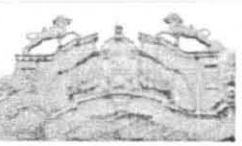


Addis Ababa  
University  
Since 1950



**Addis Ababa University**  
**College of Natural Science**  
**School of Graduate Studies**  
**Center for Food Science and Nutrition**  
**Studies on the Microbiological, Nutrient Composition and**  
**Antinutritional Contents of Fermented Maize/Sorghum/Teff**  
**Flour Blended with Soybean**

By  
**Betre Getahun**

A Thesis Submitted to the School of Graduate Studies of Addis Ababa University in Partial Fulfillment of the Requirements for the Degree of Master of Science in Food Science and Nutrition

Addis Ababa  
Ethiopia  
June, 2013

## Declaration

I, the undersigned, declare that this thesis is my original work and has not been presented for a degree in any other University, and that all sources of materials used for the thesis have been duly acknowledged.

Betre Getahun Mitiku

Signature: \_\_\_\_\_

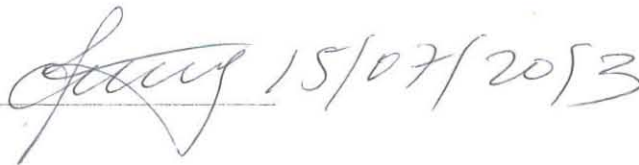
Addis Ababa, Ethiopia

Date of submission: \_\_\_\_\_

This thesis has been submitted for examination with our approval as University advisors.

Mr. Kelbessa Urga

Signature: \_\_\_\_\_

 15/07/2013

Mr. Tilahun Bekele

Signature: \_\_\_\_\_



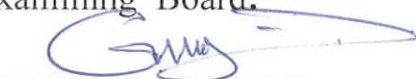
Addis Ababa University  
School of Graduate Studies  
Center for Food Science and Nutrition  
Studies on the Microbiological, Nutrient Composition and  
Antinutritional Contents of Fermented Maize/Sorghum/Teff  
Flour Blended with Soybean

A Thesis Submitted to the School of Graduate Studies of Addis Ababa  
University in Partial Fulfillment of the Requirements for the Degree of  
Master of Science in Food Science and Nutrition

By  
Betre Getahun

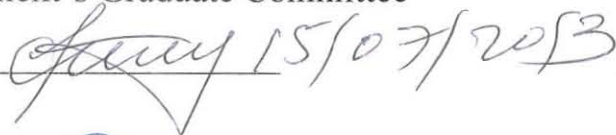
Approved by the Examining Board:

Dr. Gulelat Desse



Chairman, Department's Graduate Committee

Mr. Kebessa Urga



Advisor

Mr. Tilahun Bekele



Advisor

Dr. Getachew Addis



External Examiner

Dr. Kaleab Baye



Internal Examiner

## ACKNOWLEDGEMENT

First and foremost, I would like to thank God for the good health and strength given to me to reach this far.

I would like to express my deepest gratitude and thanks to my advisors, Mr. Kelbesa Urga and Mr. Tilhun Bekele who have extended their heartfelt advice and extraordinary suggestions beyond what I can inscribe on paper, it will remain engraved in my heart. I highly appreciate their friendly and endless encouragement and guidance until the completion of this work without which this manuscript could not have been realized.

I am deeply indebted to my friends Habitamu Addnew, Atsede Mequnint, Moges Abate with his wife Emawayish Abebaw, Yirgalem Fenta , Guta Olani, Melese Engidayehu and Yibeltal Getahun for their intensive and extensive assistance to my achievements.

I would like to extend my heartfelt thanks to Zewditu Getahun, Meseret W/yohanes, Yohans Tesfaye, Adamu Belay, Yosef beyene and Melkitu Kasaw for their kind technical, moral and material support.

Finally I would like to thank the Center for Food Science and Nutrition of Addis Ababa University and EHNRI for all rounded assistance, Amhara Agricultural Research Centers (AARC) for providing all the cereals and legumes and the School of Graduate Studies of Addis Ababa University deserve special thanks for partially financing this work.

## Table of Content

Title	Page no
Acknowledgement	i
Table of contents	ii
List of Tables	vi
List of Figures	vii
List of Abbreviations	ix
Abstract	x
1. Introduction	1
1.1. Background	1
1.2. Statement of the problem	2
1.3. Hypothesis of the thesis	3
1.4. Objective	3
2. Literature Review	5
2.1. Maize	5
2.1.1 Origin and history	5
2.1.2 Agronomic practice and economic importance of maize in Ethiopia	5
2.1.3 Nutritional composition	6
2.1.4 Antinutritional content	7
2.1.5 Uses	8
2.2. Sorghum	8
2.2.1 Origin and history	8
2.2.2 Agronomic practice and economic importance of Sorghum in Ethiopia	9
2.2.3 Nutritional composition	10
2.2.5 Antinutritional content	10
2.2.5 Uses	10

2.3. Teff	11
2.3.1 Origin and history	11
2.3.2 Nutritional composition	12
2.3.3. Uses	13
2.4 Legumes	14
2.4.1 Soybean	16
2.4.2 Proximate Composition and anti-nutrients in soybean	16
3 Fermentation	17
3.1. Microflora in fermented foods	17
3.2. Nutritional value of fermented foods	18
3.3. Health effects of fermented foods	21
3.4 . Effect of antinutritional on bioavailability of nutrients	22
4. Complementary Food	23
5 Materials and methods	25
5.1 Raw Material Collection and Transportation	25
5.2. Preparation of Maize Flour	25
5.3. Preparation of Sorghum Flour	25
5.4 Preparation of Teff Flour	26
5.5 Preparation of Soybean Flour	26
5.6 Blending	26
5.7 Fermentation	26
5.8. Microbial Analysis	29
5.8.1 Total Lactic Bacteria Counts	29
5.8.2 Total Bacteria Counts	29
5.9. Physicochemical and Functional Properties	29

5.9.1 Bulk Density (BD)	29
5.9.2 Water Absorption Capacity (WAC)	30
5.9.3 Oil-Holding Capacity (OHC)	30
5.9.4 Titratable Acidity (TA)	31
5.9.5 pH	31
5.10 Proximate Chemical Analysis	31
5.10.1 Determination of Crude Protein	31
5.10.2 Determination of Crude Fat	32
5.10.3 Determination of Crude Fiber	32
5.10.4 Determination of Moisture Content	33
5.10.5 Determination of Total Ash	33
5.10.6 Determination of Total Carbohydrate	33
5.10.7 Energy Value Calculation (Calorific Value)	34
5.10.8 Mineral analysis	34
5.11 Analysis of antinutrients	35
6.11.1 Phytic acid analysis	35
6.11.2 Tannins analysis	35
5.12 Sensory Evaluation of Complementary Gruels	36
5.13 Structure of the thesis experiment	36
5.14 Statistical analysis	37
6. Result and discussion	38
6.1 Moisture	38
6.2 Crude protein	38
6.3 Crude fat	41
6.4 Crude Fiber	42
6.5 Total ash	43

6.6	Total carbohydrates	43
6.7	Calorific value	44
6.8	Antinutritional content of blended and nonblended flours before and after fermentation	45
6.8.1	Tannins	45
6.8.2	Phytate	47
6.9	Micronutrient content of blended and non blended flour before and after fermentation	48
6.9.1	Iron	48
6.9.2	Zinc	49
6.9.3	Calcium	50
6.10	Effect of fermentation on physicochemical properties of blended and non blended flour	52
6.10.1	pH	52
6.10.2	Titrateable acidity	52
6.11	Impact of fermentation on functional properties	54
6.11.1.	Bulk density	54
6.11.2.	Water holding capacity	55
6.11.3.	Oil Absorption Capacity	57
6.12	Effect of fermentation on microbiological quality of flours	58
6.13	Sensory evaluation of gruels prepared from blended and non blended flours before and after fermentation	61
6.13.1	Taste	61
6.13.2	Odor	62
6.13.3	Color	63

6.13.4 Overall acceptability	64
7 Conclusions and recommendation	65
7.1. Conclusions	65
7.2. Recommendation	66
8 Reference	67

## List of Tables

Table 1. Non destructive nutrient determination of maize using NIR method.....	8
Table 2. Nutrient content of major Ethiopian cereals per 100gm .....	14
Table3. Amino acid contents of teff compared with other cereals .....	14
Table 4. Crude protein content (g/kg) and amino acid content (g/kg) of legumes.....	16
Table 5. Typical composition of soybean products (%).....	1
Table 6. Effect of fermentation on food and potential health benefits .....	21
Table 7. Macronutrient composition of fermented and unfermented sorghum and sorghum - soybean flours .....	39
Table 8. Macronutrient composition of fermented and unfermented maize- soybean blends.....	40
Table9. Macronutrient composition of fermented and unfermented teff - soybean blends .....	41
Table 10. Antinutritional content of blended and nonblended sorghum flour before fermentation and after fermentation .....	46
Table 11. Antinutritional content of blended and nonblended maize flour before fermentation and after fermentation .....	47
Table 12. Antinutritional content of blended and nonblended teff flour before fermentation and after fermentation .....	48
Table 13. The mineral content of blended and non blended sorghum flours before and after fermentation.....	49
Table 14. The mineral content of blended and non blended teff flours before and after fermentation .....	50
Table 15. The mineral content of blended and non blended teff flours before and after fermentation.....	51

Table 16. pH and titrable acidity for the blended and nonblended sorghum dough during the 3 days of fermentation .....	52
Table 17. pH and titrable acidity for the blended and nonblended maize dough during the 3 days of fermentation.....	53
Table 18. pH and titrable acidity for the blended and nonblended teff dough during the 3 days of fermentation.....	53
Table 19. Bulk density, water holding capacity and oil holding capacity of blended non blended sorghum flour before and after fermentation.....	55
Table 20. Bulk density, water holding capacity and oil holding capacity of blended and non blended teff flour before and after fermentation .....	56
Table 21. Bulk density, water holding capacity and oil holding capacity of blended and non blended maize flour before and after fermentation .....	58
Table 22. Microbial counts for the blended and nonblended sorghum dough during the 3 days of fermentation .....	59
Table 23. Microbial counts for the blended and nonblended teff dough during the 3 days of fermentation .....	60
Table 24. Microbial counts for the blended and nonblended maize dough during the 3 days of fermentation .....	60
Table 25. Sensory evaluation of blended and non blended sorghum before and after fermentation.....	61
Table 26. Sensory evaluation of blended and non blended maize before and after fermentation.....	62
Table 27. Sensory evaluation of blended and non blended teff before and after fermentation.....	63

## List of Figures

Figure 1. Annual production of maize in Ethiopia .....	8
Figure 2. Process flow diagram for the production of soy flour.....	27
Figure 3. Process flow diagram for the production of blended flours.....	28
Figure 4. Structure of the thesis .....	37

## List of Abbreviations

AOA C	Association of Official Analytical Chemists
AARC	Amhara Agricultural Research Centers
BD	Bulk Density
Cfu	Colony forming unit
HPC	High Performance Chromatography
GDP	Gross Domestic Product
FAO	Food and Agricultural Organization
LAB	Lactic Acid Bacteria
PER	Protein efficiency ratio
RNV	Relative Nutritional Value
TA	Titrateable Acidity
TCA	Trichloro- acetic Acid
WAC	Water Absorption Capacity
WFP	World Food Program
WHO	World Health Organization

## ABSTRACT

complementary foods prepared commercially are not available and if available, unaffordable for the poor in developing countries. Thus, there is need of preparing weaning foods from locally available raw materials such as cereal and legumes using simple technology. The objective of this paper is to study the effect of fermentation on cereals-soybean blends with respect to the nutritional quality; antinutrient, proximate & micronutrient compositions, functional & physico-chemical properties, microbiological & sensory analysis. Seed varieties of maize *QPM*, sorghum(*teshale*), teff (*Etsub*) & soybean (*Ethio-Yigozlabiya*) were collected from Amhara Agricultural Research Centers (AARC). After sample preparation, the cereal flours were blended with soybean and fermented naturally for 72 hours. The proximate analysis results before & after fermentation in (%) were 17.84, 18.6 in the case of sorghum-soybean, 16.33 and 17.35 in the case of maize-soybean and 16.5 and 19.0143 in the case of teff soybean for crude protein; 12.32 and 17.08 for sorghum-soy, 13.12 and 14.16 for maize-soy and 12.28 and 14.56 for teff-soy flours for crude fat. The total carbohydrate of blended flours before and after fermentation were 66.72 and 63.34, 70.80 and 66.48 and 68.87 and 63.60 for sorghum-soy, maize-soy and teff -soy flours respectively. Significant differences ( $P < 0.05$ ) were observed between unfermented & fermented blends in proximate compositions, with specific increment of 4.64, 6.25 and 15.21 % for sorghum-soy, maize soy and teff-soy flours for protein. Fermentation significantly ( $p < 0.05$ ) decrease antinutrient content. Micronutrient increment in(mg/100g) for Fe, Zn and Ca were 4.08 to 5.27, 2.53 to 2.96 for sorghum-soy and teff-soy flour respectively. Fermentation significantly ( $p < 0.05$ ) decreased the antinutrients which resulted in a significant( $p < 0.05$ ) increase in micronutrients. Fermentation had a significant ( $p < 0.05$ ) decreasing effect on pH. Microbiological result showed significant ( $p < 0.05$ ) reduction coliform count of Total bacterial count & increment of LAB with increasing fermentation time. Gruel prepared from the fermented blended flours were accepted with higher scores. According to the result of the study, fermentation comparably reduced antinutrients and improved the nutritional quality of the complementary foods. Natural fermentation, which is inexpensive processing method that consumers especially low & medium income families can easily afford good quality product

*Key words: Antinutrient, fermentation, micronutrient, natural fermentation, proximate composition, sorghum ,maize, teff, soybean, Weaning food.*

## 1. INTRODUCTION

### 1.1. Background

Cereals are the only sources of nutrition for one-third of the world's population especially in developing and underdeveloped nations of Sub-Saharan Africa and South-east Asia. The major cereals which constitute over 85 % of total global cereal production are rice, wheat, maize, barley and sorghum amounting to about 200 million tones of harvest annually at an average of 10% protein content, out of which a sizeable proportion goes into human consumption (Sofi *et al.*, 2009).

In Ethiopia, cereals make up 85% and 90% of the total cultivated area and total production of field crops respectively and for over 90% of modern input consumption (CSA,2000). This includes teff, barley, maize, sorghum, oats, millet and wheat. According to World Bank (2007), cereal production accounted for 30 percent of the national income with large share in rural employment (60%) and households' total food expenditure (40%).

Maize (*Zea mays* L.) is an important cereal crop in Africa serving as source of food and industrial raw material for industries such as brewery, confectionary, livestock and flour feed mills. Maize is also known to be primary provider of calories supplying 20% of the world's food calories. It also provides 15% of all food crop protein. The poor nutritive value of maize grains is due to low contents of lysine and tryptophan in the maize protein component in addition to its antinutrient content (Olakojo *et al.*, 2007).

Sorghum (*Sorghum bicolor* L. Moench) is an important staple food in many parts of Africa and Asia. It is a major source of protein, carbohydrate and calorie in the diets of large segment of population; however bioavailability is low inherently due to the presence of many antinutritional factors such as, tannins, phytic acid and proteinase inhibitors (Al Mamary *et al.*, 2002).

The cereal grain teff (*Eragrostis tef* (Zucc.) Trotter) which is endemic to Ethiopia where it is believed to have originated. Teff provides over two-thirds of the human nutrition in

Ethiopia ( Urga *et al.*, 1997 ), with a grain protein content (10-12%) similar to other cereals. Besides providing protein and calories, teff is a good source of minerals, particularly iron. It has a very high calcium content and contains high levels of phosphorus, copper, aluminum, barium and thiamine. The principal use of teff grain for human food is the Ethiopian bread “injera”, a soft porous thin pancake with a sour taste (Yigzaw *et al.*, 2001) .

Fermentation is the oldest known form of food biotechnology and it has been used for several thousands of years as an effective and low cost means to preserve the quality and safety of foods. Animal and plant tissues are subjected to the action of microorganisms and/or enzymes to give desirable biochemical changes (Sahana *et al.*, 2003). Similarly, Paredes-López *et al.* (1988) described that fermentation increases the soluble fractions of a food. The quantity as well as quality of the food proteins as expressed by biological value, and often the content of water soluble vitamins is generally increased, while the antinutritional factors show a decline during fermentation. Fermentation results in a lower proportion of dry matter in the food and the concentrations of vitamins, minerals and protein appear to increase when measured on a dry weight basis (Adams, 1990).

## **1.2. Statement of the problem**

The limited nutritional quality of cereal grains is due to their lower contents of essential amino acids, fats, minerals, and vitamins compared with animal foods. Complementary foods should contain animal sources with high biological value to foster growth and development. However, imported or commercially developed weaning foods generally are not used by low-income urban and rural households due to their high cost. They are mostly manufactured using high technologies and are sold in sophisticated packaging. Therefore, there is a need for low-cost weaning foods which can be prepared easily at home from locally available raw materials such as maize, teff, sorghum and soybean using simple technologies like fermentation that does not require complicated equipment and can be served quickly and conveniently.

In order to improve the quality of weaning diets, cereals should be blended with soyabean. Antinutritional factors in the soyabean are responsible for lowering the bioavailability of micronutrients. It is also an important fact that the amino acid composition of soyabean and the cereals ( sorghum, maize and teff) which exhibit deficiencies of the essential amino acids for the human diet, are complementary. For example, the cereals grains tend to be deficient in lysine where as soyabean is rich in the amino acid, so blending of the cereals with soyabean makes a nutritionally better complementary food than does alone. Cereals and legumes have a high nutrient content but bioavailability of some minerals are low due to the presence of antinutritional factors. Fermentation is a potentially important processing method that improves the bioavailability of minerals , thus fermentation reduces these antinutrient levels and enhance protein and carbohydrate digestibility.

### **1.3. Hypothesis**

Fermentation is one of the processes that decreases the level of antinutrients in food grains and increases the starch, protein digestibility and nutritive value. Thus, it might be possible to prepare complementary foods by blending sorghum, maize, teff and soyabean using fermentation.

## 1.4. Objectives

### General objective

The general objective of this research was to study the effect of fermentation on the microbiological, nutritional composition and anti-nutrient contents of fermented maize, sorghum and teff flour blended with soybean.

### Specific objectives

The specific objectives of the research were to:

- ✓ Evaluate how fermentation of cereals-legume blends affect proximate composition
- ✓ To study the microbiological profile of the fermented blended and non blended flours.
- ✓ To evaluate the effect of fermentation of cereals-legume blends.
- ✓ To determine physico-chemical & functional properties of fermented cereals-legume blends .
- ✓ To study the sensory acceptability of gruels prepared fermented cereals-legume blends.

## 2. LITERATURE REVIEW

### 2.1. Maize

#### 2.1.1. Origin and History

Maize (*Zea mays* L.) originated in Central America and was introduced to West Africa in the early 1500s by the Portuguese traders. The United States is the largest producer, accounting for nearly 40% of the total world production, followed by China and Brazil. It is grown on more than 96.5 million hectares in the developing world and many millions of people worldwide are dependent on maize as a staple food. Maize accounts for 15 to 56% of the total daily calories of people in about 25 developing countries (Prasanna *et al.*, 2001). In Africa, maize supplies at least one fifth of total daily calories consumed and accounts for 17% to 60% of people's total daily protein supply in 12 countries, as estimated by FAO food balance sheets (Krivanek *et al.*, 2007).

Maize was introduced to Ethiopia during the 1600s to 1700s and its production has increased over the years. In the 1980s, the total production within a year remained below 20 million quintals and maize production area exceeded slightly one million hectare only in 1987, 1988 and 1989. However, in the 1990s, maize production in Ethiopia increased: the total area and production remained over 1.3 million hectare and 23.4 million quintals from 1996-2000, respectively. The yield per hectare also increased slightly in the late 1990s. From 1995- 2000, growth rate per year for yield per hectare, maize area and total production was 3.1%, 7.1% and 11.3%, respectively ( Kebede *et al.* 1993).

#### 2.1.2. Agronomic Practice and Economic Importance of Maize in Ethiopia

In Ethiopia, maize grows from moisture stress areas to high rainfall areas and from lowlands to the highlands (Kebede *et al.*, 1993). It is cultivated mainly in the highland temperate mixed farming system, with some production in the cereal root crop mixed system of western Amhara. The majority of Ethiopia's maize comes from three regions: Oromia (61%), Amhara (20%) and SNNPR (12%) (Schneider *et al.*, 2010).

