid solution in the receiver flask with the delivery tube below the acid level. As the distillation was going on, the pink color solution of the receiver flask turned green indicating the presence of ammonia. Distillation was continued until the content of the flask was reaching the required amount. The green color solution was then titrated against 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. Ultimately the percentage of nitrogen content was estimated using the following formula:

$$\text{Total nitrogen} = \frac{(T - B) x\ N\ x 14.007\ x\ 100}{w} \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \text{Eq. } 4$$

Where:

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

W - Weight in grams of the test material

Crude protein = 5.67 * total nitrogen

3.4.3.4. Determination of crude fat (%)

Crude fat contents of unfermented and fermented flour were determined by Soxhlet extraction according to (AOAC, 2005) method, 2003.06. About 1.5 g of sample was weighed and put into a thimble. The thimble and its contents was placed in to a 50 mL beaker and dried in an oven (Memmert 854, West Germen) for 2 hrs at 102±2^{o}C. The thimble contents were transferred in to extraction unit. The sample contained in the thimble was extracted with the solvent diethyl ether in a Soxhlet extraction apparatus (FossTecator, Sweden) for 6 hrs. After the extraction completed, the extraction thimble was dried in the oven (Memmert 854 schwabach, West Germen) for 30 min at 102^{o}C ± 2^{o}C to remove moisture. Then it was removed from the oven and cooled in a desiccator. The cup and its contents were weighed. The crude fat was determined by the following formula:-

$$\text{Crude fat}(\%) = \frac{W2 - W1}{\text{Weight of sample}} \times 100\% \quad \text{......Eq.5}$$

Where:

W_1= Weight of extraction flask before extraction

W_2 = Weight of extraction flask after extraction.

3.4.3.5. Determination of crude fiber (%)

The crude fiber contents of fermented and unfermented flour samples were determined by the non-enzymatic gravimetric (AOAC, 2005), method 922.16. About 1.5 g of food sample was placed into 600mL beaker. A 200 mL of 1.25% H_2SO4 was added to each beaker and allowed to boil for 30 min by rotating and stirring. 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.

During filtration the sample was washed with hot distilled water. The final residue was washed with 1% H_2SO_4 solution, hot distilled water, 1% NaOH solution, hot distilled water and finally with acetone. Each of the crucibles with their contents was dried for 2 hr at 130°C in the oven (Memmert 854 schwabach, West Germany) and 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: -

$$\text{Crude fiber \%} = \frac{M1 - M2}{\text{weight of sample}} \times 100 \quad \text{.....Eq.6}$$

Where, M_1 = mass of crucible and residue before ignition

M_2 = mass of crucible and residue after ignition

3.4.3.6. Determination of total carbohydrates (%)

Total carbohydrate content of the samples was determined by difference method using the following mathematical expression:-

$$\text{CHO(\%)} = 100 - (\text{\%moisture} + \text{\% fat} + \text{\%Ash} + \text{\%Crude fiber} + \text{\%Crude protein}) \quad \text{.....Eq.7}$$

3.4.3.7. Determination of calorific value/energy value calculation (kcal)

Calorific value of food (kcal) was determined by multiplying each gram of protein, fat and carbohydrate obtained from laboratory analysis by their respective conversion factor.

$$\text{Caloric value} = (\text{protein} \times 4) + (\text{carbohydrate} \times 4) + (\text{fat} \times 9) \quad \text{.....Eq.8}$$

3.4.4. Determination of anti-nutritional factors

3.4.4.1. Phytate determination

Phytate determination was done as per the method described by Vaintraub and Lapteva (1988). About 0.05 g of flour sample was weighed and extracted with 10 mL of 0.2N HCl for 1 hr at an ambient temperature then centrifuged at 3000 rpm/30 min The clear supernatant was collected and 3 mL of sample solution was mixed with 2 mL of Wade reagent (0.03 % solution of $FeC_{l3}.6H_2O$ containing 0.3% sulfosalicylic acid in water) followed by homogenization and centrifugation of the solution (3000 rpm for 10 min). The absorbance was read at 500 nm measured using UV-Vis spectrophotometer (CE1021, England). The phytate concentration was calculated from the difference between the absorbance of the blank (3 mL of 0.2N HCl +2 mL of wade reagent). The amount of phytic acid was calculated using phytic acid standard curve.

Calculation:

$$\text{phytic acid in } \frac{\mu g}{g} = \frac{\text{Absorvance} - \text{Intercept}}{\text{Slope} \times \text{Density} \times \text{weight of sample}} \qquad \text{Eq.9}$$

3.4.4.2. Condensed tannin determination

Condensed tannin content of flour sample was determined by Maxson and Rooney (1972) methods. About 0.06 g of weighed sample was extracted with 10 mL of 1% HCl in methanol for 24 hrs at room temperature with mechanical shaking (Edmund Buhler, USA) then centrifuged at 3000 rpm for 5 min. One milliliter of the supernatant was mixed with 5 mL of vanillin HCl reagent which was prepared by combining equal volume of 8% concentrated HCl in methanol and 4% vanillin in methanol while waiting for 20 min until the reaction is completed. Finally, the absorbance was read at 500 nm after 20 min using UV-Vis Spectrophotometer (CE1021, England). A stock catechin solution was used as the standard (20 mg catechin dissolved in 100 mL 1% HCl in methanol) and value of tannin was expressed in mg of catechin in gram of sample. Calibration curve was constructed by using a series of 0, 0.2, 0.4, 0.6, 0.8, and 1 mL of stock solution were taken in test tubes and the volume of each test tube was adjusted to 1 mL with 1% HCl in methanol. 5 mL of Vanillin-HCl reagent was added into each test tube.

$$\text{Tannin} \frac{mg}{g} = \frac{\text{Absorvance} - \text{Intercept}}{\text{Slope} \times \text{Density} \times \text{weight of sample}} \qquad \text{Eq.10}$$

3.4.5. Determination of functional properties

3.4.5.1. Bulk densities of the flour

Bulk density of a flour sample was determined by the method of (Adeleke and Odedeji, 2010). About 50g of flour sample was placed into a 100mL measuring cylinder. The cylinder was tapped several times on a laboratory bench to a content volume. The volume of sample is recorded bulk density was calculated from the values obtained as follows:

$$\text{Bulk density } \left({}^{g}/_{cm^3}\right) = \frac{\text{weight of sample}}{\text{volume of sample after tapping}} \dots\dots\dots\dots\dots\dots\dots\dots\dots\dots\dots\dots\dots.. \text{Eq. 11}$$

3.4.5.2. Water absorption capacity

WAC of flour sample was determined by Solsulski (1962) methods. About One gram sample was mixed with 10 mL of distilled water in a weighed 15ml centrifuge tube. The tube was agitated for about 5 min in before being centrifuged at 4000 rpm for 20 min (Centrifuge model 800-1). The mixture was decanted and the clear supernatant discarded. Adhering drops of water was carefully drain off as much as quantitatively possible and the tube was reweighed, then WAC was determined by the following formula:

$$\%WAC = \frac{\text{Weight of water bound} \times 100\%}{\text{Weight of sample (dry basis)}} \dots\dots\dots\dots\dots\dots\dots\dots\dots\dots\dots\dots\dots\dots\dots\dots\dots\dots\text{Eq.12}$$

3.4.5.3. Viscosity determination

Viscosity of gruel was determined according to the method by developed (Shemilis ,2009) using a HAAKE Falling ball viscometer (D-76227 kARLSRUAE, Germany).The gruels were prepared in a glass beaker by mixing 25 g of flour and 300 ml of distilled water. The mixture of water and flour was cooked at 92^{0C} for 25 minutes. The gruel was placed in a Water bath maintained at 40°C (heating temperature). 40ml cooked gruel was poured into the measuring tube of viscometer.

The nickel iron alloy ball with 15.2 mm in diameter was placed in the measuring tube, then the falling time(se) of a ball when the ball was dropped from top to bottom of measuring tube was recorded by using a stop watch. Then the viscosity of gruel was determined by following formula:

$$\eta = K (\rho 1 - \rho 2).\ t \ldots\ldots \text{Eq.13}$$

Where:

η = viscosity of gruel (cP)

K = ball constant in $mPa.s.cm^3/g.s$

$\rho 1$ = density of the ball in g/cm3

$\rho 2$ = density of the liquid to be measured at the measuring temperature in g/cm3

t = falling time of the ball in seconds.

3.4.6. Sensory evaluation

To avoid panelist fatigue, best treatment combinations were selected from each cereals type. This was done by clustering each cereals based on the combination of fermentation period and ARF level. Then three treatments were selected from each cereals at which high energy (kcal) were recorded. Totally nine treatments were selected for sensory analysis. A total of 50 untrained females panelists were selected in this study. The use of mothers instead of the target recipient infant was necessary because of their ability to evaluate objectively the sensory characteristics of the selected starchy staple gruel to the interest of their children. The panelists were instructed about the purpose and objective of this test, the health status of the panelists was also considered during panelist selection (not suffering from colds and allergic that affect their sensitivity to the product.

