EFFECTS OF SPROUTING ON NUTRIENT COMPOSITIONS AND FUNCTIONAL PROPERTIES OF *Eleusin coracana* AND *Citrullus lanatus* FOR PRODUCTION OF SNACKS

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ABSTRACT

Food processing, particularly sprouting induces activation of hydrolytic enzymes that increases nutrient availability in food crops. Consumers interest for healthy and protein rich snack foods initiated the need to research for cheap, locally grown crops and fruits as alternative source of nutrients. Eleusin coracana (Finger millets) and Citrullus lanatus (water melon seeds) are reported to be rich source of proteins and could be used as alternative sources of protein in snack foods. This study was therefore designed to determine the effects of sprouting on nutrient compositions and functional properties of finger millets and water melon seeds and to determine the nutrient quality of the sprouted seed flours in snacks. Finger millets and water melon seeds were sprouted separately for nine days. Sprouts were evaluated for proximate, vitamins, anti-nutrients and functional properties. Bread and cookie were produced from composite flour blends of (white flour, sprouted finger millets and water melon seeds) in the following ratios; 100:0:0, 80:20:0, 70:20:10, 60:30:10 and 50:30:20. Highest protein contents were observed in seeds sprouted for six days and ranged from 12.34 to 22.61% and 22.38 to 28.63% for finger millets and water melon seeds respectively. There was a significant difference ($p<0.05$) in vitamin B_3, B_9 C and E. Anti-nutrients of the sprouts were in lower concentrations. Sprouts chosen for composite flour mixes were those that coincided with maximum protein and vitamin values. Sprouting improved functional properties of composite flour mixes. Protein contents of bread and cookie samples were enhanced with increased level of substituted sprouted seed flours. Sensory scores indicated that panelist expressed preference for snacks produced from composite flour ratios 100:0:0, 80:20:0 and 70:20:10. Cost values for bread and cookie had close ratings, however, bread and cookies substituted with sprouted flour blends may be recommended for consumption. Shelf life of bread ranged from 4-7 days while cookies lasted for 6-10days before spoilage occurred. The total aerobic bacterial and fungal counts ranged from 1.0×10^2 cfu/g to 1.8×10^6 cfu/g and 2.5×10^1 cfu/g to 1.2×10^3 cfu/g for bread, 3.7×10^5 cfu/g to 1.6×10^6 cfu/g and 3.0×10^1 cfu/g to 1.4×10^3 cfu/g for cookie. Sprouting proved to be of beneficial use for improving nutrient quality and functionality of finger millets and water melon seed flours. Enrichment of sprouted finger millets with water melon seed blends improved protein quality in snacks. Thus, may possibly be used in confectionaries for preparing protein rich and gluten free foods following additional nutritional studies.

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CHAPTER ONE

1.0 INTRODUCTION

1.1 Background to the Study

Sprouting is an important aspect of food processing in the world. It improves nutritive values and enhances sensory properties of foods following their unique enhancing features such as colour, flavour and bioactive components (Gan *et al*., 2019). Sprouted seeds are consumed in form of ready-to-eat sprouts, or may be further processed into flour for making food products. The use of sprouted food crops for making human-based food products has been reported. Falcinelli *et al.* (2018) reported the use of sprouted wheat in making whole grain bread. Abioye *et al.* (2018) reported the use of sprouted finger millet and African yam bean for the production of good quality cookies. Snacks are desirable for nutrient improvements and protein fortification due to their good eaten quality.

The consumption of snack foods such as bread and cookie has greatly increased especially in developing countries. In Nigeria, bread has become one of the most widely consumed nonindigenous food product, because of their ready-to-eat convenience and long shelf life. White flour which has been the major ingredient for production of snacks over the years are carbohydrate based food raw materials and protein deficient. Efforts have been made to promote the use of composite flours for production of snack foods thereby decreasing the use of white flour as well as producing protein-rich snacks (Akubor and Ukwuru, 2005; Ubbor and Akobundu, 2009; Udeme *et al.,* 2014; Atobatele and Afolabi, 2016; Taiwo *et al*., 2017). Composite flour is a mixture of flour from locally grown crops such as cereals, legumes or tubers to replace a proportion of white flour. In several parts of the world, the successful uses of composite flour for snacks

have been reported. Ubbor and Akobundu, (2009) reported the use of composite flour of watermelon seed, cassava and wheat in making cookie. Udeme *et al*. (2014) also reported the successful use of composite flour of wheat and potato flour blends in bread making. The authors reported that better nutritional quality baked products could be derived from composite flour. The use of cereals and legumes for composite flour has received significant attention because legumes are rich in lysine, an essential amino acid that is insufficient in most cereal crops (Olagunju and Ifesan, 2013). Also due to concerns for metabolic disorders resulting from consumption of white flour snack foods, consumers are shifting interest to the use of locally grown crops which are rich in protein and gluten free for production of cereal-based human foods, including snacks.

Finger millet (*Eleusin coracana*) is an important cereal crop popularly known as "African wonder plant" and has considerable potentials to be used as industrial raw material for making cereal-based human foods. It is called "Tamba" in hausa or "Raji" in Indian and serve as food staple for most African countries including Nigeria (Obilana and Manyas, 2002; Saleh *et al*., 2013). Its flour serve as source of energy and other valuable nutrients such as Protein, Dietary fibre, Minerals (especially calcium and potassium an essential macro-nutrient necessary for growing children, pregnant women and aged), and vitamins (Jideani, 2012; Saleh *et al*., 2013; Devi *et al.,* 2014; Gull *et al*., 2015; Kumar *et al.,* 2016; Vinoth and Ravindran, 2017). The grain is gluten-free with low-glycemic index thus, beneficial for those suffering from metabolic disorder such as celiac diseases, diabetes, and obesity.

Water melon **(***Citrullus Lanatus)* is a tropical fruit crop, highly nutritious and thirst quenching (FAO, 2011). The seeds are obtained from the rind of water melon fruit when cut open. The seeds are rich source of protein, amino acid (lysine an essential amino acid which is readily digestible), fats, dietary fibre, vitamins and minerals (magnesium,

calcium, potassium, iron, phosphorus and zinc (Braide *et al*., 2012). Water melon seeds contain high fat contents which has made them more efficient in baking (El-Adawy, 2001; Nasr and Abufoul, 2004). Enrichment of cereal-based baked foods with water melon seed has been reported. Olorode *et al*. (2017) reported the use of seeds from water melon, pawpaw and golden melon as substitutes for wheat in making cookies. Recently, blends of water melon seed were fortified with rice and murunchi flour for production of "masa" a traditional food staple popularly consumed in northern Nigeria. Addition of various proportions of sprouted finger millets and water melon seeds flour(s) in white flour could serve as a way of increasing the nutritive values in terms of protein and vitamins as well as lowering the gluten levels in white flour, thereby improving protein quality in snacks and decreasing the effects of metabolic disorder.

This study was therefore undertaken to determine the effect of sprouting on nutrient compositions and functional properties of finger millet and water melon seeds and to determine the nutrient quality of the sprouted seed flours in snacks. The microbiological qualities of the snack were also determined.

1.2 Statement of the Research Problem

Sprouting is not a common house-hold food processing method and consumer's knowledge on the use of sprouting technique as an alternative method to improve nutrient quality of food raw materials in developing countries including Nigeria is very poor.

More so, the fabrication of protein rich snacks has squeezed due to total dependence on use of white flour a major ingredient for making snacks in most industries, but is majorly carbohydrate based, protein deficient and high in gluten components. Consuming white flour snacks has significant effects on metabolic disorders. Hence,

consumers are shifting interest to use of locally grown crops which are rich sources of protein and gluten free for making cereal-based foods, including snacks

Most promising protein rich and gluten free local food crops such as finger millets and water melon seeds are given less attention and are underutilized. The primary challenge to wide consumption includes lack of informed knowledge on use of the crop and anti-nutrient components in seeds (Aremu and Ibirinde, 2012; Abioye *et al*., 2018). This has limited their consumption.

1.3 Justification for the Study

Sprouting improves nutrient availability, reduces anti-nutrients and improves functionality (Oghbaei and Prakash, 2016). Sprouting finger millets and water melon seeds will help increase their nutritive values in terms of proteins and vitamins, reduce anti-nutrient components and improve their functionality.

Finger millets are good sources of protein, gluten free and have potentials for making nutritive food. This has made them desirable for fortification in confectionaries. Water melon seeds are good source of proteins, excellent functional properties and efficient in baking. Enrichment of sprouted finger millet flour with water melon seed blends in a proportion of white flour will increase nutrient values and lower gluten levels in flour. This will also improve protein quality in snacks and decrease the effects of metabolic disorders.

Several studies have been done on use of sprouted grains/cereals in making cereal-based baked food. Little or no attempt has been made on determining the effects of sprouting on nutrient compositions of finger millets and water melon seeds, and supplementing a portion of white flour with the sprouted seeds in snack foods. This has justified the use

of composite flour of sprouted finger millets and water melon seeds for making bread and cookie.

1.4 Aim and Objectives of the Study

The aim of this study was to determine the effects of sprouting on nutrient compositions and functional properties of finger millet and water melon seeds, and to determine the nutrient quality of the sprouted seed flours in snacks.

The specific objectives of this study are to determine;

i. the drying rates of sprouted finger millet and water melon seeds.
ii. the effects of sprouting on nutrient composition and functional properties of finger millet and water melon seeds.
iii. the effects of graded levels of sprouted finger millet and water melon seed (flours) ratio on the quality attributes of snacks made from the flour.
iv. the shelf life and microbiological qualities of the resulting snack products
v. the Sensory evaluation and cost on quality attributes of the resulting snack products

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Millets

Millets are grain crops belonging to the family *Poaceae* of the monocotyledon group. They are excellent sources of carbohydrates, protein, fatty acids, minerals, vitamins, dietary fibre and polyphenols. They provide most of the nutrients required for normal functioning of human body. Millets have several nutraceutical, nutritional and health promoting functions especially the high fibre content and nature of starch help in reducing the risk of diabetes and other health related diseases. In the inner ecosystem, millets act as prebiotic feeding micro-flora and help to hydrate our inner body colon to keep us from being constipated (Obilana and Manyasa, 2002; Yang *et al*., 2012).

Epidemiological studies have indicated that consumption of whole grain cereals/millets and their products regularly, can protect against the risk of cardiovascular disease, diabetes, gastrointestinal cancer and a range of other disorders by decreasing the levels of triglycerides and C-reactive protein (Mckeown, 2002). Niacin in millets helps to lower cholesterol levels. Millets show high antioxidant activity, non-allergenic and gluten free. Hence, millets are advisable for patients with celiac problems (Chandrasekara and Shahidi, 2010). Millets largely contribute to food and nutritional security of the country, and are major staples for millions of people especially in areas where it is being cultivated in the world.

Millets are classified into major and minor millets according to the areas grown and size of grains. The major millets are Sorghum (jowar) and pearl millet (bajra). The minor millets include finger millet (raji/African millet), kodo millet, foxtail millet (kangni/Italian millet), proso millet (common millet), barnyard millet and fonio millets.

All these millets fits wide range of cropping systems and adapt to changing environmental conditions especially during unpredicted seasons or tropical rainy season. They are grown mostly in low fertile soils in which most cereals fail to give substantial yield (Adekunle, 2012).

2.2 Finger Millet

2.2.1 Plant portrayal of finger millet

Finger millet *(Eleusine coracana*) is an important grain crop belonging to the family *Poaceae*, and originated in Ethiopia (Shiihii *et al*., 2011). It is known as ragi in some parts of India or Tamba in northern parts of Nigeria, particularly in Kaduna State (Jideani *et al*., 1996; Takhellambam *et al*., 2016). The grain is widely cultivated in subtropical and tropical regions of India and Africa where it serves as staples (Obilana and Manyas, 2002; Saleh *et al*., 2013). It is a dwarf plant with characteristics finger like terminal inflorescence. The height of a matured finger millet plant ranges from 30-150 cm, and the seeds are very small as mustard. It is better adapted to areas with higher rainfall (600mm-1,200mm) especially acidic soils and matures within 100-130 days depending on the variety and growing conditions. It can adjust itself to various agro climatic conditions which make it have highest productivity among other millets (Gopalan *et al*., 2002). The millet is a small seeded (1.2-1.8 diameter) minor grain crop with brick red-coloured seed coat (testa), embryo and endosperm. The presence of five layered testa reflects its uniqueness among other millets such as foxtail millet, pearl millet, kodo millet and proso millet. Among the various varieties of finger millet such as brown, yellow, white, red or violet colour, the red coloured are cultivated extensively all over the world.

The grain is comparatively resistant to storage insect pest which makes the crop an important source of food during famine as the grain can be stored for about 50 years

without much loss due to deterioration. Although, the grain does not enter the international markets as an item of trade, but it is an important crop in the areas of adaptation. Pictorial demonstration of finger millet grain is seen in the Plate I.

Plate I: Finger Millet Grains.

Source: (Gopalan *et al*., 2002).

2.2.2 Classification of finger millet

Table (2.1) below shows the classification of finger millets according to their Rank, Scientific names and Common names.

Table 2.1: Classification of Finger Millets According to Rank, Scientific and Common Names

Rank	Scientific names and common names
Kingdom	**Plantae**- plant
Subkingdom	**Tracheobionta**- Vascular plants
Superdivision	**Spermatophyte**- Seed plants
Division	**Magnoliophyta**- Flowering plants
Class	**Liliopsida**- Monocotyledons
Subclass	**Commelinidae**
Order	**Cyperales**
Family	**Poaceae/ Gramineae**- Grass family
Genus	***Eleusine* Gaertn**- goose grass
Species	***Eleusine coracana***- Finger millet

Source: Natural Resources Conservation Service

2.2.3 Nutrient compositions of finger millets

Finger millet being indigenous minor millet is rich in quality protein (6 –13 %), Prolamin is the major fraction on finger millet protein, being 24.6 to 36.2 % of total protein (Lupien, 1990). Antony and Chandra (1998) reported a 99.1 mg soluble protein per 100 g in finger millet. The prolamin fraction contains higher proportion of glutamic acid, proline, valine, isoleucine, leucine and phenylalanine but low, arginine and glycine. Finger millet contains 44.7 % essential amino acids (Mbithi-Mwikya *et al*., 2000) of the total amino acids which is higher than the 33.9 % essential amino acids in FAO

reference protein. The total content of carbohydrates in finger millet has been reported to be in the range of 65 to 75 % (Pore and Magar, 1979; Hulse *et al*., 1980; Joshi and Katoch, 1990; Bhatt *et al*., 2003). Carbohydrate contents in finger millets are slowly digested and absorbed than those present in other cereals thus, leading to a reduction in postprandial glucose.

The millet also contains about 2.5 to 3.5 % minerals. It has the highest amount of calcium (344 mg) and potassium (408 mg) compared to other cereals and millets (Devi *et al.,* 2014; Kumar *et al.,* 2016; Vinoth and Ravindran, 2017). The iron content of finger millet ranged from 3.3 to 14.8 mg. Calcium deficiency may lead to bone and teeth disorder, while iron deficiency may lead to anaemia. These deficiencies can therefore be overcome by introducing finger millet in our daily diets. Finger millets are excellent sources of vitamins especially the vitamins B, (water soluble vitamins) and vitamin E. but available data are very meagre. The crude fat content in finger millet is within the range of 1.3 to 1.8 % (Malleshi and Klopfenstein, 1998; Lupien, 1990; Bhatt *et al*., 2003; Singh *et al*., 2003). It is high in polyunsaturated fatty acids, the major fatty acid in finger millet includes oleic acid, palmitic acid and little amount of linolenic acid. About 100 grams of finger millet have an average of 336 Kcal of energy in them. It is non-acid forming, and easily digested.

Finger millets are gluten free and hence safe for people suffering from gluten allergy and celiac diseases (Chandrasekara and Shahidi 2010). The grain contains phytates (0.48 %), tannins (0.61 %), phenolic compounds (0.3 – 3 %), 10 - 15 % dietary fibre and trypsin inhibitory factors which are regarded as “anti-nutrients” due to their metal chelating and enzyme inhibition activities, but can contribute to antioxidant activity, an important factor in resisting aging and metabolic diseases (Thompson, 1993).

Among cereals such as wheat, rice, barley and maize, finger millet grains have higher contents of polyphenols. These phenolics compound are not stored in the grain but are located in the outer aleurone layer, testa and pericarp of fruit which form the main components of the bran fraction and exist as free, soluble conjugates and insoluble bound forms (Viswanath *et al*., 2009; Shobana, 2009). The polyphenol contained in the seed coat make it show high antifungal and antibacterial activity compared to the whole flour extracts. The major biochemical benefits of polyphenols towards antifungal activity is that it forms free radicals due to oxidation of microbial membranes and cell components forms irreversible complexation with nucleophillic amino acids resulting to inactivation of enzymes. The major bound phenolics present in finger millets are ferulic acid (64 – 96 %) and p-coumaric acid (50 – 99 %). Flavonoids and tannins present in millet seed coat have multiple functions; they act as reducing agents (free radical terminators), metal chelators, oxygen quenchers and physical barrier to fungal invasion thus, providing resistance to grain against fungal attack.

The millets have high radical-scavenging activity compared to other cereals such as wheat, rice, barley and other millets. It is one of the most nutritious and healthy millets that supply a major portion of calories and protein to human body especially in people of low socio-economic groups (Kennedy *et al*., 2006).