# Food Science scan

## Part 2: Planting

all other cereal crops, though it is  
g its importance in terms of wide  
one of the high priority crops to  
*t al.*, 2001). According to FAO  
and African average yields, but  
tween 1993 and 2008, Ethiopia  
ception in 2006 when it yielded  
ained well over the East African  
achieving as much as 186% in

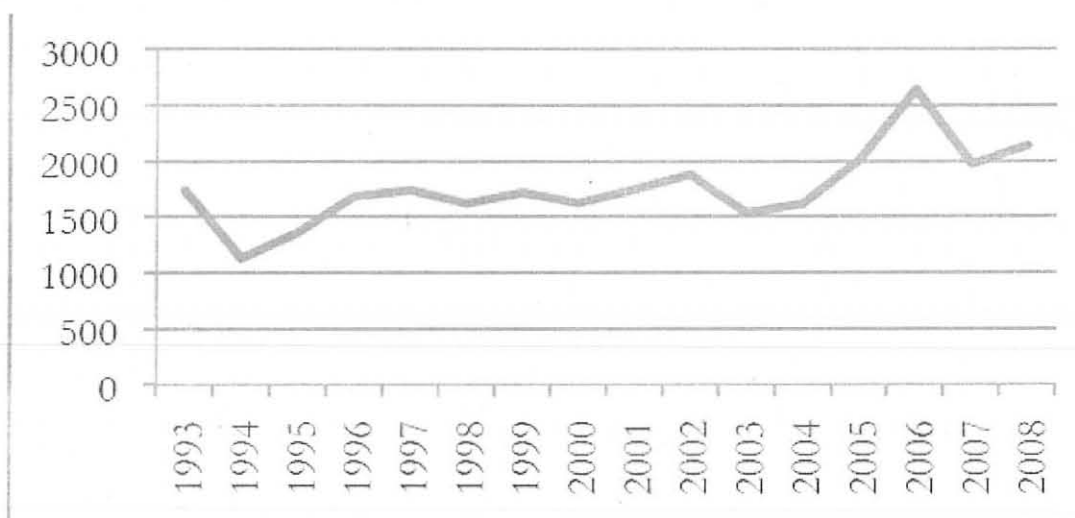
a long rainy season from May to  
the areas a small amount is produced in the short rainy period from  
February to May. Farmers in the western region also plant maize on low lands using  
residual moisture in January and harvest in June/July. This mainly solves the food  
shortage in the main season (Girma *et al.*, 2001). Moreover, maize plays a central role in  
Ethiopia's food security. It is the lowest cost source of cereal calories, providing 1½  
times and two times the calories per dollar compared to wheat and teff, respectively. An  
effective maize sector could propel Ethiopia's food production to quickly reduce the  
national food deficient and keep pace with growing population.

### 2.1.3. Nutritional Composition

Maize is a major cereal crop for human nutrition, worldwide. With its high content of  
carbohydrates, fats, proteins, some of the important vitamins and minerals, maize  
acquired a well-deserved reputation as a nutritive-cereal. Several million people,  
particularly in the developing countries, derive their protein and calorie requirements  
from maize (FAO, 2005).

Table 1 summarizes the results of Non-destructive Nutrient Determination of Maize  
Using NIR Method. Carbohydrate was the major component of maize, followed by water,  
protein, and fat. The major component of amino acids was lysine (Budiastira *et al.*, 2006).

The variation of carbohydrate, water and fat content were high, while the variation of amino acids content was low .The mineral composition of grains of different maize varieties is reported for Na (540.30-620.41 ppm), K (2915-3471 ppm), Ca (410-590 ppm), Fe (38.02-56.14 ppm), Zn (37.05-52.4 ppm), Mg (985.2-1125.3 ppm) and Cu (11.02-14.25 ppm) (Ullah1 *et al.*, 2010).



Source; FAOSTAT, 2000

Figure 1; Annual production of maize from 1993-2008

#### 2.1.4. Anti Nutritional content

Phytate content (9.8 mg/g) of raw yellow maize was found to be greater than in Malawian white maize flour (7.1 mg/g) and pounded white maize flour (6.98 mg/g). On the other hand, phytate content in fermented yellow maize was 3.8 mg/g compared well with 3.06 mg/g reported by Hotz and Gibson (2001). Trypsin and  $\alpha$ -amylase inhibitors dropped by 42 and 17 % respectively after fermentation. Trypsin inhibitor is a small protein, which inhibits the digestive enzyme trypsin. Hoffmann *et al.* (2003), reported the inactivation and proteolysis of trypsin inhibitor protein by *in vitro* microbial fermentation of soybean in rumen fluid.

### 2.1.5. Uses

Maize provides nutrients for humans and animals and serves as a basic raw material for the production of starch, oil, protein, alcoholic beverages, food sweeteners, and more recently, fuel. The green plant, made into silage, has been used with much success in the dairy and beef industries. After the grain is harvested, the dried leaves and upper part, including the flowers, are used to provide relatively good forage for ruminant animals owned by many small farmers in developing countries. The erect stalks, which in some cultivars are strong, have been used as long-lasting fences and walls. The husks are also used to make various craft items.

Table 1: Nutrient composition of maize

Composition	Mean (%)	Standard of Deviation (%)	Maximum (%)	Minimum (%)
Protein	9.04	0.79	10.33	7.27
Fat	4.62	1.34	8.23	2.79
Water	8.16	2.13	12.65	3.95
Carbohydrate	76.95	2.51	82.45	72.65
Methionine	0.18	0.038	0.296	0.110
Lysine	0.13	0.028	0.193	0.087
Tyrosine	0.29	0.042	0.359	0.179
Threonine	0.25	0.034	0.332	0.163
Arginine	0.31	0.056	0.428	0.144
Leucine	0.54	0.079	0.692	0.387

Source; Non-destructive Nutrient Determination of Maize Using NIR Method (FAO 2009)

## 2.2. Sorghum

### 2.2.1. Origin and History

Sorghum (*Sorghum bicolor* (L.) Moench), a tropical plant belonging to the Poaceae family, is one of the most important cereal crops in the world (Anglani, 1998). It is generally, although not universally, considered to have first been domesticated in North Africa, possibly in Nile or Ethiopia regions as recently as 1000BC (Kimber, 2000). Doggett, (1988) suggested that sorghum was domesticated and originated in the northeast quadrant of Africa, most likely in the Ethiopian-Sudan border regions. The presence of

wild and cultivated sorghums in Ethiopia reveals that Ethiopia is the primary centre of origin and centre of diversity (Mekibeb, 2009).

The cultivation of sorghum played a crucial role in the spread of the Bantu (black) group of people across sub-Saharan Africa (Jordan *et al.*, 1982). Ethiopia, the primary centre of origin for sorghum, where the crop was domesticated (Vavilov, 1951) and it is characterized by a diversity of climate, physiography, soils, vegetation, farming systems and socio-economic conditions (Ayana, 2001). Today, sorghum is cultivated across the world in the warmer climatic areas. It is quantitatively the world's fifth largest most important cereal grain, after wheat, maize, rice and barely (FAO, 2009).

### **2.2.2. Agronomic Practice and Economic Importance of Sorghum in Ethiopia**

World annual sorghum production is over 60 million tones, of which Africa produces about 20 million tone (Jordan *et al.*, 1982). This makes sorghum, quantitatively the second most important cereal grain in Africa after maize. It can be seen that sorghum production takes place across the continent, with the northern African countries of Nigeria, Sudan, Ethiopia and Burkina Faso accounting for nearly 70% of Africa's production. Sorghum is one of the major cereal crops consumed in Ethiopia and the total consumption of sorghum closely follows the global pattern of output, since most of it is consumed in the countries where it is grown (FAO, 1995). It is one of the main staples for the poorest and most food insecure Ethiopian people .

Sorghum is crucially important to food security in Ethiopia as it is uniquely drought resistant among cereals and can withstand periods of high temperature. annual rainfall is in the range 500-700 mm per year. Hence, most of the countries in Africa where sorghum is a significant arable crop are arid and areas at risk of desertification (FAO, 2005). It has seen that sorghum is also an important crop in east Africa where overall there is good rainfall. This is related to the fact that the rain in sub-tropical Africa is intermittent and characterized by brief periods of very high rainfall. In fact sorghum is not only drought-resistant, it can also withstand periods of water-logging (FAO, 2005).

Sorghum is a crop dominated by resource-poor smallholders and typically produced under adverse conditions in the eastern and northwest parts of the country, where there is low rainfall. The good rainfall distribution during the *meher* main rainy season in 2011, coupled with additional area planted in northern part of the country by new settlers, led to an increase in production in 2011/12, considerably more than earlier forecast. In 2012/13, however, both planted area and resultant production are forecast to drop, due to the late start of the short *belg* rainy season, delaying land preparation.

### 2.2.3. Nutritional Composition

Sorghum is one of the cereals that constitute a major source of carbohydrate, proteins, calories, minerals for millions of people in Africa and Asia (Miller,1996). It contains ash carbohydrate (74.68%), Protein (12.25%), Fiber (1.71%), Fat (4.2 %), (1.75%), (Mohammed *et al.*, 2011). Like other grains, sorghum protein is generally low in the essential amino acids such as lysine and threonine (Murty *et al.*, 2001)

### 2.2.4 Anti nutritional Content

Sorghum is also rich in minerals content but its nutritional quality is dictated by its chemical composition and presence of considerable amount of antinutritional factors such as tannin, phytic acid, polyphenoles and trypsin inhibitors with bioavailability of less than 1% for some forms of iron to greater than 90% for sodium and potassium(El-Sheikh *et al.*, 2000). The reasons for this are varied and complex, since many factors interact to determine the ultimate bioavailability of a nutrient ( Miller, 1996). . Sorghum is found to contain many antinutritional factors such as, tannins, phytic acid, proteinase inhibitors and cyanogenic glycosides (Cossak *et al.*, 1983).

### 2.2.5. Uses

Sorghum in Africa is processed into a very wide variety of attractive and nutritious traditional foods, such as semi-leavened bread, couscous, dumplings and fermented and non-fermented porridges. It is the grain of choice for brewing traditional African beers. Sorghum is also the grain of 21<sup>st</sup> century Africa. New products such as instant soft

porridge and malt extracts are great successes. In the competitive environment of multinational enterprises, sorghum has been proven to be the best alternative to barley for lager beer brewing (FAO.2009). Sorghum accounts for an average ten percent of daily calorific intake of households living in the eastern and northwest areas of the country. About three-quarters of the sorghum grain in Ethiopia is used for making *injera* (FAO.2010). Another 20 percent is used for feed and for local beer production, with the remainder held for seed. The entire plant is utilized, with sorghum stalks used for house construction and cooking fuel, and leaves used for animal fodder (FAO.2005).

### 2.3. Teff

#### 2.3.1. Origin and History

A number of investigators have speculated on the origins of teff, using morphological, cytological, and/or biochemical characters and have suggested a total of 14 wild *Eragrostis* species as potential progenitors of the crop. Jones *et al.*, (1978) examined morphological and cytological aspects of 41 *Eragrostis* species and concluded that *E. pilosa* was most similar to teff but that *E. aethiopica* also bore striking similarities to the cultigen. Costanza *et al.*, (1979) examined the relationships among 36 accessions of teff, two *E. pilosa* accessions, and *E. aethiopica* using morphometric methods, and they concluded that *E. pilosa* was far more similar to teff than was *E. aethiopica*.

Teff (*Eragrostis teff* [Zucc.] Trotter) is an allotetraploid ( $2n = 4x = 40$ ) cereal crop grown primarily in Ethiopia. Ethiopia is the center of thought to be the origin and diversity of teff. It is entirely cultivated only in Ethiopia as food crop sometime between 4000 B.C. and 1000 B.C. and distributed to several other countries in the 19th century, and it is now cultivated as a forage grass in Australia, India, Kenya and South Africa (Costanza *et al.*, 197).

### 2.3.2. Nutritional Composition

The grain of teff is used to make a variety of food products, including *injera*, a spongy fermented flatbread that serves as the staple food for the majority of Ethiopians. Chemical composition analysis showed that teff has comparable nutritional content with the major crops: maize, barley, wheat and sorghum, cultivated in Ethiopia. Tables 2 and 3 present nutritional and amino acid contents of teff in comparison with other major Ethiopian cereal crops respectively Jansen *et al.*(1962).

Teff is especially rich in mineral nutrients such as calcium, phosphorus and iron compared to maize, barley, wheat and sorghum. There is also slight difference in nutrient content among the three teff types Nech (white), key (brown) and sergegna (mixed) . The carbohydrate content of teff is comparable with other cereal crops ranging from 73.1% (brown) to 75. 2% (mixed teff) with average value of 73.9%. Since teff is the major component of Ethiopian recipe, it provides the major requirement of energy. *Nech* (white seed color) teff has 11.1% protein content which exceeds its content for other major cereal crops. All the three teff types contain significantly higher mineral nutrients (Calcium, potassium and iron) compared to maize, sorghum, wheat and barley and also have reasonably high fiber (3.2%) and ash (2.9%) contents averaged over the three teff types. Amino acid composition of the three tef types was reported to be the same (Tadesse, 1975; Endeshaw,1989) regardless of their seed color.

### 2.3.3. Uses

Teff is used in various forms by Ethiopians. The dominant form of usage is *injera*. It is also consumed in the form of porridge and bread. Its straw is a nutritious and highly preferred feed for livestock compared to the straw of other cereals, particularly during dry season. Besides its local use, it is the major cash earning crop for the farming community as market price for both its grain and straw is higher compared to other cereal crops. It is also among the export commodity at national level.

Table 2: Nutrient content of major Ethiopian cereals per 100gm

Content item	Tef			Barley (whole)	Maize (whole)	Wheat (whole)	Sorghum (whole)
	Nech (white)	Key (Brown)	Sergegna (mixed)				
Food energy (cal.)	339	336	336	334	356	339	338
Moisture (%)	10.4	11.1	10.7	11.3	12.4	10.8	12.1
Protein (g)	11.1	10.5	7.2	9.3	8.3	10.3	7.1
Fat (g)	2.4	2.7	2.9	1.9	4.6	1.9	2.8
Carbo-hydrate (g)	73.6	73.1	75.2	75.4	73.4	71.9	76.5
Fibre (g)	3.0	3.1	3.6	3.7	2.2	3.0	2.3
Ash (g)	2.5	3.1	3.0	2.0	1.3	1.6	1.5
Calcium (mg)	156.0	157.0	140.0	47.0	6.0	49.0	30.0
Phosphorus (mg)	366.0	348.0	368.0	325.0	276.0	276.0	282.0
Iron (mg)	18.9	58.9	59.0	10.2	4.2	7.5	7.8

Source: Agren and Gibson (1967)

Table 3: The amino acid content of teff compared to other cereals

Amino acid	Tef	Barley	Maize	Rice	Sorghum	Wheat	Pearl millet	FAO pattern	Whole egg
Lysine	3.68	3.48	2.67	3.79	2.02	2.08	2.89	4.20	6.60
Laoleucine	4.00	3.58	3.68	3.81	3.92	3.68	3.09	4.20	7.50
Leucine	8.53	6.67	12.5	8.22	13.3	7.04	7.29	4.80	9.40
Valine	5.46	5.04	4.45	5.50	5.01	4.13	4.49	4.20	7.20
Phe-alanine	5.69	5.14	4.88	5.15	4.90	4.86	3.46	2.80	5.80
Trysosine	3.84	3.10	3.82	3.49	2.67	2.32	1.41	2.80	4.40
Tryptophan	1.30	1.54	0.70	1.25	1.22	1.07	1.62	1.40	1.40
Threonine	4.32	3.31	3.60	3.90	3.02	2.69	2.50	2.80	4.20
Histidine	3.21	2.11	2.72	2.50	2.14	2.08	2.08	-	2.10
Arginine	5.15	4.72	4.19	8.26	3.07	3.54	3.48	-	6.90
Methionine	4.06	1.66	1.92	2.32	1.39	1.46	31.35	2.20	3.80
Cystine	2.50						3.19	2.00	2.40

Source: Alemayehu (1990); Jansen et al. (1962)

## 2.4 . Legumes

Annual legumes are among of the most important crops on a global scale. In animal feeding, they can be used as green forage, forage dry matter, forage meal, silage, immature grain, mature grain and straw, while some species may be used for grazing too. The term grain legumes denotes exclusively annual crops cultivated for immature or mature grain, with a further division into food legumes, also known as edible legumes or pulses, used for human consumption, and feed or fodder legumes, used in animal feeding (Mihailović *et al.*, 2004). Unlike these, forage legumes are used in the form of forage and comprise both annual and perennial species, having an additional role as a source of biomass and green manure (Mihailović *et al.*, 2007).

Legumes are some of the low-priced sources of protein-rich that have been important in alleviating protein malnutrition, especially in developing countries Table 4. The compositional evaluation of leguminous seeds such as soybean, chickpea, cowpea, pigeon pea, mucuna bean, scarlet runner bean, groundnut, bambara groundnut, African yam bean and red grain have been carried out in different locations by many investigators (Aremu *et al.*, 2006).

Though legumes are important sources of dietary proteins for both human and animals, their acceptability and utilization have been limited due to the presence of relatively high concentration of some toxins referred to as antinutritional factors. Nutritional quality is affected by these factors; which interact with nutrients, such as phytate, and tannins reducing protein digestibility and amino acid absorption (Udensi *et al.*, 2005). Some researchers (Duhan *et al.*, 2000) have reported that unless these substances are destroyed by heat or some other treatments, they can exert adverse physiological effects when ingested by man and animals.

The productivity of agriculture is however low due to low level of use of improved technologies, risks associated with weather conditions, diseases and pests, and underdeveloped seed supply systems and output markets. Moreover, due to increasing population pressure, on the rainfed land area, the land holding per household is declining

over time, leading to low level of production to meet the consumption requirement of the households carbohydrates, vitamins and minerals. The growing demand in both the domestic and export markets provides a source of cash for smallholder producers.

Table 4: The nutritional composition of legumes

Species	Soybean meal	Pea	Faba bean	Common vetch	White lupin	Grass pea
Crude protein	533.0	267.0	274.0	313.0	388.0	290.0
Alanine	30.2	4.4	3.3	4.4	13.3	13.5
Arginine	40.2	16.7	13.2	6.6	37.1	22.4
Aspartic acid	76.0	32.2	33.1	25.4	48.0	34.5
Cystine				4.5	4.2	5.0
Glutamic acid	35.8	17.8	28.7	22.1	78.0	49.8
Glycine	23.5	13.3	11.0	9.9	16.8	
Histidine	12.3	10.0	7.7	16.6	9.1	8.4
Isoleucine	29.1	13.3	11.0	14.3	13.9	10.8
Leucine	44.7	17.7	18.7	21.0	28.3	18.3
Lysine	36.9	13.3	16.5	16.6	14.1	19.4
Methionine	7.8	4.4	3.3	4.4	2.9	2.5
Phenylalanine	31.3	16.7	14.3	14.3	15.5	12.1
Proline	8.9	6.7	4.4	4.4	16.3	
Serine	64.8	31.1	28.7	19.9	21.3	13.7
Threonine	17.9	6.7	7.7	12.1	13.6	11.1
Tryptophan				2.4		2.1
Tyrosine	20.1	8.9	8.8	9.9	15.9	8.1
Valine	33.5	18.9	15.4		14.4	

Source: Crude protein content (g kg<sup>-1</sup>) and amino acid content (g kg<sup>-1</sup>) in grain dry matter of some feed grain legumes (Mihailović *et al.*, 2007)

#### 2.4.1 Soybean (*Glycine max*)

Soybean (*Glycine max*) a grain legume, is one of the richest and cheapest sources of plant protein that can be used to improve the diets of millions of people, especially the poor and low income earners in developing countries because of its nutritional quality, attractiveness and functional properties (Iwe, 2003). Soybean is mainly cultivated for its seeds which are used commercially as human food, livestock feed and for the extraction of oil.

#### 2.4.2 Proximate Composition and Antinutrients in Soybean

Nutritionally, soybean protein resembles animal protein more closely than other vegetable proteins, oilseeds and legumes. Soybean protein constitutes about 40% of the total solids and plays a very important role in the enrichment of cereal-based food products (Fukushima, 1999). It is also a rich source of vitamin, minerals and is relatively low in crude fibre (Duke, 1981). Soybean is one such protein sources, which when used partially to replace or complement corn starch in the preparation of custard would help tremendously in improving the nutritional quality of the product. Soyabeans are valued for their high protein content which varies between 38% and 42% and for their oil which is mainly used for cooking purposes (Kent, 1985).

The average proximate composition of soyabean products is presented in Table 5. Solvent extracted soybean meal is made usually by hexane extraction. Certain times alcohol extraction is also used. The value of fiber given, is for extraction of non dehulled seeds. Dehulling of seeds prior to extraction reduces the fiber content to about 50% and antinutrients (Tacon, 1990).

The nutritional quality of soya bean grain depends on many factors. Mature soya bean grain contains a number of anti-nutritional components with various level of biological activity. The most important among them are protease inhibitors. Their presence prohibits the utilization of raw soya bean as food and feed and requires heat treatment or fermentation in order to reduce or fully inactivated the antinutrients. The improvements in decreasing or eliminating trypsin inhibitors lead to enhancement of nutritional quality. The other important anti-nutrient present in soya bean grain is phytate. It is not digested by monogastric animals, so it does not provide monogastrics with sufficient phosphorus and minerals and it leads to phosphorus runoff causing phosphorus pollution of ground water from animal wastes (Bilyeu *et al.*, 2008).