The samples were coded in three digits and kept far apart to avoid crowding and for independent judgment. The complementary food thin porridge (gruels) was served to the panelists in white and transparent glass cups. Panelists were asked to rinse their mouth with tap water that was provided to them, before the next serving. The panelists were asked to rank the gruel on the basis of Appearance, aroma, taste, mouth-feel, consistency and overall acceptability using a five point hedonic scale**,** where:

5 = like extremely

4 = like moderately

3 = neither like nor dislike,

2 = dislike moderately and

1= dislike extremely (Muhimbula *et al*., 2011).

3.4.7. Data analysis

The effect of type of starchy staple, fermentation time and levels of ARF on physic-chemical properties of the flour and sensory properties of gruel was determined using a three-way analysis of variance. For each response, the validity of model assumptions, namely normal distribution and constant variance of the error terms assumptions, were verified by examining the residuals as described in Montgomery (2009). For those responses on the effect of factors was significant ($P < 0.05$), multiple means were compared and completed using the Tukey's studentized range test at the 5% level of significance, and letter groupings were generated. The Analysis of Variance (ANOVA) was done using SAS statistical software packages version) 9.2 (SAS®, 2002).The following model for factorial RCBD was used.

$$Yijk=\mu+\pi i+bj+tb(ij)+Ck+tc(ik)+bc(jk)+tbc(ijk)+£(ijk)$$

Where:

Yijk-is the response for the Y_{ijk} observation

μ - is the overall population mean of the response

π_i-is the treatment effect of the i^{th}level of factor A

b_j- is the effect of j^{th}level of factor B

$tb_{(ij)}$-is the interaction effect of the i^{th} level factor A and j^{th}level factor B

C_k-is the effect of the k^{th} level of factor C

$tc_{(ik)}$ - is the interaction of the factor A and factor C

$bc_{(jk)}$-is the interaction effect ofj^{th}level factor B and k^{th} level factor C

$tbc_{(ijk)}$ -is the overall interaction effect of the three factors.

£ (ijk)-is error term of the i^{th},j^{th} treatment and k^{th} replication

4. RESULTS AND DISCUSSION

The results of pH, titratable acidity, proximate compositions (moisture protein, fat ash, fiber, carbohydrate and energy), anti-nutritional factors (phytic acid and tannin), functional properties (bulk density water absorption capacity and viscosity) and sensory attributes are presented using tables and discussed as follows.

4.1. Effect of fermentation and amylase rich flour on pH and titratable acidity of oat, barley and teff based complementary food

4.1.1. pH

There was a highly significant ($P<0.01$) difference in pH due to the interaction effect of cereals type, fermentation time and ARF (Appendix Table 1). Fermentation time and ARF level were found to cause a gradual reduction in mean pH. The drop in pH value was probably the result of microbial activity on selected starchy staple flours converting some of the carbohydrates in to organic acids such as lactic acid, and acetic acids **(**Marko *et al.*, 2014).The change in pH from 0h without ARF to 48h with 5% ARF resulted in a pH drop from 6.40 to 4.20, 5.94 to 3.42 and 6.29 to 3.89 for oats, barley and teff flour, respectively (Table 2).

Similar results were found by Singh *et al.*, (2012) who reported that the pH drops from 5.2 to 3.73, 5.63 to 3.40 and 5.76 to 3.5 for sorghum, pearl millet and maize flour respectively after 36h fermentation period. These results also agreed with the result obtained by Mihiret (2009) who reported that increment of fermentation period significantly drops the pH value of Gobyie and 76T1#23 sorghum cultivars from 6.30 to 4.08 and 6.41 to 4.15, respectively.

Increasing fermentation time from 0h to 48h resulted in a sharp drop in pH with a corresponding increase in titratable acidity has been reported in lactic acid fermentation of various food grains (Elyas *et al*., 2002; Ejigui *et al*., 2005; Abdelhaleem *et al*., 2008; Shimelis and Rakshit, 2008). According to these authors, the production of lactic acid during fermentation has attributed to the decrease in pH. Khetarpaul, (2007) reported that fermented foods having acidic pH are microbiologically safe and can be stored for a long time.

Table 2. Interaction effect of type of cereal, fermentation time and amylase rich flour on mean value of pH

Amylase rich flour	Type of cereal by fermentation time								
	Oats			Barley			Teff		
	0h	24h	48h	0h	24h	48h	0h	24h	48h
0%	6.40^{a}	5.81^{f}	4.84^{l}	5.94^{e}	5.22^{k}	3.76^{r}	6.29^{b}	5.70^{g}	4.30^{n}
2%	6.10^{d}	5.44^{j}	4.71^{m}	5.82^{f}	4.82^{l}	3.61^{s}	5.81^{f}	5.53^{i}	4.00^{p}
5%	5.91^{e}	5.19^{k}	4.20^{o}	5.63^{h}	4.31^{n}	3.42^{t}	6.20^{c}	4.69^{m}	3.89^{q}
	Grand mean=5.09			LSD(0.05) = 0.049			CV (%) = 0.29		

Means sharing the same letter(s) are not significantly different at P=0.05 as established by LSD test.

4.1.2. Titratable acidity (TA)

Analysis of variance showed that the interaction effect among cereal type, fermentation time and ARF highly significantly (P< 0.01) affect the titratable acidity of selected fermented cereal flours (Appendix Table 1).Accordingly the highest TA value (0.80%) was recorded for barley flour fermented for 48h at 5% ARF level followed by 48h fermented teff flour at 5% ARF level (0.79%) which was statistically similar with 48h fermented oats flour at 5% ARF level (0.77%). while lowest value was obtained from unfermented oats, barley and teff flours at 0h, 0% ARF level (0.09%), which was statistically similar with unfermented teff flour with 2% ARF(Table 3).

The present study shows that TA of selected fermented starchy staple flours sharply increased as fermentation period and ARF levels increased. This observation might be due to the production of acid by lactic acid bacteria produced during the fermentation period. The result obtained from this experiment is in agreement with Anthony and Babatunde (2014) who reported that TA of fermented millet-soybean blend flour increased sharply with fermentation time. Similarly Meseret (2011) reported that titrable acidity of fermented QPM-soybean blend flour increased sharply with fermentation time and particle size distribution. Such increase in acidity due to microbial activity has been well documented in cereals, millets and cereal pulse mixtures fermented with endogenous grain microflora or with pure cultures (Ali *et al*., 2003).

In many traditional fermented products, the drop in pH and an increase in TA were a means for protection from many food borne pathogens (Shimelis and Rakshit, 2008). Especially in complementary foods, such physicochemical properties of fermented foods is highly desirable, for the fact that children are most of the time highly vulnerable for food pathogens due to their physiological conditions (Jay, 2000; Wambugu *et al*., 2002). According to Elyas *et al*. (2002), the increased acidity and low pH as a result of fermentation enhances the keeping quality of fermented foods, by inhibiting microbial growth and also contributing to the flavor of the processed food.

Table 3. Interaction effect of type of cereal, fermentation time and amylase rich flour on mean value of titratable acidity

	Type of cereal by fermentation time								
Amylase rich flour	Oats			Barley			Teff		
	0h	24h	48h	0h	24h	48h	0h	24h	48h
0%	0.09^{m}	0.44^{i}	0.69^{cd}	0.09^{m}	0.41^{i}	0.64^{de}	0.09^{m}	0.42^{i}	0.69^{cd}
2%	0.15^{kl}	0.57^{fg}	0.72^{bc}	0.18^{k}	0.52^{gh}	0.72^{c}	0.12^{lm}	0.51^{h}	0.72^{c}
5%	0.20^{jk}	0.63^{e}	0.77^{ab}	0.25^{j}	0.59^{ef}	0.80^{a}	0.25^{j}	0.59^{ef}	0.79^{a}
	Grand mean=0.47			LSD(0.05) = 0.053			CV(%)= 3.55		

Means sharing the same letter(s) are not significantly different at P=0.05 as established by LSD test.

4.2. Effect of fermentation time and ARF on the proximate composition of oat, barley and teff flours

4.2.1. Moisture content

The combined effect of the cereal type, ARF level and fermentation time resulted in a highly significant difference in the average moisture contents of selected cereals flour (P<0.01) (Appendix Table 2).The result presented in (Table 4) shows the moisture content of the flour samples ranged from 3.67% to 6.17%. Accordingly the maximum moisture content was recorded from 24h fermented Oat flour at 2% ARF followed by unfermented barley flour with 2% ARF. On the other hand the minimum value was obtained from 48h fermented teff flour at 2% ARF followed by 48h fermented Teff flour at 0% ARF. This was statistically similar with 24h fermented Teff flour without ARF and 48h fermented barley flour with 5% ARF.