Table 2.2: Nutrient Compositions of Millets and Other Cereals (mg/100 g)

Parameters/food types	Wheat	Sorghum	Rice	Finger millets	Pearl millet	maize
Protein	11.6	10.4	7.9	12.2	11.8	9.2
Fat	2.0	3.1	2.7	1.5	4.8	4.6
Ash	1.6	1.6	1.3	2.6	2.2	1.2
Crude fibre	2.0	2.0	1.0	3.6	2.3	2.8
Carbohydrate	71.0	70.7	76.0	72.6	67.0	73.0
Calcium	30	25	33	350	42	26
Iron	3.5	5.4	1.8	3.9	11.0	2.7
Thiamin	0.41	0.38	0.41	0.62	0.38	0.38
Riboflavin	0.10	0.15	0.04	1.19	0.21	0.20
Niacin	5.1	4.3	4.3	3.9	2.8	3.6

Source: Hulse *et al.,* 1980; Saleh *et al*., 2013.

2.2.4 Health benefits of finger millets

Health benefits associated with finger millet include delayed nutrient absorption, increased faecal bulk, transit time and fermentability characteristics, lowering of blood lipids, prevention of colon cancer, barrier to digestion and mobility of intestinal contents (Tharanathan and Mahadevamma, 2003). Finger millet contains a functional fibre fraction known as resistance starch (RS), this escapes enzymatic digestion, imparts beneficial effects by preventing several intestinal disorders (Annison *et al*., 1994). It

also provides benefits such as the production of desirable metabolites, including short-chain fatty acids in the colon, especially butyrate, which stabilizes colonic cell proliferation as a preventive mechanism for colon cancer. The dietary fibre, phenolics, minerals and vitamins embedded in the outer layer of finger millet seed coat, forms the part of the food and offer their nutritional and health benefits.

Finger millet is also recognized in management of various physiological disorders such as diabetes mellitus. Diabetes mellitus is characterized by hyperglycemia with alterations in carbohydrate, protein and lipid metabolism. Lower incidence of diabetes has been reported in finger millet-consuming population (American Diabetes association, 2005; Shobana *et al*., 2009; Kim *et al*., 2012). This may be due to higher fibre content and antinutritional factors in whole finger millets which are known to reduce starch digestibility and absorption (Lakshmi and Sumathi, 2002). Finger millet helps in management of hypertension, vascular fragility, hypercholesterolemia, prevention of oxidation of low density lipoproteins (LDLs) and improves gastrointestinal health (Devi *et al*., 2014). Lee *et al*. (2010) reported that finger millets help in reducing cardiovascular disease by reducing plasma tryglycerides in hyperlipidemic rats. Daily consumption of finger millet and its products can protect against the risk of cardiovascular, gastrointestinal cancers and other health diseases (McKeown, 2002).

Finger millets are gluten free, hence suitable for individuals suffering from celiac disease (Taylor *et al.,* 2006; Taylor and Emmambux, 2008; Chandrasekara and Shahidi, 2011a, 2011b), Celiac disease is a lifelong immune-mediated disorder, usually triggerd by the ingestion of gluten in genetically susceptible individuals. However, Finger millet has a potential to be used for producing foods for celiac patients. The calcium contents in finger millet helps in strengthening bones for growing children and aging adults.

Regular consumption of finger millets is beneficial for bones health and helps in reducing diseases such as osteoporosis at bay and could help to reduce risk of fracture. The millets are good source of antioxidant and phenolics. It has however been established that phytates, phenols and tannins can contribute to antioxidant activity in health, aging and metabolic diseases (Bravo, 1998). Methanol extracts of finger millet and kodo millet have been found to inhibit glycation and cross-linking of collagen (Hedge *et al*., 2002). Hence, there is need for potential usefulness of finger millet in protection against aging. Viswanathan *et al*. (2009) reported that seed coat extract of finger millet showed higher antimicrobial activity against bacillus cereus and aspergillus flavus than whole flour extracts. This indicated that finger millet seed coat can be used as natural antioxidant and food preservative.

Finger millet has long been used as a remedy for many infections including leukemia. Recently seed purified extract of finger millet gained the importance of anti-proliferative activity on K562 chronic myeloid leukemia because finger millet seeds contain a bi-functional complex of amylase-trypsin inhibitor more commonly called RBI (ragi bi-functional inhibitor), that inhibits amylase and trypsin simultaneously. Basically, RBI is a monomeric protein made of 122 amino acid containing five intra-molecular disulfide bonds and the gene responsible for the encoding. RBI has been cloned from finger millet seeds and expressed functionally in *Escherichiacoli* (Shivarag *et al*., 1981; Campos *et al*., 1983; Maskos *et al*., 1996; Sen *et al*., 2011; Sen and Dutta, 2012). The plant protease inhibitors (PIs) are multifunctional proteins required in diverse biological processes, such as redness, infection, extra- cellular matrix degradation, blood coagulation, apoptosis, tumor invasion and cancer metastasis and play very significant role in human health and disease management (Mosolove *et al*., 2001; Park and obha, 2004; Pandy *et al*., 2007). Finger millets can be recommended for

conditions of blood pressure, liver disorders, asthma, and heart weakness. It can also be recommended to lactating mothers in condition of lack of milk production, malnutrition, degenerative diseases and premature aging at bay.

2.3 Water Melon (*Citrullus lanatus)*

2.3.1 Plant description of watermelon

Water melon **(***Citrullus lanatus)* is a warm-season fruit crop belonging to the family *cucurbitaceae*. The fruit contain 92 % water, low in calories, highly nutritious and thirst quenching. It is an excellent source of lysine, (an essential amino acid readily digestible). Contains citrulline (an amino acid the body uses to make arginine) (Oyeleke *et al*., 2012), vitamins and important antioxidants (Braide *et al.,* 2012). It is one of the most popularly grown fruit during summer and mostly grown for fresh consumption of the flesh of a matured fruit. Water melon are sensitive to cold temperatures hence, thrives best in warm areas with sunlight and an average temperature ranging from 18 to 35 ^{o}C and a pH of 5 to 6.6 for optimum growth (FAO, 2011). The crop matures within three months after planting and yield from 5 to 72 t/ha. It grows as a vine and individual plants produce both male and female flowers and fruit size ranges from 2 to 14 kg, depending on the variety. The leaves are dark green and hearts shaped with three to seven lobes per leaf and are produced on trailing vines. The fruit shape and appearance are varied, ranging from round to cylindrical or oval. The fruit is usually large and smooth. The edible flesh may be pink, yellow or red depending on the varieties with many flat black seeds throughout. There are over 50 varieties of water melon grown worldwide with a wide range of sizes, shapes and colours. The most commonly produced varieties are Charleston Gray strains, Crimson sweet, Jubilee, All sweet, Sangria, triploid seedless and black diamond types (Maynard, 2001). Pictorial description of various varieties of water melon fruit and flesh colours are seen in Plate II.

Plate II: Water Melon Fruit and Flesh Colours

Source: Maynard, (2001)

2.3.2 Classification of water melon (*Citrullus lanatus)*

Table (2.3) below shows the classification of water melon (*Citrullus lanatus*) according to Rank, Scientific names and Common names.

Table 2.3: Classification of Water Melon Ranging from Rank, Scientific and Common Names

Rank	Scientific names and common names
Kingdom	**Plantae**- plants
Subkingdom	**Tracheobionta**- Vascular plants
Superdivision	**Spermatophyte**- Seed plants
Division	**Magnoliophyta**- Flowering plants
Class	**Magnoliopsida-** Monocotyledons
Subclass	**Delleniidae**
Order	**Cucurbitales**
Family	**Cucurbitaceae**- Cocumber family
Genus	***Citrullus* Schrad**- Water melon
Species	***Citrullus lanatus***- Water melon

Source: Natural Resources Conservation Service

2.3.3 Water melon seeds

Water melon seeds are obtained from the rind of water melon fruit when cut open. The seeds are highly nutritious and calorie free. They are rich sources of protein, minerals, Vitamins B, C and E, fats, carbohydrates and phytochemicals (Braide *et al*., 2012). Water melon seeds are known to have several economic benefits particularly in countries where cultivation is on the increase. The seeds are widely distributed and less utilised. In many countries, melon seeds are major soup ingredients and are used to thicken soups. Water melons seeds have been reported to have good baking qualities

and can be milled into flour to prepare snacks. Oils from the seeds have been used as a useful raw material in food and cosmetic industries (Jensen *et al*., 2011). Water melon seeds have been reported to possess several medicinal effects such as; anti-diabetic, Cardio protective effects, Anti-obesity and arthritic effects as well as anti-ulcerogenic effects (Alok *et al.,* 2011).

Pictorial description of wet and dried water melon seeds is seen Plate III

Plate III: Wet and Dried Varieties of Water Melon Seeds

Source: Jensen *et al*., (2011).

2.4 Nutrients in Food

2.4.1 Carbohydrates

Carbohydrates are polyhydroxy aldehydes or ketones. They are structural components of cell wall in plants, and exoskeleton of animals, they form part of RNA and DNA from which ribose and deoxyribose are linked by N-glycosidic bonds to purine and pyrimidine bases and forms the integral features of protein and lipids (glycoproteins and

glycolipids). Carbohydrates are one of the major classes of food. They serve as source of fuel and energy to humans. Carbohydrates comprise two major classes: simple carbohydrates and complex carbohydrates. Simple carbohydrates include monosaccharides and disaccharides. Monosaccharides are simple sugars or monosaccharide units. They are the simplest form of carbohydrate in that they cannot be reduced in size to smaller carbohydrate units by hydrolysis. The most abundant monosaccharide in nature and certainly the most important nutritionally is the 6-carbon sugar glucose. Disaccharides consist of two monosaccharide units joined by covalent bonds. Complex Carbohydrates/Oligosaccharides consist of short chains of monosaccharide units that are also joined by covalent bonds. Among the oligosaccharides, trisaccharides occur most frequently in nature. Polysaccharides are long chains of monosaccharide units. The major polysaccharides of interest in nutrition are glycogen, found in certain animal tissues, starch and cellulose, both of plant origin. All these polysaccharides consist of only glucose units. The major sources of carbohydrate foods are roots, tubers and cereals crops (Wardlaw *et al*., 2004).

2.4.2 Dietary fibre

Dietary fibres refer to a type of carbohydrate and lignin that cannot be digested by the human digestive system. They are found intact and intrinsic in edible plants foods such as cereals, fruits grains, vegetables etc. Dietary fibres are divided into two major classes; which include; soluble and insoluble fibre. The soluble fibres are pectins and gums while the insoluble forms are cellulose, hemicellulose and lignin. The soluble form dissolves in water in the intestinal tract to produce a gel which slows down movement of food through the intestine. The insoluble forms are not soluble in water. They stimulate peristalsis, rhythmic muscular contractions of the intestine which moves the digester along the digestive tract. Dietary fibres help to reduce the risk of

gastrointestinal problems such as constipation and diarrhoea. It helps to also slow down the rate at which carbohydrates are digested and absorbed. Water soluble fibres lower the levels of cholesterol in humans (Wardlaw *et al*., 2004)

2.4.3 Lipids

Lipids are bi-molecules which are soluble in organic solvents such as ether, chloroform, and acetone. They contain hydrocarbons and constitute protein and carbohydrates to make up the building block of the structure and function of the living cells. Lipids are divided into four groups these include, simple, compound (complex), derived and miscellaneous. Ths simple lipids includeds esters of fatty acids with various alciohols such as glycerol or cholesterol. Examples are tryglycerols, waxes cholesteryl esters and vitamin A and D esters. Compound lipids are esters of fatty acids in combination with both alcohols and other groups. Examples are phospholipids, glycolipids, cerebrosides, lipoprotein, sulfolipids and lipopolysaccahrides. Derived lipids are hydrolysis products of simple or combined lipids. Examples are fatty acids, monoacylglycerols and diacyglycerols. Straight chain and ring containing alcohols, sterols and steroids, while misllenous lipids include wax lipids, squalene, carotenoids, and vitamin Eand K. Lipids plays vital role in dietary constituents due to their high energy values, fat-soluble vitamins and essential fatty acids present in the fat of natural foods. The essential fatty acids cannot be synthesised by the body and are required for maintaining the body structure. Fatty acids are a rich source of energy. They are stored in adipose tissue where they act as thermal insulator in the subcutaneous tissue and around certain organs. Their mitochondrial oxidation makes up large amounts of acetyl CoA for TCA cycle catabolism, and in situations of low carbohydrate intake or use, as occurs in starvation or diabetes, the rate of fatty acid oxidation increases significantly with concomitant acetyl CoA accumulation. This causes an increase in the level of the ketone bodies

organic acids that can be deleterious through their disturbance of acid base balance but that also are beneficial as sources of fuel to tissues such as muscle and brain in periods of starvation. Dietary lipid has been implicated in atherogenesis, the process leading to development of the degenerative cardiovascular disease called atherosclerosis, cancer and obesity. Elevated levels of cholesterol (LDL) results in increased risk of chronic disease, while high levels of cholesterol (HDL) have been associated with decreased risk of coronary heart diseases (Wardlaw *et al.,* 2004).

2.4.4 Proteins

Protein is one of the primary biomolecules present in cells and tissues. The polymerization of L –amino acids through the synthesis of peptide bonds contribute to the formation and structural frame work of proteins. Proteins in foods become available for use by the body after they have been broken down into their component amino acids. Nine of these amino acids are considered essential; therefore, the quality of dietary proteins correlates with their content of these indispensable amino acids. In the body, proteins play many vital roles including functions in structural capacities, and as enzymes, hormones, transporters, and immunological protectors. Among other roles, an important concept in protein metabolism is that of amino acid pools, which contain amino acids of dietary origin plus those contributed by the breakdown of body tissue. The amino acids comprising the pools are used in a variety of ways; for synthesis of new proteins, growth and replacement of existing body proteins, for oxidation as a source of energy and synthesis of glucose, ketones, or fatty acids. The liver is the primary site of amino acid metabolism (Wardlaw *et al*., 2004).

2.4.5 Vitamins

Vitamins are complex organic compounds required by the body in trace amount for the maintenance and growth of living organisms. They are classified into two major group based on their solubility namely, water soluble and fat soluble vitamins. Water soluble vitamins include vitamins B complexes and C, whereas, vitamin A, D, E, and K are fat soluble. The Human body is unable to synthesize vitamins so, their intake through diet is necessary as they play a highly significant role in growth and development of the human body. Severe or even fatal diseases can be caused by deficiency of these vitamins. Vitamin deficiencies are associated with specific disease like, vitamin A associated with blindness, vitamin B_1 with beriberi, vitamin B_3 with pellagra, vitamin B_6 with anemia, vitamin C with scurvy and vitamin D with rickets (Asensi-Fabado and Munne´-Bosch, 2010). The different classification of vitamins is seen in Figure 2.1 below.

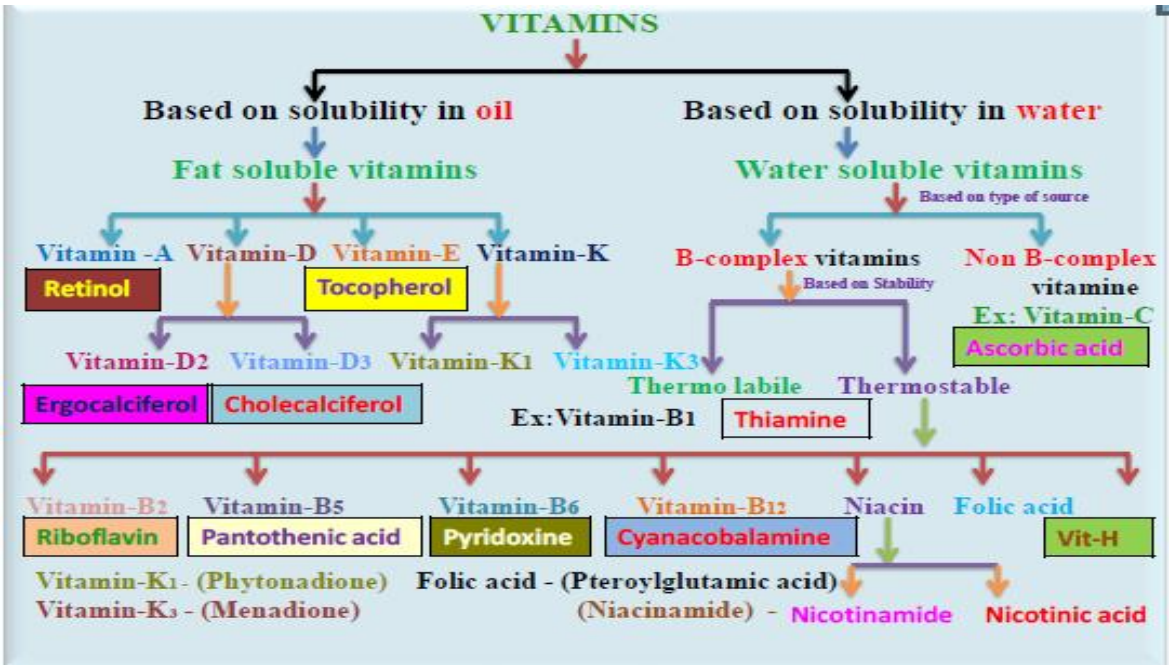

Figure 2.1: Classification of vitamins according to their solubility

Source: (McDowell, 2000; Asensi-Fabado and Munne´-Bosch, 2010)

2.4.5.1. Vitamin A

Vitamin A can be obtained both from plant and animal sources. Vitamin A from animal sources is known as retinoid while those from plant sources are called provitamin A carotenoids. Retinol, retinal, retinoic acid and retinyl esters are biologically active forms of retinoids. α-carotene, β-carotene and β-cryptoxanthin are the plant originated provitamin A-carotenoids which are converted into preformed vitamin A retinoids during digestion in the human body (WHFoods, 2017). Vitamin A plays a vital role in Vision; the retinol is oxidized to its aldehyde and retinal which complexes with a molecule in the eye called opsin. Within the photoreceptor cells of the retina are the rods which detect small amounts of light and are specialized for motion. The cones are specialized for colour vision in bright light. The both rods and cones possess specialized outer segment disks that contain high amounts of rhodopsin and iodopsin respectively. When a photon of light hits the complex the retinal changes from the 11-cis retinol form to the all-trans retinol form. These are initiating a chain of events which results in the transmission of an impulse up to the optic nerve. These compounds are often referred to as the "Visual pigment". Photoreceptor cells detect light and undergo a series of reactions which send signals to the brain where they are deciphered as a particular visual image. Vitamin A gives support to immune system and inflammatory systems, cell growth and development, antioxidant activity and promotes proper cell communication (WHFoods, 2017).