Table 5: Typical composition of soybean products (%)

	Moisture	Protein	Lipids	Fibre	Ash
Solvent extracted soybean meal	11	45	1.2	6.1	6.1
Full fat soybean meal	10.0	38.0	18.0	5.0	4.1
Soya protein concentrate	8	84	0.5	0.1	3.5

Source: Tacon, (1990)

### 3. Fermentation

Campbell-Platt (1987) has defined fermented foods as those foods which have been subjected to the action of micro-organisms or enzymes so that desirable biochemical changes cause a significant modification to the food. However, to the microbiologist, the term "fermentation" describes a form of energy-yielding microbial metabolism in which an organic substrate, usually a carbohydrate, is incompletely oxidized, and an organic carbohydrate acts as the electron acceptor (Adams, 1990).

#### 3.1 Microflora in Fermented Foods

By tradition, lactic acid bacteria (LAB) are the most commonly used microorganisms for preservation of foods. Their importance is associated mainly with their safe metabolic activity while growing in foods utilizing available sugar for the production of organic acids and other metabolites. Their common occurrence in foods and feeds coupled with their long-lived use contributes to their natural acceptance as GRAS (Generally Recognised As Safe) for human consumption (Aguirre & Collins, 1993). In Europe, mould-ripened foods are primarily cheeses and meats, usually using a *Penicillium* species (Leistner, 1990).

Odunfa & Komolafe (1989), studied the micro-organisms present in fermented food made in Ghana called dawadawa. According to their study, after 24 h of fermentation, predominantly were *Bacillus* sp. with small numbers of (0,3%) *Staphylococcus* sp., after 36 h 60% *Bacillus* sp., 34% *Staphylococcus* sp. and after 48 h 56% *Bacillus* sp. and 42%

*Staphylococcus* sp. In Trahanas, a fermented food prepared in Greece from a mixture of milk and wheat flour, *Streptococcus lactis*, *Streptococcus diacetylactis*, *Leuconostoc cremoris*, *Lactobacillus lactis*, *Lactobacillus casei*, *Lactobacillus bulgaricus* and *Lactobacillus acidophilus* were found to play the major role in producing acid and aroma (Lazos *et al.*, 1993).

### 3.2. Nutritional Value of Fermented Foods

Generally, a significant increase in the soluble fraction of a food is observed during fermentation. Table 6 shows the importance of fermentation. The quantity as well as quality of the food proteins as expressed by biological value, and often the content of water soluble vitamins is generally increased, while the Antinutritional factors show a decline during fermentation (Paredes-López & Harry, 1988). Fermentation results in a lower proportion of dry matter in the food and the concentrations of vitamins, minerals and protein appear to increase when measured on a dry weight basis (Adams, 1990). Combination of cooking and fermentation improved the nutrient quality of all tested sorghum seeds and reduced the content of antinutritional factors to a safe level in comparison with other methods of processing (Obizoba & Atii, 1991). In bambara nut milk tannin content could be reduced by fermentation (Obizoba & Egbuna, 1992).

A study on the effect of fermentation of cowpea (*Vigna unguiculata*) on the nutritional quality of cowpea meal showed that 72h fermentation increased the content of protein, ash and lipid levels while decreasing the levels of tannin and phytate (Nnam, 1995). Thiamin and riboflavin were reduced significantly during fermentation. A decrease in protein content was observed during the first 2 days of fermentation and thereafter the decrease was not significant (Gupta *et al.*, 1998). Vaishali *et al* (1997) who studied effect of natural fermentation on *in vitro* zinc bioavailability in cereal-legume mixtures found that fermentation increased the zinc solubility (2-28%) and the zinc uptake by intestinal segment (1-16%) to a significant level.

The protein efficiency ratio (PER) of wheat was found to increase on fermentation, partly due to the increase in availability of lysine. A mixture of wheat and soybeans in equal amounts would provide an improved pattern of amino acids. The fermentation process raised the PER value of the mixture to a level which was comparable to that of casein (Hesseltine & Wang, 1980). Fermentation may not increase the content of protein and amino acids unless ammonia or urea is added as a nitrogen source to the fermentation media (Reed, 1981). The relative nutritional value (RNV) of maize increased from 65% to 81% when it was germinated, and fermentation of the flour made of the germinated maize gave a further increase in RNV to 87% (Lay & Fields, 1981).

Fermented milk products in general showed an increase in folic acid content and a slight decrease in vitamin B12 while other B vitamins were affected only slightly (Alm, 1982) in comparison to raw milk. The levels of vitamin B12, riboflavin and folacin were increased by lactic acid fermentation of maize flour, while the level of pyridoxine was decreased (Murdock & Fields, 1984). Fermented whole onion plant retained 97% of vitamin A activity, while fermented egg plant only retained 34% of the vitamin A activity (Speck *et al.*, 1988). Kefir made from ten different kefir grain cultures showed significant (>20%) increase for pyridoxine, cobalamin, folic acid and biotin and reduction exceeding 20% for thiamine, riboflavin, nicotinic acid, and pantothenic acid depending on the culture used.

The mineral content is not affected by fermentation unless some salts are added to the product during fermentation or by leaching when the liquid portion is separated from the fermented food. Sometimes, when fermentation is carried out in metal containers, some minerals are solubilised by the fermented product, which may cause an increase in mineral content (Harland & Harland, 1980). Phytate content in locust bean seeds was lowered from 0.51 mg/g to 0.31 mg/g by fermentation (Eka, 1980). Natural lactic fermentation of maize meal decreased phytate phosphorus by 78% (Chompreeda & Fields, 1984). The reduction of phytate content during dough fermentation for whole grain flour was about 50% (Roos *et al.*, 1990).

Fermentation by *Saccharomyces diastaticus* followed by *Lactobacillus brevis* completely eliminated phytic acid from pearl millet flour (Khetarpaul & Chauhan, 1991). In bambara nut milk tannin content could be reduced by fermentation (Obizoba & Egbuna, 1992).

Table 6; Effect of fermentation on food and potential health benefits

Effect on food	Potential health benefit
Break down of starch by amylases	Reduces bulk and increases energy intake
Reduction of phytic acid	Improved absorption of minerals and protein
Decrease in pH	Improved absorption of minerals 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)

### 3.3. Health Effects of Fermented Foods

One of the reasons for the increasing interest in fermented foods is its ability to promote the functions of the human digestive system in a number of positive ways. This particular contribution is called probiotic effect. Fermented milks in the diet contribute for the prevention of certain diseases of the gastrointestinal tract and promotion of healthy day to day life. A fermented food product or live microbial food supplement which has beneficial effects on the host by improving intestinal microbial balance is generally understood to have probiotic effect (Fuller, 1989).

During fermentation of beans for preparation of tempe, the trypsin inhibitor is inactivated, and the amount of several oligosaccharides which usually cause flatulence

are significantly reduced (Hesseltine, 1983). Bean flour inoculated with *Lactobacillus* and fermented with 20% moisture content, showed a reduction of the stachyose content (Duszkiewicz-Reinhard *et al.*, 1994).

In an *in vitro* study the ability of 23 strains of lactic acid bacteria isolated from various fermented milk products on the bacterial cells to bind cholesterol was investigated. No cholesterol was found inside the cells (Taranto *et al.*, 1997). Poppel and Schaafsma (1996) have also reported the ability of yoghurt to lower the cholesterol in serum by controlled human trials. Possible role of lactic acid bacteria in lowering cholesterol concentration. Apart from this, there are interesting data on anticarcinogenic effect of fermented foods showing potential role of lactobacilli in reducing or eliminating procarcinogens and carcinogens in the alimentary canal (Reddy *et al.*, 1983; Shahani, 1983; Mital & Garg, 1995).

Some lactic acid bacteria which are present in fermented milk products, are found to play an important role in the immune system of the host after colonization in the gut (De Simone, 1986). The mechanism of this effect is not clearly known, but it is speculated that the lactobacilli, their enzymes or the metabolic products present in the fermented food product may act as antigens, activating production of antibodies.

### **3. 4. Effect of Antinutrients on Bioavailability**

Anti-nutrients have been defined as substances which by themselves, or through their metabolic products arising in living systems, interfere with food utilization and affect the health and production of animals (Makkar, 1993). Tannins are members of the naturally occurring active nutrients known as polyphenols. Tannins are reported to interact with proteins (both enzymes and non-enzyme proteins) to form tannin-protein complexes resulting in inhibition of digestive enzymes (Al Mamary *et al.*, 2002). Phytic acid is found in plant seeds and many roots and tubers. Phytic acid is found to form complex with minerals in physiological pH, leading to lower mineral bioavailability (Cossak *et al.*, 1983). In addition phytic acid has been shown to inhibit trypsin (Singh *et al.*, 1982).

Trypsin inhibitors are widely spread in plant kingdom, especially in legumes and cereals. They inhibit proteolytic enzymes in digestive system (Liener *et al.*, 1980). and cause pancreatic hypertrophy and poor growth performance (Osman *et al.*, 2003). Several processing methods such as heat- treatment, soaking, germination, fermentation and radiation were found to reduce these antinutritional factor levels and enhance protein and carbohydrates digestibility (Elmaki *et al.*,1999). Sorghum, like legume and oil seed meals has some limitations, due to the presence of antinutritional factors, such as trypsin and amylase inhibitors, phytic acid, and tannins. These compounds are known to interfere with protein, carbohydrates and mineral metabolism (Elmaki *et al.*,1999).

#### **4. Complementary Food**

According to the WHO definition, Complementary foods are any food other than breast milk given to a breastfeeding child. A complementary food is normally a semi-solid food that is used in addition to breast milk and not only to replace it. complementary foods are mostly prepared in the form of thin porridges or gruels (Ikujenlola *et al.*, 2003). Development of complementary foods is guided by: 1) high nutritional value to supplement breastfeeding, 2) acceptability, 3) low price, and 4) use of local food items. During formulation of any complementary foods made from locally available raw materials, the techniques of food processing, storage and distribution; socioeconomic status; cultural and religious factors; sensory properties; and food quality and safety issues should be taken in to account ( Amuna *et al.*, 2000).

In developing countries, commonly-used complementary foods are prepared from flours of starchy staples, cereals and legumes, such as rice, millet, sorghum, or maize (rarely wheat). Cereals and legumes are the most common first complementary foods. Gruels/thin porridges prepared from cereals and legumes play an important role as a complementary food but their nutrient density is often low, especially deficient in essential nutrients. Cereals have low content of proteins and fat while legumes are high in Protein and fat. The presence of high concentration of crude fiber and absorption

inhibitors (antinutritional factors like phytic acid and condensed tannin) is major factors reducing their nutritional benefits (Wharton, 1989).

During cooking/reconstitution process of staple-based complementary foods, the starch granules swell and bind a large volume of water, resulting in gruels of high viscosity. Gruels of suitable feeding consistency contains a great amount of water and large in volume relative to its contents of solid matter. If solids in gruels are increased to improve the nutrient and energy density, the gruel will be too thick and viscous for a small child to eat easily. This high volume or high viscosity characteristics of complementary foods referred to as dietary bulk, is responsible for the occurrence of malnutrition in areas where cereals and starchy staples are the major foods ( Kulkarni *et al* 19991).

## 5. MATERIALS AND METHODS

### 5.1 Raw Material Collection and Transportation

Three cereals namely Sorghum, variety *Teshale*, Maize, variety BHQPY-545, Teff variety *Estub* and Soybean variety *Ethio-Yigozilabiya* were collected & transported from Amhara Agricultural Research Centers (AARC) during the 2012/13 crop season. Sample preparation was done in Addis Ababa University, Center for Food Science and Nutrition. Proximate composition, Physico-chemical, anti-nutritional and microbiological analyses were conducted at the Ethiopian Health & Nutrition Research Institute and Center for Food Science and Nutrition Addis Ababa University.

### 5.2. Preparation of Maize Flour

Preparation of maize flour was performed according to the method suggested by Ahima (2005). The maize was first sorted to remove defective grains, stones, soil, and other debris. The grains were then soaked for about 30 min in order to simplify the washing process. It was done using distilled water to get rid of foreign matters and then dried in the oven at a temperature of 60<sup>0</sup>C for 8h. Then, it was milled and sieved using 1mm sieve.

### 5.3. Preparation of Sorghum Flour

Sorghum flour was prepared according to the method described by Ihekoronye (1999). During preparation, two kilograms of sorghum grains, which were free from dirties, damaged and contaminated grains were weighed, cleaned and soaked in tap water for 18h. During soaking, the water was changed occasionally at intervals of 6h to prevent fermentation. Thereafter, the soaked grains were drained and dried on tray dryer (60<sup>0</sup>C) for 8h. After that, the dried grains were milled (attrition mill) and sieved through a 1mm mesh sieve. Sorghum flour produced was finally packaged in sealed polyethylene bags for blending and preparation of complementary food formulations.

#### **5.4 Preparation of Teff Flour**

Stones and other debris were removed from the Teff. Grains were milled (attrition mill) and sieved through a 1mm mesh sieve

#### **5.5 Preparation of Soybean Flour**

Defective grains (with holes), stones, dried pods and other debris were removed from the soybeans. The beans were then washed and soaked in distilled water 5:1v/w for 15 hour according to Assefa (2008). The soaked beans were placed in a sieve and allowed to drain. It was then lowered into a container containing boiled water for about 20 min (Ahima, 2005). This was done to make dehulling easier, and to inactivate enzymes' activities. The hulls were removed manually and washed repeatedly using distilled water. The dehulled beans were then dried using tray dryer (Biosec dryer) until the moisture content reached 11 - 13%. Then after, it was roasted using an electric oven for 8 min at a temperature of 120<sup>0</sup>C until it gets brown to further reduce anti-nutritive factors and improve the flavor of the final product. The roasted soybeans were milled into flour to obtain smooth and consistent particle sizes and sieved through 1mm as shown in figure 2.

#### **5.6 Blending**

The Soyabean flour was added to maize, teff and sorghum flour in the ratio 30:70 (w/w) (Bressani and Elias, 1974) as shown in Figure 3.

#### **5.7 Fermentation**

Natural fermentation was carried out by mixing each blended and non blended flours with distilled water (1:2 w/v). Two hundred gram blended and non blended flours were mixed with 400 ml distilled water in a 1000 ml beaker and were left at room temperature. Samples were withdrawn at periods of 0, 24, 48 and 72 h for microbiology analysis. After the end of fermentation each samples were mixed with a glass rod and transferred

to aluminum dishes, and dried using freeze drier. Dried samples were finely ground and stored in polyethylene bags at 4°C for subsequent analysis.

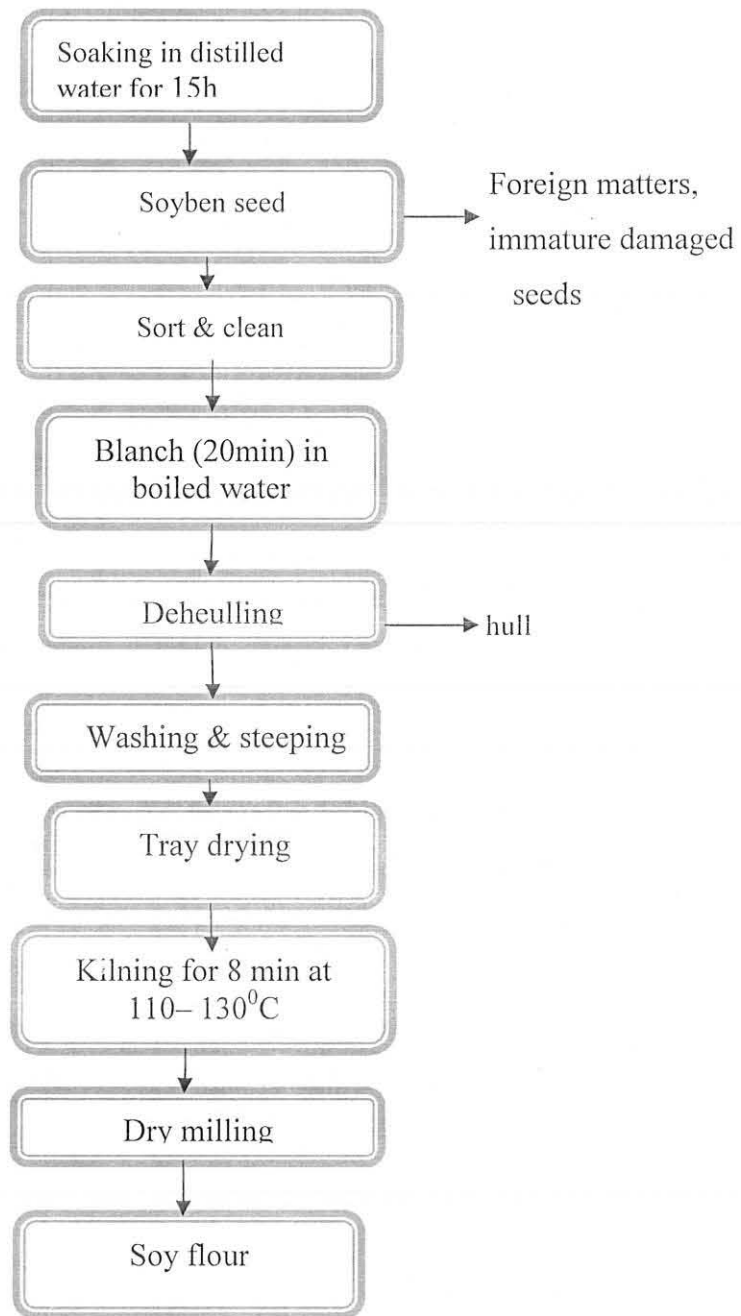


Figure 2. Process flow diagram for the production of soy flours

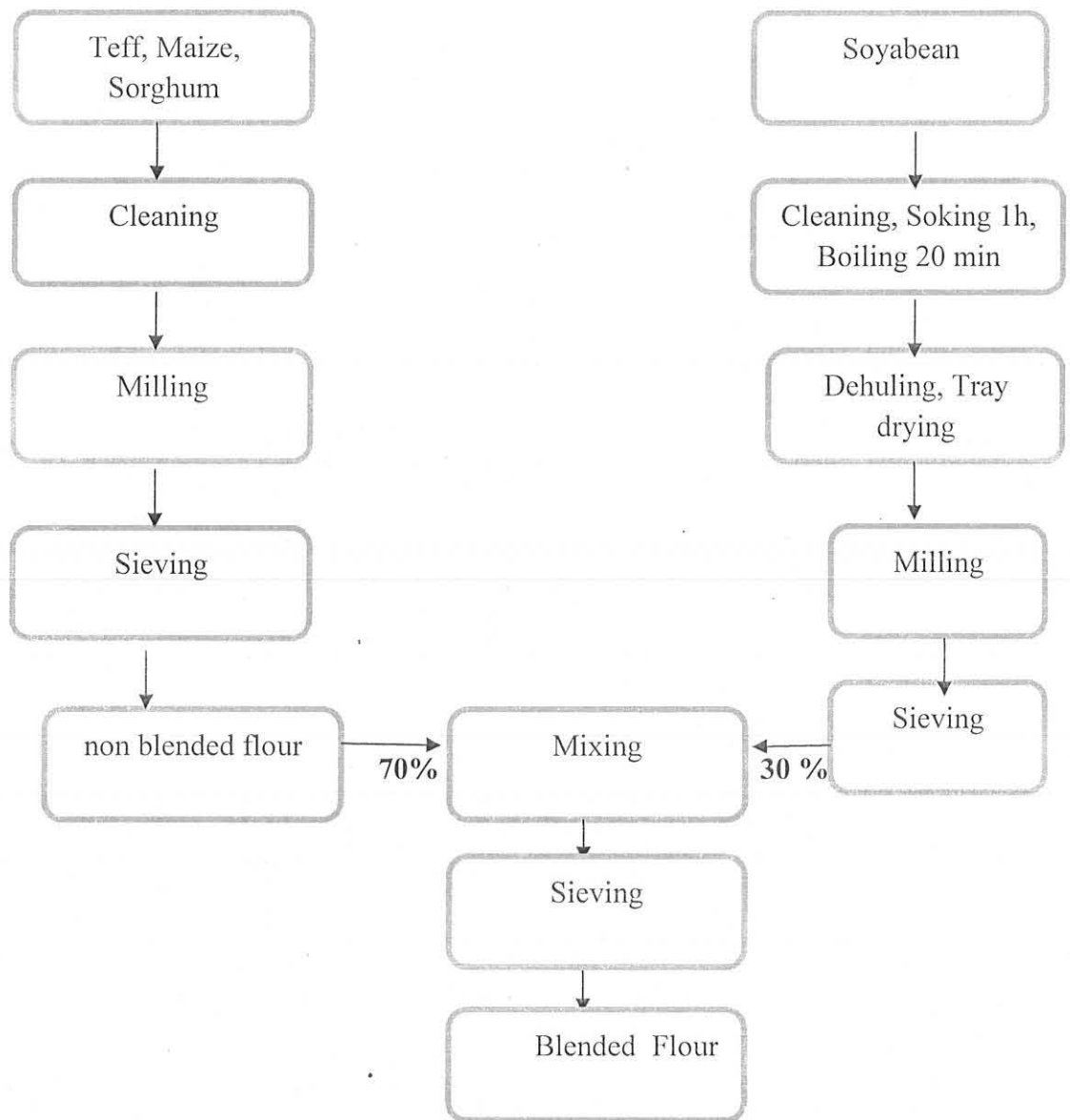


Figure 3. Process flow diagram for the production of blended flour.