Table 4. Interaction effect of type of cereal, fermentation time and amylase rich flour on mean values of moisture content

	Type of cereal by fermentation time								
Amylase rich flour	Oats			Barley			Teff		
	0h	24h	48h	0h	24h	48h	0h	24h	48h
0%	5.00[efg]	5.50[bcde]	4.67[fgh]	5.83[abc]	5.00[efg]	4.17[hij]	5.50[bcde]	4.00[ij]	3.83[j]
2%	5.00[efg]	6.17[a]	4.50[ghi]	6.00[ab]	5.17[def]	5.17[def]	4.50[ghi]	4.50[ghi]	3.67[j]
5%	5.00[efg]	5.17[def]	4.50[ghi]	5.50[bcde]	5.50[bcde]	4.00[ij]	5.17[def]	5.33[cde]	5.67[abcd]
	Grand mean=4.96			LSD(0.05) = 0.61			CV (%) = 3.88		

Means sharing the same letter(s) are not significantly different at P=0.05 as established by LSD test.

Variation in fermentation time and ARF has nothing to do with moisture content. However, the relative significant difference in moisture content may be attributed to a variation in treatment during the drying process of the fermented samples. The recommended moisture content for fermented cereal flours is less than 10% (Olorunfemi *et al.*, 2006).The moisture contents of selected cereal flours before and after fermentation obtained in this study were below 10% as can be seen from (Table 4). Those values are in agreement with the values obtained from Shimelis (2009) who reported that the moisture contents of sorghum flour after 48h germination period is less than 10%.

The result of current study was within the range of Nelson (1992) result who indicated moisture content as a quality factor for prepared cereals flours which should have 3-8% moisture content. It also very close to that of Amankwah *et al*.,(2009) who reported that the maximum moisture content after formulating complementary food from fermented maize, rice, soybean and fishmeal is 5.47% and 6.87% respectively. Such low moisture content of flours prevents microbial activity and extends the shelf life of the flour Kikafunda, (2006). Alozie *et al*., (2009) also reported that low moisture content in food samples increases the storage periods of the food products while high moisture content in foods encourage microbial growth; hence, food spoilage.

4.2.2 Total ash

Mean ash contents of selected fermented cereal flours showed a highly significant difference ($P<0.01$) due to the interaction effect among the cereal type, fermentation time and ARF level (Appendix Table 2). Forty eight hour fermented Teff flour at 2% ARF, 48h fermented Teff flour without ARF and 24h fermented Teff with 5% ARF attained the highest average total ash contents. On the contrary, the lowest mean values were obtained from unfermented oat flour at 2% ARF level which was statistically similar with 24h fermented oats flour at 2% ARF level (Table 5).

Table 5. Interaction effect of type of cereal, fermentation time and amylase rich flour on total ash content

	Type of cereal by fermentation time		
Amylase	Oats	Barley	Teff

rich flour	0h	24h	48h	0h	24h	48h	0h	24h	48h
0%	1.81^{ij}	1.80^{ij}	1.89^{h}	2.20^{f}	2.20^{f}	2.40^{d}	2.67^{b}	2.60^{c}	2.70^{a}
2%	1.75^{m}	1.77^{lm}	1.89^{h}	2.31^{e}	2.30^{e}	2.21^{f}	2.60^{c}	2.67^{b}	2.71^{a}
5%	1.78^{kl}	1.89^{h}	1.79^{jk}	2.1^{g}	2.30^{e}	2.30^{e}	2.60^{c}	2.70^{a}	2.67^{b}
	Grand mean=2.24			LSD(0.05) = 0.021			CV (%) = 0.29		

Means sharing the same letter(s) are not significantly different at P=0.05 as established by LSD test.

A variation in fermentation time and ARF levels have not uniformly increases or decreases the total ash contents of selected starchy staple flours as fermentation time and ARF level increases as compared to unfermented flour samples. However, highly significant variation was observed among the three way interaction. The observed variation in total ash content might be due to the nature of variation in mineral contents of selected cereals and leaching of some minerals in fermentation medium during fermentation periods. The level of ash in food is an important nutritional indicator for mineral density. These minerals may include calcium, potassium, phosphorus, iron, sodium, zinc, and magnesium and others at varying amounts (Mosha, and Vicent, 2005).

The results of this study are in concurrence with the report of Afoakwa *et al*., (2010) who reported that fermentation medium and ARF level had not dramatically increase or decrease total ash contents of cowpea fortified nixtamalized maize. This contradicts with the report of Mihiret (2009) who reported that ash content of fermented sorghum (76T1#23 Cultivar) decreases from 1.30% to 0.94%.as fermentation time increased from 0h to 48h. The total ash contents of all samples in present study was less than 5% which is in agreement with the recommendation of WHO/FAO (2004) reports that total ash contents of complementary food is (<5g/100g).

4.2.3 Crude protein

The crude protein content of selected starchy staple flour samples in the present study ranged from 8.12% to 16.82%. Analysis of variance showed that the interaction among cereals type, fermentation time and ARF highly significantly affect (P<0.001) the crude protein content of selected fermented cereal flours (Appendix Table 2). Accordingly, the maximum result for crude protein contents of selected cereal flour was recorded from 48 h fermented Oat flour at 5% ARF concentration (16.82 %) whereas the minimum value was obtained from unfermented teff flour at 0% ARF concentration (8.12%). This was statistically similar with unfermented teff flour at 2% ARF (8.34 %) (Table 6).

Table 6. Interaction effect of type of cereal, fermentation time and amylase rich flour on mean value of crude protein

	Type of cereal by fermentation time								
Amylase rich flour	Oats			Barley			Teff		
	0h	24h	48h	0h	24h	48h	0h	24h	48h
0%	11.10^{j}	12.63^{ef}	14.38^{c}	8.78^{pq}	10.33^{lm}	12.42^{fg}	8.12^{r}	9.93^{mn}	11.80^{hi}
2%	11.35^{ij}	12.95^{e}	15.61^{b}	8.97^{p}	11.03^{jk}	12.34^{fg}	8.34^{qr}	10.57^{kl}	12.10^{gh}
5%	11.83^{hi}	13.66^{d}	16.82^{a}	9.22^{op}	11.77^{hi}	12.99^{e}	9.53^{no}	11.66^{hi}	12.61^{ef}
	Grand mean=11.58			LSD(0.05) = 0.496			CV (%) = 1.35		

Means sharing the same letter(s) are not significantly different at P=0.05 as established by LSD test.

As both fermentation time and ARF concentrations increased, the amount of crude protein content of the flour samples increased. This increase in protein content as fermentation time increased may be due to shift in dry matter content through depletion during fermentation by action of the fermenting microorganisms. Values obtained from this study were comparable with the values reported by Meseret (2011) who reported that the crude protein contents of fermented QPM-soybean blend increased as fermentation periods increased.

Abdelhaleem *et al*., (2008) reported that the observed increment in protein content after fermentation was probably due to shift in dry matter content through depletion during fermentation by action of the fermenting microorganisms. Similarly, Onyango *et al*.,(2004) and Cui *et al*., (2012) also found out that the increase in protein content after fermentation was attributable to a decrease in carbon ratio in the total mass. Microorganisms utilize carbohydrates as an energy source and produce carbon dioxide as a by-product. This causes concentration of nitrogen in the fermented product, and thus the proportion of the proteins in the total mass increases.

Mugocha (2001) reported that fermentation is believed to increase total protein content. This is due to the decrease in starch and sugar as a result of hydrolysis by bacteria enzymes with the formation of volatile products such as lactic acid, acetic acid, carbon dioxide, and ethanol. This leads to changes in proportions of nutrient components.

Similarly Ade-Omowaye *et al*., (2003) and Osundahunsi (2006) reported that increment in protein content of the fermented cereal flours could be attributed to the breakdown of nutrients of substrates by natural microorganisms in fermented cereals. Chavan *et al*., (2006) also reported that natural fermentation in traditional African food preparation is an effective method of improving the protein and complex carbohydrate digestibility of cooked cereals**.**

4.2.4. Crude fat

Crude fat contents of the cereal flour samples showed highly significant difference ($P<0.01$) due to the three way interaction effect of cereal type, fermentation time and amylase rich flour (Appendix Table 2). According to the results presented in (Table 7), the maximum crude fat was recorded from unfermented oat flour with 0% ARF (4.55%) which was statistically similar with unfermented oat flour with 2% ARF (4.43%). On the other hand , minimum value was attained from 48h fermented barley flour with 5% ARF level(1.63%) which was statistically similar with fermented barley flour at 48h with 2% ARF(1.73%)

Table 7. Interaction effect of type of cereal, fermentation time and amylase rich flour on mean value of crude fat

	Type of cereal by fermentation time		
Amylase	Oats	Barley	Teff

rich flour	0h	24h	48h	0h	24h	48h	0h	24h	48h
0%	4.55[a]	3.64[c]	2.97[hi]	3.31[ef]	2.25[n]	1.85[pq]	3.53[cd]	2.83[jk]	2.60[lm]
2%	4.43[a]	3.27[f]	2.91[ij]	3.13[g]	2.02[o]	1.73[qr]	3.46[d]	2.74[kl]	2.53[m]
5%	4.14[b]	3.10[gh]	2.70[kl]	2.94[hij]	1.95[op]	1.63[r]	3.42[de]	2.68[l]	2.19[n]
	Grand mean=2.91			LSD(0.05) = 0.139			CV (%) = 1.51		

Means sharing the same letter(s) are not significantly different at P=0.05 as established by LSD test.