The Daily intake requirement of vitamin A for young males is 900 micrograms, for females 700 microgramsand for children 300-400 micrograms (WHFoods, 2017). Major sources of vitamin A include; dark leafy green vegetables, sweet potato, and animal products such as glandular meat, liver, egg yolk and red palm oil (rich in Provitamin A), Vitamin A is highly responsible for maintaining a normal surface of the eye (cornea)

and deficiency leads to drying of the eye surface resulting to a condition called Xerophthalmia. This can lead to blue cloudiness of the eye followed by ulcer formation. Poor dietary habits, malnutrition owing to improper balance between dietary consumption causes vitamin A deficiency. This is very high in patients with a medical history of cystic fibroses, sprue, and inflammatory bowel diseases etc. Some symptoms of vitamin A deficiency also occur in protein-energy malnutrition, not regarding whether vitamin A intake is adequate.

2.4.5.2 Vitamin D

Vitamin D is known for the treatment of rickets. It is required to maintain normal blood levels of calcium and phosphate that are in turn needed for the normal mineralisation of bone, muscle contraction, nerve conduction and general cellular function in all cells of the body. Vitamin D is very important for human body but exact intake dose is variable and has inconsistent pattern because exposure of sunlight also produce vitamin D in human skin. Ultraviolet B (UVB) wavelength of sunlight strikes the body and resultantly 7-dehydrocholesterol in human body is converted into cholecalciferol which is preliminary form of vitamin D. However, amount of cholecalciferol produced by UV-B is unpredictable due to type and nature of skin pigments, skin health and nature of sunlight. There are seven different types of vitamins D, however the most important are vitamin D_1 a molecular compound containing lumisterol and calciferol in 1:1 ratio. Vitamin D_2 is ergocalciferol, derived from ergosterol. And vitamin D_3 is cholecalciferol obtained form 7- dehydrochloesterol. Food sources are very limited which provide vitamin D but amount of this vitamin in body can be increased by increasing the exposure to sunlight (WHFoods, 2017).

Vitamin D increases the calcium absorption from food and reduces the losses through urine. It also plays an important role in enhancing the absorption of calcium and phosphorus from the intestine and helps to maintain calcium homeostasis. Vitamin D is also involved in maturation of white blood cells which plays vital role in immunity responses. Dietary intake is about 15 micrograms (600 International Unit) in daily basis for teenager and adult males and females (WHFoods, 2017). Dietary sources of vitamin D are required only when there is an inadequate exposure to sunlight. Vitamin D may be obtained from natural plant sources and supplied through fortified food such as bread, cereal, margarine, milk, pastries, yogurt, egg yolks etc. (WHFoods, 2017). Vitamin D deficiency cause the malformation and softening of bones a disease known as osteoporosis, osteomalacia or rickets. Research have shown that Vitamin D deficiency is reported to be increasing since 1980s due to increased use of sunscreens and decreased exposure to the sunlight (Faurschou *et al*., 2012).

2.4.5.3 Vitamin E

Vitamin E is an important naturally occurring antioxidant. It occurs in diets as a mixture of several closely related compounds known as tocopherol and tocotrienols. The active component of vitamin E is α-tocopherol. Being an antioxidant, vitamin E protects the membrane fats from oxidative damage and maintains cellular functioning. This vitamin also protects the food from oxidative damage during storage and processing (WHFoods, 2017). Vitamin E plays a significant role in preventing low-density lipoprotein (LDL) cholesterol from oxidative damage caused by free radicals. Deficiencies of vitamin E make the LDL cholesterols prone to oxidative damage and convert them into oxidized LDL. Oxidized LDL accumulates in the blood vessels and cause hardening of arteries known as atherosclerosis. Vitamin E also protects oxygen rich blood from free radicals and the heart during sex which is also called sex vitamin. Vitamin E acts as a

neuroprotector as well as anti-sterility factor which is essential for fertility of the male and the birth process of the female. Daily recommended intake of vitamin E is 15 milligrams for males and females of adult age while 5 milligrams for children (WHFoods, 2017). Vitamin E can be found in fortified cereals, seed oils like sunflower, legumes, whole grains, and green leafy vegetables (WHFoods, 2017). Deficiency of vitamin E is associated with cancer, heart attack, stroke, fibrocystic breast disease, epilepsy, diabetes, parkinson's disease, cataract, alzheimer's disease and increased erythrocyte haemolysis (WHFoods, 2017).

2.4.5.4 Vitamin K

Vitamins K are blood clotting factors. It was derived from German word “koagulation” which means blood clotting (Shearer *et al*., 2012; Shearer and Newman, 2014). The three biologically active forms of vitamin K are vitamin K_1 (phylloquinone) found in green vegetables, vitamin K_2 (menaquinone) which is synthesized by intestinal bacterial and vitamin K_3 (menadione). Vitamin K_1 is most prevalent and needed for photosynthesis of plants. K_2 is synthesized from K_1 and K_3 form by bacteria and other microorganisms. In human body, K_2 is synthesized by biological conversion of K_1 and K_3. And vitamin K_2 is not present in preformed form in plants but it is produced by fermenting bacteria through transformation of K_1 into K_2 (Hirota *et al*., 2013). Key functions of vitamin K are photosynthesis, antioxidants and energy generation by electron movement (Kurosu and Begari, 2010). Vitamin K plays a vital role in blood clothing. Blood clotting is very complex process because there is involvement of twenty different proteins for completion of clotting process and four of these proteins require vitamin K for their activity (Shearer *et al*., 2012; Shearer and Newman, 2014). In young person’s there is rare incidence of vitamin K deficiency but frequent in newborns because placenta prevent the extensive movement of vitamin K to fetus. Severe

gastrointestinal and liver diseases provoke the incidence of vitamin K deficiency (Shearer *et al*., 2012; Shearer and Newman, 2014).

Vitamin K especially K_1 and K_2 are important for bone health and its deficiency increases the risk of bone fracture. Osteoclasts are special type of the cells involved in demineralization of bones and make the minerals available for other body functions but too much demineralization can harm the bones. Vitamin K keeps the osteoclasts cells under controls and induces the programmed cell death to avoid the extensive production of osteoclast cells and to keep the demineralization under control (Atkins *et al*., 2009; Shearer and Newman, 2014). Under-carboxylatedosteocalcin is a disorder in which risk of bone fracture is increased especially hip fracture. This disorder is caused by lower carboxylation of osteocalcin protein. Vitamin K especially K_1 and K_2, regulate the carboxylation of osteocalcin proteins and strengthen the bones. Daily dietary requirement for vitamin K intake is 80 micrograms. WHFoods recommended 90 micrograms daily required intake for females and 120 micrograms for males and 55 micrograms for children (WHFoods, 2017). Vitamin K is widely available in our daily diets. Specifically, dark green leafy vegetables, eggs, meat, fish, dairy, and fermented plant foods are rich sources of vitamin K_1 and K_2. Vitamin K_3 is not naturally present in the dietary foods (Shearer *et al*., 2012; Shearer and Newman, 2014; WHFoods, 2017). Deficiency of vitamin K is quite rare, as it is widely distributed in many sources of food and also produced by the intestinal bacteria. However, vitamin K is associated with blood clotting disorders, coronary artery disease, osteoporosis, liver disease, cancer, crohn's disease, celiac disease, cystic fibrosis and ulcerative colitis, blood in urine and intestinal bleeding (Shearer *et al*., 2012; Shearer and Newman, 2014)

2.4.5.5 Vitamin B_1 (Thiamin)

Vitamin B_1 is a member of vitamin B complex also known as thiamin. Thiamin plays very important role in human health. They are involved in energy production especially in carbohydrates and fats metabolisms. (Kala and Prakash, 2003). Thiamin is a coenzyme for three multienzyme complexes that catalyses oxidative decarboxylation reactions; pyruvate dehydrogenase in carbohydrate metabolism, alpha-ketoglutarate dehydrogenase in citric acid cycle and branched chain keto acid dehydrogenase involved in the metabolism of valine, leusine and isoleusine. This vitamin is central in energy metabolism and its deficiency can seriously impair the energy metabolism. And because the body requires much energy to function properly, deficiency of vitamin B_1 can seriously affect the nervous system, digestion and heart. Structure development and integrity of brain cells are also dependent on vitamin B_1. The active form of thiamin; pyrophosphate is also a coenzyme for transketolase in the pentose phosphate pathway. Thiamin triphosphate plays key role in nerve conduction as it phosphorylates and activates chloride channel in the nerve membrane. Daily intake requirement for females is 1 milligram whereas for males it is 1.2 milligram (WHFoods, 2017). Dietary sources of vitamin B_1 are oil seeds, legumes, vegetables sweet potatoe and whole grains (WHFoods, 2017). Vitamin B_1 deficiency is associated with beri-beri, wernicke's encephalopathy, liver failure and pulmonary hypertension (WHFoods, 2017). Symptoms of mild vitamin B_1 deficiency include; constipation, loss of appetite, nausea, mental depression, irritability and fatque. Parkinson's, Alzheimer's and alcohol-related brain diseases are also linked with vitamin B_1 deficiency. In case of diabetes, gastrointestinal disease, heart failure, there is higher risk of vitamin B_1 deficiency especially in elderly persons (Keogh *et al.,* 2012).

2.4.5.6 Vitamin B_2 (Riboflavin)

Vitamin B_2 also known as riboflavin is a precursor of adenine dinucleotide (ADP) and flavin mononucleotide (FMN) both of which are coenzymes in a wide variety of redox reactions. Higher level of vitamin B_2 turns into yellow colour in urine. This is the only vitamin which provides us with visual clue of its abundance. Vitamin B_2 also plays a key role in energy metabolism (WHFoods, 2017). It promotes iron metabolism and its deficiency may increase the risk of anemia as iron is an important element for red blood cell production. Glutathione is most important antioxidant which provides antioxidative protection to body, and this antioxidant is recycled in the human body by vitamin B_2. Riboflavin is also widely used as food additives due to its intense yellow color. About 1.3 milligram for males and 1 milligram vitamin B_2 for females is recommended on daily basis (WHFoods, 2017). Dietary sources of vitamin B_2 are majorly dairy and cereal products. Other sources include; vegetables, peas and nuts (WHFoods, 2017). Riboflavin deficiencies are not fatal as there is enough conservation of tissue riboflavin. However, deficiency of these vitamins increases the risk of migraine headache, anaemia, high homocysteine, congestive heart failure, parkinson's disease, cataract, glossitis, scaly dermatitis and hypertension (WHFoods, 2017).

2.4.5.7 Vitamin B_3 (Niacin)

Vitamin B_3 also known as niacin comprised of nicotinic acid, nicotinamide and numerous enzymatic forms. Niacin is efficient in energy production and antioxidative defense for human health (Lanska, 2010). Nicotinamide adenine dinucleotide (NAD) and nicotinamide adenine dinucleotide phosphate (NADP) are two distinct forms of vitamin B_3 and essentially involve in production of energy from dietary proteins, carbohydrates and fats metabolisms (Leskova, 2006). These coenzymes are hydrogen donors in many redox reactions and cellular respiration. Niacin is important in energy

metabolism. Starch is synthesized from niacin and stored in liver and muscles as energy source. NAD, NADP and other niacin containing enzymes are also used as defense mechanisms for free radicals and protect the tissues from oxidative damage. Daily vitamin B_3 requirement for males is 16 milligrams whereas, for teenager females is 14 milligrams (WHFoods, 2017). Dietary sources for vitamin B_3 are animal proteins, legumes, sprouted cereals and vegetables (WHFoods, 2017). The most detectable disease associated with vitamin B_3 is Pellagra. Vitamin B_3 deficiency is also associated with alcoholism and numerous diseases like, high cholesterol, osteoarthritis, reynaud's disease, acne vulgaris, schizophrenia and type-1 diabetes (Lanska, 2010).

2.4.5.8 Vitamin B_5 (Pantothenic acid)

Pantothenic acid is derived from Greek word “pantothen” which means "on all sides". Pantothenic acid is largely incorporated into Coenzyme A (CoA) which plays important role in energy metabolism, Krebs cycle, fatty acid synthesis and oxidation. Pantothenic acid is widely distributed in natural foods. Daily vitamin B_5 requirement for human body is 10 milligrams. However daily intake requirement is also variable depending upon the age, gender and health conditions of the individuals (WHFoods, 2017). Dietary sources for vitamin B_5 are mushrooms, sweet potato, green leafy vegetables and fruits (WHFoods, 2017). Vitamin B_5 deficiency is associated with multiple nutrient deficiencies. However, deficiency of this vitamin is associated with chronic fatigue, vomiting, burning cramps, abnormal distress, high cholesterol and diabetes-related foot ulcers (WHFoods, 2017).

2.4.5.9 Vitamin B_6 (Pyridoxine)

Vitamin B_6 exists in different forms. They are; pyridoxine, pryridoxamine, pyridoxal, pyridoxal 5'-phosphate, pyridoxine-5'-phosphate and pyridoxamine 5'-phosphate (WHFoods, 2017). Pyridixal phosphate is the most active form of vitamin $B_{6.}$ It is a

coenzyme for many enzymes involved in amino acid metabolism, particularly transamination and decarboxylation. Vitamin B_6 is involved in red blood cell production, carbohydrate metabolism, liver detoxification, brain and nervous system health (Combs, 2007). Hemoglobin is oxygen career protein in blood and its function is dependent on heme. Heme production is dependent on availability of vitamin B_6. This vitamin is also involved in production of messaging molecules in brain and nervous system, known as neurotransmitter. Dopamine, GABA and serotonin are key neurotransmitters whose biosynthesis is dependent on vitamin B_6. Vitamin B_6 is also involved in the metabolism of tryptophan, other amino acids, proteins and development of immunity. Daily requirement of vitamin B_6 is about 1.3 milligrams in young males, and 1.2 milligrams in young females (WHFoods, 2017). Dietary requirement of vitamin B_6 is increased during pregnancy, lactation and may increase with age. Vitamin B_6 is widely distributed in vegetables, whole grains, animal proteins, green vegetables and fortified cereals (WHFoods, 2017). Interference in function of vitamin B_6 through genetic mutation induces a rare and distinct type of condition known as pyridoxine-dependent epilepsy. In this condition brain is under developed and epileptic seizures are prevailing during infancy (Combs, 2007; Gregory *et al*., 2013). Vitamin B_6 deficiency is also associated with rare types of anaemia known as sideroblasticanemias and depression however; this association of depression and vitamin B_6 is further pronounced by folic acid deficiency (Combs, 2007; Gregory *et al*., 2013).

2.4.5.10 Vitamin B_9 (Folate)

Folates have been reported to have critical importance for human health. There are different forms of folates which are present in foods, they include; dihydrofolates, methylfolates, polyglutamylfolates and monoglutamylfolates (Crider *et al*., 2011). The most active form of folic acid is tetrahydrofolates. One carbon tetrahydrofolat

derivatives is required in biosynthetic reactions like synthesis of choline, serine, glycine, methionine and purines. The basic role of folate is to support the brain health. Messengers molecules are produced which are used by nerves to send the signals in whole body. Tetrahydrobiopterin cycle (BH4) revealed that there is close association between folates and neurotransmitters especially serotonin and dopamine. BH4 and folates used same mechanism to cross the brain blood barrier. Folates also support the cardiovascular system and nervous system in human (Crider *et al*., 2011). Folates regulate the homocysteine level in blood and this amino acid is marker for cardiovascular diseases as higher level of homocysteine showed the increased risk of cardiovascular diseases. Higher level of homocysteine in blood is known as hyperhomocysteinemia whereas, optimum folates in blood especially in the form of 5-methyltetrahydrofolate (5-MTHF) lower the level of homocysteine in blood hence, it can be concluded that higher intake of folates can control the risk of cardiovascular diseases (Hayden and Tyagi, 2004; Crider *et al*., 2011). Folate supplementation may also help to reduce the risk of developing certain cancers. Young males and females require 400 microgram folates on daily basis (WHFoods, 2017). Dietary sources for folates are green leafy vegetables, fruits, orange juices, cereals and fortified bread (WHFoods, 2017). Folate is important for production of red blood cells and its deficiency along with deficiency of copper, iron, vitamin B_6 and vitamin B_{12} can destroy the production of red blood cells. Folate deficiency in females during pregnancy induced the problems in neural tube which severely affects the nervous system of foetus. Defects in neural tube can also lead to the loss of pregnancy in females. Risk of cancer is also lowered in human by higher intake of folates, especially the risks of breast cancer in females (Crider *et al.,* 2011).