## **5.8. Microbial Analysis**

Microbial counts were made on selective media after decimal dilution of the samples using the spread plate method as described below.

### **5.8.1 Total Lactic Acid Bacteria Counts**

At 24 h intervals (0, 24, 48, and 72 h) 1ml of fermenting meal was homogenized in 9.0 ml of sterile 0.1% saline water for 30s. The mixture was serially diluted in saline water Maynell and Meynell (1970). Colony-forming units (cfu) were determined from the 10 fold dilutions using the spread plate method. One ml of the appropriate dilutions was mixed with molten MRS agar (Oxoid), aseptically pour-plated in duplicates and incubated at  $37 \pm 2^\circ\text{C}$  for 48 h in anaerobic jars with Anaerogen (Oxoid, Basingstoke, Hampshire, England). Counts were expressed as colony forming units (cfu/ ml (Malleshi *et al.*, 1989).

### **5.8.2 Total Bacteria Counts**

At 24 h intervals (0, 24, 48, and 72 h), 1ml of fermenting meal was homogenized in 9.0 ml of sterile 0.1% saline water for 30s. The mixture was serially diluted in saline water by the method of Maynell and Meynell (1970) Colony-forming units (cfu) were determined from the 10 fold dilutions using the spread plate method. One ml of the appropriate dilutions was mixed with molten PCA agar (Oxoid), aseptically pour-plated in duplicates and incubated at  $42^\circ\text{C}$  for 48 h and counts were expressed as colony forming units (cfu) /ml (Malleshi *et al.*, 1989).

## **5.9. Physicochemical and functional properties**

### **5.9.1 Bulk density**

Bulk density was determined according to the method given by (Chau *et al.*,2003). A graduated cylinder (10 ml), previously weighed, and filled with sample to 10 ml by constant tapping, until there is no further change in volume. The content was weighed,

and from the difference in weight, the bulk density of sample was calculated as grams per milliliter.

$$\text{Bulk density} = \text{weight/volume}$$

### 5.9.2 Water Absorption Capacity (WAC)

WAC which gives an indication of the amount of water available for gelatinization was determined according to method used by Solsulski (1962). 1.0 g of each sample was added to 10 ml ( $V_1$ ) distilled water in a weighed 25 ml centrifuge tube. The tube was agitated for about 5 min before being centrifuged at 4000 rpm for 20 min and the volume of the supernatant was noted in a 10ml graduated cylinder ( $V_2$ ). The difference in volume was taken as the water absorbed by the sample ( $V_3$ ). The WAC was expressed as milliliters of water held per gram of sample.

$$\text{WAC} = (V_1 - V_3) / \text{weight of sample} * \text{density of water}$$

### 5.9.3 Oil-Holding Capacity (OHC)

Oil absorption capacity was determined according to the method of Beuchat (1977). 1g of the sample flour was measured and mixed with 10ml ( $V_1$ ) oil (soybean oil) in a 15ml centrifuge tube and stirred for 2 min. The samples were allowed to stand at room temperature for 30 min, centrifuged at 5000rpm using a centrifuge for 30 min, and the volume of the supernatant was noted in a 10ml graduated cylinder ( $V_2$ ). The difference in volume was taken as the oil absorbed by the sample ( $V_3$ ). Density of oil was taken as 0.895g/ml. The OHC was expressed as milliliters of vegetable oil held per gram of sample (Chau *et al.*, 2003).

$$\text{OHC} = (V_1 - V_3) / \text{weight of sample} * \text{density of oil}$$

#### 5.9.4 Titratable Acidity (TA)

Twenty grams of the fermenting samples were collected 24 interval for 72 h into sterile bottles and mixed with 100 ml of distilled water. The mixture was allowed to stand for 15 min, shaken at 5 min interval and filtered with whatman No. 4 filter paper and 10 ml were pipetted from the filtrate obtained above, into conical flask and then titrated against 0.1N NaOH to phenolphthalein end point. The percentage titratable acidity was calculated by multiplying the titre value by 0.09 (Vasconcelos *et al.*, 1990).

#### 5.9.5 pH

Twenty grams of the fermenting samples were collected 24 interval for 72 h into sterile bottles and mixed with 100 ml of distilled water. The mixture was allowed to stand for 15 min, shaken at 5 min interval and filtered with whatman No. 4 filter paper, the pH of the filtrate was measured using the pin electrode of pH meter (Vasconcelos *et al.*, 1990).

### 5.10. Proximate Chemical Analysis

#### 5.11.1 Determination of Crude Protein

Protein content was determined according to AOAC, (2000) using the official method 979.09. A digestion flask containing about 1 g of sample, to which 6 ml of acid mixture (conc. Sulphuric acid ) and about 3g of catalyst mixture (K<sub>2</sub>SO<sub>4</sub> and Selenium) was added and exposed to about 370 °C in order to allow digestion. Then, distillation took place by adding 25 ml of 40% NaOH and using 25 ml of boric acid with 10 drops of indicator solution. Finally, the distillate will be titrated with standardized 0.1N HCl to a reddish color. Then, crude protein content will be estimated using the formula:-

$$\text{Total nitrogen} = \frac{((V_2 - V_1) * N * 14.007 * 100)}{W}$$

Where, V<sub>2</sub> = Volume in ml of standard sulfuric acid solution used in the titration for the test material.

V<sub>1</sub> = Volume in ml of standard sulfuric acid solution used in the titration for the blank determination.

N = Normality of standard sulfuric acid.

W = Weight in grams of the test material.

N.B. Crude protein content percent per weight = total nitrogen \* 6.25 for maize, teff and sorghum and total nitrogen 5.71 for soybean flour and 5.98 for the blend.

### 5.10.2 Determination of Crude Fat

A clean and dried thimble containing about 2 g of dried sample and covered with fat free cotton at the bottom and top was placed in the extraction chamber. Then, extraction took place for at least 4h according to (AOAC, 2000) official method 450.1. The crude fat content was determined by the formula:-

$$\text{Weight of fat (Wf)} = \text{Wa} - \text{Wb}$$

Where:

Wa = Weight of extraction flask after extraction.

Wb = Weight of extraction flask before extraction.

$$\text{Crude fat content [g/100]} = (\text{Wf [100 - moisture, \%]} / \text{Wd})$$

Where:

Wd = Dried sample obtained after determination of moisture.

### 5.10.3 Determination of Crude Fiber

Crude fiber analysis was conducted using the method of AOAC, (2000) official method 962.09. About 1.5g weighed sample was transferred into a 600 ml beaker and about 200 ml 1.25% sulfuric acid will be added and boiled for 30 min. Recording was taken by placing a watch glass over the mouth of the beaker. After 30 min heating by gently keeping the level constant with distilled water, 20 ml of 28% KOH was added and again boiled gently for further 30 min. Subsequently, washing was conducted with 1% sulfuric acid, NaOH and acetone. Then, it was filtered and dried it in the electric oven at 130 °C for 2h. Furthermore, it was cooled at room temperature for 30 min in a desiccator and weighed, then it was transferred to a crucible muffle furnace for 30 min ashing at 550 °C. Finally, it was cooled again in a desiccators and re-weighed. The crude fiber content was determined by using the formula:-

$$\text{Crude fiber content [(g/100)]} = [(((w1-w2) * (100 - m)) / w3)]$$

Where,

w1 = Crucible weight after drying

w2 = Crucible weight after ashing

w3 = Dry weight

m= % moisture of the sample

#### 5.10.4 Determination of Moisture Content

Moisture of the flour was determined according to (AOAC, 2000) using the official method 925.09. A clean dried and covered flat aluminum dish was weighed and about 5g of the sample was transferred to the dish. The dish was placed in the oven (memmert 854 Schwabach, West Germany) at 105 °C for 5h and cooled in desiccators and re-weighed. Then, the moisture content was estimated by the formula:-

$$\text{Moisture content [\%]} = \left[ \frac{\text{((weight of fresh sample-weight of dry sample)/ (weight of fresh sample))}}{1} \right] * 100$$

#### 5.10.5 Determination of Total Ash

A dry porcelain dish containing about 2.5g sample was placed in a muffle furnace at 550 °C for 5h and allowed to cool in desiccators and weighted, the ash content was determined by (AOAC, 2000) using the official method 923.03 and applying a simple formula:-

$$\text{Total ash [\%]} = \left[ \frac{\text{((w2-w)/ (w1-w))}}{1} \right] * 100$$

Where:

w = Weight in grams of empty dish

w1 = Weight in grams of the dish plus the dried test material

w2 = Weight in grams of the dish plus ash

#### 5.10.6 Determination of Total Carbohydrate

Total carbohydrate content of the samples including crude fiber was determined by subtraction of the above tested parameters from 100%.

$$\text{Total carbohydrates [\%]} = 100 - [\% \text{Moisture} + \% \text{Protein} + \% \text{Fat} + \% \text{Ash}]$$

### 5.10.7 Energy Value Calculation (Calorific Value)

Energy value (calorific value) was quantified using an indirect calculation method. The three groups of nutrients, which provide the body with energy, are carbohydrates, fats and proteins. One gram of carbohydrate (C) was assumed to give 15.71KJ energy; one gram of fat (F) 37.71KJ energy and one gram of protein (P) 16.76KJ. The energy values for one gram of the three groups of nutrients which provides the body with energy was calculated by using specific values of Atwater factors for protein, fat, and total carbohydrate as recommended by Mahgoub (1999).

$$\text{Total energy (kcal/100g)} =$$

$$[(\% \text{ available carbohydrates} \times 4) + (\% \text{ protein} \times 4) + (\% \text{ fat} \times 5.13).$$

### 5.10.8 Mineral analysis

Zinc, Iron and calcium were determined using atomic absorption method of Osborne & Voogt (1978). The ash obtained after dry ashing at 550 °C was treated with 7 ml of 6N HCl to wet it completely and 15 ml of 3N HCl was added and the dish was heated on the hot plate until the solution just boils. Then, it was cooled and filtered. 10 ml of 3N HCl was added to the dish and heated until the solution just boils. Finally, it was cooled and filtered into the graduated flask. Using atomic absorption spectrophotometer (Varian, spectra-10/20, Australia) a calibration curve was prepared by plotting the absorption or emission values against the metal concentration in mg/100g. Reading was taken from the graph which depicted the metal concentrations that correspond to the absorption or emission values of the samples and the blank. The metal contents will be calculated by using the formula:

$$\text{Metal content [(mg/100g)]} = [(((A-B) \times V) / 10W)]$$

Where,

W = Weight of sample in (g)

V = Volume of extract (ml)

A = Concentration of sample solution (µg/ml)

B = Concentration of blank solution ( $\mu\text{g/ml}$ )

## 5.11 Analysis of antinutrients

### 5.11.1 Phytic acid analysis

About 0.1500 g of fresh samples was extracted with 10 ml 2.4% HCl in a mechanical shaker for 1 hour at a room temperature. The extract was centrifuged at 3000 rpm for 30 minute (Dynac II centrifuge, Clay Adams, Bacton, Dickinson and company, USA). The clear supernatant was used for phytate estimation. One ml of Wade reagent (containing 0.03% solution of  $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$  and 0.3% of sulfosalicylic acid in water) was added to 3 ml of the sample solution (supernatant) and the mixture was mixed on a Vortex for 5 seconds. The absorbance of the sample solutions were measured at 500 nm using UV-VIS spectrophotometer. A series of standard solutions were prepared containing 0, 5, 10, 20 and 40  $\mu\text{g/ml}$  of phytic acid (analytical grade sodium phytate) in 2.4% HCl. Three ml of the standard solution was added into 15ml of centrifuge tubes. Three ml of water was prepared to serve as standard blank. One ml of the Wade reagent was added to each test tube and the solution was mixed on a Vortex mixer for 5 seconds. The mixture was centrifuged for 10 minutes and the absorbance of the solutions (both the sample and standard) was measured at 500 nm by using deionized water as sample blank.

$$\text{Phytic acid in } \mu\text{g/g} = \left\{ \left[ \frac{(\text{absorbance-intercept})}{(\text{slope} \cdot \text{density} \cdot \text{weight of sample})} \right] \cdot 10 \right\} / 3$$

### 5.11.2 Tannins analysis

About 2.0000 g of fresh sample was weighed in screw cap test tubes . The samples were extracted with 10 ml of 1% HCl in methanol for 24 hours at room temperature with a mechanical shaking. After 24 hours shaking, the solution was centrifuged at 1000 rpm for 5 minutes. One ml of supernatant was taken and mixed with 5 ml of Vanillin-HCl reagent (prepared by combining equal volume of 8% concentrated HCl in methanol and 4% Vanillin in methanol).

D-catechin was used as standard for condensed tannin determination. Forty mg of D-catechin was weighed and dissolved in 1000 ml of 1% HCl in methanol, which was used as stock solution. Exactly 0, 0.2, 0.4, 0.6, 0.8 and 1 ml of stock solution was taken in test tubes and the volume of each test tube was adjusted to 1.0 ml with 1% HCl in methanol. Five ml of Vanillin-HCl reagent was added into each test tube. After 20 minutes, the absorbance of the solutions and the standard solution were measured at 500 nm by using deionized water as blank, and the calibration curve was constructed from a series of standard solution using SPSS Version 15. Concentration of tannin was read in mg of D-catechin per gm of sample (Maxson and Rooney 1972).

$$\text{Tannin in } \mu\text{g/g} = [(\text{absorbance-intercept})/(\text{slope}*\text{density}*\text{weight of sample})]*10$$

### **5.12 Sensory Evaluation of Complementary Gruels**

The 12 complementary foods were prepared from blended and non blended flours into gruels and subjected to sensory evaluation to test their acceptability using a five point hedonic scale, where 1= dislike extremely, 2 = dislike moderately, 3 = neither like nor dislike, 4 = like moderately and 5 = like extremely. A total of 30 untrained panelists (nursing mothers) were used in this study for three days. Each panelist were given two cups and teaspoon in the morning and two in the afternoon for use in the sensory evaluation. The panelists were provided with clean water to rinse their mouth in between testing of the gruels to avoid carry over effect. Each panelist evaluated the gruels for color, odor, taste and overall acceptability.

### **5.13 Structure of the thesis experiment**

The overall framework of experiments of the thesis is shown in fig. 4. It generally shows sample collection & preparation, blend formulation, processing methods, sample analysis and performance evaluation of the product.

### 5.1 4 Statistical analysis

The data were subjected to analysis of variance in a completely randomized design using the method of (Snedecor *et al.*, 1967). Significance difference was accepted at  $p < 0.05$  levels.

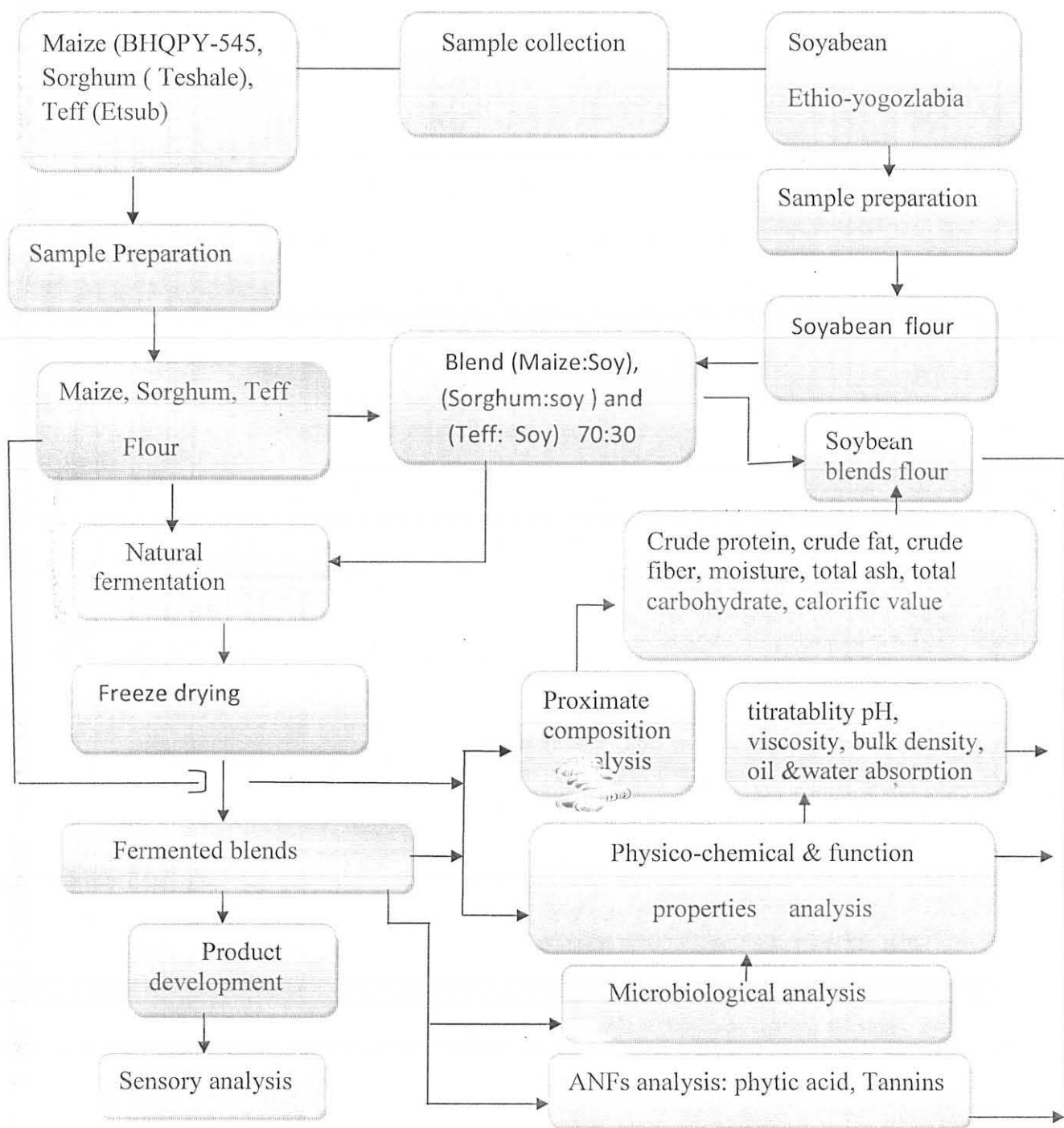


Figure 4. Structure of the thesis

## 6. RESULTS and DISCUSSION

### 6.1 Moisture

The removal of moisture generally increases concentrations of nutrients and can make some nutrients more available Amankwah *et al.* (2009) . The moisture contents of both the blended and non nonblended flours before and after fermentation obtained in this study were below 10% Table 7, 8 and 9. Such low moisture content of flours prevents microbial activity and extends the shelf life of the flour (Kikafunda, 2006). According to WFP (2006), specification the maximum requirement of moisture content of maize-soya blend is 10%, therefore the values obtained in this experiment for all the blends and non blended flours are in agreement with WFP value.

The low moisture observed for the blends and nonblended flours before fermentation are good indicator of their potential to have longer shelf life. This is in line with the findings of Adebayo *et al* (2012). It is believed that materials such as flour and starch containing more than 12% moisture have less storage stability than those with lower moisture content. The present study values are within the range to the values (3.30 to 8.45 %) reported from the production of legumes belended weaning food Egounlety, (2002) but lower than reported results of Kanu *et al.*,(2009) from production and evaluation of breakfast cereal-based porridge mixed with sesame and pigeon peas for adults (9.8%).

### 6.2 Crude protein

The crude protein content of sorghum-soyabean, teff-soyabean and maize -soyabean flours before fermentation (17.84, 16.33 and 16.5%) are significantly ( $p<0.05$ ) lower than that of fermented blended flours (18.67, 17.35 and 19.01%) respectively as it indicated in Table 7, 8, and 9. Similarly, the fermented non blended flours are significantly ( $p<0.05$ ) higher than that of unfermented non blended flours.