The crude fat content of current study was gradually decreased as fermentation time and ARF concentration increased. The observed decrease in fat content of selected starchy staple flour during fermentation might be attributed to the increased activities of the lipolytic enzymes during fermentation, which hydrolyze fats to fatty acids and glycerol. The result of this study can be confirmed with the findings of Shimelis, (2009) who reports crude fat contents of sorghum based complementary food was decreased as germination time was increased.

Mbata *et al*., (2006) and Fasasi *et al*., (2007) also reported as fermentation time increased the crude fat content of fermented weaning food flour was decreased. The decrease in fat content at the end of fermentation time may have resulted from oxidation during fermentation and also could be attributed to the use of lipid by the microorganisms to obtain energy for their metabolic activities during fermentation.

4.2.5. Crude fiber

There was a highly significant (P<0.01) difference in crude fiber contents accountable to the interaction effect of cereal type, fermentation time and ARF level (Appendix Table 2). The result presented in Table 8 describes that, the maximum value for crude fiber contents of present study was recorded from unfermented barley flour with 0% ARF (5.96%) followed by the statistically similar result recorded from unfermented barley flour with 2% ARF (5.78%). On the other hand, the minimum results were obtained from fermented Teff flour at 48h with 5% ARF (1.58%) which was statistically similar with fermented Teff flour at 48h with 2% ARF (1.72%).

Table 8. Interaction effect of type of cereal, fermentation time and amylase rich flour on mean value of crude fiber

Type of cereal by fermentation time

Amylase rich flour	Oats			Barley			Teff		
	0h	24h	48h	0h	24h	48h	0h	24h	48h
0%	4.56^{de}	3.91^{i}	3.15^{klm}	5.96^{a}	4.82^{c}	4.35^{fg}	3.21^{k}	2.53^{p}	1.85^{rs}
2%	4.16^{gh}	3.56^{j}	3.10^{lmn}	5.78^{ab}	4.75^{cd}	4.21^{fg}	2.92^{no}	2.26^{q}	1.72^{st}
5%	4.38^{ef}	3.33^{k}	2.98^{mn}	5.61^{b}	4.59^{d}	3.96^{hi}	2.74^{o}	1.99^{r}	1.58^{t}
Grand mean=3.63				LSD(0.05) = 0.198			CV (%) = 1.71		

Means sharing the same letter(s) are not significantly different at P=0.05 as established by LSD test.

The crude fiber content of current study decreased significantly as fermentation time and ARF concentration increased which was comparable with the result of Helen (2010) who reported that crude fiber contents of fermented finger millet based complementary food was decreased as fermentation time increased. This reduction in crude fibre may be due to the enzymatic breakdown of the fiber during fermentation by lactic acid bacteria which utilized them as carbon source and converted to microbial biomass, thereby reducing the fiber content of fermented cereals flours (Anthony and Babatunde, 2014).

Mugocha, (2001) reported that the expected decrease in fiber content during fermentation could be attributed to the partial solubilisation of cellulose and hemi-cellulosic types of material by microbial enzymes. The result of this study regarding the effect of fermentation and ARF on fiber contents are contradictory with the report of Amankwah *et al*. (2009) states that crude fiber contents increased during fermentation of corn meal.

Olorunfemi *et al.* (2006) reported that food with high fiber content tends to cause indigestion in babies. Hence, samples with low fiber content were rated good as potential complementary foods. Fermentation is a process which is promising to meet crude fiber standards in the preparation of complementary foods from locally available cereals. According to WHO (2004) specification, the maximum requirement is 5%. Therefore, the experimental values of the present study after fermentation are close to this value.

4.2.6. Total Carbohydrate

Analysis of variance showed that the interaction effect among cereal type, fermentation time and ARF highly significantly affect the total carbohydrate contents of selected fermented cereal flours (P < 0.01) (Appendix Table 2).According to the results presented in (Table 9), total carbohydrates content of the unfermented oat, barley and teff samples without ARF were 73.01%, 74.92%, and 76.98% for oats, barley and teff flours, respectively. Forty eight hours with 5% ARF fermentation caused a significant reduction of total carbohydrate in fermented oats (71.20%) and teff flour (75.28) while 48h fermented barley flour with 2% ARF (74.33%) showed significant reduction of total carbohydrate in barley flour as compared to unfermented flour. The carbohydrate content decreased as fermentation time and ARF concentrations increased but there are still slight fluctuations especially for Teff flour samples.

This may be due to the increasing or decreasing mean value of other proximate composition (moisture, ash, protein, fat and fiber) by the effect of ARF and fermentation process. The decreased carbohydrate content may be attributed to conversion of carbohydrate to glucose and used by fermenting microorganism as energy source (Anthony and Babatunde, 2014).

Table 9. Interaction effect of type of cereal, fermentation time and amylase rich flour on mean value of total carbohydrate

	Type of cereal by fermentation time								
ARF	Oats			Barley			Teff		
	0h	24h	48h	0h	24h	48h	0h	24h	48h
0%	73.01^{ijk}	72.53^{jk}	72.84^{jk}	74.92^{fgh}	75.41^{d}	74.82^{de}	76.98^{c}	78.11^{ab}	77.22^{c}
2%	73.32^{hij}	72.28^{kl}	72.01^{lm}	73.81^{ghi}	74.72^{def}	74.33^{efg}	78.18^{a}	76.43^{c}	77.28^{bc}
5%	72.88^{jk}	72.95^{jk}	71.20^{m}	74.63^{defg}	73.89^{fgh}	75.12^{de}	76.55^{c}	76.47^{c}	75.28^{d}
	Grand mean=74.68			LSD(0.05) = 0.85			CV (%) = 0.36		

Means sharing the same letter(s) are not significantly different at P=0.05 as established by LSD test.*ARF=Amylase rich flour*

The decrease in total carbohydrate could be due to particularly starch and soluble sugars are principal substances for fermenting microorganisms. Therefore degradation and subsequent decrease in starch content are expected to occur (Ejigui *et al*., 2005). The result of this study regarding the effect of fermentation and ARF on total carbohydrate contents are in agreement with the report of Mihiret(2009) and Helen(2010) who stated that minimum carbohydrate content was recorded from 48h fermentation of sorghum based complementary food and black cultivar finger millet flour respectively .All mean value of fermented selected starchy staples of current study meets the carbohydrate content ≥65g/100g recommended by WHO/FAO(2004) for complementary foods.

4.2.7. Energy

The result of the present study indicated that the interaction effect among cereals type, fermentation time and different levels of ARF on calorific values were observed to be highly significantly different ($p<0.01$) (Appendix Table 2). The results presented in Table 10 explain that the calorific values of cereal flour samples varied from 359.33 to 380.26 kcal/100g. The highest gross energy content was observed in 48 h fermented Teff flour with 2% ARF (380.26 kcal/100g) followed 48h fermented Teff with 0% ARF (379.50 kcal/100g). While the lower energy content was observed in unfermented barley flour with 2% ARF (359.33kcal/100g) which is statistically similar with 24h fermented barley flour with 5% ARF.

Table 10: Interaction effect of type of cereal, fermentation time and amylase rich flour on mean values of energy

	Type of cereal by fermentation time								
ARF	Oats			Barley			Teff		
	0h	24h	48h	0h	24h	48h	0h	24h	48h
0%	377.26^{bcd}	373.34^{fghi}	376.03^{cdef}	361.61^{mno}	363.17^{lm}	365.5^{lkl}	372.18^{hij}	377.60^{abcd}	379.50^{ab}
2%	378.49^{abc}	370.38^{j}	376.68^{cde}	359.33^{o}	361.24^{mno}	362.28^{mn}	377.24^{bcd}	372.66^{ghij}	380.26^{a}
5%	376.06^{cdef}	374.10^{efgh}	376.42^{cde}	361.89^{mno}	360.20^{no}	367.11^{k}	375.06^{defg}	376.61^{cde}	371.31^{ij}

Grand mean=371.21	LSD(0.05) = 2.78	CV (%) = 0.24

Means sharing the same letter(s) are not significantly different at P=0.05 as established by LSD test. *ARF=Amylase rich flour*

As the fermentation time and ARF increased, the calorific value of barley and teff flour also increased but there are still slight fluctuations within ARF concentration but in the case of oats flour the calorific values was decreased. This may be due to the increasing or decreasing values of the three energy source food staffs (protein, fat and carbohydrate) by the effect of ARF and fermentation process. Marko *et al., (*2014) reported that energy value decreased as a result of the total carbohydrate and lipid reduction during the fermentation of cereal substrates.