2.4.5.11 Vitamin B_{12} (Cobalamin)

Vitamin B_{12} is a cobalt containing vitamin. Various forms of vitamin B_{12} include; cyanocobalame, methyl cobalamin and 5-deoxycobalamin. Vitamin B_{12} plays an important role in energy metabolism and other biological processes but differently from vitamin B complexes. The vitamin has some unique storage functions as it can be stored in the body for many years whereas, most of other B vitamins cannot be stored for such a long time (WHFoods, 2017). Vitamin B_{12} is also very important for cardiovascular health of human. It is involved in production of red blood cells which are oxygen carrier throughout the blood stream with the help of haemoglobin pigment. Succinyl-CoA is the building block for haemoglobin and this building block is dependent on the vitamin B_{12}. Vitamin B_{12} prevents the increase in level of homocysteine. It actually converts homocysteine into methionine with the help of methionine synthase enzyme to control the level of homocysteine (WHFoods, 2017). S-adenosylmethionine (SAM) is also simultaneously recycled parallel of homocysteine conversion by vitamin B_{12}. Vitamin B_{12} plays a key role in methyl metabolism. SAM is known as universal donor of methyl group. Some neurotransmitters are dependent on methyl transferases enzymes for synthesis and these enzymes are dependent on methyl groups for synthesis. In brain health, Vitamin B_{12} is necessary co-factor for DNA synthesis which is a necessary molecule for every life. Vitamin B_{12} combined with vitamin B_6 and B_9 are necessarily needed for DNA synthesis (WHFoods, 2017). Citric acid cycle is central cycle for aerobic energy production, while, succinyl-coA is key molecule in citric acid cycle. Vitamin B_{12} helps to maintain the supply of succinyl-CoA for citric acid, so this vitamin has key role in aerobic energy metabolism. Vitamin B_{12} also helps to maintain the bone health as incidence of osteoporosis may increase with deficiency of this vitamin. Younger adults require 2.4 microgram vitamin B_{12} on daily

basis for both males and females (WHFoods, 2017). Dietary sources for vitamin B_{12} are sardines, salmon, shrimps, dairy foods, mushrooms and breakfast cereals (WHFoods, 2017). Vitamin B_{12} deficiency is associated with blood homocysteine, atrophic gastritis, pernicious anemia, fatigue, neuropathy, migraine, asthma, depression, memory loss, mascular degeneration, alzheimer's disease, kidney disease and multiple sclerosis (WHFoods, 2017).

2.4.5.12 Biotin

This was formally known as coenzyme R, vitamin H, and vitamin B7 but now called biotin. It is widely distributed in diets as biocytin, which is released on proteolysis. It is synthesized by intestinal flora. Biotin plays a key role in sugar and fat metabolism (WHFoods, 2017). It functions in the transfer of carbondioxide in reactions like acetyl CoA carboxylases, pyruvate carboxylase, ppropionyl CoA carboxylase and methylcrotonyl CoA-carboxylase. Daily intake of biotin value is about 30 micrograms for both males and females (WHFoods, 2017). Dietary sources of biotin include vegetables, yogurt and fruits (WHFoods, 2017). Biotin deficiency is associated with impairment of insulin. Skin rash is also caused by biotin deficiency as this is needed for fat deposition in the skin. These fats are prerequisite for keeping the skin moist and supple whereas, in case of deficit fats, skin becomes irritated and flaky or scaly. Some other problems associated with Vitamin B_{12} deficiency include; cradle cap, dermatitis, hair loss, diabetes, brittle nails, pregnancy issues and seizures (WHFoods, 2017).

2.4.5.13 Vitamin C (Ascorbic acid)

Vitamin C exists in two forms; ascorbic acid and dehydroascorbic acid, both of which have vitamin C activity. Vitamin C is most familiar nutrient and commonly used nutritional supplement (WHFoods, 2017). Vitamin C is a cofactor for mixed function oxidases, and acts as a reducing agent in hydroxylation of proline and lysine, synthesis

of carnitine and norepinephrine. Hydroxylation of proline and lysine are required for protocollagen to cross link properly into normal collagen fibrils, hence, vitamin C is required for the maintenance of normal connective tissue and wound healing. Vitamin C has antioxidant properties and protects the lens of the eyes, molecules circulating in blood stream and genetic material (DNA) from harmful effects of free radicals. This vitamin also transforms iron into a form which can be easily absorbed into intestines. Vitamin C is needed for collagen production which is structural component of human body. Ascorbic acid acts as a non-enzymatic reducing agent. It aids in absorption of iron by reducing it to ferrous state in the stomach. It spares vitamin A, E, and some B vitamins from oxidation. It enhances utilization of folate by aiding the conversion of folate to tetrahydrofolates and its derivatives. Neurotransmitters are also dependent on vitamin C. These neurotransmitters involve in signaling of feelings, thoughts, and commands throughout the brain and nervous system. Vitamin C is also prerequisite for synthesis of serotonin, a hormone needed for proper functioning of endocrine system, nervous system, digestive system and immune system. Dietary recommended intake for vitamin C is about 75 milligrams on daily basis for younger males and females (WHFoods, 2017). Dietary sources for vitamin C are fruits, brussels sprouts, strawberries, sea vegetables, garlic and peppermint (WHFoods, 2017). The most severe form of vitamin C deficiency is scurvy. Scurvy is associated with decreased wound healing, hemorrhage and anemia (WHfoods, 2017). Vitamin C deficiency is also associated with common cold, asthma, capillary fragility, decreased immunocompitence, gout, gingivitis, musculoskeletal injury, seasonal allergies and increased high blood pressure (WHFoods, 2017).

2.5 Anti-nutrient Components in Food

Anti-nutrients are primarily associated with compounds of natural or synthetic origin. They are chemical compounds synthesized in natural foods by normal metabolism of species and different mechanisms. They are frequently related to plant-based, raw diets and are naturally synthesized in plants (Gemede and Ratta, 2014). They interfere with the absorption of nutrients, reduce nutrient intake, digestion, and utilization and may produce other adverse effects. However, anti-nutrients cannot be totally eliminated once they have been introduced to the body. In this regard, the biochemical effects of the anti-nutritional factors are an object of research interest (Cheeke and Shull, 1985; Aletor, 1993; Fu *et al*., 2002). Most of the secondary metabolites, acting as anti-nutrients, elicit very harmful biological responses, while some of them are widely applied in nutrition and as pharmacologically-active agents (Oakenfull *et al*., 1989; Soetan, 2008). The major anti-nutrients found in plant-based foods are phytates, cyanides, tannins, saponins, lectins, oxalates, etc. Anti-nutrients in vegetables, whole grains, legumes and nuts are a concern only when a person's diet is composed exclusively of uncooked plant foods. Modern biotechnology`s techniques such as germination, sprouting, and malting could reduce the level of certain allergens and anti-nutrients in food. Some of the antinutriensts present in plant based foods are discussed below.

2.5.1 Phytates

Phytates occur in several vegetable products. Seeds, grains, nuts and legumes store phosphorus as phytic acid in their husks in the form of phytin or phytate salt. The presence of phytates may affect bioavailability of minerals, solubility, functionality and digestibility of proteins and carbohydrates (Salunkhe *et al*., 1990). Phytic acid is most concentrated in the bran of grains (Wcislo and Szarlej-Wcislo, 2014). In legumes, phytic acid is found in the cotyledon layer and can be removed prior to consumption

(Nissar *et al*., 2017). The digestive enzyme phytase can unlock the phosphorus stored as phytic acid. In the absence of phytase, phytic acid can impede the absorption of other minerals like iron, zinc, manganese, copper, magnesium and calcium by binding to them (leading in highly insoluble salts that are poorly absorbed by the gastrointestinal tract leading to lower bioavailability of minerals (Masum *et al*., 2011). Phytates also inhibit digestive enzymes like pepsin, trypsin and amylase (Kumar *et al*., 2016).

2.5.2 Tannins

Tannins are a major group of antioxidant polyphenols found in food and beverages. They either bind or precipitate proteins and various organic compounds including amino acids and alkaloids. Tannin-protein complexes may cause digestive enzymes inactivation and reduction in protein digestibility caused by protein substrate and ionisable iron interaction (Salunkhe *et al*., 1990). Tannins are oligomers of flavan-3-ols and flavan-3, 4-diols that are concentrated in the bran fraction of legumes (Ngozi, 2014). Grapes and green tea are rich in this water-soluble polyphenol (Chu *et al*., 2015). Tannins exhibit antinutritional properties by impairing the digestion of various nutrients and preventing the body from absorbing beneficial bioavailable substances (Hendek and Bektaş, 2018).

2.5.3 Oxalates

Oxalic acid can form soluble or insoluble salts or esters called oxalates that are commonly found in plants such as leafy vegetables or synthesized in the body (Akwaowo *et al.,* 2000). Oxalates may be present in plants as soluble salts (potassium, sodium) or as insoluble salts (calcium, magnesium, iron) Insoluble salts cannot be processed out of the urinary tract once processed through the digestive system. Calcium oxalate can have a deleterious effect on human nutrition and health by accumulating kidney stones (Olawoye and Gbadamosi, 2017). Oxalate also binds with nutrients,

rendering them inaccessible to the body. Cruciferous vegetables (kale, radishes, cauliflower, broccoli), as well as chard, spinach, parsley, beets, rhubarb, black pepper, chocolate, nuts, berries (blueberries, blackberries) and beans are some of the foods with high amounts of oxalates (Mamboleo, 2015). Most people consume normal amounts of oxalate rich foods, while people with certain conditions, such as enteric and primary hyperoxaluria, need to lower their oxalate intake. In sensitive people, even small amounts of oxalates can result in burning in the eyes, ears, mouth, and throat; large amounts may cause abdominal pain, muscle weakness, nausea, and diarrhoea (Natesh, 2018).

2.5.4 Saponins

Saponins are steroids or triterpene glycoside compounds characterized by either bitter or astringent taste. Some saponins (steroid or triterpenoids) can be used for food while others are toxic. Saponins with bitter taste are toxic in high concentrations and can affect nutrient absorption by inhibiting both metabolic and digestive enzymes as well as by binding with nutrients such as zinc. Saponins are naturally occurring substances with various biological effects. In the presence of cholesterol, saponins exhibit strong hypocholesterolemic effect (Ikewuchi, 2012). They also lead to hypoglycemia (Barky *et al*., 2017) or impair the protein digestion, uptake vitamins and minerals in the gut, as well as lead to the development of a leaky gut (Johnson *et al*., 1986).

2.5.5 Trypsin inhibitors

Trypsin inhibitors occur in a wide range of foods like chickpeas, soybeans, red kidney beans, adzuki beans, mung beans and other representatives of the Leguminoseae, Solanaceae, and Gramineae families (Tibe *et al*., 2007). About ten percent of the world's dietary protein is derived from grain legumes (Klupšaitė and Juodeikienė, 2011).

Trypsin inhibitors contribute to the loss of trypsin and chymotrypsin in the gut, thus preventing protein digestion.

2.5.6 Cyanogenic glycosides

Cyanogenic glycosides are natural plant toxins present in several plant based foods. They composed of an a-hydroxynitrile of aglycone and a sugar moiety, mainly D-glucose and derived from five protein-amino acid which include; Valine, Leusine, Isoleusine, Phenylalanine and Tyrosine and from non proteinogenic amino acid Cyclopentenyl glycine. Cyanide is formed from the hydrolysis of Cyanogenic glycosides that occur during crushing of the edible plant material either during processing or consumption of the food crop. Cyanides are widely distributed in nature and occur in very low concentrations in blood. It is an inhibitor of cytochrome oxidase and interferes with aerobic respiratory system (Natesh, 2018).

2.6 Sprouting Technology

Sprouting is a natural germination process which involves the transition of a seed from dormant state to a more active state. During the process of seed germination, hydrolytic enzymes such as amylases are developed which causes endosperm modification and increased nutritional properties (Nelson *et al*., 2013; Di Gioia *et al*., 2017; Gan *et al*., 2019). Sprouting has been found to improve bioavailability of micronutrients by reducing the levels of anti-nutrients and enhancing digestibility in food crops (Hübner and Arendt, 2013). The term “sprouting” is used synonymously with “germination” and “malting” (H¨ubner and Arendt, 2013; Hassani *et al*., 2016; Singh and Sharma, 2017). Germination of a seed as seen in Figure 2.2 below begins with the uptake of water and ends with the appearance of the radicle (Nonogaki *et al*., 2010). The germination process is grouped into three phases; in phase I, the seed is steeped in water and cell material becomes fully hydrated. The endogenous metabolisms necessary for mobilizing

reserve material and radicle growth is activated in phase 2, and the radicle emerges at the end of phase 2. While in phase 3 the major mobilization of reserved material occurs and seedlings starts to grow. Germination process is terminated at phase 3 (Nonogaki *et al*., 2010). This therefore forms the basis for producing sprouted grains and shoot.

"Sprouts" (Regulation (EC) No 208/2013) are products gotten from the development of seeds either in water or non- water medium and are usually harvested before the emergence of true leaves. Sprouted grains according to (AACC, 2008; ESSA, 2016) contains its original bran, embryo and endosperm and does not exceed its kernel length nor loses its available nutrients.

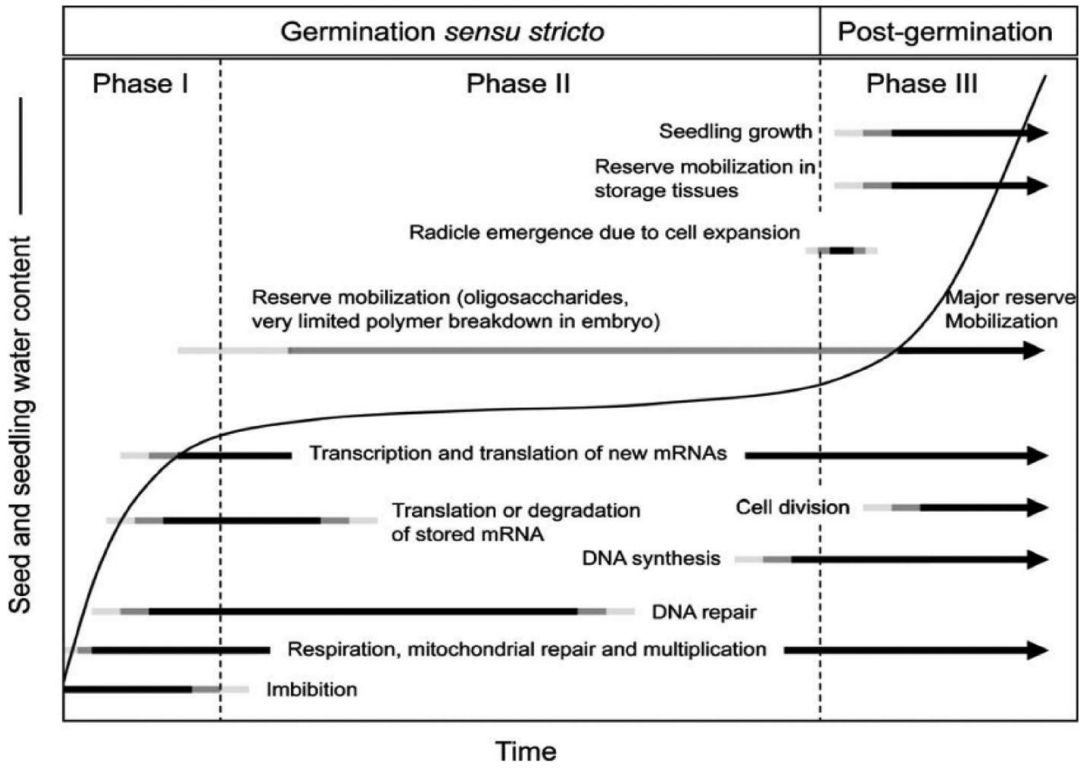

Figure 2.2: Physical and metabolic events occurring during sprouting.

Source: Nonogaki *et al*., 2010 with permission from Plant Science.

2.6.1 Uses of sprouted seeds in human nutrition

Sprouting of seeds has been in existence for quite a very long time particularly in the Eastern countries where seedlings are consumed as an important component of culinary history. In the 1980s, consumption of sprouted seeds raised popularity in the western countries due to consumers demand for dietetics and healthy foods. Sprouting gives peculiar characteristics to foods such as unique color, rich flavor and appreciable content of bioactive components. It also enhances sensory properties of confectionaries and other food products (Oghbaei and Prakash, 2012). Sprouted seedlings may be consumed in form of ready-to-eat sprouts or further processed by drying, roasting or popping (Hübner and Arendt, 2013). Recently, interest in sprouted seeds has been focused majorly on low processing and additive-free which are thus beneficial for human health. A possible example is the addition of sprouted cereals to whole white flour to enriched bakery products (Falcinelli *et al*., 2018). Dried sprouted cereals have also been used for making noodles, pasta, cakes, unleavened bread, porridge and functional beverages (Shingare and Thorat, 2013). Sprouted grains could be milled into flour to be used as supplements in infants and animal feeding system (Dal Bosco *et al*., 2015; Mattioli *et al*., 2016).

2.6.2 Impacts of sprouting on nutrient quality

2.6.2.1 Impacts of sprouting on starch content in oat grains

Sprouting initiates the de novo synthesis of starch-degrading enzymes, such as α-amylase and α-glucosidase, in the scutellum and aleurone cells of grains (Ayernor and Ocloo, 2007; Saman *et al*., 2008; Duke, 2009; Xu *et al*., 2012). Sprouting decreases starch contents by 5 to 15 % in brown rice sprouted for 1 to 3 days at 30 to 35 ^{o}C (Xu *et al*., 2012; Chungcharoen *et al*., 2015), and in barley sprouted for 4 days at 17 ^{o}C (Vinje *et al*., 2015). The activities of enzymes leads to partial hydrolysis of starch into glucose,

maltose, maltotriose and dextrins (Ayernor and Ocloo, 2007; Saman *et al*., 2008), thereby increasing the sugar content (Agu and Palmer, 1997; Ayernor and Ocloo, 2007; Coulibaly and Chen, 2011; Xu *et al*., 2012). The sugars formed during sprouting of grains serves as a source of energy for the development of embryo. Sprouted cereals are generally better digestible as a result of their enzymatically damaged starch granules, thin cell walls, and higher content of readily available sugars, making them especially suitable for the production of foods for infants and the elderly (Correia *et al*., 2008; Tizazu *et al*., 2010; Srivastava *et al*., 2015).