Generally the values of protein of blended flours that are obtained before fermentation and after fermentation are higher than the minimum protein requirement (14%) of WFP

specification for corn-soya blend. The crude protein values are within the range to the values (16.00% – 19.97%) reported by the authors Lalude *et al.* (2006) of a weaning food from sorghum and oil - seeds and higher than (7.68% – 8.56%) of Amankwah *et al.* (2009), for maize-soybean weaning blend. The findings are in agreement with the value (17.7%) reported by Griffith *et al.* (1998) weaning food from selected cereal and legumes. The crude protein content values of the blended flours before and after fermentation are higher than that of minimum recommended protein (10%) FAO/WHO (1994).

Table 7: Macronutrient composition of fermented and unfermented sorghum and sorghum - soyabean flours

Sample	Moisture %	Protein %	Fat %	Fiber %	Ash %	Carbohyd- rate%	Energy Kcal/100gm
FSS	3.31 <sup>a</sup> ± 0.14	17.27 <sup>a</sup> ± 0.12	14.08 <sup>a</sup> ± 0.81	3.51 <sup>a</sup> ± 0.6	2.48 <sup>a</sup> ± 0.01	62.34 <sup>a</sup> ± 0.45	423.38 <sup>a</sup> ± 1.07
SSU	4.95 <sup>b</sup> ± 0.19	16.40 <sup>b</sup> ± 0.8	11.32 <sup>b</sup> ± 0.62	3.86 <sup>b</sup> ± 0.7	2.36 <sup>a</sup> ± 0.04	63.72 <sup>b</sup> ± 0.43	438.07 <sup>b</sup> ± 2.87
FS	4.71 <sup>c</sup> ± 0.15	11.52 <sup>c</sup> ± 0.05	3.38 <sup>c</sup> ± 0.33	3.37 <sup>a</sup> ±0.8	1.68 <sup>c</sup> ± 0.03	76.04 <sup>c</sup> ± 0.04	367.68 <sup>c</sup> ± 1.52
SU	5.71 <sup>d</sup> ± 0.13	10.26 <sup>d</sup> ± 0.29	2.99 <sup>d</sup> ± 0.21	4.13 <sup>d</sup> ±0.1 7	1.54 <sup>c</sup> ± 0.08	76.03 <sup>c</sup> ± 0.03	367.81 <sup>c</sup> ± 0.14

Values in the same column with different superscripts are significantly different ( $p < 0.05$ ). All values are means of duplicate  $\pm$ SD.

FSS= fermented sorghum with soyabean,

SSU=unfermented sorghum with soyabean,

FS= fermented sorghum

SU= unfermented sorghum flour

Table 8: Macronutrient composition of fermented and unfermented maize- soyabean blends

Sample	Moisture %	Protein %	Fat %	Fiber %	Ash %	Carbohydrate %	Energy Kcal/100 gm
FMS	3.91 <sup>a</sup> ± 0.13	17.35 <sup>a</sup> ± 0.00	14.16 <sup>a</sup> ± 0.44	4.11 <sup>a</sup> ± 0.04	2.45 <sup>a</sup> ± 0.03	66.48 <sup>a</sup> ±0.1 8	425.73 <sup>a</sup> ± 2.06
MSU	7.82 <sup>b</sup> ± 0.29	16.33 <sup>b</sup> ± 0.12	13.12 <sup>b</sup> ± 0.26	3.72 <sup>b</sup> ± 0.01	2.16 <sup>a</sup> ± 0.06	70.8 <sup>b</sup> ± 0.34	453.79 <sup>b</sup> ± 15.2
FM	4.45 <sup>c</sup> ± 0.23	8.36 <sup>c</sup> ± 0.32	4.60 <sup>c</sup> ± 0.15	2.38 <sup>c</sup> ± 0.29	1.04 <sup>c</sup> ± 0.14	75.67 <sup>c</sup> ±1.0 8	373.17 <sup>c</sup> ± 2.97
MU	7.61 <sup>b</sup> ± 0.53	7.38 <sup>d</sup> ± 0.34	4.26 <sup>d</sup> ± 0.01	2.42 <sup>d</sup> ± 0.32	.93 <sup>c</sup> ± 0.15	80.05 <sup>d</sup> ±0. 26	369.35 <sup>d</sup> ± 0.86

Values in the same column with different superscripts are significantly different ( $P < 0.05$ ). All values are means of duplicate  $\pm$ SD.

FMS= fermented maize with soyabean,

MSU= unfermented maize with soyabean,

FM= fermented maize

MU= unfermented maize flours.

Table 9: Macronutrient composition of fermented and unfermented teff - soyabean blends

Sample	Moisture %	Protein %	Fat %	Ash %	Fiber %	Carbohydrate %	Energy
FTS	3.31 <sup>a</sup> ± 0.14	19.01 <sup>a</sup> ± 0.12	14.56 <sup>a</sup> ± 0.66	3.50 <sup>a</sup> ± 0.02	4.52 <sup>a</sup> ± 0.07	63.60 <sup>a</sup> ± 0.53	405.12 <sup>a</sup> ± 0.75
TSU	8.31 <sup>b</sup> ± 0.13	16.50 <sup>b</sup> ± 0.12	12.28 <sup>b</sup> ± 0.33	2.83 <sup>b</sup> ± 0.00	3.32 <sup>b</sup> ± 0.04	68.87 <sup>b</sup> ± 0.25	405.64 <sup>a</sup> ± 0.94
FT	4.69 <sup>c</sup> ±0.12	8.52 <sup>c</sup> ± 0.18	4.16 <sup>c</sup> ± 0.07	2.35 <sup>c</sup> ± 0.09	3.42 <sup>c</sup> ± 0.17	83.82 <sup>c</sup> ± 0.27	390.72 <sup>c</sup> ± 0.00
TU	6.70 <sup>d</sup> ± 0.15	7.19 <sup>d</sup> ± 0.17	3.01 <sup>d</sup> ± 0.00	2.38 <sup>c</sup> ± 0.05	3.64 <sup>bc</sup> ± 0.04	87.42 <sup>d</sup> ± 0.01	393.86 <sup>d</sup> ± 0.14

Values in the same column with different superscripts are significantly different ( $P < 0.05$ ). All values are means of duplicate  $\pm$ SD.

FTS=fermented teff with soyabean,

MTU= unfermented teff with soyabean

FT= fermented teff

TU= unfermented teff flour.

### 6.3 Crude fat

The crude fat content of sorghum-soyabean, maize-soyabean and teff-soyabean blended flours before fermentation (12.32, 13.12 and 12.28 ) are significantly ( $P < 0.05$ ) lower than that of the fermented (17.08, 14.16 and 14.56%) respectively as it referred from Table 7, 8 and 9. This is due to the superior quality of soybean over maize, sorghum and teff in terms of fat content. The crude fat content of non blended sorghum and teff fermented flours (3.37 and 4.16%) are significantly ( $p < 0.05$ ) higher than that of unfermented (2.99 and 3.01%) respectively. Whereas the crude fat content of fermented

maize (4.60%) is not significantly ( $p>0.05$ ) different from the unfermented maize (4.26%) as it referred from table 8.

Crude fat values of the maize - soyabean flour before and after fermentation are higher than the value (12%) reported by Egounlety (2002), for nutritive value of high-protein-energy legume-blended weaning flour. The crude fat values of the blended flours before and after fermentation are higher than the crude fat value (9.87%) reported by Lalude & Fashakin (2006), for weaning foods from sorghum and oil – seeds. According to the findings of Amankwah *et al.* (2009), the fat content of formulation of weaning food from fermented maize, rice, soybean and fishmeal is (9.38% and 8.75%). Thus, all experimental values of the blended flours are higher than this values. The crude fat content of the blended flours before and after fermentation are higher than the value of WFP specification for the minimum requirement of 6% fat of corn-soya blend. The findings for the blended flours are comparable with the value of famix ( $\geq 7\%$ ) and higher than that of the value (3.67%) reported by Shimelis, (2009) on sorghum based weaning food. Experimental values are within the range with the value (9.0% and 21 %) of Nutrend- commercially sold Nigerian weaning food.

#### **6.4 Crude fiber**

Weaning foods with low fiber content is very important for children considering their low gastric capacity since they have to consume more to get satisfied to meet their daily energy requirement (Eka and Edijala, 1972). The crude fiber content of sorghum-soyabean blend before fermentation is not significantly ( $p>0.05$ ) different from that of the fermented blend. Similarly, the value of crude fiber of maize -soyabean blend before fermentation is not significantly ( $p>0.05$ ) different from that of the fermented blend. Whereas the fermented teff - soyabean blend is significantly ( $p<0.05$ ) higher than that of the unfermented blend as it indicted in Table 7. 8 and 9.

The crude fiber content of non blended maize and teff flour before fermentation are significantly ( $p>0.05$ ) different from the values after fermentation. Whereas the crude fiber values of sorhum-soyabean blend before fermentation significantly ( $P< 0.05$ ) higher

than that of the value of fermented. All experimental values are below from that of the maximum requirement (5%) of WFP specification.

### 6.5 Total ash

According to Fouzia (2009), ash content is an indirect indicator of the mineral level of food stuffs. As it can be seen from the Table 7, the ash content of fermented sorghum-soyabean blend is not significantly ( $p>0.05$ ) different from the unfermented sorghum -soyabean blend. Similarly the ash content of maize -soyabean blend before fermentation is not significantly ( $p<0.05$ ) different from that of the fermented blend Table 8. As shown in Table 9, the value of ash for the fermented teff-soyabean blend (3.58) is significantly ( $p<0.05$ ) higher than that of the unfermented blend. Similarly, the ash content of non blended sorghum, maize and teff flours before fermentation (1.68, 1.04, 3.50 %) are not significantly ( $P> 0.05$ ) different from the fermented (1.78, 1.3, and 3.68 %) respectively. The ash content of blended fours are comparable with the values reported by Egounlety (2002), for nutritive value of high-protein-energy legume-blended complementary flours and higher than the value (.98 %) reported by Edema *et al.* (2005) for maize-soybean blend

### 6.6 Total carbohydrates

The amount of total carbohydrate of nonblended sorghum flour before fermentation (77.03%) is not significantly ( $p>0.05$ ) different from the fermented (77.04%). The nonblended maize and teff flours (80.05 and 87.42%) are significantly ( $P< 0.05$ ) higher than the fermented (75.67 and 83.82%) respectively. The values for the sorghum-soyabean, maize-soyabean and teff soyabean blended flours before fermentation (66.72, 70.8% and 68.87%) are significantly ( $P<0.05$ ) higher than that of the fermented (63.34, 66.48 and 63.60%), respectively as shown above Table 7- 9.

In general, fermentation of cereals leads to a decrease in the level of carbohydrates as well as some non-digestible poly and oligosaccharides (Katongole, 2008). This was in agreement with the findings of Ezeji and Ojimekwe (1993) who reported a decrease in

carbohydrate content with increase in soybean flour fortification. The observed decrease in carbohydrate during fermentation could be attributed to their utilization in the fermentation process as energy sources (Fasasi, 2009).

All the experimental values of blended flours before and after fermentation are comparable with the value (63.21%) reported by Mbata *et al.* (2009), for fermented maize flour and Bambara groundnut-maize blended flour. The calorific values for the blended flours before and after fermentation are lower than the value obtained from Famix (70%) but slightly higher than the values (60.85 and 61.99) reported by the authors Amankwah *et al.* (2009), for different blend ratio in the formulation of weaning food from fermented maize, rice, soybean and fishmeal. All the experimental values of nonblended flours before and after fermentation are comparable with the average value of (75%) for the production of sorghum based weaning food reported by (Shimelis, 2009). Bolaji *et al.* (2010) reported that the total carbohydrate content of maize – soybean blend for the production of Ogi is (71.76%); that is somewhat higher than the value of the current study. These results obtained in this study are within the range (63.11-88.39%) of results reported by Egounley (2002), for nutritive value of high-protein-energy legume-blended weaning flours.

### 6.7 Calorific value

Source of calories for an infant's diet are protein, fat and carbohydrates (Amankwah *et al.*, 2009). The caloric value of sorghum-soyabean and maize-soyabean flours before fermentation (438.07 and 453.79Kcal/100gm) are significantly ( $p < 0.05$ ) higher than that of the fermented (423.38 and 425.73 %) respectively as it shown in Table 7. The energy value of teff -soyabean flour before fermentation (405.64Kcal/100gm) is not significantly ( $p > 0.05$ ) different from the fermented blend (405.12Kcal/100gm). Similarly, the caloric value of maize and teff flours after fermentation (373.17 and 390.72Kcal/100gm) are significantly ( $p < 0.05$ ) higher than that of unfermented (369.35 and 393.35Kcal/100gm) respectively as shown above from table 8 and 9. This is due to the higher carbohydrate content of unfermented flours than that of the fermented blends which their carbohydrate is utilized by the microorganisms.

On the other hand, the increasing effect of fermentation on the value of protein and fat content of the blend surpass the decrease in total carbohydrate. Thus, the calorific value of sorghum flour before fermentation (367.68KCal) is not significantly different ( $p>0.05$ ) with fermented sorghum (367.81). The calorific value of the blended flour are within the range (395 to 509 KCal.) of previous studies such as (Griffith *et al.*, 1998) of weaning food. The values obtained from the study are in agreement with the value ( $>398.9$  KCal.) “Nutrend” (Nestle, Nigeria-weaning diet) obtained commercially and the experimental value (441 KCal.) obtained from the author Lalude & Fashakin, (2006). The values for the blended and nonblended flours are higher than that of WFP specification minimum requirement (380KCal.) for the weaning food from corn-soya blend (CSB) as reported by (Onilude,1999).

## **6.8 Antinutritional content of blended and non blended flours before and after fermentation**

### **6.8 .1 Tannins**

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 soybeans 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 antinutritional potential caused by the presence of tannins is of little concern (Shimelis & Rakshit, 2006). The values of tannins for sorghum, maize and teff flours before fermentation (20.90 3.72 and 10.7 mg/100gm) are significantly ( $p<0.05$ ) higher than that of the fermented (17.33, 1.31 and 8.59mg/100gm) respectively Table 10, 11 and 12.

Similarly, the values for sorghum- soyabean, maize-soyabean and teff-soyabean flours before fermentation (25.69, 31.75 and 35.59mg/100gm ) are significantly ( $p<0.05$ ) higher than that of the fermented (18.43, 28.28 and 25.91mg/100gm) respectively Table 10.

The observed reduction in tannin during fermentation could be attributed to the action of the enzymes released by microorganisms during fermentation.

Table.10: Antinutritional content of blended and nonblended sorghum flour before fermentation and after fermentation

Sample	Tannin	Phytate
FSS	18.43 <sup>a</sup> ±0.66	167.93 <sup>a</sup> ±0.68
SSU	25.69 <sup>b</sup> ±0.34	268.87 <sup>b</sup> ±0.57
FS	17.33 <sup>c</sup> ±0.04	180.56 <sup>c</sup> ±2.14
SU	20.90 <sup>d</sup> ±1.38	360.74 <sup>d</sup> ±4.67

Values in the same column with different superscripts are significantly different (p < 0.05). All values are means of duplicate ±SD.

FSS= fermented sorghum - soyabean,

SSU= unfermented sorghum- soyabean,

FS= Fermented sorghum

SU=unfermented sorghum

Table.11: Antinutritional content of blended and nonblended maize flour before fermentation and after fermentation

Sample	Tannin	Phytate
FMS	14.14 <sup>a</sup> ±0.26	158.43 <sup>a</sup> ±2.21
MSU	31.75 <sup>b</sup> ±0.36	219.68 <sup>b</sup> ±0.7
FM	1.31 <sup>c</sup> ±0.14	131.96 <sup>c</sup> ±2.16
MU	3.72 <sup>d</sup> ±0.02	242.18 <sup>d</sup> ±0.72

Values in the same column with different superscripts are significantly different ( $p < 0.05$ ). All values are means of duplicate  $\pm$ SD.

FMS=fermented maize with soyabean,

MSU=unfermented maize with soyabean,

FM=fermented maize

MU= unfermented maize flour.

### 6.8.2 Phytate

Phytate present in raw materials and foods of plant origin are suggested to be a major factor responsible for lowering the bioavailability of minerals and some proteins (Shimelis & Rakshit, 2006, Reddy *et al.*, 1989, Lesteinne *et al.*, 2005).

Phytate for sorghum, maize and teff nonblended flours before fermentation (360.74, 242.32 and 269.20 and 100gm) are significantly ( $p < 0.05$ ) higher than that of the fermented (180.56, 231.96 and 153.06mg/100gm) respectively as indicated in Table 10-12. Similarly, the values sorghum-soyabean, maize-soyabean and teff-soyabean flours before fermentation (268.87, 219.68 and 272.73mg/100gm) are significantly ( $p < 0.05$ ) higher than that of the fermented (167.93, 158.68 and 188.21mg/100gm) respectively. The observed reduction in phytate during fermentation could be attributed to the action of the enzyme phytase released by microorganisms during fermentation. Phytic acid as powerful chelating agent reduces the bioavailability of divalent cations by the formation

of insoluble complexes (Sandberg, 2002, Weaver and Kanna, 2002, Oberlease .1983, Leshenne,*et al.*,2005).

Table.12:Antinutritional content of blended and nonblended teff flour before fermentation and after fermentation

Sample	Tannin	Phytate
FTS	25.91 <sup>a</sup> ±1.39	188.21 <sup>a</sup> ±1.12
TSU	35.59 <sup>b</sup> ±2.11	471.25 <sup>b</sup> ±1.08
FT	8.59 <sup>c</sup> ±0.5	153.06 <sup>c</sup> ±1.44
TU	10.70 <sup>d</sup> ±0.49	269.20 <sup>d</sup> ±0.32

Values in the same column with different superscripts are significantly different ( $p < 0.05$ ). All values are means of duplicate ±SD.

FTS=fremented teff with soyabean,

MTU= unfermented teff with soyabean,

FT= fermented teff

TU=unfermented teff flour.

## 6.9 Micronutrient content of blended and non blended flour before and after fermentation

### 6.9 1. Iron

The iron content of the blended and nonblended sorghum and maize flours before fermentation (4.08, 2.5, 3.6, and 1.93 mg/100gm ) are significantly ( $p<005$ ) lower than of the fermented (5.27, 2.97, 6.82 and 2.29 mg/gm ) Table 13 and 15. Whereas, fermentation decreases the iron content of teff-soyabean blend from 54.40mg/100gm to 48.39gm/100gm as it is indicated in Table 14. The values of iron for the blended sorghum and teff flour are higher than that of WFP minimum specification (3.25 mg/100g) for the manufacture of corn soya blend for infants. The iron content of the blended and nonblended flours before and after fermentation are higher than from the values 1.78 to

2.01 mg/100g) reported by Shimelis (2009), for sorghum based weaning food. The Fe content of the blended sorghum, maize and teff is higher than the value (2.5 mg/100g ) reported by (Lalude & Fashakin, 2006), for Nutrend – commercial weaning food from Nigeria.

Table 13. The mineral content of blended and non blended sorghum flours before and after fermentation

Sample	Fe mg/100gm	Zn mg/100gm	Ca mg/100gm
FSS	5.27 <sup>a</sup> ±0.13	2.34 <sup>a</sup> ±0.01	259.38 <sup>a</sup> ±10.42
SSU	4.08 <sup>b</sup> ±0.56	2.88 <sup>b</sup> ±0.06	129.93 <sup>b</sup> ±1.15
FS	4.57 <sup>c</sup> ±0.12	1.86 <sup>c</sup> ±0.02	42.48 <sup>c</sup> ±0.41
SU	3.62 <sup>d</sup> ±0.26	2.73 <sup>b</sup> ±0.28	48.93 <sup>d</sup> ±2.81

Values in the same column with different superscripts are significantly different ( $p < 0.05$ ). All values are means of duplicate  $\pm$ SD.

FSS=fermented sorghum with soyabean

SSU=unfermented sorghum with soyabean,

F S=fermented sorghum and

SU=unfermented sorghum flour.

### 6.9.2 Zinc

The zinc content of the blended and nonblended sorghum flour before fermentation (2.88 and 2.73mg/100gm) are significantly ( $p < 0.05$ ) higher than that of the fermented (2.34, and 1.86mg/100gm), respectively as it is referred from table 13. . Similarly, the zinc content of blended and nonblended teff flour (2.79 and 264mg/100gm) are significantly ( $p < 0.05$ ) higher than that of the fermented (2.04 and 2.15mg/100gm) respectively as it is indicated from table 14 . Whereas, for the blended flour of maize before fermentation (2.01mg/100gm) is not significantly ( $p > 0.05$ ) different from the

fermented (1.17mg/100gm) as it is shown in table 15. The zinc content of the blended flours is lower than that of WFP minimum specification (5 mg/100g) for the manufacture of corn soya blend for infants.