The gross energy of fermented oat and teff flour observed in this study were higher than the gross energy value reported by Helen (2010) 340 to 370kcal/100g.However, Beruk *et al.,* (2015) reported a higher gross energy (386.75 to 394.83 kcal/100g) for QPM based complementary foods. The energy content obtained in the current study is less than the recommended energy content of 400-420kcal/100g for infant complementary foods (WHO/FAO, 2004). This is due to the less protein and fat contents of the raw materials used in fermentation process of the current study.

4.3. Effect of fermentation time and ARF concentration on phytate and tannin contents

4.3.1. Phytate

There was a highly significant (P<0.01) interaction effect of the three factors on the phytate content of samples (Appendix Table 3). According to the result on Table 11, unfermented teff flour with 0% ARF records the highest phytate content (175.07mg/100g) of the current study, while the least phytate content was recorded from 48h fermented oat flour with 5% ARF (18.63 mg/100g). Phytate is naturally present in many foods especially cereals and legumes. When above a certain level, phytate reduce the availability of minerals and solubility, functionality and digestibility of proteins (Akubo and Badifu, 2004).

In general, the phytate contents decreased as ARF levels and fermentation periods increased. This is due to fermentation can induce phytate hydrolysis via the action of microbial phytase enzymes, which hydrolyze phytate to lower inositol phosphates. This results in line with the finding of Meseret (2011) who reported that the phytate contents of fermented QPM-soybean blend flour was decreased as fermentation time increased from 0h to 48 .The result of current study also in agreement with the findings of Fagbemi *et al.,* (2005) who reported that fermentation is the most effective processing technique that reduced phytic acid in the cereal flours.

Table 11. Interaction effect of type of cereal, fermentation time and amylase rich flour on Phytate (mg/100g) contents of selected starchy staple

	Type of cereal by fermentation time								
Amylase rich flour	Oats			Barley			Teff		
	0h	24h	48h	0h	24h	48h	0h	24h	48h
0%	88.59^{gh}	52.37^{mn}	34.66^{op}	108.90^{e}	86.75^{ghi}	68.76^{jkl}	175.07^{a}	143.43^{bc}	102.86^{ef}
2%	75.92^{ijk}	45.04^{no}	27.64^{pq}	105.15^{ef}	80.14^{hij}	62.66^{lm}	153.81^{b}	133.49^{cd}	88.59^{ghi}
5%	64.92^{klm}	40.30^{no}	18.63^{q}	93.00^{fg}	72.64^{jkl}	41.55^{no}	150.12^{b}	122.48^{d}	76.35^{hijk}
Grand mean=85.75			LSD(0.05) = 12.56			CV (%) = 4.60			

Means sharing the same letter(s) are not significantly different at P=0.05 as established by LSD test.

Most of the reduction of phytate in the present study occurred during the 48 hour of fermentation with 5% ARF. According to Khetarpaul and Chauhan (2007) Lactic acid fermentation decreased content of phytic acid in the fermented cereal product. This may be due to the prevailing pH which is considered to be an optimum pH for microbial phytase activity, since all enzymes have a specific pH in which they function most proficiently. It has been stated that phytate is insoluble at pH 6; the microbial phytase activity is inhibited below pH 5.0 (Abdelhaleem *et al*., 2008). It has been suggested by different researchers the loss of phytate during fermentation could be a result of the activity of native phytase and/or the fermentative microflora (Elyas *et al*., 2002; Shimelis and Rakshit, 2008).

4.3.2. Condensed tannin

The condensed tannin contents of current study showed a highly significant difference (P<0.01) due to interaction effect of the three factors (Appendix Table 3). The unfermented oats flour with 0% ARF shows highest mean value of tannin content (42.89 mg/100g) while the least tannin content was recorded from teff flour with 5%ARF and 48h of fermentation (0.84mg/100g) which is statically similar with 48h fermented teff flour with 2% ARF (2.52 mg/100g) (Table 12). Booth *et al.,* (2001) reported that among the total of 0.25% tannin content, the tannin activity is high in hull (11.06mg/g and lower in endosperm (0.16mg/g) part of grains which is easily reduced by de hulling process during preparation of complementary foods.

Table 12. Interaction effect of type of cereals, fermentation time and amylase rich flour concentration on mean value of condensed Tannin (mg/100)

	Type of cereal by fermentation time								
ARF	Oats			Barley			Teff		
	0h	24h	48h	0h	24h	48h	0h	24h	48h
0%	42.89^{a}	26.99^{cd}	18.06^{fg}	18.55^{efg}	12.92^{hijk}	9.15^{ijklmn}	8.49^{jklmno}	5.39^{nopq}	3.51^{opq}
2%	33.31^{b}	23.46^{de}	14.63^{gh}	15.15^{gh}	11.98^{hijkl}	7.90^{klmno}	7.26^{lmnop}	4.69^{nopq}	2.52^{pq}
5%	29.97^{bc}	20.43^{ef}	13.50^{ghi}	14.02^{ghi}	10.66^{hijkl}	5.33^{nopq}	6.31^{mnop}	4.16^{nopq}	0.84^{q}
Grand mean=13.78			LSD(0.05) = 5.09			CV (%) = 11.62			

*Means sharing the same letter(s) are not significantly different at P=0.05 as established by LSD*test.ARF=Amylase rich flour

The current study result was far lower than that of Mihiret (2009) who reported that tannin content of fermented sorghum cultivar ranged from 17.95mg/100g to34.25mg/100g and 33.10mg/100g to 80.06mg/100g for gobyie and 76T1#23 cultivars respectively. However tannin content of both cultivars was decreased as fermentation time increased which is similar with the present study. Reduction in tannin contents due to fermentation might have been caused by the activity of polyphenoloxidase or tanniase of fermenting microflora on tannins (Fagbemi *et al.,* 2005*)*.Tannins form insoluble complexes with proteins thereby decreasing the digestibility of proteins (Uzoechina, 2007). Tannins also decrease palatability, cause damage to intestinal tract, and enhance carcinogenesis (Makkar and Becker, 1996).

During the preparation of many fermented foods, tannins are reduced before the fermentation step because of their presence in the seed coats of the raw ingredients. According to previous researchers, dehulling and cooking eliminated more than 90% of tannins in pulse and cereals because of their predominance in the seed coats. In several fermented foods, the seed coat or testa is removed from the substrate before fermentation so the anti-nutritional potential caused by the presence of tannins is of little concern (Shimelis & Rakshit, 2008).

4.4. Functional Properties of Fermented Selected Starchy Staples Flour

4.4.1. Bulk density

The result of the current experiment showed that the interaction effect among cereal types, fermentation time and amylase rich flour on bulk density of selected cereal flours was found to be highly significant (P<0.01) (Appendix Table4). Bulk density values ranged from 0.66g/ml to 0.99g/ml. Accordingly, the maximum result for bulk density of selected cereal flours was recorded from unfermented Teff flour at 0% ARF concentration (0.99g/ml) whereas the minimum value was obtained from 48h fermented barley flour at 5% ARF concentration (0.66g/ml) (Table13).

Table 13. Interaction effect of type of cereal, fermentation time and amylase rich flour on mean value of bulk density

	Type of cereal by fermentation time								
Amylase rich flour	Oats			Barley			Teff		
	0h	24h	48h	0h	24h	48h	0h	24h	48h
0%	0.92^{c}	0.78^{j}	0.74^{lm}	0.89^{d}	0.82^{i}	0.73^{nm}	0.99^{a}	0.89^{d}	0.75^{kl}
2%	0.86^{ef}	0.76^{k}	0.72^{mn}	0.87^{e}	0.76^{k}	0.70^{op}	0.96^{b}	0.84^{ghi}	0.71^{no}
5%	0.84^{fgh}	0.75^{kl}	0.70^{op}	0.85^{efg}	0.75^{kl}	0.66^{q}	0.92^{c}	0.82^{hi}	0.69^{p}
	Grand mean=0.80			LSD(0.05) = 0.0197			CV (%) = 0.77		

Means sharing the same letter(s) are not significantly different at P=0.05 as established by LSD test.

Bulk density values of flour samples decreased gradually with increase in fermentation period and amylase rich flour. Values obtained from this study were comparable with the values reported by Meseret (2011) and Singh *et al.,* (2012) who reported that the bulk density of fermented QPM-soybean blend decreased as fermentation periods increase from 0.79g/ml to 0.67g/ml and bulk density of fermented cereal flour from 0.75g/ml to 0.60g/ml respectively. The decrease in bulk density of fermented flour would be an advantage in the preparation of low bulk weaning foods for infants (Elkhalifa *et al.*, 2005). Similar report was reported by Singh *et al.*, (2012) which promote fermentation as a useful and traditional method for the preparation of low bulk weaning foods.