2.6.2.2 Impacts of sprouting on protein content in oat grains

The content of protein in cereal grains differs and ranges from 8 % to 16 % of dry mass (dm). During sprouting, metabolic enzymes such as proteinases are activated; this leads to release and synthesis of some amino acids and peptides. As a consequence, nutritional quality of proteins is enhanced. Although sprouting results in protein hydrolysis, it does not cause any significant changes in total protein content (Chung *et al*., 1989; Vis and Lorenz, 1998; Kaur *et al*., 2002; Van Hung *et al*., 2011; Ogbonna *et al*., 2012; Xu *et al*., 2012; C´aceres *et al*., 2014; Swieca and Dziki, 2015). However, Endogenous peptidase activity levels is low in healthy cereal grains and increases considerably after 1 day of sprouting (Kolodziejczyk *et al*., 2004; Elmoneim *et al*., 2010; Faltermaier *et al*., 2015). Reports has shown that a 2 to 5 fold increase in peptidase activity was observed in sorghum (Elmoneim *et al*., 2010), and wheat (Faltermaier *et al*., 2015) when sprxouted for 3 to 7 days at 15 to 27 ^{o}C. Endopeptidases secreted from the aleurone and scutellum layers of cereal grains during sprouting are essential for seedling development as they degrade storage proteins and activate functional proteins (An example is β-amylases) (Faltermaier *et al*., 2015). Some studies have reported a significant decrease of 3 to 10 % of protein content in sprouted sorghum (Elmaki *et al*.,

1999; Afify *et al*., 2012), while other studies have reported an increase in protein content of 5 to 10 % in sprouted barley (Donkor *et al*., 2012; Teixeira *et al*., 2016) and sorghum (Donkor *et al*., 2012). The decrease in protein content was attributed to leaching of water-soluble peptides in the steeping water (Elmaki *et al*., 1999; Afify *et al*., 2012), while the increase may probably be attributed to loss of carbohydrates through respiration (Mbithi-Mwikya *et al*., 2000; Tizazu *et al*., 2010). However, studies have reported the relative differences in protein content between sprouted and non-sprouted cereal grains to be smaller than 10 %, indicating that sprouting generally does not substantially affect total protein content.

2.6.2.3 Impacts of sprouting on lipid content in oat grains

Lipase catalyzes the degradation of triglycerides to glycerol and free fatty acids (Peterson, 1999; Kubicka *et al*., 2011). The glycerol and free fatty acids are mainly converted to sucrose, which is sent to the scutellum for use by the rootlet and shoot. The lipid catabolism releases energy and carbon sources required for the biochemical and physicochemical changes during seedling growth (Chung *et al*., 1989; Elmaki *et al*., 1999; Coulibaly and Chen, 2011; Kubicka *et al*., 2011). Kubicka *et al*. (2011) reported a 1.2 to 2.3- fold increase in lipase and lipoxygenase activity levels during sprouting of cereal grains due to their de novo synthesis in the aleurone and/or scutellum. However, mature oat grains have high levels of lipase activity that remain unchanged or even decline during sprouting, which is unique among the cereals (Peterson, 1999). An 8 to 15 % decrease in lipid contents occurs in millets sprouted for 3 days at 37 ^{o}C, in oat sprouted for 6 days at 16 ^{o}C and in barley sprouted for 5 days at 22 ^{o}C as a result of the lipase activities (Chung *et al*., 1989). It may be concluded that increase in sprouting temperatures leads to a higher lipid breakdown.

2.6.2.4 Impacts of sprouting on dietary fibre in oat grains

Several health effects have been ascribed to dietary fibre consumption, some of which are related to their solubility, viscosity, particle size, and/or water-holding capacity. Dietary fibre components are embedded in plant cell walls of cereal grains. As a result of sprouting, these cell wall polysaccharides are hydrolyzed by de novo synthesized enzymes (Krahl *et al*., 2010; Teixeira *et al*., 2016). Sprouting induces changes in composition and contents of insoluble and soluble dietary fibre (Krahl *et al*., 2010; Teixeira *et al*., 2016). These changes can be used to modify the dietary fibre content. In general, the impact of process conditions on solubilization of dietary fibre depends on the cereal cultivar, since differences in hydration rate affect enzyme synthesis and activity (C´aceres *et al*., 2014; Teixeira *et al*., 2016). Teixeira *et al*. (2016) reported that sprouting does not significantly affect the total fibre content in barley grains when sprouted for 3 days at 15 ^{o}C, while Koehler *et al*. (2007) showed that fibre decreases in wheat during the first 2 days of sprouting at 15 to 20 ^{o}C, while it remains fairly constant at higher temperatures (25 ^{o}C and 30 ^{o}C). It is therefore concluded that soluble dietary fibre content increases 3 to 4 fold due to sprouting, while the insoluble dietary fibre content decreases, especially during long sprouting times (5 to 7 days) (Koehler *et al*., 2007).

2.6.2.5 Impacts of sprouting on antinutreint, mineral redistribution and bioaccessibility in oat grains

About 85 % of phosphorus (P) in bran is stored as phytic acid (inositol-hexaphosphate IP6) which occurs as granules embedded in protein-rich globoid structures found mainly in aleurone cells (Raboy, 2003; Schlemmer *et al*., 2009). Most phytic acid is chelated by (divalent) cations, such as those of iron (Fe), zinc (Zn), calcium (Ca), manganese (Mn), magnesium (Mg), and copper (Cu), to result in phytates (Schlemmer *et al*., 2009). The

bioavailability of these phytates and their availability for absorption in the human gastrointestinal tract as well as their distribution to human organs and tissues through the blood stream only amounts to 5 to 25 % , since they are poorly digested and absorbed, as humans lack intestinal phytase enzymes (Bouis *et al*., 2011). During germination of cereal grains, phytases are activated, synthesized and secreted to make phosphate, mineral elements and myoinositol available for plant growth and development (Raboy, 2003). Thus, controlled grain sprouting increases the bioaccessibility of mineral elements. Mature cereal seeds have relatively low endogenous phytase activity levels, which substantially increase during sprouting. These enzyme activity levels vary with the cereals and the sprouting conditions. Studies have reported a 3 to 10 fold increase in phytase activity levels in rice, rye, barley, sorghum and oats when sprouted for 4 to 5 days at 15 to 25 ^{o}C (Azeke *et al*., 2011; Lemmens *et al*., 2018). However, the phytase activity decreases relatively fast (by 20 to 50 %) at sprouting temperatures exceeding 20 ^{o}C after reaching its maximum level. This may be ascribed to enzyme degradation by activated peptidases and/or by product inhibition due to the liberated phosphates (Haraldsson *et al*., 2004; Sung *et al*., 2005; Ou *et al*., 2011). The increase in endogenous phytase activity as a result of sprouting leads to phytate hydrolysis and thus, to release of bound minerals and improved mineral bioaccessibility. Generally, higher endogenous phytase activity levels in a cereal result in more extensive phytate hydrolysis. This explains that long sprouting times (3 to 5 days) are needed to lower the phytate concentration by more than 30 % (Bartnik and Szafranska, 1987; Centeno *et al*., 2001; Tian *et al*., 2010; Azeke *et al*., 2011; C´aceres *et al*., 2014). Azeke *et al*. (2011) reported only a 5 %, 10 %, 9 % and 9 % decrease in phytate content due to 2 days of sprouting at 24 to 28 ^{o}C, while the content decreased by 57 %, 47 %, 34 %, and 37 % in rice, sorghum, wheat, and millet, respectively, due to 5

days sprouting. Cereal phytases have optimum temperature and pH of 37 to 55 ^{o}C and 4.5 to 6.0 respectively, thus, increasing the sprouting temperatures to those optimal for phytase action can enhance phytate breakdown.

During steeping, minerals can be lost due to leaching. The loss is relatively high (about 30 %) when the cereal grains are steeped at 30 ^{0}C prior to germination (Afify *et al*., 2011). The elements passing to the seedling originate mainly from the scutellum and aleurone, while the concentration of minerals in the starchy endosperm remains largely unchanged. The most mobile mineral ions during seed sprouting are those of Potassium, Calcium and Zinc while Manganese and irons are less mobile (Lu *et al*., 2013). Lemmens *et al*. (2018) found that Zinc was also detected in the pericarp tissues after sprouting, but it especially accumulates in the developing coleoptile and radicle, suggesting that Zinc ions play a key role in their meristemic tissues. Sprouting has been found to cause a 15 % decrease in phytate content in wheat leading to an increase in iron and Zinc bioaccessibility from 4.6 to 14.1 % and from 2.5 to 14.6 %, respectively (Lemmens *et al*., 2018). A 30 % decrease in phytate content in sorghum as a result of sprouting has been found to increase the iron and Zinc bioaccessibilities from 10 to 20 % and from 8 to 15 %, respectively (Afify *et al*., 2011).

Antinutreints such as tanins and trypsin inhibitors can reduce protein digestibility by forming complexes with proteolytic enzymes in cereals (Ogbonna *et al*., 2012). Protein content in sorghum is 15 to 25 % less digestible than other cereal proteins because it contains condensed tannins and trypsin inhibitors (Chung *et al*., 1989; Afify *et al.,* 2012; Swieca and Dziki, 2015), which limit digestibility of proteins. Studies have reported a decrease in tannin content of 8 to 60 % when sprouting sorghum and millet for 2 to 7 days at 20 to 30 ^{o}C (Mbithi-Mwikya *et al*., 2000; Ogbonna *et al*., 2012). This has been attributed to leaching of tannin into the steeping medium, to the activity of polyphenol

oxidases, and to polymerization of tannins, resulting in loss of solubility (Mbithi-Mwikya *et al*., 2000). In contrast, tannin content increased 25 % to 30 % when sprouting wheat and sorghum for 3 to 5 day at 20 to 32 ^{o}C (Ahmed *et al*., 1996; Hithamani and Srinivasan, 2014; Yang *et al*., 2016). This increase in tannin content has been explained as resulting from de novo synthesis (Ahmed *et al.,* 1996; Yang *et al*., 2016). Sprouting sorghum for 4 days at 37 ^{o}C reduced trypsin inhibitor content by about 40 %, this may be due to increased action of specific peptidases (Mbithi-Mwikya *et al*., 2000; Ogbonna *et al*., 2012). It can therefore be concluded that controlled sprouting of cereals under optimal conditions reduces phytase activity and improves in vitro mineral bioaccessibility.

2.6.2.6 Impacts of sprouting on vitamin content in oat grains

The vitamin content in cereal grains (vitamin B, C and E) is essential for seedling growth and development and increases during sprouting due to biosynthesis. The changes which occur in vitamin content of sprouted grains largely depend on the type of grain and conditions of steeping or sprouting. Vitamin E a well-known antioxidant is mainly located in the embryo of cereals and scavenges free radicals within cell membranes (Fardet *et al*., 2008). Tocopherols are synthetized and stored within the embryo and transported to the rootlets and acrospires during seed sprouting (Fardet *et al*., 2008). Reports showed that Haraldsson *et al*. (2004) found no changes in the total level of all tocopherol and tocotrienol compounds in barley first sprouted for 4 days at 15 ^{o}C and from which in a next step, the rootlets were removed prior to analysis. This shows that the duration of sprouting is the main determinant for the increase in vitamin E content. Moongngarm and Saetung (2010) and Watanabe *et al*. (2004), indicating that relatively long sprouting times are needed to allow de novo synthesis of this vitamin.

The vitamin B contents in cereals generally increases as a result of sprouting and supports the seedling growth and development. However, a 1.2 to 5.5 fold increase in thiamine content was found in sorghum, wheat and rice when sprouted for 3 to 4 days at 25 to 30 ^{o}C, likewise, the riboflavin content in sorghum, barley, millet and wheat when sprouted for 4 days at 17 to 28 ^{o}C were doubled and increased from 0.07 to 0.25 mg/100 g of dm (Malleshi and Klopfenstein, 1998; Plaza *et al*., 2003). A 1.3 to 1.5 fold increase in niacin and pyridoxal contents was observed in wheat, millet and sorghum when sprouted for 3 to 4 days at 25 ^{o}C (Malleshi and Klopfenstein, 1998; Plaza *et al*., 2003; Coulibaly and Chen, 2011). However, Watanabe *et al*. (2004) and Moongngarm and Saetung (2010) observed that the thiamine, riboflavin, niacin, and pyridoxine contents remain unchanged or even decrease when brown rice is steeped or sprouted for only 1 day. The authors concluded that de novo synthesis of vitamins is only initiated in later sprouting stages and that water-soluble vitamins can leach into the steeping water. The B vitamins are also transported to the rootlets and acrospires during sprouting, which, in turn, mean that discarding them decreases the vitamin B content of sprouted grains (Malleshi and Klopfenstein, 1998). Reports showed that a 40 to 50 % loss in thiamine concentration was observed in sprouted sorghum (Malleshi and Klopfenstein, 1998) and brown rice (Moongngarm and Saetung, 2010) when the developing embryo is removed. It is possible that thiamine, one of the most heat labile B-vitamins, is degraded during the drying process.

Sprouting also stimulates de novo synthesis of folate because of the increased demand for methyl groups as the metabolic activities of the developing seedling increases (Jabrin *et al*., 2003; Jagerstad *et al*., 2005; Kariluoto *et al*., 2006). Several researchers showed a 4 to 6 fold increase in folate to a final content ranging from 1.5 to 3.0 mg/kg in wheat and rye when sprouted for 4 to 6 days at 18 to 25 ^{o}C. (Kariluoto *et al.,* 2006;

Koehler *et al*., 2007). Vitamin C content in cereal grains is usually undetectable or very low. During sprouting, however, vitamin C is synthesized de novo (Lintschinger *et al*., 1997; Malleshi and Klopfenstein, 1998; Yang *et al*., 2001; Coulibaly and Chen, 2011) leading to 5 to 55 mg/100 g in sprouted wheat, millet and sorghum (Lintschinger *et al*., 1997; Malleshi and Klopfenstein, 1998; Yang *et al*., 2001; Coulibaly and Chen, 2011). Lintschinger *et al*. (1997) reported that rinsing wheat sprouts with hot water at 70 ^{o}C decreased vitamin C content by 40 to 60 %. Hence, process conditions need to be carefully chosen to preserve this vitamin as it is one of the most heat- and light-unstable vitamins.

The relatively high levels of vitamins B and E in sprouted cereals can significantly contribute to daily intakes of these vitamins when consumed. Adults can meet the required daily allowance (RDA) value of folate by daily consuming 100 to 150 g of sprouted wheat or rye grains. 100 g of sprouted cereals can account for 20 to 35 % of the RDA of niacin, 5 to 30 % of the RDA of riboflavin, and pyridoxal and to 3 to 36 % of the RDA of vitamin E (EFSA, 2006, 2015). However, there is great variation in the thiamine content in sprouted cereals. Consuming 100 g of sprouted wheat, brown rice, oats, millet, or barley contributes up to 10 to 50 % of the RDA of thiamine, while consuming 100 g of sprouted sorghum contributes up to 20 to 80 % of its RDA. Sprouted grains only contribute little to the daily intake of vitamin C, since consuming 100 g of sprouted cereals meets about 3 to 10 % of its RDA. Therefore, it can be concluded that short steeping times with limited amount of water, followed by long sprouting times, and mild drying are most favourable to obtain elevated vitamin content in sprouted grains.

2.6.2.7 Impacts of processing on the safety and quality of sprouts

Apart from fresh consumption of sprouted seeds, sprouts can be further extracted for juices or dried and milled into soluble powder to enriched flour products. During powder production the drying process plays the most important role to preserve the biochemical quality of sprouts. Several drying methods can be used including hot air oven, microwave and vacuum oven dryings (Singh, 2017). Generally, the quantitative analysis of antioxidant component of microwave dried wheat grass powder (600 W, 15 min) showed the highest mean value of total phenol content, chlorophyll and scavenging ability (Singh, 2017). Notwithstanding, seedlings rich in phytochemicals, should be dried at low temperatures. Also, powders from wheatgrass juice can be encapsulated to avoid undesirable odor and protect the phytochemicals, using maltodextrin and whey protein (Akbas *et al.,* 2017). Fermentation is also a common technique for cereal processing. This can be used to better preserve or promote nutritionally interesting compounds (Shokoohi *et al*., 2014).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Materials

3.1.1 Reagents and chemicals

All chemicals and reagent used were of analytical grade manufactured by British Drug House (BDH) Limited, England and Sigma Aldrich Chemical Company Incorporation, Milwaukee, Wisconson, (USA), Clariant (Canada) and Merck (Germany).

3.1.2 Equipment

Analytical mill (Cole Parmar, IL, USA), Air oven (model 334), Hitachi liquid chromatography coupled with a photodiode array detector (UV-DAD) Hitachi L-2455, fluorescence detector (FLD) L-2485, pump Hitachi L-2130, column oven Hitachi L-2350 and auto sampler Hitachi L-2200, and Jenway model 6000 electronic Spectrophotometer.

3.2 Research Methodology

3.2.1 Sample collection and preparation

Finger millet grains and water melon seeds were purchased from a local market in Kaduna State, Nigeria. Refined white flour was purchased from the local market in Minna, Niger State, Nigeria.

3.2.1.1 Sprouting, drying and processing of samples

Sprouting technique was carried out following the methods reported by Abioye *et al*. (2018). One hundred grams (100 g) each of finger millets and water melon seeds was weighed and sorted to remove stones and other extraneous materials. Spoilt or cracked seed were discarded and the remaining seeds were washed and soaked in distilled water for about 2 hours. The soaked seeds were washed several times with distilled water

until very clean. The seeds were placed on a sterile germinating plastic plate lined with filter paper and allowed to sprout at 25 ^{o}C from 0 to 9 days. Sprouted seeds were collected three days interval and dried in an air oven at 45 ^{o}C until water was dried off. Weight loss was calculated by subtracting the initial weight from the final weight and a graph of weight loss was plotted against drying time to obtain the kinetics of drying. The dried samples were milled to powder using analytical mill (Cole Parmar, IL, USA) at high speed (20,000 rpm), sieved through a mesh pore size of 600 microns and packaged in an air tight container until further analysis.