Table 14. The mineral content of blended and non blended teff flours before and after fermentation

Sample	Fe mg/100gm	Zn mg/100gm	Ca mg/100gm
FTS	48.39 <sup>a</sup> ±0.95	2.04 <sup>a</sup> ±0.72	199.66 <sup>a</sup> ±65.18
TSU	54.39 <sup>b</sup> ±0.47	2.79 <sup>b</sup> ±0.02	247.95 <sup>b</sup> ±3.11
FT	73.54 <sup>c</sup> ±3.73	2.15 <sup>a</sup> ±0.02	207.76 <sup>c</sup> ±1.85
TU	69.59 <sup>c</sup> ±1.89	2.64 <sup>b</sup> ±0.08	200.22 <sup>d</sup> ±3.10

Values in the same column with different superscripts are significantly different ( $p < 0.05$ ). All values are means of duplicate  $\pm$ SD.

FTS=fermented teff with soyabean

MTU=unfermented teff with soyabean,

FT= fermented teff

TU=unfermented teff flour.

### 6.9.3 Calcium

The calcium content of sorghum- soyabean flour before fermentation is significantly ( $p < 0.05$ ) lower than that of the fermented as it is indicated from Table 13. fermentation significantly ( $p < 0.05$ ) decreases the calcium content of non blended sorghum and maize flours from (48.93mg/100gm and 15.54 mg/100gm) to (42.48 mg/100gm and 10.03 mg/100gm) respectively. Similarly, there is significant ( $p > 0.05$ ) decrease in the calcium content of the blended maize and teff flours from (178.44 mg/gm and 247.95mg/100gm) to (133.76 and 199.66 .93mg/100gm), respectively as it is shown in Table 14 and 15.

The Ca content of the blended sorghum, maize and teff flours are higher than the value (22 mg/100g) as it is reported by (Lalude & Fashakin, 2006), for Nutrend – commercial weaning food from Nigeria. The Ca content of the blended maize flour is higher than that of (45.1 mg/100g) reported by Edema *et al.* (2005) for maize-soybean blend. The Ca content of the blended flours before and after fermentation are higher than that of the value reported by Bolaj *et al.*, (2010) for the production of Ogi. All the experimental values of blended flours before and after fermentation are higher than from the values (17 to 25 mg/100g) reported by Shimelis (2009), for sorghum based weaning food. Generally, calcium content of the blends can fit the minimum requirement (130 mg/100g) of WFP specification for the manufacture of corn soya blend for infants.

Table 15. The mineral content of blended and non blended teff flours before and after fermentation

Sample	Fe mg/100gm	Zn mg/100gm	Ca mg/100gm
FMS	2.96 <sup>a</sup> ±0.04	2.01 <sup>a</sup> ±0.02	133.76 <sup>a</sup> ±0.79
MSU	2.53 <sup>b</sup> ±0.61	1.72 <sup>a</sup> ±0.38	178.44 <sup>b</sup> ±0.00
FM	2.29 <sup>c</sup> ±1.56	1.17 <sup>b</sup> ±0.06	10.03 <sup>c</sup> ±0.60
MU	1.93 <sup>c</sup> ±0.056	1.82 <sup>a</sup> ±0.15	15.54 <sup>d</sup> ±2.99

Values in the same column with different superscripts are significantly different ( $p < 0.05$ ). All values are means of duplicate  $\pm$ SD.

FMS= fermented maize with soyabean,

MSU=unfermented maize with soyabean

FM= fermented maize

MU=unfermented maize flour.

## 6. 10 Effect of fermentation on physicochemical properties of blended and non blended flour

### 6.10.1 pH

Fermentation significantly affect ( $p < 0.05$ ) the pH values of blended and nonblended flours. As fermentation time increases the pH value decreased (6.71, 4.71, 4.07 and 4.1) for sorghum-soyabean, (6.7, 4.4, 4.3 and 4.1) for maize- soyabean and (6.69, 4.52, 4.24 and 4.21) for teff-soy flour. The same is true for nonblended flour and (6.53, 5.33, 4.01 and 4 in the case of sorghum), (6.4, 4.23, 4.13 and 3.91 in the case of maize) and (6.61, 4.29, 4.08 and 4.04 in the case of teff) at (0, 24, 48 and 72 hour) fermentation time respectively as it is shown in Table 16, 17 and 19.

Table 16; pH and titrable acidity for the blended and nonblended sorghum dough during the 3 days of fermentation

Sample	pH				TA			
	0 h	24 h	48h	72h	0h	24h	48h	72h
FSS	6.71 <sup>a±</sup> 0.00	4.71 <sup>b±</sup> 0.00	4.07 <sup>c±</sup> 0.02	4.02 <sup>c±</sup> 0.00	0.85 <sup>d±</sup> 0.02	1.53 <sup>e±</sup> 0.02	2.32 <sup>f±</sup> 0.01	2.53 <sup>g±</sup> 0.03
FS	6.53 <sup>a±</sup> 0.03	5.33 <sup>b±</sup> 0.01	4.01 <sup>c±</sup> 0.01	4.00 <sup>c±</sup> 0.00	0.74 <sup>d±</sup> 0.03	1.15 <sup>e±</sup> 0.04	1.93 <sup>f±</sup> 0.04	2.25 <sup>g±</sup> 0.04

Values in the same row with different superscripts are significantly different ( $P < 0.05$ ). All values are means of duplicate  $\pm$ SD.

FSS=fermented sorghum with soyabean

FS=fermented sorghum

### 6.10.2 Titratable acidity

Titratable acidity significantly ( $p < 0.05$ ) increased as can be seen in table 16, 17 and 18. According to Amankwah *et al.* (2009) the pH decreased from 6.5-4.3 and titratability increased from 0.4-2.9 for sorghum- legume based complementary foods. Thus, the

experimental values are comparable with the above report. According to Akinrele *et al.* (1970), the metabolic activities of microorganisms during fermentation reduce the pH and increase titratable acidity. Such trend has also been reported by Bolaj *et al.*, (2010 ) where pH of fermented maize-cowpea weaning blends ranged between 4.0 - 5.3. Mensah *et al.* (1991) reported that fermented foods with low pH have some antimicrobial activities and as a result, exhibit longer shelf life.

Table 17; pH and titratable acidity for the blended and nonblended maize dough during the 3 days of fermentation

Sample	pH				TA			
	0h	24 h	48h	72h	0h	24h	48h	72h
FMS	6.70 <sup>a</sup> ± 0.00	4.4 <sup>b</sup> ± 0.00	4.31 <sup>c</sup> ± 0.02	4.10 <sup>d</sup> ± 0.00	0.88 <sup>e</sup> ± 0.02	1.54 <sup>f</sup> ± 0.02	2.34 ± 0.01	2.53 <sup>h</sup> ± 0.03
FM	6.4 <sup>a</sup> ± 0.03	4.23 <sup>b</sup> ± 0.01	4.13 <sup>c</sup> ± 0.01	3.91 <sup>cd</sup> ± 0.00	0.76 <sup>e</sup> ± 0.03	1.15 <sup>f</sup> ± 0.04	1.93± 0.04	2.23 <sup>h</sup> ± 0.04

Values in the same row with different superscripts are significantly different (p <0.05). All values are means of duplicate ±SD.

FMS=fermented maize with soyabean

FM=fermented maize

Table 18; pH and titratable acidity for the blended and nonblended teff dough during the 3 days of fermentation

Sample	pH				TA			
	0h	24 h	48h	72h	0h	24h	48h	72h
FTS	6.69 <sup>a</sup> ± 0.01	4.52 <sup>b</sup> ± 0.00	4.24 <sup>c</sup> ± 0.01	4.21 <sup>cd</sup> ± 0.01	0.98 <sup>e</sup> ± 0.02	1.68 <sup>f</sup> ± 0.02	2.33 <sup>g</sup> ± 0.01	2.53 <sup>h</sup> ± 0.03
FT	6.61 <sup>a</sup> ± 0.02	4.29 <sup>b</sup> ± 0.08	4.08 <sup>c</sup> ± 0.02	4.04 <sup>cd</sup> ± 0.02	0.79 <sup>a</sup> ± 0.03	1.15 <sup>b</sup> ± 0.04	1.90 <sup>c</sup> ± 0.04	2.22 <sup>d</sup> ± 0.04

Values in the same row with different superscripts are significantly different ( $p < 0.05$ ). All values are means of duplicate  $\pm$ SD

FTS=fermented teff with soyabean,

FT= fermented teff

## **6.11 Impact of fermentation on functional properties**

### **6.11.1 Bulk density**

Fermentation has been used by various workers to remove the antinutritional factors as well as to improve the nutritional level and have also helped in reducing the bulk density of reconstituted gruels. Fermentation significantly ( $p < 0.05$ ) decreases the bulk density of blended and non blended flours as it is shown in the Table 16,17 and 18. The bulk density of the blended and nonblended flours after fermentation are in agreement with the values reported by Lalude & Fashakin (2006), for weaning food from sorghum and oil – seeds and nutrend – Nigerian commercial weaning food. Similarly the study values are comparable with value (0.68) reported by (Mesfin, 2007). The bulk density obtained in this study are higher than the value (0.54) reported by Cuevas-Rodri'guez *et al.* (2005) for nutritional quality of tempeh flour. The experimental values for blended and nonblended flours before and after fermentation are higher than the value reported by (Edema *et al.* 2005). starch and starch granules absorb a lot of water to reach gelatinization peak (Perez Consesa et al., 2002).

Table 19; Bulk density, water holding capacity and oil holding capacity of blended and non blended sorghum flour before and after fermentation

Sample	BD g/ml	WAC ml/gm	OH (ml/gm)
FSS	.56 <sup>a</sup> ±0.01	3.08 <sup>a</sup> ±0.04	2.02 <sup>a</sup> ±0.03
SSU	.95 <sup>b</sup> ±0.56	1.5 <sup>b</sup> ±0.03	1.91 <sup>b</sup> ±0.03
FS	.71 <sup>c</sup> ±0.01	3.41 <sup>c</sup> ±0.02	1.99 <sup>abc</sup> ±0.01
SU	.95 <sup>cd</sup> ±0.06	1.59 <sup>bd</sup> ±0.02	1.89 <sup>bd</sup> ±0.06

Values in the same column with different superscripts are significantly different ( $P < 0.05$ ). All values are means of duplicate  $\pm$ SD.

FSS=fermented sorghum with soyabean

SSU=unfermented sorghum with soyabean,

FS= fermented sorghum

SU=unfermented sorghum flour

#### 6.11.2. Water holding capacity

The water absorption capacity is an index of the maximum amount of water that a food product would absorb and retain (Marero et al., 1988; Mosha and Lorri, 1987). The water absorption of the fermented blended flours are significantly ( $p>0.05$ ) lower than that of unfermented blends Table 19, 20 and 21. Similarly, WAC of the fermented nonblended flours significantly ( $p>0.05$ ) higher than that of the fermented nonblend flours. This due to the decrease in carbohydrate and the increase in protein and fat content during fermentation. The high water absorption capacity observed from the unfermented flours is because of the high percentage of carbohydrate in the form of starch and starch granules absorb a lot of water to reach gelatinization peak (PerezConsesa et al., 2002).

The water absorption of maize and maize-soy flours before and after fermentation are higher than that of the values reported by Edema *et al.* (2005), for commercially sold maize and Maize-soy flour. In the this study, the unfermented and fermented blended

flours are observed to contain incomparable amount of water absorption with (1.34) reported by Emmanuel, *et al.* (2010), for cowpea – fortified food. This is because addition of soybean increases the water absorption of cereals based weaning foods. High water content in a food reduces the energy density and increases the bulk of the food, and if the water content is too high, it will negatively influence energy intake. Lower water absorption is desirable for making thinner gruels that will enhance more in-take of nutrients (Kulkani et al., 1991).

Table 20; Bulk density, water holding capacity and oil holding capacity of blended and non blended teff flour before and after fermentation.

Sample	BD g/ml	WAC ml/gm	OHC ml/gm
FTS	.65 <sup>a</sup> ±0.03	1.84 <sup>a</sup> ±0.85	2.23 <sup>a</sup> ±0.02
TSU	.83 <sup>b</sup> ±0.00	1.02 <sup>b</sup> ±0.03	2.02 <sup>b</sup> ±0.03
FT	.62 <sup>ac</sup> ±0.00	2.10 <sup>c</sup> ±0.13	2.09 <sup>b</sup> ±0.07
TU	.97 <sup>d</sup> ±0.02	1.72 <sup>ad</sup> ±0.04	1.76 <sup>c</sup> ±0.06

Values in the same column with different superscripts are significantly different (P < 0.05). All values are means of duplicate ±SD.

FTS= fermented teff with soyabean,

TSU= unfermented teff with soyabean,

FT=fermented teff

TU=unfermented teff flour

### 6. 11. 3 Oil absorption capacity

The oil absorption capacity of fermented blended flours are significantly ( $p < 0.05$ ) higher than that of unfermented blends as it indicated from table 16, 17 and 18. Similarly, the fermented nonblended flours significantly ( $p < 0.05$ ) lower than that of the unfermented. This is due to the decrease in carbohydrate and the increase in protein and fat content during fermentation.

Therefore, fermentation had significantly ( $p < 0.05$ ) a increasing effect on the oil absorption. The values obtained from current study are higher than that of (1.22ml - 2.23ml) reported by Fouzia (2009) of extrusion cooking of full-fat soy flour. Similarly, oil absorption of fermented blended and nonblended flours are higher than the value (1.82ml, 1.44ml) reported by Assefa (2008), for different varieties of improved varieties of soybean in Ethiopia

The higher oil-binding capacity of sorghum flour suggests that this flour would be useful in formulation of foods where an oil holding property is an important consideration. According to Lahl and Braun (1994) lipid binding is dependent on the surface availability of hydrophobic amino acids. Oil absorption capacity is important as oil acts as flavor retainer and gives soft texture to food improving mouth-feel (Ubbor and Akobundu, 2009; Aremu *et al.*, 2006). Since the flours had good oil absorption capacity it suggests the presence of good lipophilic constituents and therefore may be suitable for production of sausage, soups and cakes (Aremu *et al.*, 2006; Kinsella, 1979).

Table 21; Bulk density, water holding capacity and oil holding capacity of blended and non blended maize flour before and after fermentation

Sample	BD g/ml	WAC ml/gm	OHC ml/gm
FMS	.67 <sup>a</sup> ±0.00	3.05 <sup>a</sup> ±0.08	2.12 <sup>a</sup> ±0.12
MSU	.84 <sup>b</sup> ±0.00	2.19 <sup>b</sup> ±0.16	2.03 <sup>b</sup> ±0.17
FM	.71 <sup>ac</sup> ±0.00	2.06 <sup>bc</sup> ±0.06	1.94 <sup>c</sup> ±0.04
MU	.95 <sup>d</sup> ±0.06	1.89 <sup>cd</sup> ±0.02	1.84 <sup>d</sup> ±0.01

Values in the same column with different superscripts are significantly different ( $p < 0.05$ ). All values are means of duplicate ±SD.

FMS= fermented maize with soyabean,

MSU= unfermented maize with soyabean,

FM= fermented maize

MU= unfermented maize flour

#### 6.12 . Effect of fermentation on microbiological quality of blended and nonblended flours

Lactic acid bacteria are the predominant microorganisms at the end of the fermentation (Hounhouigan 1994). The highest LAB counts were obtained at 72 h fermentation for all samples Table 22-24. Pattison *et al.*, (1998) reported that high numbers of LAB in commercial sorghum beer and that this indicated the ability of the bacteria to survive and grow in an acidic environment. The low pH of the beer reportedly inhibits or kills pathogenic or most anaerobic endospore-forming bacteria, thus improving the safety of the product (Haggblade and Holzapfel, 1998).

Table 22; Microbial counts for the blended and nonblended sorghum dough during the 3 days of fermentation.

Sample	Lactic acid Bacteria Count Cfu/ml				Total Bacteria Count Cfu/ml			
	0h	24h	48h	72h	0h	24h	48h	72h
FSS	3.17 <sup>a</sup> ± 0.25	6.00 <sup>b</sup> ± 0.17	7.24 <sup>c</sup> ± 0.13	7.92 <sup>d</sup> ± 0.06	6.02 <sup>a</sup> ± 0.04	3.90 <sup>b</sup> ± 0.13	2.77 <sup>c</sup> ± 0.06	2.03 <sup>d</sup> ± 0.00
FS	2.68 <sup>a</sup> ± 0.28	4.84 <sup>b</sup> ± 0.11	6.06 <sup>c</sup> ± 0.09	6.91 <sup>d</sup> ± 0.08	5.50 <sup>a</sup> ± 0.04	4.78 <sup>b</sup> ± 0.30	4.09 <sup>c</sup> ± 0.16	3.92 <sup>cd</sup> ± 0.06

Values in the same row with different superscripts are significantly different ( $P < 0.05$ ). All values are means of duplicate  $\pm$ SD.

FSS= fermented sorghum with soyabean

FS= fermented sorghum flour

During the onset of fermentation (0, 24, 48 and 72 h). Aerobic Bacteria plate Count (APC) significantly ( $p < 0.05$ ) decreases as fermentation time increases as it is indicated from Table 22, 23 and 24. This result indicated that as fermentation time increased, the microorganism, decreases. The expected decrease or elimination of aerobic bacteria is in agreement with the findings reported by Mbata *et al.* (2009) for fermented maize flour fortified with bambara groundnut as complementary food. When water is added to flour, the micro-population in the flour begins to grow and metabolize. This process is the basis of the preparation of cereal gruels which are common complementary foods in developing countries.

Table 23; Microbial counts for the blended and nonblended teff dough during the 3 days of fermentation.

Sample	Lactic acid bacteria count Cfu/100gm				Total bacterial count Cfu/100gm			
	0h	24h	48h	72h	0h	24h	48h	72h
FTS	2.37 <sup>a</sup> ± 0.25	5.00 <sup>b</sup> ± 0.17	6.54 <sup>c</sup> ± 0.13	6.92 <sup>d</sup> ± 0.06	7.02 <sup>e</sup> ± 0.04	4.90 <sup>f</sup> ± 0.13	2.77 <sup>g</sup> ± 0.06	2.03 <sup>h</sup> ± 0.00
FT	2.68 <sup>a</sup> ± 0.28	5.34 <sup>b</sup> ± 0.11	6.06 <sup>c</sup> ± 0.09	7.50 <sup>d</sup> ± 0.08	5.50 <sup>e</sup> ± 0.04	4.78 <sup>f</sup> ± 0.30	3.09 <sup>g</sup> ± 0.16	3.02 <sup>h</sup> ± 0.06

Values in the same row with different superscripts are significantly different ( $p < 0.05$ ).

All values are means of duplicate  $\pm$ SD.

FTS=fermented teff with soyabean

MTU=unfermented teff with soyabean,

Table 24: Microbial counts for the blended and nonblended maize dough during the 3 days of fermentation.

Sample	Lactic acid bacteria count Cfu/100gm				Total bacterial count Cfu/100gm			
	0h	24h	48h	72h	0h	24h	48h	72h
FMS	2.37 <sup>a</sup> ± 0.25	5.00 <sup>b</sup> ± 0.17	6.54 <sup>c</sup> ± 0.13	6.92 <sup>d</sup> ± 0.06	7.02 <sup>f</sup> ± 0.04	4.90 <sup>g</sup> ± 0.13	2.77 <sup>h</sup> ± 0.06	2.03 <sup>i</sup> ± 0.00
FM	2.68 <sup>a</sup> ± 0.28	5.34 <sup>b</sup> ± 0.11	6.06 <sup>c</sup> ± 0.09	7.50 <sup>d</sup> ± 0.08	5.50 <sup>e</sup> ± 0.04	4.78 <sup>f</sup> ± 0.30	3.09 <sup>g</sup> ± 0.16	3.92 <sup>h</sup> ± 0.06

Values in the same row with different superscripts are significantly different ( $P < 0.05$ ).

All values are means of duplicate  $\pm$ SD.

FMS=fermented maize with soyabean

FS=fermented maize flour

### 6.13 Sensory evaluation of gruels prepared from blended and non blended flours before and after fermentation

#### 6.13.1 Taste

Taste is an important parameter when evaluating sensory attribute of food. The product might be appealing and having energy density but without good taste, such a product is likely to be unacceptable. Fermentation significantly ( $p < 0.05$ ) increases the taste of the blended flours. Similarly, the fermented teff -soybean is significantly ( $p < 0.05$ ) more tasted than that of unfermented. Fermented maize soybean flour (4.67) is not significantly ( $p > 0.05$ ) different from unfermented (4.47). For the nonblended flours, the values indicates that the fermented flours are not significantly ( $p < 0.05$ ) different from the unfermented as it is shown in Table 25- 27.