Gernah *et al*. (2011) also reported that the bulky nature of cereal based weaning foods is the other discouraging factor for many infants from consuming it. According to Peleg and Bagley (1983), bulk density depends on the combined effects of interrelated factors such as the intensity of attractive inter-particle forces, particle size, and number of contact points. Bulk density is also important in infant feeding where less bulk is desirable. The low bulk density of flour would an advantage in the use of the flour for preparation of complementary foods (Akubor *et al.,* 2013). The differences in the particle size may be the cause of variations in bulk density of flours. Bulk density is an indication of the porosity of a product which influences package design and could be used in determining the type of packaging material required, material handling and application in wet processing in the food industry (Kinsella, 1987).

4.4.2. Water absorption capacity (WAC)

Results of the current investigation for water absorption capacity of selected starchy staples signified that the interaction effect among the cereal types, fermentation period and amylase rich flour were showed a highly significant difference ($P<0.01$) (Appendix Table 4). The values ranged from 61.33% to 143.12% as presented in Table 14. The maximum WAC value was recorded from unfermented barley flour and at 0% ARF concentration (143.12%), whereas the minimum result was recorded from fermented teff flour at 48h fermentation period and at 5% ARF concentration (61.33%).

Table 14. Interaction effect of type of cereal, fermentation time and amylase rich flour on mean on water absorption capacity

ARF	Type of cereal by fermentation time								
	Oats			Barley			Teff		
	0h	24h	48h	0h	24h	48h	0h	24h	48h
0%	136.30^{b}	116.15^{fg}	94.86^{l}	143.12^{a}	128.00^{d}	102.14^{k}	121.00^{e}	105.30^{j}	81.00^{n}
2%	129.00^{d}	109.11^{i}	85.70^{n}	136.00^{b}	121.00^{e}	94.48^{l}	117.00^{f}	94.78^{l}	73.00^{p}
5%	120.70^{e}	101.15^{k}	74.40^{p}	132.10^{c}	115.15^{g}	85.51^{n}	113.00^{h}	88.67^{m}	61.33^{q}
	Grand mean =106.66			LSD(0.05) = 1.6			CV (%) = 0.47		

Means sharing the same letter(s) are not significantly different at P=0.05 as established by LSD test

This high water absorption capacity of unfermented barley flour was because of more fiber content than oats and teff flour. According to Rosell *et al*., (2006) as cited in Nasser *et al*., (2008), the differences in water absorption is mainly caused by the greater number of hydroxyl group which exist in the fiber structure and allow more water interaction through hydrogen bonding. Water absorption capacity mean values of selected cereals flour was decreased significantly with increase in fermentation periods and amylase rich flour.

The decrease in water absorption with increasing ARF concentration and fermentation time is due to the breakdown of the starch by amylolytic enzymes (α-amylase and β -amylase) present in the ARF, which were suspected to be produced in the aleurone layer during the malting process. Lawal and Adebowale (2004) reported that chemical composition that enhances the WAC of flour is carbohydrate, since this component contains hydrophilic parts such as polar or charged side chains. Similarly Kaur and sing (2005) reported that flour with higher WAC have more hydrophilic constituents, such as polysaccharides.

The result of this study is in agreement with the report of Singh *et al.*, (2012) who reported that the water absorption capacity of selected fermented cereal flours (sorghum, pearl millet and maize) was decreased from 141% to 77% as fermentation period is increased from 0 h to 36 h. On other hand, Okerie and Bello (1988) as cited in Singh *et al.*, (2012), water absorption capacity is a useful indication of flour or isolates whether it can be incorporated into aqueous food formulations especially those involving dough handling and it gives an indication of the amount of water available for gelatinization. Lower water absorption capacity is desirable for making thinner gruels.

Water absorption characteristic represents ability of the product to associate with water under conditions when water is limiting such as dough's and pastes (Akubor *et al.*, 2013). Water absorption capacity of flour may depend on the higher polar amino acid residues of proteins that have an affinity for water molecules (Yusuf *et al*., 2008).

4.4.3. Viscosity

The viscosity of gruels shows highly significant ($p<0.01$) difference due to the interaction effect of cereal type, fermentation time and ARF concentration (Appendix Table 4). According to the results presented in Table 15, the viscosity of gruel samples varied from 235cP to 1016.33cP. The highest mean viscosity was recorded for a gruel prepared from unfermented barely flour with 0% ARF while the least mean value was obtained from 48h fermented teff flour with 5% ARF followed by 48h fermented teff flour with 2% ARF.The viscosity of gruels decreased significantly with increased period of fermentation and ARF in all samples and reached 436.33, 493.67 and 235 cP within 48h fermentation and 5% ARF for oats, barley and teff, respectively.

This study is in agreement with Afoakwa *et al.*, (2009) who reported that, addition of maize and millet malts to fermented maize was most effective in lowering the viscosity of maize based porridges. Wambugu *et al.*, (2006) reported that production of porridges from malted and fermented flours mixes resulted in lower viscosities.

Table 15. Interaction effect of type of cereal, fermentation time and amylase rich flour on mean value of viscosity

	Type of cereal by fermentation time								
	Oats			Barley			Teff		
ARF	0h	24h	48h	0h	24h	48h	0h	24h	48h
0%	650.00[bcde]	577.00[defg]	465.33[fghij]	1016.33[a]	740.00[bcd]	749.67[bcd]	523.67[efgh]	426.67[ghij]	337.67[ijk]

2%	653.67^{bcde}	538.33^{efgh}	433.67^{ghij}	785.00^{bc}	791.67^{bc}	729.00^{bcd}	497.33^{efghi}	402.00^{ghijk}	307.33^{jk}
5%	621.00^{cdef}	506.67^{efghi}	436.33^{ghij}	825.33^{b}	758.67^{bc}	493.67^{efghi}	484.33^{efghij}	360.33^{hijk}	235.00^{k}
	Grand mean=568.36			LSD(0.05) = 178.85			CV (%) = 9.88		

Means sharing the same letter(s) are not significantly different at P=0.05 as established by LSD test, *ARF=amylase rich flour.*

The decrease in viscosity during fermentation may be due to increased α-Amylase activity from ARF flour. This enzyme can hydrolyze amylose and amylopectin to dextrins and maltose, thus reducing the viscosity of thick cereal porridges (Gibson *et al*., 2006). Mosh *et al*., (2004) reported that the viscosity of infant feed must be less than 3000 cP. Therefore, the results of this study were far lower than this value.

The gruels viscosity exceeded upper viscosity limit (3000 cP) need to be diluted with water to make them suitable (for their viscosity) for infant feeding. According to Shimelis (2009) viscosity is the most important determinant of energy density. Complementary porridges prepared in developing countries are known with high viscosity which can limit the energy and nutrient need of children. John *et a*l., (2008) also reported that the high rates of malnutrition in developing countries are due to the bulkiness and high viscosity of the predominant cereal-based complementary foods.

4.5. Sensory evaluation of gruels prepared from selected starchy staples flours

Sensory analysis or sensory evaluation is a scientific discipline that is done by the use of human senses for the purposes of evaluating consumer acceptance of the product products. The discipline requires panels of human assessors, on whom the products are tested, and recording the responses made by them. Organoleptic properties are the aspects of food or other substances as experienced by the senses, including taste, sight, smell, and touch (Falola *et al.,* 2011). The sensory analysis of variance result is summarized in the Appendix Table 5 show that there were highly significant difference (P<0.01) among the samples prepared from selected grain flours in terms of appearance, aroma, taste, mouth feel, consistency and overall acceptability.

4.5.1. Appearance

The appearance of the thin porridge (gruel) made from 24h fermented oat flour with 5% ARF was preferred by the panelists which was followed by unfermented oat flour with 2% ARF, 24h fermented barley flour with 0% ARF, unfermented teff flour with 2% ARF and 24h fermented teff flour with 0% ARF respectively. They were statistically similar with each other (Table 16).

Whereas the gruel prepared from 48h fermented barley flour with 5% ARF was least preferred which was flowed by both 48h fermented Oat and Teff flour with 2%ARF they were statistically similar with each other.