3.2.2 Proximate analysis of test samples

Proximate composition of moisture, crude protein, total ash, crude fibre, fat and total carbohydrate contents were determined according to the method reported by Onwuka (2005).

3.2.2.1 Determination of moisture contents in samples

Two grams (2 g) of sprouted sample was weighed into a clean and dried crucible (W_1). The crucible with its content (W_2) was transferred into an oven and dried at 100 ^{o}C for 6 hours and 105 ^{o}C for another 6 hours. The sample was allowed to cool in a desiccator for 30 minutes. After which dry weight of sample and crucible (W_3) was recorded and percentage moisture was calculated as follows

$$\% \, Moisture = \frac{\text{W1}-\text{W2 X 100}}{\text{Weight of sample}} \qquad 3.1$$

Where

W_1 = Initial weight of crucible + Sample

W_2 = Final weight of crucible + Sample

3.2.2.2 Determination of ash contents in samples

Empty crucible was placed in a muffle furnace at 550 ^{0}C for an hour, cooled in desiccator and weight of empty crucible was recorded (W_1). Two grams of sprouted sample was weighed into the pre-heated crucible (W_2) and allowed to char for 2 hours. A white ash was obtained, indicating complete oxidation of all organic matter in the sample. Sample was allowed to cool in a desiccator and reweighed (W_3). Percentage ash was calculated as follows

$$\% \, Ash = \frac{\text{Difference in weight of ash X 100}}{\text{Weight of sample}} \qquad 3.2$$

Where,

W_1 = weight of empty crucible

W_2 = weight of crucible + sample before drying /ashing

W_3 = weight of crucible + ash.

3.2.2.3 Determination of crude protein contents in samples

Kjeldahl method was used to determine protein content of flour sample. Sprouted flour sample (0.2 g) was weighed and wrapped in a whatman filter paper and placed in the Kjedahl digestion flask. 6 mL of concentrated H_2SO_4 and Kjeldah1 catalyst mixture of (10 g Na_2SO_4+5 g $CuSO_4$+ 0.05 g selenium) were added to the flask and swirled for thorough content mixture and then digested till the mixtures become clear or turned light green. The digested sample was cooled and diluted with 100 mL distilled water in a standard volumetric flask. 10 mL of diluted sample plus 10 mL of 40 % NaOH was transferred into Markham distillation apparatus. Distillation was continued for at least 10 minutes and NH_3 produced was collected as NH_4OH in conical flask containing 5 mL of 4 % boric acid solution with few drops of methyl red indicator until about 70 mL of distillate was collected. The distillate was titrated against standard 0.1 N HCI

solutions until a pink colour was observed. Percentage crude protein was calculated as follows;

% Crude Protein = 6.25* x % N (* = Correction factor)

$$\% \, Nitrogen = \frac{(a-b) \text{ X } 0.1 \text{ X } 0.014 \text{ X V X } 100}{\text{Weight of sample X V}} \quad 3.3$$

Where;

a = Titre value of digested sample

b = Titre value of blank sample

v = Volume taken for distillation (100 mL)

w = weight of flour sample (mg)

0.014 – Milli equivalent weight of Nitrogen

3.2.2.4 Determination of crude fat contents in samples

Crude fat in sprouted seeds sample was determined using Soxhlet apparatus. Two gram of sample was wrapped in filter paper, placed into a thimble of known weight (W_1) and sample plus thimble was placed in a Soxhlet extraction tube. 300 mL of petroleum ether was poured into a 500 mL round bottom ground joint flask placed on a heating mantle. The soxhlet apparatus was assembled and allow refluxing for 24 hours. After which the thimble with content was removed, placed in an oven to dry at 50 ^{0}C for 24 hours, cooled in a desiccator and weighed (W_3). Percentage crude fat was determined as follows;

$$\% \, Crude \, fat = \frac{100 \, (W2-W3)}{W2-W1} \quad 3.4$$

Where;

W1 = weight of sample

W2 = weight of sample plus thimble

W3 = weight of sample after extraction

3.2.2.5 Determination of crude fibre contents in samples

Two grams of seed sample was defatted with petroleum ether. Sample was then transferred into a 250 cm^3 Erlenmeyer flask and boiled under reflux for 30 minutes with 200 mL of a solution containing 1.25 % of H_2SO_4. The solution was filtered through several layers of cheese cloth on fluted funnel, and subsequently washed with boiling water until the washings are no longer acidic. The residue was transferred into a beaker and boiled for 30 minutes with 200 mL of solution containing 1.25 % NaOH solution. The residue was filtered and washed thoroughly until the washings were no longer alkaline. The sample was transferred into a crucible and dried in an oven at 105 ^oC the crucible with its content was incinerated at 550 ^oC for 30 minutes, cooled and weighed. The loss in weight after incineration was expressed as percentage crude fibre.

$$\% \, Crude \; fibre = \frac{(\text{Wc} + \text{sample before ignition}) - Wc + ash \; X \; 100}{\text{Initial weight of sample}} \qquad 3.3$$

Where;

Wc= weight of crucible

3.2.2.6 Determination of carbohydrate contents in samples

Total carbohydrate content was determined by difference following the methods reported by Nielsen (2002). The total amount of crude protein, crude fat, moisture, ash and crude fibre of each sample was added and subtracted from 100. The value obtained was the percentage carbohydrate content of the sprouted seeds.

$$\% \text{ carbohydrates} = 100 - (\% \text{ moisture} + \% \text{ ash} + \% \text{ protein} + \% \text{ fat} + \% \text{ fibre})$$

3.2.3 Determination of vitamin contents in samples

The following vitamins (vitamins B_1, B_2, B_3, B_6, B_9, C, and E) were determined using standard analytical methods (Association of Official Analytical Chemists (AOAC), 2011)

3.2.3.1 Determination of water soluble vitamin contents in samples

Chromatograms of water soluble vitamins (B_1, B_2, B_3, B_6, B_9, and C) were determined using Hitachi liquid chromatography coupled with a photodiode array detector (UV-DAD) Hitachi L-2455, fluorescence detector (FLD) L-2485, pump Hitachi L-2130, column oven Hitachi L-2350 and auto sampler Hitachi L-2200. The analysis was presented in steps as follows.

Preparation of dilute solutions

The dilute solution of sodium dihydrogen phosphate ($NaH_2PO_4(2H_2O)$) in concentration 0.05 M of pH 6.3 was obtained by dissolving 7.8 g $NaH_2PO_4(2H_2O)$ in 900 mL High performance liquid chromatography (HPLC) water in 1 L volumetric flask. The pH of the resulting solution was adjusted to 6.3 through drop wise addition of 1 M sodium hydroxide and the volume was completed to 1000 mL with HPLC water.

Preparation of standard stock solutions

Standard stock solution of vitamins (B_1, B_2, B_3, B_6, B_9 and C) in concentration of (2000 mg/L) for all vitamins was obtained by dissolving 0.2 g of each vitamin in 40 mL diluted solution in 100 mL volumetric flask and was completed to mark with dilute standard solutions.

Preparation of working standard solutions

Working solutions (0.001 – 1000 mg/L) for all vitamins were prepared by diluting suitable volume of standard stock solution with diluted solution. To construct the

calibration curve, five replications (20 µL) of each standard solution was injected immediately after preparation into column. Relative peaks area of (vitamin and methyl paraben) were measured.

Sample preparation

Samples were prepared in two steps;

Step I: Six gram of sprouted seeds sample was weighed and divided into three parts into a glass test tube containing 5 mL methanol. The mixture was sonicated for 25 minutes in an ultrasonic bath, and centrifuged for 25 minutes at 515 rpm. The supernatant was transferred into a 25 mL beaker and sonicated for another 120 minutes at 35 ^{o}C in the dark to evaporate methanol. 0.1 mL NaOH was added to dissolve the residue.

Step II: Two mL of prepared HPLC water was added to the solid precipitate in the three glass test tubes, it was shaken for 10 minutes in an ultrasonic bath and 0.1 mL H_3PO_4 was added to each tube, shaken for 20 minutes and centrifuged for 25 minutes at 515 rpm. The supernatant was transferred to the residue which was dissolved in 25 mL beaker resulting from the first step. It was sonicated for 10 minutes and transferred into a 10 mL volumetric flask and diluted to the mark with diluted solutions of vitamins. 2.5 mL from the diluted solutions and 0.2 mL methyl paraben (1 g/L) was transferred into a 5 mL volumetric flask, diluted to mark with diluted solutions and filtered through a 0.22 µm Millipore filter.

Chromatographic conditions

The column chromatographic analysis was carried out according to the methods of AOAC (2011). A C18 BDS column (10 cm x 4.6 mm; 3 µm) was used. The mobile phase was filled with 5.84 mM of hexane-1-sulfonic acid sodium and acetonitrile in a

ratio of 95:5 with 0.1 % triethylamine as solvent (A) at an adjusted pH 2.5 (1 M orthophosphoric acid was used for adjusting the pH). Solvent (B) was 0.1 % triethylamine of pH 2.5 with hexane-1-sulfonic acid sodium and acetonitrile in 50:50 ratios at 5.84 mM molar concentration. The column was operated at flow rate of 1.6 mL/min at 40 ^{o}C with a 20 µL injected volume. Starting with 100 % solvent A. A gradient elution was performed for 5 minutes until the composition of the mobile phase was 50 % of A and 50 % of B. detection was performed for UV-DAD detector at the following wavelengths; 246 nm, 267 nm, 260 nm, 290 nm, 282 nm and 361 nm for vitamins C and B1, B2, B3, B6, B9, and B12 respectively. The FLD Detector was programmed at λex = 296 nm, λem = 390 nm for vitamin B6 during the first 3 minutes and changes to λex = 450 nm, λem = 530 nm for vitamin B2 from 3 to 4.6 minute.

3.2.3.2 Determination of vitamin E content in samples

Vitamin E was determined by the methods of (Marck index). One gram of sprouted sample was weighed and 10 mL of methanol was added. The sample was homogenized and then filtered. 0.4 mL of the extract was taken and 7.6 mL of colour developer (containing 0.84 g NaH_2PO_4, 1.24 g $(NH4)_2MoO_4$, 8.15 mL H_2SO_4, and 250 mL methanol) was added to the sample. The sample was incubated at 90 ^{o}C for 1 hour. Absorbance of sample was read at 695 nm on Spectrophotometer using 21 D Spectronic models. Concentration of vitamin E was extrapolated from standard curve that was prepared.

3.2.4 Determination of anti-nutritional components in samples

The following anti-nutrients; saponins, tannins, phytates, oxalates and cyanogenic glycosides were determined in sprouted samples.

3.2.4.1 Determination of saponin contents in samples

The spectrophotometric method reported by Oloyede, 2005 was used for the determination of saponin. Samples (0.5g) was added to 20 mL of 1NHCl and was allowed to boil for 4 hours. It was cooled and filtered. Petroleum ether (50 mL) was added to the filtrate for ether layer and evaporated to dryness. Acetone ethanol (5 mL) was added to the residue, and 0.4 mL of each was taken into 3 different test tubes. Six mL of ferrous sulphate reagent was added into the different text tubes followed by 2 mL of concentrated H_2SO_4. It was thoroughly mixed after 10 minutes and the absorbance was read at 490 nm using Jenway model 6000 electronic spectrophotometer and standard saponin was used to establish the calibration curve.

3.2.4.2 Determination of tannin contents in samples

The folin Denis spectrophotometric method as reported by Onwuka (2005) was used for tannins determination. One gram of each sample was measured into a 50 mL beaker containing 10 mL distilled water and shaken. The mixture was allowed to stand for 30 minutes at 37 ^oC. After 30 minutes, the mixture was centrifuged and extract obtained. Two point five mL of the supernatant was transferred into a 50 mL volumetric flask. Similarly, 2.5 mL of standard tannic acid solution was transferred into a separate 50 mL volumetric flask. One mL of folin-Denis reagent was added into each flask followed by 2.5 mL of saturated Na_2CO_3 solution. The mixture was made up to mark in the flask (50 mL) and incubated for 90 minutes at 37 ^oC and absorbance was measured at 250 nm using Jenway model 6000 electronic Spectrophotometer. Tannin contents was calculated as follows;

$$\% \, Tannin = \frac{\left(\frac{An}{As}\right) X \, C \, X \, 100}{W \, X \, Vf} \qquad 3.6$$

Where;

An = absorbance of test sample, As = absorbance of standard solution

C = concentration of standard solution, W = weight of sample, Vf = total volume of extract.

3.2.4.3 Determination of phytate contents in samples

Phytate was determined according to the methods reported by Lolas and Markakis (1975) and Essien *et al*. (2005). Two grams of each sample was measured into a 250 mL conical flask. Sample was soaked with 100 mL of 2 % concentrated HCl for 3 hours. After 3 hours, the mixture was filtered and 50 mL of the filterate was placed in a 250 mL beaker and 107 mL of distilled water was added. Ten mL of 0.3 % ammonium thiocyanate was added to the same moles as indicator, and titrated with Fe_3Cl solution which contained 1.95 mg iron per mL. the titration was continued until solution turned brown which persist for about 5 minutes.

3.2.4.4 Determination of oxalate contents in samples

Oxalate was determined according to the methods reported by Day and Underwood (1986) and Onwuka (2005). For each sample (1 g) was weighed into a 100 mL volumetric flask. 75 mL of $3NH_2SO_4$ was added and stirred for 1 hour. The mixture was filtered using whatman filter paper No. 1. 25 mL was measured out of the filtrate and titrated against 0.1 N $KMnO_4$ solution. A pink colour was observed, which persisted for at least 30 seconds. Oxalate content was calculated as follows;

$$\text{Mg 100 g oxalate} = \frac{\text{T X (Vme)(Df)}X\ 105}{\text{(ME)X Mf}} \qquad 3.7$$

Where;

T = the titre of KMnO4 (mL), Vme is the volume-mass equivalent (1 cm3 of 0.05 M KMnO4 solution is equivalent to 0.00225 g anhydrous oxalic acid),

Df = the dilution factor VT/A (2.5 where VT is the total volume of titrate (300 mL) and A is the aliquot used (125 mL),

ME = the molar equivalent of KMnO4 in oxalate (KMnO4 redox reaction) and Mf is the mass of flour sample used.

3.2.4.5 Determination of cyanogenic glycoside contents in samples

Alkaline picrate method as reported by Onwuka, (2005) was used to determine cyanide content in sample. Five grams of flour sample was dissolved in 50 mL of distilled water in a conical flask and the extraction was allowed to stand over-night for cyanide to be extracted. The extract was filtered and filtrate was used for cyanide determination. 1 mL of filtrate was mixed with 4 mL alkaline picrate in a test tube and incubated in a water bath for about 5 minutes. Reddish brown colour was developed and absorbance was taken at 490 nm, cyanide content was extrapolated from cyanide standard curve.

3.2.5 Determination of functional properties of sprouted seeds flour blends

3.2.5.1 Determination of percentage flour dispersibility

Dispersibility of flour samples was determined by the method reported by Kulkarni *et al*. (1991). For each sample, 10 g was suspended in 200 mL measuring cylinder and was made up to 100 mL mark of the measuring cylinder. The set-up was stirred vigorously and allowed to settle for 3 hours. The volume of settled particles was recorded and subtracted from 100. The difference was reported as percentage dispersibility.

$$\text{Dispersibility} = 100 - \text{volume of settled particles}$$

3.2.5.2 Determination of flour bulk density

The bulk density (BD) of flour sample was determined using the methods reported by Onwuka (2005). Ten grams of the sample was weighed into 50 mL graduated measuring cylinder. The sample was packed by gently tapping the cylinder on the bench top about 10 times from a height of 5 cm and the volume of the sample was recorded.

$$\text{Bulk density (g/ mL)} = \frac{\text{Weight of Sample}}{Volume\ of\ Sample\ after\ tapping} \qquad 3.8$$

3.2.5.3 Determination of water absorption capacity (WAC) and oil absorption capacity (OAC) of flour blends

The method as reported by Onwuka (2005) was used to determine the WAC and OAC of flour samples. One gram of the flour sample was weighed into a 15 mL centrifuge tube and suspended in 10 mL of water/oil. It was shaken on a platform tube rocker for 1 minute at 37 ^{o}C. The sample was allowed to stand for 30 minutes and centrifuged at 1200 x g for 30 minutes. The volume of free water/oil was read directly from the centrifuge tube.

$$\text{WAC/OAC (\%)} = \frac{\frac{\text{Amount of water}}{\text{oil added}} - \frac{\text{free water}}{\text{oil}} \text{X} \frac{\text{density of water}}{\text{oil}} \text{X100}}{weight\ of\ sample} \qquad 3.9$$

3.2.5.4 Determination of flour viscosity

Viscosity of flour sample was determined according to the methods reported by Krishnan *et al*. (2011). One gram of the flour sample was dissolved in 100 mL of distilled water and heated to hydrate for about 30 minutes at 95 ± 1 ^{o}C with continuous stirring. The slurry was allowed to cool and the viscosity was measured in Brookfield viscometer (Model RV, Brookfield Engineering, Inc., USA) using spindle number Q3 rotating at 12.0 rpm and the cold paste viscosity was measured in centipoise (cP).