Table 25; Sensory evaluation of gruels prepared from blended and non blended sorghum before and after fermentation

Sample	Taste	Color	Odor	Overall acceptability
FSS	4.47 <sup>a</sup> ±0.51	4.67 <sup>a</sup> ±0.48	4.80 <sup>a</sup> ±0.48	4.57 <sup>a</sup> ±0.56
SSU	3.37 <sup>b</sup> ±0.61	4.37 <sup>b</sup> ±0.57	4.00 <sup>b</sup> ±0.66	3.97 <sup>b</sup> ±0.67
FS	3.97 <sup>c</sup> ±0.89	4.33 <sup>b</sup> ±0.48	3.13 <sup>c</sup> ±0.69	3.93 <sup>b</sup> ±0.83
SU	3.87 <sup>cd</sup> ±0.82	4.50 <sup>c</sup> ±0.51	3.10 <sup>c</sup> ±0.77	3.53 <sup>c</sup> ±1.12

Values in the same column with different superscripts are significantly different ( $P < 0.05$ ). All values are means of duplicate ±SD.

FSS= fermented sorghum with soyabean,

SSU=unfermented sorghum with soyabean,

FS= fermented sorghum

SU=unfermented sorghum flour

### 7.13.2 Odor

Odor is an integral part of taste and general acceptance of the food before it is put in the mouth. It is therefore an important parameter when testing acceptability of formulated foods. Results of sensory evaluation indicated that the fermented blended flours are significantly ( $p < 0.05$ ) preferred than that of unfermented blends. Similarly the smell of nonblended sorghum, maize and teff flours before fermentation (3.13, 3.33 and 2.97) are not significantly ( $P < 0.05$ ) different from the fermented (3.7, 4.07 and 2.7) respectively as it is shown in table 25- 27. Generally, the fermented blended flours scored significantly higher ( $P < 0.05$ ) in terms of odor than the rest of samples by panelists. This might be due to the production of flavors during fermentation by the microorganisms that enhance the odor of the fermented flours to be preferred by the panelists with respect to in their odor.

Table 26; Sensory evaluation of gruels prepared from blended and non blended maize before and after fermentation

Sample	Taste	Color	Odor	Overall acceptability
FMS	4.67 <sup>a</sup> ±0.48	4.67 <sup>a</sup> ±0.55	4.87 <sup>a</sup> ±0.35	4.53 <sup>a</sup> ±0.71
MSU	4.47 <sup>b</sup> ±0.57	4.30 <sup>b</sup> ±0.53	4.20 <sup>b</sup> ±0.13	4.07 <sup>b</sup> ±0.82
FM	4.33 <sup>c</sup> ±0.48	4.03 <sup>c</sup> ±0.49	3.84 <sup>a</sup> ±0.77	4.17 <sup>b</sup> ±0.75
MU	4.30 <sup>c</sup> ±0.51	4.77 <sup>a</sup> ±0.43	3.81 <sup>a</sup> ±0.66	3.20 <sup>d</sup> ±0.99

Values in the same column with different superscripts are significantly different ( $P < 0.05$ ). All values are means of duplicate ±SD.

FMS= fermented maize with soyabean

MSU=unfermented maize with soyabean,

FM=fermented maize

MU=unfermented maize flour

### 7.13.3 Color

Color is an important attribute in food choice and acceptance. Outcome of sensory evaluation indicated that some samples were similar in appearance while others differed significantly as it is indicated from Table 25- 27. The fermented blended flours for all samples are significantly ( $p < 0.05$ ) preferred than that of unfermented flours. Whereas, the nonblended flours before and after fermentation are similar in appearance.

Table 27; Sensory evaluation of gruels prepared from blended and non blended teff before and after fermentation

Sample	Taste	Color	Odor	Overall acceptability
FTS	3.60 <sup>a</sup> ±0.86	4.63 <sup>a</sup> ±0.55	4.33 <sup>a</sup> ±0.61	3.53 <sup>a</sup> ±1.12
TSU	2.00 <sup>b</sup> ±0.83	4.24 <sup>b</sup> ±0.53	3.67 <sup>b</sup> ±0.76	4.07 <sup>b</sup> ±0.82
FT	3.13 <sup>c</sup> ±0.73	4.03 <sup>c</sup> ±0.49	2.83 <sup>c</sup> ±0.53	3.13 <sup>c</sup> ±0.78
TU	3.16 <sup>c</sup> ±0.93	4.70 <sup>a</sup> ±0.43	2.93 <sup>c</sup> ±0.65	2.93 <sup>c</sup> ±1.41

Values in the same column with different superscripts different ( $P < 0.05$ ). All values are means of duplicate ±SD.

FTT=fermented teff with soyabean,

TSU=unfermented teff with soyabean,

FT=fermented teff

TU=unfermented teff flour.

### 7.13.4 Overall acceptability

Generally, the gruels prepared from the blended flours are highly acceptable by panelists with mean scores (4.57, 4.53 and 4.07) for sorghum-soy, maize -soy and teff-

soyabean flours respectively as it indicate in table 25- 27. Their acceptability levels were significantly higher ( $P < 0.05$ ) than the rest of samples. Formulations TU and MU are significantly( $p < 0.05$ ) disliked by panelist as compared to the rest of samples, The acceptability of the gruels prepared from blended fermented flours might be due to the advantages of fermentation process, which causes changes in food quality including texture, flavor, appearance, nutrition and safety. The benefit of fermentation process may include improvement in palatability and acceptability by developing improved flavors and textures (Sahana & Fauzia, 2003).

## **8. Conclusions and Recommendation**

### **8.1. Conclusions**

This study aimed in formulating the complementary food that provide protein-energy requirement using the staple cereal, maize, sorghum and teff that are rich in essential limiting amino acids, lysine & tryptophan, and protein rich soybean. The research was mainly focused on investigating the effect of fermentation process on the formulated complementary food.

In the present work, it was demonstrated that fermentation process significantly changed the nutritional value of the complementary food by reducing antinutrients such as tannin and phytate. The reduction of these and other antinutritional factors that are not included in this study, but expected to be reduced during fermentation can lead us to the increment of the bioavailability of micronutrients.

Moreover, fermentation increases the crude protein, crude fat and calorific value of blended flours. Therefore, maximum of 15% increment of protein for teff- soy blend. On the hand, fermentation significantly decreases the bulk density and moisture content which helps for the shelf life of the complementary food. The total microbial count decreases as fermentation time increases and the LAB count also increases. The response of the panelists shows that fermented complementary foods are more accepted than unfermented.

Utilization of simple equipments, such as utensils in home makes fermentation process suitable for low-income families living in rural areas. Hence, fermentation is a promising food processing method for weaning food preparation, especially in developing countries.

Generally fermentation comparably reduced or eliminated antinutrients and increase proximate composition and decrease the antinutrient content which increase the bioavailability of minerals.

## 9.2. Recommendation

During the process of undergoing this research paper, there had been some constraints and results. Based on this, the following recommendations are made.

- Further study should be conducted on nutritive values by animal (*in vivo* protein digestibility test) to further check and compare the quality of fermented weaning blends with the results of this thesis work.
- It is also recommended for researches to conduct other more functional properties such as emulsion activity & stability; foaming capacity & stability; and water solubility index. In addition to this, physic-chemical properties like seed density, swelling coefficient, swelling capacities & indices.
- There should be immediate use of the complementary foods prepared from the products because of high content of fats that may go to oxidation.

## 8. Reference

- Adams, M.R. (1990). Topical aspects of fermented foods. *Trends in Food Science & Technology* 1, 141- 144.
- Adebayo, A., and Prolin, O. (2012). Evaluation of complementary food formulated from fermented sorghum, walnut and ginger, *J. Appl. Biosci.*, 4, 433-441.
- Agren, G. and Gibson, R. (1968). *Food composition table for use in Ethiopia*. I. ENI, Addis Ababa Ethiopia.
- Aguirre, M. & Collins, M.D. (1993). Lactic acid bacteria and human clinical infecti *Journal of Applied Bacteriology* 75, 95-107.
- Ahima, K. J. (2005). Formulation of weaning food using composite of maize groundnut an soybean and assessing its nutritional effect using animal model. A thesis submitted to the department of biochemsitry and biotechnology in partial fulfillment of the requirement for the degree of Master of Science (Food Science and Technology).
- Akinele, T. and. Omotola, B. 1986. Energy and protein Intake of Infants and Children from the low income group of Ibadan. *Nutr. Res.*, 26: 129-37.
- Akpanunam, M.A. and Sefa-Dedeh, M.(1995). Energy and protein Intake of Infants. *S., Food and Nutrition Bulletin*, 16(1) Pp 75 – 80
- Alemayehu, A. (1990). Studies on the nutritional composition of tef [*Eragrostis tef*(Zucc)Trotter] and the interactive influence of Environmental and genotype. *PhD thesis*, University of London, London, UK
- Al Mamary, M., Al-Hobori. M., Al-Aghbari, A. and Al-Obeidi, A.( 2002). *In vivo* effect of dietary sorghum tannins on rabbit digestive enzymes and mineral absorption. *Nutrition Research* 21:1393-1401
- Amankwah, E.A., Barimah, J., Acheampong, R., Addai L.O., and Nnaji, C.O. (2009). Effect of Fermentation and Malting on the Viscosity of Maize-Soyabean Weaning Blends, *Pak. J. Nutr.* 8 (10): 1671-1675.

- Amuna, P., Zotor, F., Sumar, S., and Chinyanga, Y. T.(2000). The role of traditional cereal/legume/fruit-based multimixes in weaning in developing countries. *J. Nutr. Food Sci.* 30(3): 116-122.
- Alm, L. (1982): Effect of fermentation on B-vitamin content of milk in Sweden. *Journal of Dairy Science* 65, 353-359.
- Anglani, C. 1998. Sorghum for human food: a review. *Plant Foods and Human Nutrition* 52: 85 89.
- AOAC. (1998). Official methods of Analysis. (16th ed.) Arlington, VA: Association of Official Analytical Chemists.
- AOAC. (2000). Association of Official Analytical Chemists. Official methods of Analysis (Vol.II 17<sup>th</sup> edition) of AOAC International. Washington, DC, USA.
- Aremu, M. O., Olaofe, O., and Akintayo, E., T.( 2006). A comparative study on the chemical composition and amino acid composition of some Nigerian under-utilized legume flours. *Pak. J. Nutr.* 5(1):34-38.
- Assefa, Yimer, (2008). Effect of processing on some antinutritional factors of improved soybean (glycine max) varieties grown in Ethiopia. A Thesis Submitted to the School of Graduate Studies of Addis Ababa University in Partial Fulfillment of the Requirement for Degree of Master of Science in Food Engineering.
- Ayana, A.( 2001). Genetic diversity in sorghum (*Sorghum bicolor* (L.) Moench) germplasm from Ethiopia and Eritrea. PhD Thesis, Addis Ababa University, Addis Ababa, Ethiopia
- Beuchat, L. R.(1977). Functional and electrophoretic characteristics of succinylated peanu flour protein. *J. Agric. Food Chem.* 25, 258–261.
- Bolaji, O. A., Olubunmi, O. A., Samuel, A. G. (2010). Quality assessment of selected cereal soyabean mixtures in ogi production. Department of Microbiology, Federal University of Technology, P.M.B 65 Minna, Nigeria., *J. Sci.*8(3):68-81.
- Bressani R, Elias LG (1974) Legume foods. In Atschul AM (ed), *New protein foods*, Vol. 4: 231 –297. New York: Academic Press.
- Birch, G.G., Spencer, M., and Cameron, A.G. (1980). *Food science* (2nd and., pp.29-37). Oxford, UK. Pergamon Press Ltd.

- Budiastra, I., W., Andrianyta, H., and Purwadaria, H., k., (2000). Non-destructive Nutrient determination of maize using NIR Method.
- Butler, L.G., Price, M.L., Brotherton J.E. (1982). Vanillin assay for Proanthocyanidins (Condensed Tannins): Modification of the Solvent for Estimation of the Degree of Polymerization. *J. Agric. Food Chem.*, 30: 1087 -1089.
- Campbell-Platt, G. (1987). *Fermented foods of the world - a dictionary and guide*. London, Butterworths. ISBN: 0 407-00313-4.
- Cawley, R., and Mitchello, T. A. (1968): Inhibition of wheat alpha amylase by bran phytic acid. *J. Sci. Food and Agric.* 19:106–109.
- (CSA). (2000). Statistical Abstracts, various annual reports.
- CSA. (2005). Agricultural sample survey 2004/2005. Volume I: Report on area and production of crops (private peasant holdings, meher season): Statistical Bulletin 331, May 2005.
- Chau, C.F., and Huang, Y., L. (2003). Comparison of the chemical composition and physico-chemical properties of different fibers prepared from the peel of *Citrus sinensis* L. cv. Liucheng. *Journal of Agricultural and Food Chemistry* 51, 2615-2618.
- Chavan, J.K., Kadam, S.S., Salunkhe, D.K. (1989). Chickpea. In: Salunkhe, D.K., Kadam, S.S. (Eds), *CRC Handbook of World Food Legumes: Nutritional Chemistry, Processing Technology and Utilization, Vol. I*. CRC Press, Inc., Boca Raton, Florida, USA, pp. 247–288.
- Chi, L., De-jia, L., and Chun-quan, L. (2012). Effect of fermentation on the nutrient value of maize article first published online, DOI. 11. 1111/j.1365-2621. 2011.02904.x
- Chompreeda, P., T. & Fields, M.L. (1984). Effects of heat and fermentation on the extractability of minerals from soybean meal and corn meal blends. *Journal of Food Science* 49, 566- 568.
- Claus, D. C. (1992). A standardized gram staining procedure. *World J. Microbiol. Biotechnol.* 8: 451-452.
- Cossak, Z. T. and Asa, A. (1983). Effect of protein source on bioavailability of zinc in human subjects. *Nutrition Research* . 3:23-26.
- Costanza, S., DeWet, J., and Harlan, J. (1979). Literature review and taxonomy of *Eragrostis tef*(tef). *Econ. Bot.*, 33:413 – 424.

- Cuevas-Rodríguez, E.O., Verdugo-Montoya, N.M., Angulo-Bejarano, P.I., Milán-Carrillo, J., Mora-Escobedo, R., Bello-Pérez, L.A., Garzón-Tiznado, J.A., and Reyes-Moreno, C. (2006). Nutritional properties of tempeh flour from quality protein maize (*Zea mays* L.). *LWT* 39, 1072–1079. Available online at [www.sciencedirect.com](http://www.sciencedirect.com).
- Diamond, J. (1997). Location, location, location: the first farmers. *The American Association for the Advancement of Science. Science* 278, 1243–1244.
- De Simone, C. (1986): Microflora, yoghurt and the immune system. *International Journal of Immunotherapy*, Suppl. 19-23.
- Doggett, H. (1988). Sorghum, 2nd ed. Longman Scientific and Technical, New York, N.Y.
- Edema, M. O. and Sann, I. (2006). Micropopulation of fermenting maize meal for sour maize bread production in Nigeria. *Niger. J. Microbiol.* 20(2): 937-946.
- Duhan, A., Khetarpaul, N. and Bishnoi, S. (2000). Changes in phytates and HCl-extractability of calcium, phosphorus and iron of soaked, dehulled, cooked and sprouted pigeon pea cultivar. *Plant Food Human Nutri.* 57:275-274.
- Duke, J.A. (1981): Handbook of Legumes of World stability of fortified cereal products. *Journal of Food Technology* 30(4): 56-62.
- Duszkiewicz-Reinhard, W., Gujska, E, & Khan. K. (1994): Reduction of stachyose in legume flours by lactic acid bacteria. *Journal of Food Science* 59, 115-117.
- Edema, M. O, Sanni, L. O. & Sanni, A. I. (2005). Evaluation of maize-soybean flour blends for sour maize bread production in Nigeria. *African J. Biotechnol. Vol. 4* (9), pp. 911-918.
- Egounlety, M., 2002. Production of legume-fortified weaning food. *Food Res. Inter.* 35: 233-23
- Eka, O.,U. (1980). Effect of fermentation on the nutrient status of locust beans. *Food Chemistry* 5, 303-308.
- Elmaki, B., H., Babiker, E., E. and El Tinay, A., H. (1999). Change in chemical composition, rain malting, starch, tannin content and protein digestibility during germination of Sorghum, *Journal of Food Science* 57,134-156.
- El-sheikh, E.A., EFadul, I.,A. and El Tinay, A. (2000). Effect of cooking on

antinutritional factors and in vitro protein digestibility of faba bean grown with different nutritional regimes. *Food Chem.* 68: 211-212. sorghum cultivars. *Food Chem.* 64:331-336.

Endeshaw, B. (1989). Lysine and other essential amino acids in various fractions of major tef seed proteins. In: Cereals of the semi arid tropics. IFS, Cameroon. Pp230 – 240.

Fadlallah,L. Omima ,E., Abdullahi, H., Tinay,E and Elfadil,E. (2010). Effect of fermentation on biochemical characteristics of sorghum flour supplemented with chickpea flour . *Journal of Applied Sciences Research*, 6 (7). pp. 860-865.

Ezeji, C. and P.C. Ojmelukwe, 1993. Effect of fermentation on the nutritional quality and functional properties of infant food formulations prepared from bambara-groundnut, fluted-pumpkin and millet seeds. *Plant Foods for Human Nutrition*, 44: 267-276.

Food and Agricultural Organization (FAO).(20050. FAOSTAT agricultural database. Available on line <http://faostat.fao.org/faostat/>.

FAOSTAT agricultural database. Available on line <http://faostat.fao.org/faostat/>.

(FAO). (2009). FAOSTAT agricultural database. Available on line <http://faostat.fao.org/faostat/>.

FAO. (2006). Agriculture database. Available at: <http://apps.fao.org/page/collections?subset=agriculture>.

FAO. (2009). *Rice and narrowing the yield gap*. Retrieved from <http://www.fao.org/rice/2004/en/f-sheet/factsheet5.pdf>

FAO/WHO, 1994. Joint FAO/WHO Food standards programme Codex Alimentarius Commission, Foods for special dietary uses (including foods for infants and children) Volume 4.

Fouzia Muhsin, (2009). Effect of extrusion cooking on some quality attributes of full-fat soy flour and soy protein concentrate from locally grown Soybean (*Glycine Max L.*) varieties, A Thesis Submitted to the School of Graduate

- Studies of Addis Ababa University in Partial Fulfillment of Requirements for the Degree of Masters of Science in Chemical Engineering (Food Engineering).
- Frey, D., Oldfield, R. J. and Bridger, R.C.A. (1979). Colour atlas of pathogenic fungi, London. Wolfe Medical Publication Ltd.
- Fuller, R. (1989). Probiotica in man and animals. *Journal of Applied Bacteriology* 66, 109-116.
- Fukushima, D. (1999). Recent progress of soybean protein foods: Chemistry, Technology and Nutrition. *Food Review Int.* 7(3): 323-352.
- Girma, A., Mosisa, W., Dereje, B., Abdissa, G., and Demissew, K. (2001). Participatory analysis of off-season maize production in Eastern and Western Wollega zones. (In press).
- Griffith, L.D., Castell-Perez, M.E., and Griffith, M.E. (1998). Effects of Blend and Processing Method on the Nutritional Quality of Weaning Foods Made from Select Cereals and Legumes, published by the American Association of Cereal Chem., Inc, Vol. 75, No. 1, p.p 105-112.
- Gupta, U., Rudramma, R. & Joseph, R. (1998). Nutritional quality of lactic fermented bitter gourd and ferngrek leaves. *int. J. of Food Science and Nutrition* 49, 101-108.
- Haggblade, S., Holzapfel, W.H. (1989). Industrialisation of Africa's indigenous beer brewing. In: Steinkraus, K.H. (Ed.), *Industrialization of Indigenous fermented foods*, 33. Marcel Dekker, New York, pp. 191-283.
- Harland BF & Harland J (1980). Fermentative reduction of phytate in rye, white, and whole wheat breads. *J. food Chemistry* 57, 226-229.
- Harlan, J. (1971). Agricultural origins: centers and non centers. *Science* 174, 468-474.
- Hesseltine, C,W. (1983). The future of fermented foods. *Nutrition Reviews* 41, 293-301.
- Hesseltine, C,W. & Wang, H.,L. (1980). The importance of traditional fermented foods. *J. Bio- Science* 30, 402-404.
- Hoffmann, E.,M., Muetzel,,S., and Becker,,K.(2003). The fermentation of soybean meal by rumen microbes in vitro reveals different kinetic features for the inactivation and the degradation of trypsin inhibitor protein. *Anim. Feed Sci. Technol.*,106:187-197.
- Hounhouigan, D.J., 1994. Fermentation of maize (*Zeamays L.*) meal for *mawe* production