The appearance of the gruels prepared from unfermented and 24hrs fermented grain flour samples was the most preferred than 48hrs fermented flour. This is due to gruels from unfermented and 24hrs fermented flour were white while 48hfermented were dark brown. The results obtained in this investigation was consistent with findings obtained by Kikafunda *et al*.,(2006), who reported that the most preferred porridge in terms of appearance was the most white in color, while the least preferred was the creamiest in color

Table 16. Effect of fermentation and ARF on Sensory mean scores for samples (Gruels) evaluated by untrained panels

Samples	F(h)	ARF	Appearance	Aroma	Taste	Mouth feel	Consistency (Viscosity)	Overall acceptability
	0	2	4.38a	4.16ab	4.02ab	3.98ab	2.10b	4.00ab
Oats	24	5	4.44a	4.52a	4.42a	4.36a	3.90a	4.42a
	48	2	2.14c	2.28e	2.38cd	2.14e	4.38a	2.64d
Barley	0	5	3.60b	3.34cd	3.48b	3.18cd	1.64b	3.28cd
	24	0	4.06ab	4.00ab	3.72b	3.72abc	3.76a	3.80abc
	48	5	1.88c	2.20e	1.94d	2.02e	4.02a	2.66d
Teff	0	2	3.90ab	3.68bc	3.58b	3.52bc	2.26b	3.40bc
	24	0	3.90ab	4.18ab	4.06ab	3.76abc	3.92a	3.98ab
	48	2	2.26c	2.80de	2.64c	2.70de	4.16a	3.34bc
CV (%)			29.43	29.25	32.86	35.30	34.05	31.08
LSD(5%			0.62	0.63	0.69	0.72	0.71	0.68

Means sharing the same letter(s) are not significantly different at P=0.05 according to LSD Test. F(h)= fermentation time, ARF= Amylase rich flour.)

The gruels prepared from 48hr fermented flour was less white probably due to formation of brown color (melanoidins) through a maillard reaction when the starch reacted with proteins probably during oven drying following fermentation. The presence of sugars from starch hydrolysis might also lead to color changes in the presence of proteins upon exposure to high temperature as reported by (Kikafunda *et al.*, 2006).

4.5.2. Aroma

The aroma of gruel prepared from 24h fermented with 5% ARF oats flour was most preferred by panelists followed by 24h fermented teff flour, unfermented oats flour and 24h fermented barley flour. While Gruels prepared from 48h fermented barley flour with 5% ARF and 48h fermented oat flour with 2% ARF was least preferred (Table 16) which was statistically similar with 48h fermented Teff flour. Gruels prepared from 48h fermented selected starchy staple of current experiment were least preferred by untrained panelists. This is due to the production of volatile compound such as lactic acid, butyric acid and alcohol by microbial activities which increased as the fermentation progressed may account for the perceived changes in aroma of the gruels fermented for different length of time (Oyewole and Ogundele, 2001).

Katongole (2008) reported that during cereal fermentations, several volatile compounds are formed which contribute to a complex blend of flavors in the products. The presence of aroma representatives; diacetyl, acetic acid and butyric acid make fermented cereal based products more appetizing.

4.5.3. Taste

The highest scores for taste were obtained from gruels prepared from 24h fermented oats flour with 5%ARF, followed by 24h fermented teff flour and unfermented oats flour, whereas the least taste scores was obtained from 48h fermented barley flour with 5% ARF, which is statistically similar with 48h fermented oat flour with 2% ARF.The taste of thin porridge prepared from 48h fermented selected starchy staple flour of present study was least preferred as compared to unfermented and 24h fermented flour (Table 16). This is due to higher acidity resulted from the prolonged fermentation time. This study is in agreement with the report of Mihret (2009) who reported that gruels prepared from 48h fermented sorghum flour were least accepted by sensory panelists.

4.5.4. Mouth feel

Mouth feel is the sensation produced by a material taken in the mouth, perceived principally by the senses of taste, and also by the general pain, tactile, and temperature receptors in the mouth. According to the result presented in Table 16 , gruel prepared from 24h fermented oat flour with 5% ARF and unfermented oat flours scored highest mean value of mouth feel while least mean score for mouth feel was obtained from 48h fermented barley flour with 5% ARF, followed by 48h fermented oats flour and 48h fermented teff flour respectively.

4.5.5. Consistency

The consistency of thin porridge prepared from both 24h and48h fermented selected cereal flours were mostly preferred while gruel prepared from all unfermented cereal flours was least accepted (Table 16). This could be due to the viscous nature of the all gruels prepared from unfermented cereal flour samples.

In other way the gruels prepared from 24h and 48h fermented flour were less thick (low in viscosity) than unfermented cereals flours this might be due to activity of fermentation microbes that convert starch to sugar and makes the gruels or porridge less thick. Thus, starch degradation reduces the viscosity of thick cereal porridges without dilution with water while simultaneously enhancing their energy and nutrient densities (Michaelsen *et al.,* 2008).

An infant needs a gradual transition from fluid to solid foods which allow the infant to develop feeding skills. Early in weaning, the infant will reject very viscous foods by spitting them out (Kabeir *et al*., 2004). During weaning inadequate intake of energy and protein occurs due to traditionally prepared complementary foods, which are bulky in nature (have high viscosity but low energy density) and limit the infant’s ability to eat enough (Agostoni *et al*., 2008).

4.5.6. Overall acceptability

Overall acceptability of gruels prepared from 24h fermented oat flour with 5% ARF was most accepted, which was followed by unfermented oat flour with 2% ARF and 24h fermented Teff flour respectively. For acceptability, gruels prepared from 48h fermented Oat flour with 2% ARF and 48h fermented barley flour with 5% ARF were least accepted respectively which is statistically similar with unfermented barley flour with 5% ARF (Table 16). The microbial activities and organic acid production which increased as the fermentation progressed may account for the perceived changes in the taste, aroma and overall acceptability of the gruels fermented for different lengths of time.

In this study the result shows that the panelist like highly significantly ($p<0.01$) the overall acceptability of the gruel prepared from selected starchy staple flour that was subjected to 24h fermentation differ from 48h fermentation. Sahana & Fauzia (2003) reported that fermentation may include improvement in palatability and acceptability by developing improved flavors and textures.

5. SUMMARY AND CONCLUSIONS

5.1. Summary

This study was aimed in developing value added products that provide low viscosity, low dietary bulkiness, and low anti-nutrient content, high energy and nutrient density, therefore, increased food intake and bioavailability of nutrients for 6-24 months old children.

The present work mainly focused on investigating the effect of fermentation process and ARF on the physicochemical and sensory properties of complementary food from oat, barley and teff. The combined effect of cereal type, fermentation time and ARF levels significantly affect pH, TA, proximate composition, anti-nutritional factors, functional properties and sensory properties of products from oat, barley and teff. Variation in fermentation time with addition of ARF resulted in decrease in pH, crude fat, crude fiber, total carbohydrate, phytic acid, tannin, bulk density, water absorption capacity. On the other hand, combined effect of fermentation time and ARF level resulted in significant increase in the TA, crude protein and calorific value.

Fermentation and ARF had highly significantly ($P<0.01$) affect the sensory analysis evaluated by untrained sensory panelists. Therefore, gruel prepared from 24h fermented oat flour with 5% ARF, unfermented oat flour with 2% ARF and 24h fermented teff flour were acceptable with higher mean scores of 4.42, 4.00 and 3.98 respectively for overall acceptability. The panelists ranked that appearance, aroma, taste, mouth feel and overall acceptability of the gruel prepared from selected cereal flours that were subjected to 48h fermentation differ from others and was least accepted. In contrast, gruels prepared from oat, teff and barley flours that was subjected to 48h fermentation period was acceptable with higher mean score 4.38, 4.16 and 4.02, for consistency respectively.

5.2. Conclusion

In the present work, it was demonstrated that combined effect of ARF and fermentation process significantly improved the nutritional value and energy density of the complementary food from oat, barley and teff by reducing water absorption capacity, bulk density, viscosity and anti-nutritional factors. Finally, it can be concluded from the sensory analysis that the gruels prepared from 24h fermented oat, barley and teff flours were mostly preferred by panelist with high mean score value than unfermented and 48h fermented flour in terms of appearance, aroma, taste, mouth feel, consistency and overall acceptability. Therefore fermentation period of 24h is recommended for the production of nutritious and acceptable complementary food prepared from fermented oat, barely and teff flours. Fermentation with addition of ARF can therefore be employed to further improve the nutritive value and functionality of energy-dense products from oat, barley and teff flours for the production of semi-solid porridges suitable for use as complementary foods for infant feeding.

6. FUTURE LINE OF WORK

Based on current research findings, observation and experiences the following research topics are recommended for immediate or future consideration.

- Mineral determination of the fermented oat, barley and teff flour.
- Microbial analysis and shelf life of the fermented oat, barley and teff flour.
- Analysis of starch and other functional properties such as emulsion activity and stability; foaming capacity and stability; and water solubility index.
- There should be awareness creation on the value addition of oat, barley and teff based complementary food accompanied with simple processing methods like combined effect of fermentation and ARF deep in the society by demonstrating developed products like fermented oat, barley and teff based complementary foods, for lower class families especially in rural areas.