3.2.6 Determination of amylose and amylopectin content in sprouted seeds flour samples

Spectrophotometric method as reported by Chrastil (1987) with few modifications was used to determine amylose contents in flour sample. A flour sample (0.032) was weighed into a 50 mL volumetric flask. DMSO (1 mL) was added to the flask and gently stirred at a low speed on a vortex mixer. The tubes were capped and heated in a water bath for about 30 minutes until the sample was completely dispersed and mixture was smooth. Sample was allowed to cool. A 5 mL of solution (containing 0.08 g of iodine, 0.27 g of potassium iodide and 250 mL of distilled water) was added to the sample and made up to a volume of 50 mL with distilled water. A blue coloration was observed and absorbance was measured at 600 nm wavelength against the reference solution. The amylose content was determined using equation from a standard curve with amylose.

$$\text{Amylose contents (x)} = \frac{\text{Y}-0.2138}{0.0168} \qquad 3.10$$

Amylopectin content was determined by subtracting the amylose content from 100

$$Amylopectin\ \% = 100 - amylose\ content \qquad 3.11$$

3.2.7 Formulation of composite flour

Composite flour was formulated using D-optimal mixture design (Design Expert 9.0) as reported by Abioye *et al*. (2018) with few modifications. The design was based on 100 % white flour, 80 % white flour and 20 % sprouted finger millet flour, 70 % white flour, 20 % sprouted finger millet flour and 10 % sprouted water melon seed flour, 60 % white flour, 30 % sprouted finger millets and 10 % sprouted water melon seed flour, and 50 % white flour, 30 % sprouted finger millets and 20 % sprouted water melon seed flour. The generated mixes are shown in Table 3.1.

Table 3.1: Formulation of Composite Flour

Parameters/Sample (%)	White flour	Sprouted finger millet flour	Sprouted water melon seed flour
A	100	-	-
B	80	20	-
C	70	20	10
D	60	30	10
E	50	30	20

Key; A: 100 % White flour, B: 80 % white flour, 20 % sprouted finger millet flour, C: 70 % White flour, 20 % sprouted finger millet flour, 10 % sprouted water melon seed flour, D: 60 % White flour, 30 % sprouted finger millet flour, 10 % sprouted water melon seed flour, E: 50 % White flour, 30% sprouted finger millets flour, 20 % sprouted water melon seed flour.

3.2.8 Formulation of bread recipes

Bread recipe was formulated according to the methods reported by Bugusu *et al*. (2001) The formulation was based on 100 % white flour, 80 % white flour and 20 % sprouted finger millet flour, 70 % white flour, 20 % sprouted finger millet flour and 10 % sprouted water melon seed flour, 60 % white flour, 30 % sprouted finger millets and 10 % sprouted water melon seed flour, and 50 % white flour, 30 % sprouted finger millets and 20 % sprouted water melon seed flour. The generated recipes are shown in Table 3.2.

Table 3.2: Formulation of Bread Recipe from Composite Flour Mixes

Parameters/ sample (%)	WF %	SFMF %	SWMSF %	Yeast mg	Salt mg	Sugar %	Baking butter %	Water mL
A	100	-	-	2	2	2	1	55
B	80	20	-	2	2	2	1	58
C	70	20	10	2	2	2	1	60
D	60	30	10	2	2	2	1	63
E	50	30	20	2	2	2	1	65

Key; WF: white flour, SFMF: sprouted finger millet flour, SWMSF: sprouted water melon seed flour. A: 100 % WF, B: 80 % WF, 20 % SFMF, C: 70 % WF, 20 % SFMF, 10 % SWMSF, D: 60 % WF, 30 % SFMF, 10 % SWMSF, E: 50 % WF, 30 % SFMF, 20 % SWMSF.

3.2.9 Bread preparation

Composite flour mix was formed into dough, kneaded, cut, and baked using the methods as reported by Oyeyinka *et al*. (2014) with few changes in measurements. The flow chart for bread dough preparation is presented in Figure 3.1.

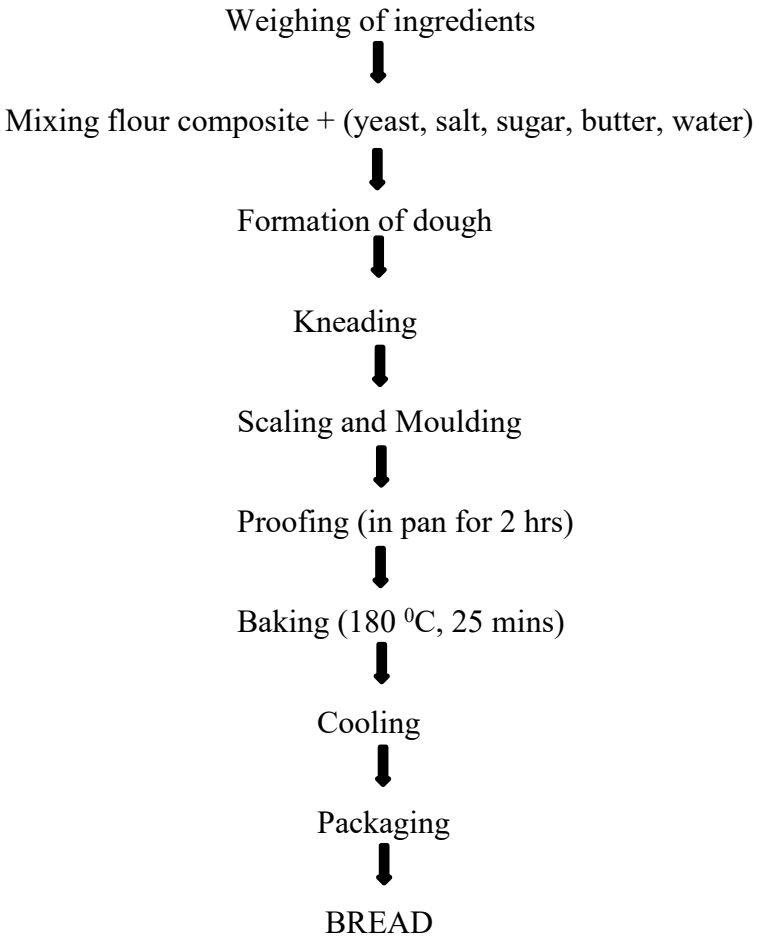

Figure 3.1: Flow chart for bread preparation.
Source; Oyeyinka *et al*. (2014).

3.2.10 Formulation of cookie recipes

Cookie recipes were prepared following the methods reported by McWatters *et al*. (2003). The formulation was based on 100 % white flour, 80 % white flour and 20 % sprouted finger millet flour, 70 % white flour, 20 % sprouted finger millet flour and 10 % sprouted water melon seed flour, 60 % white flour, 30 % sprouted finger millets and 10 % sprouted water melon seed flour, and 50 % white flour, 30 % sprouted finger millets and 20 % sprouted water melon seed flour. The generated recipes are shown in 3.3.

Table 3.3: Formulation of Cookie Recipe from Composite Flour Mixes

Parameter/ Sample%	WF	SFMF	SWMSF	White sugar teaspoon	Salt teaspoon	Eggs	Baking butter teaspoon	Baking powder teaspoon
A	100	-	-	½	½	1	1	½
B	80	20	-	½	½	1	1	½
C	70	20	10	½	½	1	1	½
D	60	30	10	½	½	1	1	½
E	50	30	20	½	½	1	1	½

Key; WF: white flour, SFMF: sprouted finger millet flour, SWMSF: sprouted water melon seed flour. A: 100 % WF, B: 80 % WF, 20 % SFMF, C: 70 % WF, 20% SFMF, 10 % SWMSF, D: 60 % WF, 30 % SFMF, 10 % SWMSF, E: 50 % WF, 30 % SFMF, 20 % SWMSF.

3.2.11 Cookie preparation

The cookies were prepared using the methods as reported by Caserani *et al*. (2008). The flow chart for cookie preparation is presented in Figure 3.2 below;

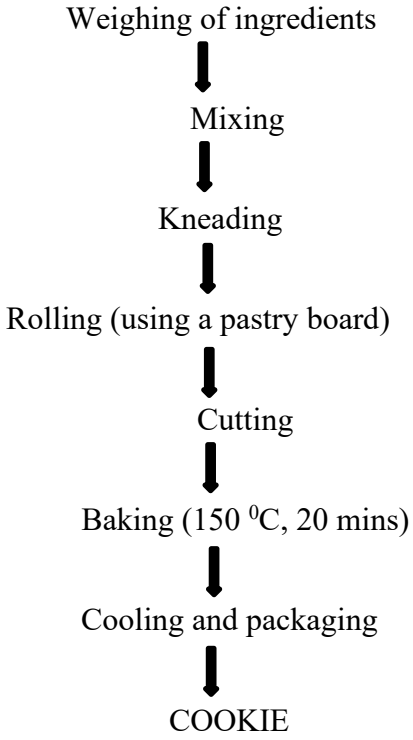

Figure 3.2: Flow chart for cookie preparation.
Source: Caserani e*t al*. (2008).

3.2.12 Microbiological analysis of bread and cookie samples

The total viable bacterial counts and fungi counts (yeast and mould counts) were carried out on the bread and cookies samples to determine the microbial load of the samples following the methods as reported by Udeme *et al*. (2014). Bread and cookies samples were prepared by mashing and mixing in peptone water. Sub-samples were diluted and 0.1 mL aliquots were spread on nutrient agar (NA), and MacConkey agar (MCA) plate for the separation of aerobic viable bacteria, coliforms, and fungi, respectively. The agar plates were incubated at 37 ^{0}C for 24 – 48 hours. Colonies were counted and expressed as colony forming units per gram (cfu/g) of samples. All counts were done in duplicate

using the Stuart scientific colony counter. Observed colonies were sub-cultured repeatedly on media used for primary isolation to obtain pure cultures.

3.2.13 Characterization and identification of isolates in bread and cookie samples

The bacterial isolates were characterized using Gram reaction and biochemical tests. They were identified by comparing their characteristics with those of known taxa as outlined in Bergey's Manual of Systematic Bacteriology Krieg *et al*. (1994). The fungal isolates were characterized based on macroscopic and microscopic examination and identified using the scheme of Alexopoulus and Mims (1979).

3.2.14 Storage of bread and cookie samples

Bread and cookie samples were stored under ambient temperature (26 – 33 ^{0}C) and observed for about 14 days. Visual observations for mold growth were carried out on the samples stored.

3.2.15 Sensational test of bread and cookie products

Sensory evaluation of bread and cookie was performed 24 hours after baking following the methods as reported by Meilgaard *et al.* (2007). A panel of ten trained and semi-trained panelist (using a questionnaire) were used to evaluate the appearance, colour, aroma, taste, crumb texture, crispiness, and overall acceptance of the bread and cookie using a Hedonic scale set up and sensory scores were analyzed statistically.

3.2.16 Cost on quality attributes of bread and cookie products

Bread and cookie samples were subjected to cost rate (i. e cost of production versus sales price), and cost effectiveness was calculated in percentage.

3.3 Statistical Data Analysis

Statistical analyses were performed using SPSS (version 2.2.0) and one-way analysis of variance (ANOVA). Values were considered statistically significant at ($p < 0.05$).

CHAPTER FOUR

4.0 RESULTS AND DISCUSSION

4.1 Results

4.1.1 Results of sprouting on weight loss and drying rate of finger millets and water melon seeds

The results of the effects of sprouting on weight loss and drying rate of finger millets and water melon seeds are presented in Figure 4.1a and 4.1b. There was a significant difference ($p<0.05$) in drying rate of sprouts particularly, those sprouted for six (6) days had better drying rate.

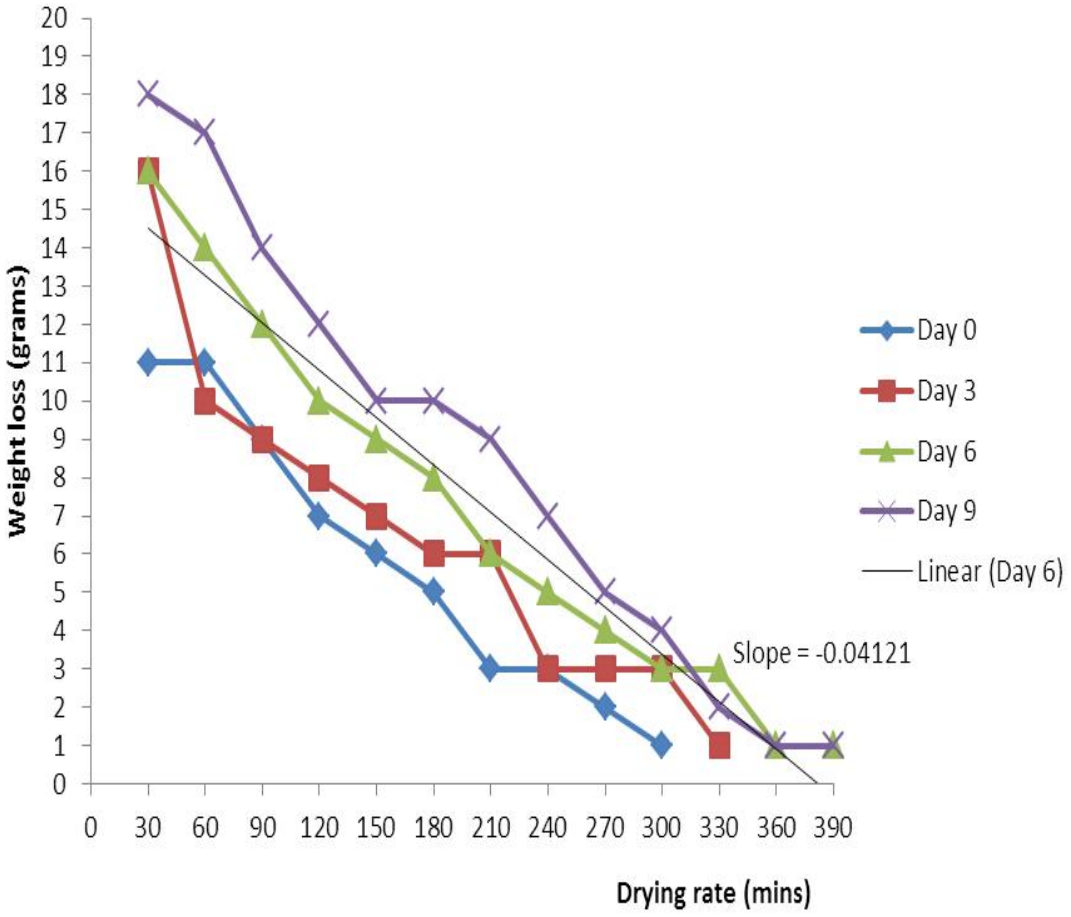

Figure 4.1a: Weight Loss against Drying Time for Sprouted Finger Millets.

Key: Day 0= non- sprouted seeds, Day 3= seeds sprouted for three days, Day 6= seeds sprouted for six days, Day 9= seeds sprouted for nine days.

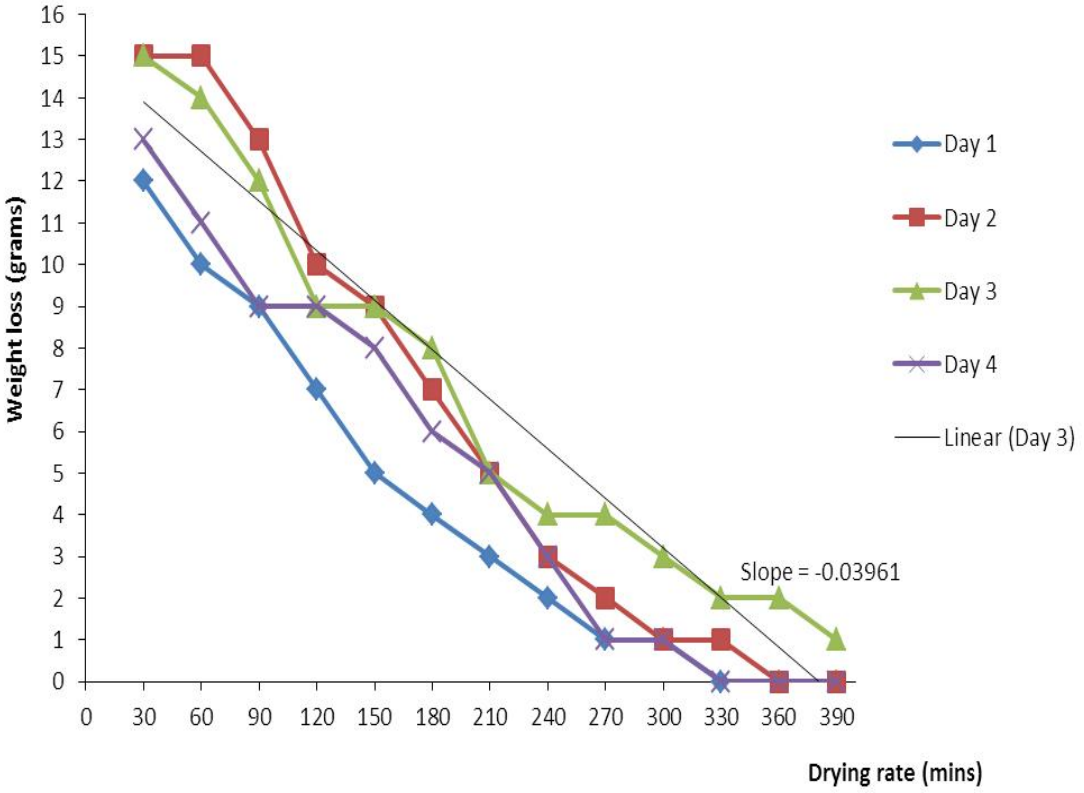

Figure 4.1b: Weight Loss against Drying Time for Sprouted Water Melon Seeds.