- in Benin; Physical, chemical and microbiological aspects: *PhD Thesis*, Agricultural University, Wagening, The Netherlands.
- Hotz, C. and Gibson, R.,S. (2001). Assessment of home-based processing methods to reduce the phytate content and phytate/zinc molar ratio of white maize (*Zea mays*). *J. Agric. Food Chem.*,49: 692-698.
- Hotz, C., Gibson, R.,S. and Temple, A .,L. (2001). home based method to reduce phyatate content and increase zinc bioavailability in maize-based complementary diets. *Int. J. Food Scie. Nutr.*,52:133-142.
- Ihekoronye, A.I. (1999). *Manual of Small-scale Food Processing*. Macmillan Publishers Ltd,London. Pp. 108 – 212.
- Ikujenlola, V.A. and Fashakin, J. B.(2003). The physico-chemical properties of a complementary diet prepared from vegetable proteins. *J. Food Agri. Env. 4: 23-26*.
- Iwe, M.O. (2003). Science and Technology of Soybean. Rejoint Communication Services Ltd., Enugu. Pp.324-342.
- Jansen, G., Dimaio, L. and Hause, N. (1962). Amino acid composition and lysine supplementation of tef. *J. Agric.Food Chem.* 10: 62 – 64.
- Jones, B., M., Ponti, G., J., Tavassoli, A. and Dixon, P., A (1978). Relationships of the Ethiopian cereal t'ef (*Eragrostis tef* (Zucc.) Trotter):evidence from morphology and chromosome number. *Annals of Botany* 42: 1369–1373.
- Jordan, W.,R. and Sullivan, C.,Y.(1982). *Reaction and resistance of grain sorghum to heat and drought*. In 'Sorghum in the Eighties, Vol 1,' (J.V. Mertin, ed), ICRISAT, Patancheru, India pp 131-142.
- Kanu, P.J Sandy E.H, B.A. Joseph Kandeh , Bahsoon J.P and Huiming, Z 2009. Production and Evaluation of Breakfast Cereal-Based Porridge Mixed with Sesame and Pigeon Peas for Adults. *Pakistan Journal of Nutrition* 8 (9): 1335-1343.
- Kebede, M., Gezahegne, B., Benti, T., Mosisa, W., Yigzaw, D., and Assefa, A. (1993). Maize production trends and research in Ethiopia, pp. 4-12. In: Benti Tolessa and J.K. Ransom(eds. ), *Proceedings of the First National Maize Workshop of Ethiopia*, IAR/CIMMYT, and Addis Ababa, Ethiopia.
- Kent, N.,L. (1985). *Technology of Cereal*. An introduction for students of Food Science

- and Agric. (3rd edition). Pergamen press, oxford. pp.193-202.
- Khetarpaul, N. & Chauhan, B.,M. (1991). Sequential fermentation of pearl millet by yeasts and lactobacilli - effect on the antinutrients and in vitro digestibility. *Plant Foods for Human Nutrition* 41, 321-327.
- Kimber, C.,T.(2000). Origins of domesticated sorghum and its early diffusion into India and China. In 'Sorghum: Origin, History, Technology, and Production', (C. Wayn,Smith and R.A. Frederiksen, eds), John Wiley & Sons, New York pp 3-98.
- Kulkani, K.D., D.N. Kulkani and U.M Ingle, 1991. respiratory distress syndrome. *Crit. Care Med.*, 24: Sorghum malt-based weaning food formulations: 241-246.
- Radha, C., Kumar, P.,R. and Prakash ,V. (2007). Properties, functional properties, nutritive value. *Food Nutr. Bull.*, 13: 322-329.
- Kreger-van Rij N.,J.,W. (1984). *The Yeast: A Taxonomic study*, 3rd ed, Elsevier Science Publishers. Amsterdam.
- Krivanek, A.F., Groote, H.,D. Gunaratna, N.,S. Diallo, A.,O. and Friesen, D. (2007). Breeding and Disseminating Quality Protein Maize (QPM) for Africa. In review; submitted to the *Afri. J. of Biotechnol*, 56,234-242.
- Kulkarni, K. D., Kulkarni, D. N. and Ingle. U. M.( 19991). Sorghum malt-based weaning food formulations: Preparation, functional properties, and nutritive value. *Food Nutr. Bull.*1991; 13(4).
- Lalude, L.O., and Fashakin. J.B. (2006). Development and nutritional assessment of a weaning food from sorghum and oil – seeds. *Pakistan. J. Nutr.* 5 (3): 257-260.
- Latta, M. and Eskin,M.(1980). A colorimetric method for phytate determination. *J.Agric.Food Chem.*, 28: 1313-1315.
- Lay, M.G. & Fields, M. L. (1981). Nutritive value of germinated corn and corn fermented after germination. *Journal of Food Science* 46, 1069-1073.
- Lazos, E.S., Aggelousis, G. & Bratakos ,M. (1993). The fermentation of trahanas: a milk-wheat flour combination. *Plant Foods for Human Nutrition* 44, 45-62.
- Leistner, L. (1990).Mould-fermented foods: recent developments. (In 'Proceedings of the

- International Conference on Biotechnology & Food' [see FSTA (1991) 23 4B18].  
*Food Biote* 45.
- Lestienne, I., Rivier, C.M., Verniere, C.I., Rochette, I., and Treche, S. (2005). The effects of soaking of whole, dehulled and ground millet and soybean seeds on phytate degradation and Phy/Fe and Phy/Zn molar ratios. *Int. J. Food Sci. Tech.* 40(4): 391-399.
- Lesheme, I Caporiccio, B., Besancon, P., Rochette, I., Treche, S., (2005): Relative contribution of Phytate, Fibers and Tannins to low Iron and Zinc in vitro solubility in pearl millet (*Pennisetum glaucum*) flour and grain fractions. *J. Agric. Food Chem.*, 53 (21):8342-8348.
- Liener, I. E. and Kakade, M. L. (1980). Protease inhibitors. In Liener, I. E. (ed.) *Toxic Constituents of Plant Foodstuffs*. Academic Press, New York, pp. 7-71.
- Mahgoub, S.E.O., 1999. Production and evaluation of weaning foods based on sorghum and legumes. *Plants Foods Hum. Nutr.*, 54: 29-42
- Makkar, H. (1993). Anti-nutritional factors in foods for livestock. In: Gill, M., Owen, E., Pollot, G.E., Lawrence, T.L.J. (Eds), *Animal Production in Developing Countries*. Occasional publication No. 16. British Society of Animal Production, pp: 69-85.
- Mallesh NG, Daodu MA, Chandrasekhar A (1989). Development of weaning food formulations based on malting and roller drying of sorghum and cowpea. *Int. J. Food Sci. Technol.* 24: 511-519.
- Marero, L.M., Pajumo, E.M. Librando, E.C. (1988). Technology of weaning food formulation prepared from germinated cereals and legumes. *J. Food Sci.*, 53, 1391 – 1395.
- Maxson, E.D., Rooney, L.W. Evaluation of methods for tannin analysis in sorghum grain. *Cereal Chem.* 1972, 49: 719–729.
- Mbata, I., Ikenebomeh, M. J. and Alaneme, J. C.(2009). Studies on the microbiological nutrient composition and antinutritional contents of fermented maize flour fortified with bambara groundnut (*Vigna subterranean L*). *African J. Food Sci.* (3) 65-171.
- Mekibeb, F.(2009). Farmers' breeding of sorghum in the center of diversity, Ethiopia. I: Socio-ecotype differentiation, varietal mixture and efficiency. *Maydica* 54: 25-27.
- Meyell, G. G. and Meynell, E. (1970). *Theory and Practice in Experimental*

- Bacteriology. 2nd Edn, Cambridge Univ. Press, U.K. P. 347.
- Mihailovic, V., Mikic, A., and Ćupina, B. (2004). Botanical and agronomic classification of fodder pea (*Pisum sativum* L.). *Acta Agriculturae Serbica*, IX, 17, special issue, 61-65.
- Mihailovic, V., Mikic, A., and Ćupina, B. Manojlovic, M., Krstic, Đ., Čabilovski, R., Vasiljevic, S. and Halmajan, H., V. (2007). Potential of annual legumes for forage and green manure production. *Scientifical Papers, Faculty of Agriculture, Timișoara, Romania*, XXXIX, I, 249-254.
- Miller, E. (1996). Minerals. In: *food chemistry* chapter 4 (O.R. Fennema, ed.). Marcel Dekker Inc. New York. Basel. Hong Kong.
- Ministry of Economic Development and Cooperation (MEDaC) (1999). Survey of the Ethiopian Economy. Review of post reform developments (1992/93-1997/98).
- Mital, B.K. & Garg, S. K. (1995). Anticarcinogenic, hypoholesterolemic and antagonistic activities of *Lactobacillus acidophilus*. *CRC Critic. Rev. Microbiol.* 21, 175-214.
- Mohammed, H., Isam, A., Mohamed, A. and Elfadil, B. (2011). *Nutritional Evaluation of Sorghum Flour (Sorghum bicolor L. Moench) During Processing of Injera* International Journal of Biological and Life Sciences 7:111-123.
- Mohiedeen, I. E. Tinay, A. H, E. Elkhalya, A. E. O. E., Babiker, E and Mallasiy, O. L. (2010). Effect of fermentation on in vitro protein digestibility, protein fractions and amino acids composition of maize (*Zea mays* Linnaeus) cultivars. *Elec. J. Env. Agricult. Food.Chem.* 9: 838-847.
- Monjula, S. and John, E. (1991). Biochemical changes and *in vitro* protein digestibility of endosperm of germinating *Dolichos lablab*. *J. Sci. Food Agric.* 55 :429-438.
- Murdock, F.A. & Fields, M.L. (1984). B-vitamin content of natural lactic acid fermented cornmeal. *Journal of Food Science* 49, 373-375.
- Murty, D, S. and Renard, C. (2001). Sorghum. In *crops in tropical Africa*. Raemaekers, R. H (ed.). pp 68-96. Brussels. Belgium.
- Mosha, A.C., Lorri, W.S.M. (1987). High-nutrient-density weaning foods from germinated cereals. In: Alnwick D, Moses S, Schmidt OG, eds. Improving young child feeding in Eastern and Southern Africa. Nairobi, New York, Stockholm:

IDRC, UNICEF, SIDA, 288-99.

Nnam, N.M. (1995). Evaluation of nutritional quality of fermented cowpea (*Vigna unguiculata*) flours. *Ecology of Food and Nutrition* 33, 273-279.

Oberleas, D., (1983): Phytate content in cereals and legumes and methods of determination. *a Cereal.*, 28: 352-35

Obizoba. I.C. & Atii, J.V. (1991). Effect of soaking, sprouting, fermentation and cooking on nutrient composition and some anti-nutritional factors of sorghum (*Guinea*) seeds *Plant Foods for Human Nutrition* 41, 203-212.

Obizoba, I.C. & Egbuna, H.I. (1992). Effect of germination and fermentation on the nutritional quality of bambara nut (*Voandzeia subterranea* L. Thouars) and its product (milk). *Plant Foods for Human Nutrition* 42, 13-23.

Odunfa, S.A. & Komolafe, O. B. (1989). Nutritional characteristics of *Staphylococcus* species from fermenting African locust bean (*Parkia biglobosa*). *Nahrung* 33, 607-615.

Olanajo, S.A., Omuetti, O., Ajomale, and Ogunbodede, B.A. (2007). Development of Quality Protein Maize: biochemical and agronomic evaluation, tropical and subtropical agro ecosystem, 7:97-104, Institute of Agricultural research and Training, Obafemi Awolowo University.

Onilude, A.A., Sanni, A.I. and Ighalo, M.I. (1999). Effect of process improvement on the physico-chemical properties of infant weaning food from fermented composite blends of cereal and soybeans. *Plant Foods for Hum. Nutr.* 54: 239-250.

Osman, M.A. (2009). Effect of germination on nutritional quality of pearl millet. *Journal of Agriculture and Technology* 3(8):1-7.

Osman, M., A. Reid, M. R. and Weber, C.W. (2003). The effect of feeding tepary bean (*Phaseolus acutifolius*) proteinase inhibitors on the growth and pancreas of young mice. *Pakistan J. of Nutrition* 2:111-115.

Paredes-López, O. & Harry, G.,I. (1988). Food biotechnology review: traditional solid-state fermentations of plant raw materials - application, nutritional significance and future prospects. *CRC Critical Reviews in Food Science and Nutrition* 27, 159-187.

- Perez-Consesa, D., G. Ros and M.J. Periago, 2002 Protein quality of infant cereals during processing. *J. Cereal Sci.*, 36: 125-133.
- Poppel, G.,Y. & Schaafsma,G. (1996). *Cholesterol lowering by a functional yoghurt*. Food Ingredients Europe, Conference proceedings, 31-32.
- Prasanna, B.,M., vasal, S.,K., kassahun, B.,and Singh, N.,N. (2001). Quality Protein Maize,direc torate of maize research, Indian agricultural research institute. New Delhi 110 012, In. current sci., vol. 81, no. 10, 25: 1308- 1319.
- Rashid, S. (2010). Staple Food Prices in Ethiopia. In *Variation in staple food prices: Causes, consequence, and policy options, Maputo,Mozambique, 25-26 January 2010*. Retrieved from [http://programmes.comesa.int/attachments/186\\_AAMP%20Ethiopia%20backg round % 20 paper% 20\(5%20Jan\).pdf](http://programmes.comesa.int/attachments/186_AAMP%20Ethiopia%20backg%20round%20paper%20(5%20Jan).pdf)
- Reddy, B.S., Ekelund, G., Bohe, M., Engle, A. & Domellöf, L. (1983). Metabolic epidemiology of colon cancer: Dietary pattern and fecal sterol concentrations of three populations. *Nutr. Cancer* 5, 34-40.
- Reed, G. (1981). Use of microbial cultures: yeast products. *Food Technology* 35, 89-94.
- Rice, A.L., L. Sacco, A. Hyder and R.E. Black, 2000, Malnutrition As An Underlying Cause Of Childhood Deaths Associated With Infectious Diseases In Developing Countries. *Bull. World Health Organ*, 78: 1207-1221.
- Roos, U., Caneva, P., Escher, F. & Amadò, R. (1990). Influence of fermentation on the phytic acid content of conventionally produced bread. *Processing and Quality of Foods, Vol. 2, Food Biotechnology: Avenues to Healthy and Nutritious Products*. ed. Z. e. al. ISBN 1 85166 496 3, pp 2.105 2.110.
- Robertson, L.G., Monredon, F.D., Dysseler, P., Guillon, F., Amado,R. and Thibault, F.J. (2000). Hydration properties of dietary fibre and resistant starch: a European collaborative study. *Lebensm. Wiss. -Technology*. 33: 72-79.
- Sahana, P., and Fauzia, H. (2003). Fermented Cereal from Indigenous Raw Materials, Food Microbiology section, Institute of Food Science and Technology, Bangladesh Council of Scientific and Industrial Research, Bangladesh, *Pak. J. of Nutr.* 2 (5): 289-291
- Sandberg, A.S. 2002. Bioavailability of minerals in legumes. *Br. J. Nutr.* 88(3): S281-S285.

- Schneide, K. & Anderson, L.( 2010). Yield Gap and Productivity Potential in Ethiopian Agriculture : Staple Grains & Pulses *Prepared for the Farmer Productivity Team, of the Bill & Melinda Gates Foundation*, EPAR Brief No. 98.
- Singh, M. and Kirkorian, A. D. (1982). Inhibition of trypsin activity *in vitro* by phosphate. *Journal of Agriculture Food Chemistry* 30:700- 703.
- Shahani, K.M. (1983). Nutritional impact of lactobacillic fermented foods. *Nutrition and the Intestinal Flora*. ed. B. Hallgren ISBN 91 22 00593 5.
- Shimelis Kenaw. (2009). Development of rice based baby foods, a thesis submitted in partial fulfillment of the requirements for the MSc Degree in Food Engineering, Department of Chemical Engineering, Faculty of Technology, and Addis Ababa University.
- Singh, U., Kherdekar, M. S. and Jambunathan, R. (1982). Studies on Desi and Kabuli chickpea (*Cicer arietinum* L) cultivars. The levels of amylase inhibitors, levels of oligosaccharide and *in vitro* starch digestibility. *J. Food Sci.* 47: 510-512.
- Sneath, P. A., Mair, N.S., Sharpe, M.E. and Holt, J.G. (ed)(1986). *Bergey's Manual of Systematic Bacteriology*, Vol. 2. Williams and Wilkins Co Baltimore.
- Snedecor, G.W, and Cochran, W.G. (1967). *A statistical method*. Iowa State University Press, Ames, Iowa, USA.
- Sofi, P.A., Wani, S.A., Rather, A. G., and Wani, S.H. (2009). Quality Protein Maize (QPM): Genetic manipulation for the nutritional fortification of maize, *J. of Plant Breeding and Crop Sci.* Vol. 1(16).pp.244-253.
- Solsulski, F.W. (1962). The centrifuge method for determining flour absorptivity in hard red spring wheats. *Cereal Chem.* 39: 344-347.
- Speek, A.J., Speek, S. S. & Schreurs, W.H. (1988). Total carotenoid and beta-carotene contents of Thai vegetables and the effect of processing. *Food Chem.* 27, 245-257.
- Stanbury, P.F., Whitaken, A., and Hall, S.J. (2003). *Principles of Fermentation Technology*, 2<sup>nd</sup> edn, Elsevier Science Ltd.
- Tadesse, E. (1975). Tef (*Eragrostis tef*) cultivars. Morphological and classification. Part II. *Agricultural Experimental Station Bulletin*, 66, Addis Ababa University, College of Agriculture, Dire Dawa, Ethiopia.

- Tanno, K., and Willcox, G. (2006). The origins of cultivation of *Cicer arietinum* L. and *Vicia faba* L.: early finds from Tell el-Kerkh, north-west Syria, late 10th millennium B.P. *Vegetation History and Archaeobotany* 15(3), 197–204.
- Taranto, M.P., Sesma, F., Pesce de Ruiz, H. A. & Valdez, G.F.d. (1997). Bile salts hydrolase plays a key role on cholesterol removal by *Lactobacillus reuteri*. *Biotechnology Letters* 19, 845-847.
- Tavassoli, A.( 1986). The cytology of *Eragrostis* with special reference to *E. tef* and its relatives. Ph.D. dissertation, London University, London, UK.
- Udensi, E. A., Onwuka, G. I. and Oyewer, C. R. (2005). Effect of autoclaving and boiling on some antinutritional factors in *Mucuna sloanie*. *Nigeria Food J.* 23:53-58.
- Ullah, u., Muhammad, A., and Arifa, F.(020100). Chemical and Nutritional Properties of Some Maize, (*Zea mays* L.) Varieties Grown in NWFP, Pakista *Pakistan Journal of Nutrition* 9 (11): 1113-1117.
- Uraga, K., and Narasimha, H.V. (1997). Effect of natural fermentation on the HCl-extractability of minerals from Teff (*Eragrostis tef*). *Bull. Chem.Soc. Ethiop.* 11, 3-10.
- Vaintraub, I.A., and Lapteva, N.A. (1988).Colorimetric determination of phytate in unpurified extracts of seeds and the products of their processing. *Anal. Biochem.*, 175: 227 -230.
- Vaishali, V.A., Medha, K.G., & Shashi. A.C. (1997). Effect of natural fermentation on in vitro bioavailability in cereal-legume mixtures. *Int. J. of Food Science and Technology* 32, 29-32.
- Varela, G., Moreiras, O., Carbajal, A. and Campo, M. (1995). *National Study in Nutrition and Alimentation 1990/91*; Publicaciones del Instituto Nacional de Estadística: Madrid,
- Vasconcelos, A. T., Twiddy, D. R., Westby, A. and Reilly, A.J.A.( 1990). Detoxification of Cassava during Gari Processing. *International Journal of Food Science and Technology* 25: 198-203.
- Vavilov, N.I. (1951). The origin, variation, immunity and breeding of cultivated plants. *Chronica Botanica* 13: 1-366.
- Weaver, C.M. and Kanna, S. ( 2002). Phytate and mineral bioavailability In: N.R. Reddy

- and S.K.Sathe, editors food phytates. CRC press boca raton. P.211-224.
- Wharton, B. (1989). Weaning and child Health. *Annu. Rev. Nutr.* **9**: 377-94.
- World Bank (2006). Ethiopia at a glance. Retrieved from [http://devdata.worldbank.org/AAG/eth\\_aag.pdf](http://devdata.worldbank.org/AAG/eth_aag.pdf).
- World Bank (2007). Explaining Sources of Food Price Inflation in Ethiopia: A Just in Time Policy Note
- Yigzaw, Y., Gorton1, L., Akalu, G.and Solomon, T.(2001). Fermentation of teff (*Eragrostis tef*), grass-pea (*Lathyrus sativus*), and their mixtures: Aspects of nutrition and food safety, Lathyrus Lathyrism Newsletter 2
- Zohary, D. and Hopf, M. (1973) Domestication of pulses in the old world. *Science* 182, 887-894.

### Annex

Sensory evaluation score card using nine five point hedonic scale

Panelist code/name: \_\_\_\_\_ sample code: \_\_\_\_\_ date: \_\_\_\_\_

Sensory perception (score)	Sensory quality attributes			Overall acceptability
	Taste	Color	Odor	hedonic scale
1=dislike extremely				1=Extremely unacceptable
2=dislike moderately				2=moderately unacceptable
3=neither like nor dislike				3=neither acceptable nor unacceptable
4=like moderately				4=moderately acceptable
5=like extremely				5=Extremely acceptable