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8. APPENDICES

A) List of tables in appendices

Appendix Table 1: Mean square values of pH and TA affected by cereals type, fermentation time and Amylase rich flour and interaction

Source of variation	Degree of freedom	Mean square values	
		pH (%)	TA (%)
Block	2	0.000386^{ns}	0.001721^{**}
CT	2	3.179^{***}	0.002098^{***}
FT	2	25.32^{***}	2.178^{***}
ARF	2	1.928^{***}	0.156343^{***}
CT*FT	4	0.269^{***}	0.002335^{***}
CT*ARF	4	0.0132^{***}	0.001678^{***}
FT*ARF	4	0.2362^{***}	0.002100^{***}
CT*FT*ARF	8	0.0962^{**}	0.000995^{**}
Error	52	0.000227	0.000218
CV (%)		0.29	3.55

, **, *, significant, highly significant and very highly significant respectively; ns=non significant at P=0.05*

Appendix Table 2: Mean square values of Total ash, moisture contents, crude protein, crude fat, crude fiber, Total Carbohydrate and Calorific value affected by cereals type, fermentation time and Amylase rich flour and interaction

Source of variation	Df	Mean square values						
		Total Ash (%)	Moisture content (%)	Crude protein (%)	Crude fat (%)	Crude fiber (%)	Carbohydrate (%)	Calorific value (Kcal.)/100g
Block	2	0.000046^{ns}	0.12037*	0.3362***	0.0048^{ns}	0.0012^{ns}	0.680***	2.390*
CT	2	4.814***	1.620***	65.085***	9.848***	45.009***	129.815***	1580.30***
FT	2	0.04699***	5.176***	95.577***	12.32***	13.049***	0.908***	57.535***
ARF	2	0.00384***	0.361***	9.708***	0.655***	0.849***	2.987***	4.797**
CT*FT	4	0.00159***	1.213***	0.8220***	0.159***	0.116***	1.727***	28.220***
CT*ARF	4	0.00197***	0.856***	0.254***	0.0245**	0.026***	1.409***	12.313***
FT*ARF	4	0.01762***	0.815***	0.176***	0.013***	0.036***	1.434***	18.480***
CT*FT*ARF	8	0.01339**	1.130**	0.452**	0.023**	0.028**	1.413**	23.387**
Error	52	0.000043	0.033832	0.0123	0.00182	0.00396	0.0485	0.690
CV (%)		0.29	3.88	1.35	1.51	1.71	0.36	0.24

, **, *, significant, highly significant and very highly significant respectively; ns=non significant at P=0.05 df= degree of freedom*

Appendix Table 3: Mean square values of Phytate and Tannin affected by cereals type, fermentation time and Amylase rich flour and interaction.

Source of variation	Degree of freedom	Mean square values	
		Phytate mg/100g	Tannin mg/100g
Block	2	129.07***	24.17***
CT	2	41490.61***	2786.86***
FT	2	20354.51***	844.75***
ARF	2	2802.68***	140.56***
CT*FT	4	601.360***	141.28***
CT*ARF	4	78.32***	24.04***
FT*ARF	4	42.49**	8.92***
CT*FT*ARF	8	29.37**	4.92**
Error	52	12.21	1.73
CV (%)		4.60	11.62

, **, *, significant, highly significant and very highly significant respectively; ns=non significant at P=0.05*

Appendix Table 4: Mean square values of Bulk density, water Absorption capacity and Viscosity affected by cereals type, fermentation time and Amylase rich flour and interaction

Source of variation	Degree of freedom	Mean square values		
		BD(g/ml)	WAC (%)	Viscosity(cP)
Block	2	0.00024**	1.266**	24535.346***
CT	2	0.03206***	3423.812***	929366.901***
FT	2	0.23826***	13145.809***	291144.975***
ARF	2	0.02262***	1539.220***	48898.457***
CT*FT	4	0.00646***	18.161***	267.883ns
CT*ARF	4	0.00018***	8.167***	6088.309*
FT*ARF	4	0.00044***	33.843***	9493.216***
CT*FT*ARF	8	0.0007 **	6.058**	11751.957**
Error	52	0.000030	0.214	2329.423
CV (%)		0.77	0.47	9.88

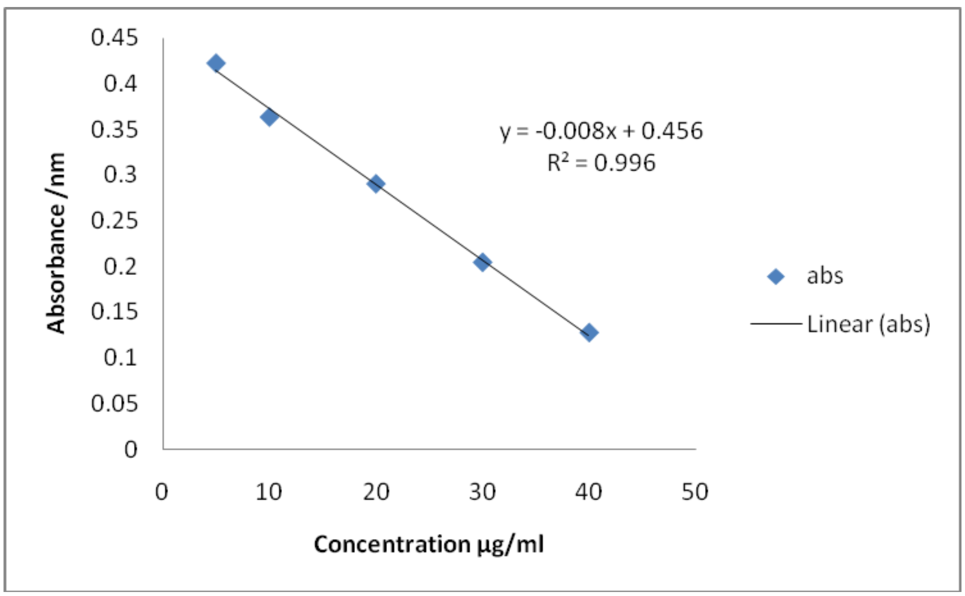
, **, *, significant, highly significant and very highly significant respectively; ns=non significant at P=0.05.BD= bulk density, WAC= water Absorption capacity.*

Appendix Table 5: Mean square values of Appearance, Aroma, Taste, mouth feel consistency

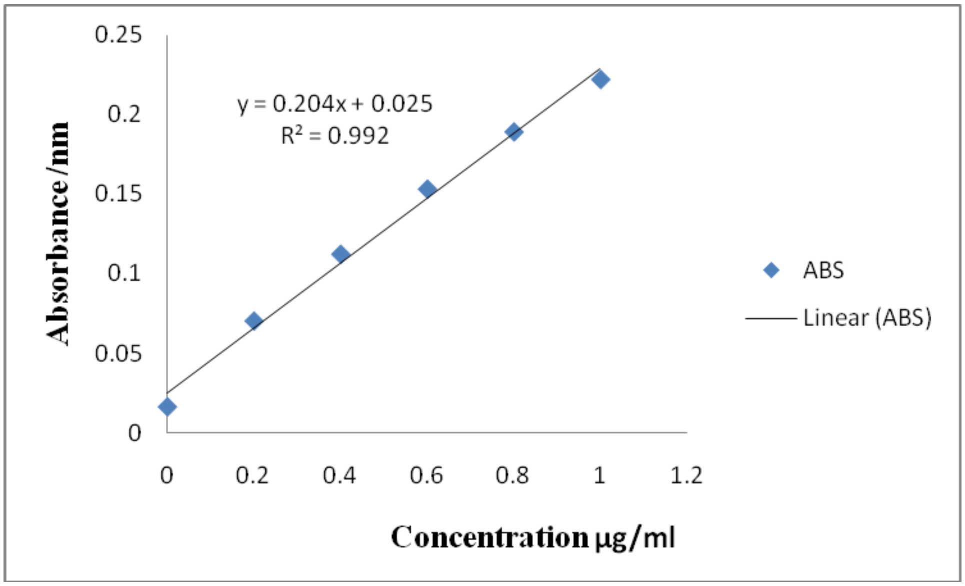
Source of variation	Degree of freedom	Mean square values					
		Appearance	Aroma	Taste	Mouth feel	consistency	Overall acceptability
Panelist	49	1.497^{*}	1.64^{**}	1.28^{ns}	1.52^{ns}	1.16^{ns}	1.52^{ns}
Treatment combination	8	51.34^{***}	36.89^{***}	35.86^{***}	33.56^{***}	53.97^{***}	18.41^{**}
Error	392	0.99	1.03	1.22	1.33	1.30	1.18
CV (%)		29.43	29.25	32.86	35.30	34.05	31.08

*, **, ***, *significant, highly significant and very highly significant respectively; ns=non significant at P=0.05*

b) List of figure in appendices



Appendix Figure 1: Calibration curve of phytate



Appendix Figure 2: Calibration curve of Tannin

C) Sensory evaluation questioner 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 4. Like

2. Dislike 5. Like Very Much

3. Neither Like or Dislike

Sample code	Appearance(color)	Aroma	Taste	Mouth feel(Texture)	Consistency (Viscosity)	Overall acceptability

General Comments /suggestion about the product