Key: Day 1= non - sprouted seeds, Day 2= seeds sprouted for three days, Day 3= seeds sprouted for six days, Day 4= seeds sprouted for nine days

4.1.2 Proximate compositions of finger millets and water melon seeds

The results of proximate compositions of sprouted and non-sprouted finger millets and water melon seeds are presented in Table 4.1a and 4.1b. Moisture contents of seeds ranged from 3.23±0.01 to 4.74±0.24 % in finger millets and 2.22±0.01 to 2.56±0.01 % in water melon seeds. There was a significant difference ($p<0.05$) in the moisture contents of the seeds. The ash contents of seeds ranged from 1.15±0.03 to 1.32±0.02 %

in finger millets and 1.37±0.01 to 1.54±0.02 % in water melon seeds. Both seeds showed no significant difference ($p>0.05$) in ash contents. The crude fibre contents of seeds ranged from 0.09±0.12 to 1.14±0.01 % in finger millets and 0.39±0.02 to 2.28±0.01 % in water melon seeds. There was a significant difference ($p<0.05$) in the fibre contents of sprouted seeds. The fat contents of seeds ranged from 1.90±0.01 to 2.12±0.01 % in finger millets and 25.83±0.01 to 27.10±0.01 % in water melon seeds. The crude protein contents of seeds ranged from 12.34±0.03 to 24.61±0.05 % in finger millets and 22.38±0.04 to 30.63±0.03 % in water melon seeds respectively. There was a significant difference ($p<0.05$) in protein contents of seeds. The total carbohydrate contents of seeds ranged from 68.34±0.08 to78.49±0.04 % in finger millets and 37.37±0.02 to55.33±1.89 % in water melon seeds. There was a significance difference ($p<0.05$) in the total Carbohydrate content of sprouted seeds.

Table 4.1a: Impact of Sprouting on Proximate Compositions of Finger Millets

Parameters/Treatment	Day 0	Day 3 (%)	Day 6	Day 9
Moisture	4.74±0.02^{c}	4.68±0.01^{b}	3.36±0.01^{a}	3.23±0.01^{a}
Ash	1.23±0.01^{b}	1.27±0.01^{b}	1.42±0.02^{c}	1.15±0.03^{a}
Crude fibre	1.14±0.01^{d}	0.67±0.07^{c}	0.24±0.09^{b}	0.09±0.12^{a}
Crude fat	2.04±0.01^{b}	2.09±0.00^{b}	2.12±0.01^{b}	1.90±0.01^{a}
Crude protein	12.34±0.03^{a}	18.14±0.04^{b}	24.61±0.05^{d}	21.93±0.02^{c}
Carbohydrates	78.49±0.04^{d}	73.14±0.02^{b}	68.35±0.08^{a}	73.68±0.03^{c}

Values are mean of triplicate determinations ± SEM Values along rows with different superscript are significantly different ($p<0.05$)

Key: Day 0= non- sprouted seeds, Day 3= seeds sprouted for three days, Day 6= seeds sprouted for six days, Day 9= seeds sprouted for nine days

Table 4.1b: Impact of Sprouting on Proximate Compositions of Water Melon Seeds

Parameters/ Treatment	Day 0	Day 3 (%)	Day 6	Day 9
Moisture	2.26±0.03[a]	2.56±1.06[b]	2.34±0.01[a]	2.22±0.01[a]
Ash	1.43±0.03[a]	1.46±0.07a[b]	1.54±0.02[b]	1.37±0.01[a]
Crude fibre	2.38±0.01[d]	1.33±0.02[c]	1.02±0.02[b]	0.69±0.02[a]
Crude fat	26.29±0.01[b]	26.52±0.03[c]	27.10±0.01[d]	25.83±0.02[a]
Crude protein	22.38±0.04[a]	24.80±0.03[b]	30.63±0.03[d]	26.43±0.02[c]
Carbohydrates	45.25±0.04[b]	55.33±1.89[a]	37.37±0.02[d]	43.54±0.02[c]

Values are mean of triplicate determinations ± SEM Values along rows with different superscript are significantly different ($p<0.05$)

Key: Day 0= non- sprouted seeds, Day 3= seeds sprouted for three days, Day 6= seeds sprouted for six days, Day 9= seeds sprouted for nine days

4.1.3 Vitamin compositions of finger millets and water melon seeds

The results of vitamin compositions of sprouted and non-sprouted finger millets and water melon seeds are represented in Table 4.2a and 4.2b respectively. Vitamin B_1 contents ranged from 0.423±0.007 to 0.767±0.009 Mg/100 g in finger millet and 1.177±0.044 to 1.647±0.030 Mg/100 g in water melon seeds. There was no significant difference ($p>0.05$) in sprouted seeds. Vitamin B_2 contents ranged from 0.217±0.020 to 1.630±0.025 Mg/100 g in finger millets and 2.643±0.057 to 4.097±0.0.038 Mg/100 g in water melon seed. There was a significant difference ($p<0.05$) in vitamin B_2 contents in sprouted seeds. Vitamin B_3 contents was highest in seeds sprouted for six days and ranged from 0.870±0.040 to 4.237±0.015 Mg/100 g in finger millets and 3.223±0.018 to 4.853±0.050 Mg/100 g in water melon seeds. Vitamin B_5 content ranged from 0.126±0.052 to 1.363±0.031 Mg/100 g in finger millets and 2.340±0.006 to 3.297±0.007 Mg/100 g in water melon seed. Vitamin B_6 also increased during sprouting.

Highest values (1.853±0.026 Mg/100 g) and (4.090±0.035 Mg/100 g) were recorded in finger millets and water melon seeds sprouted for six days. Vitamin B_9 contents ranged from 0.117±0.007 to2.390±0.006 Mg/100 g in finger millets and 1.107±0.233 to 2.697±0.044 Mg/100 g in water melon seeds. Vitamin C contents ranged from 2.180±0.012 to 3.293±0.023 Mg/100 g in finger millets and 2.440±0.015 to 3.867±0.024 Mg/100 g in water melon seeds. There was a significant difference ($p<0.05$) following sprouting in vitamin C contents. Vitamin E contents ranged from 0.867±0.020 to 3.147±0.012 Mg/100 g in finger millets and 4.070±0.017 to 5.843±0.026 Mg/100 g in water melon seeds. There was a significant difference ($p<0.05$) particularly for seeds sprouted for six days.

Table 4.2a: Impact of Sprouting on Vitamin Compositions of Finger Millets

Parameters/ Treatment	Day 0	Day 3 (Mg/100 g)	Day 6	Day 9
Vitamin B_1	$0.443±0.023^a$	$0.767±0.009^c$	$0.647±0.049^b$	$0.423±0.007^a$
Vitamin B_2	$0.217±0.020^a$	$0.687±0.007^b$	$1.630±0.025^d$	$1.440±0.006^c$
Vitamin B_3	$0.870±0.040^a$	$2.053±0.023^b$	$4.237±0.015^d$	$2.953±0.015^c$
Vitamin B_5	$0.126±0.052^a$	$0.693±0.003^b$	$1.363±0.031^d$	$0.950±0.031^c$
Vitamin B_6	$0.763±0.015^a$	$1.043±0.009^b$	$1.853±0.026^d$	$1.670±0.012^c$
Vitamin B_9	$0.117±0.007^a$	$1.760±0.031^b$	$2.390±0.006^d$	$1.940±0.015^c$
Vitamin C	$2.660±0.025^c$	$2.297±0.015^b$	$3.293±0.023^b$	$2.180±0.012^a$
Vitamin E	$0.867±0.020^a$	$2.413±0.009^b$	$3.147±0.012^d$	$2.607±0.012^c$

Values are mean of triplicate determinations ± SEM Values along rows with different superscript are significantly different ($p<0.05$)

Key: Day 0= non - sprouted seeds, Day 3= seeds sprouted for three days, Day 6= seeds sprouted for six days, Day 9= seeds sprouted for nine days.

Table 4.2b: Impact of Sprouting on Vitamin Compositions of Water Melon Seeds

Parameters/Treatment	Day 0	Day 3 (Mg/100 g)	Day 6	Day 9
Vitamin B_1	1.177±0.044[a]	1.447±0.015[b]	1.360±0.020[b]	1.647±0.030[c]
Vitamin B_2	2.643±0.057[a]	3.120±0.017[b]	4.097±0.0.038[d]	3.510±0.032[c]
Vitamin B_3	3.223±0.018[a]	3.827±0.068[b]	4.853±0.050[d]	4.207±0.092[c]
Vitamin B_5	2.340±0.006[a]	2.573±0.022[b]	3.297±0.007[d]	3.127±0.059[c]
Vitamin B_6	3.313±0.018[a]	3.777±0.028[b]	4.090±0.035[c]	3.857±0.024[b]
Vitamin B_9	1.327±0.047[b]	1.490±0.031[c]	2.697±0.044[d]	1.107±0.233[a]
Vitamin C	2.580±0.685[a]	2.440±0.015[b]	3.867±0.024[d]	2.580±0.042[c]
Vitamin E	4.070±0.017[a]	4.690±0.035[b]	5.843±0.026[d]	5.300±0.021[c]

Values are mean of triplicate determinations ± SEM Values along rows with different superscript are significantly different ($p<0.05$)

Key: Day 0= non- sprouted seeds, Day 3= seeds sprouted for three days, Day 6= seeds sprouted for six days, Day 9= seeds sprouted for nine days

4.1.4 Results of anti-nutrient compositions of finger millets and water melon seeds

Table 4.3a and 4.3b shows the result of anti-nutrient compositions of sprouted and non-sprouted finger millets and water melon seeds. The non-sprouted seeds contained higher amount of anti-nutrients as compared to the sprouted seeds. There was a significant difference ($p<0.05$) in anti-nutrient compositions of sprouted seeds. Phytate contents ranged from 132.67±38.39 mg/100 g to 479.48±59.71 mg/100 g in finger millets and 97.05±16.04 mg/100 g to 253.18±19.06 mg/100 g in water melon seeds. Tannin contents ranged from 20.00±0.19 mg/100 g to 33.11±0.63 mg/100 g in finger millets and 26.28±0.19 mg/100 g to 45.25±0.07 mg/100 g in water melon seed. The concentrations of saponin in finger millets was highest is non- sprouted seeds (965.46±70.51 mg/100 g) and lowest in seeds sprouted for nine days (511.46±45.34 mg/100 g) and water melon seeds ranged from 86.46±98.20 mg/100 g to 159.13±35.28

mg/100 g in sprouted and non-sprouted seeds. Oxalate contents ranged from 0.19±0.01 mg/100 g to 0.33±0.01 mg/100 g in finger millets and 0.10±0.01 mg/100 g to 0.50±0.03 mg/100 g in water melon seeds while, cyanide contents ranged from 113.00±45.84 mg/100 g to 181.33±25.17 mg/100 g in finger millets and 110.87±18.33 mg/100 g to 388.33±44.35 mg/100 g in water melon seeds respectively.

4.1.5 Results of amylose and amylopectin contents of finger millets and water melon seeds

The results of the effects of sprouting on amylose and amylopectin contents of finger millets and water melon seed flours is presented in Table 4.4.. The amylose and amylopectin contents ranged from 21.24±0.53 % to 40.45±0.36 %, and 59.55±0.60 % to 78.76±0.53 % in finger millets. While amylose and amylopectin contents of water melon seeds ranged from 15.23±0.31 % to 26.96±0.36 % and 73.04±0.63 % to 84.77±0.31 % respectively. A significant difference ($p<0.05$) was observed in amylose and amylopectin contents of sprouted seed flours.

Table 4.3a: Impact of Sprouting on Anti-nutrient Compositions of Finger Millets

Parameters/ Treatment	Day 0	Day 3 (mg/100 g)	Day 6	Day 9
Phytates	479.48±33.71^{a}	398.68±27.15^{b}	154.39±24.03^{c}	132.67±38.39^{d}
Tannins	33.11±12.63^{a}	24.71±10.04^{b}	21.85±08.14^{c}	20.00±8.19^{d}
Saponins	965.46±41.51^{a}	811.46±35.05^{b}	641.46±27.07^{c}	511.46±45.34^{d}
Oxalates	0.33±17.02^{a}	0.27±013.54^{b}	0.20±10.09^{b}	0.19±12.01^{c}
Cyanogenic Glycoside	181.33±30.17^{a}	143.66±13.68^{b}	120.60±25.01^{c}	113.00±18.13^{d}

Values are mean of triplicate determinations ± SEM Values along rows with different superscript are significantly different ($p<0.05$)

Key: Day 0= non - sprouted seeds, Day 3= seeds sprouted for three days, Day 6= seeds sprouted for six days, Day 9= seeds sprouted for nine days

Table 4.3b: Impact of Sprouting on Anti-nutrient Compositions of Water Melon Seeds

Parameters /Treatment	Day 0	Day 3 (mg/100 g)	Day 6	Day 9
Phytates	253.18±19.06[a]	201.97±35.83[b]	124.93±28.10[c]	97.05±16.04[d]
Tannins	45.25±8.07[a]	37.03±7.35[b]	30.14±10.01[c]	26.28±8.19[d]
Saponins	159.13±35.28[a]	120.80±23.09[b]	106.13±26.67[c]	86.46±21.20[d]
Oxalate	0.52±10.03[a]	0.32±8.02[b]	0.16±8.01[c]	0.10±10.01[c]
Cyanogenic Glycoside	388.33±44.35[a]	310.67±34.61[b]	219.40±29.01[c]	110.87±18.33[d]

Values are mean of triplicate determinations ± SEM Values along rows with different superscript are significantly different ($p<0.05$)

Key: Day 0= non - sprouted seeds, Day 3= seeds sprouted for three days, Day 6= seeds sprouted for six days, Day 9= seeds sprouted for nine days

Table 4.4: Impact of Sprouting on Amylose and Amylopectin Contents of Sprouted Seed Flours

Parameters/Treatment	Day 0	Day 3 (%)	Day 6	Day 9
Amylose (FM)	40.45±0.36[d]	33.57±0.38[c]	28.68±0.66[b]	21.24±0.53[a]
Amylopectin (FM)	59.55±0.60[a]	66.43±0.38[b]	71.32±0.66[c]	78.76±0.53[d]
Amylose (WM)	26.96±0.36[d]	21.23±0.31[c]	18.57±0.56[b]	15.23±0.31[a]
Amylopectin (WM)	73.04±0.63[a]	78.77±0.31[b]	81.43±0.56[c]	84.77±0.31[d]

Values are mean of triplicate determinations ± SEM Values along rows with different superscript are significantly different ($p<0.05$)

Key: FM= finger millet WM= water melon seeds

Day 0= non- sprouted seeds, Day 3= seeds sprouted for three days, Day 6= seeds sprouted for six days, Day 9= seeds sprouted for nine days**.**

4.1.6 Results of proximate composition of composite flour mixes

The result of the impact of sprouting on proximate compositions of composite flour mixes is presented in Table 4.5. There was a significant difference ($p<0.05$) in the crude protein content of all the composite flours samples with sample E (50 % white flour,

30 % sprouted finger millet flour, 20 % sprouted water melon seed flour) having the highest amount of protein (13.62± 1.37 %) and sample A (100 % white flour) having the least protein content (6.89±0.60 %). Sample A (100 % white flour) had the least amount of moisture (5.67±0.20 %) while sample E had the highest amount of moisture (6.77±0.32 %). The Crude fat contents of the various flour composites ranged from 1.13±0.14 to 3.39±0.65 % while the crude fibre contents ranged from 1.13±0.53 to 3.68±1.42 %. The ash contents ranged from 1.82±1.40 to 5.48±1.72 %. There was a significant difference ($p<0.05$) in total carbohydrate contents of composite flours with sample A (100 % white flour) having the highest amount of total carbohydrates (80.18±0.22 %) while Sample E (50 % white flour, 30 % sprouted finger millets, 20 % sprouted water melon seed) had the least amount of carbohydrates (64.06±1.32 %).

Table 4.5: Proximate Compositions of Composite Flour Mixes

Parameters/ Sample	A	B (%)	C	D	E
Moisture	5.67±0.20^{a}	6.65±0.05^{a}	6.44±0.03^{a}	6.41±0.10^{b}	6.77±0.32^{b}
Ash	1.82±1.40^{a}	2.03±0.17^{b}	2.71±1.38^{c}	3.58±1.70^{d}	5.48±1.72^{e}
Crude fibre	1.06±0.53^{a}	1.16±0.13^{a}	2.68±0.23^{b}	3.01±1.15^{c}	3.68±1.42^{d}
Crude fat	1.31±0.14^{a}	1.45±0.13^{a}	2.43±0.72^{b}	2.87±0.66^{b}	3.39±0.65^{c}
Crude protein	6.89±0.60^{a}	7.29±0.21^{b}	11.12±1.03^{c}	11.63±1.42^{c}	13.62±1.37^{d}
Carbohydrates	80.18±0.22^{e}	79.22±1.55^{d}	73.41±0.90^{c}	71.01±0.53^{b}	64.06±1.32^{a}

Values are mean of triplicate determinations ± SEM
Values along rows with different superscript are significantly different ($p<0.05$)

Key: A: 100 % White flour, B: 80 % white flour, 20 % sprouted finger millet flour, C:70 % White flour, 20 % sprouted finger millet flour, 10 % sprouted water melon seed flour, D: 60 % White flour, 30 % sprouted finger millet flour, 10 % sprouted water melon seed flour, E: 50% White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour.

4.1.7 Results of functional properties of composite flour mixes

The result of the functional properties of composite flour mixes is presented in Figure 4.2. No significant difference ($p>0.05$) was observed for bulk density. There was a significant difference ($p<0.05$) in percentage flour dispensability with sample E (50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour) having the highest percentage flour dispersibility (48.37±0.41), while sample A (100 % white flour) had the least percentage flour dispensability (35.78±0.03). There was a significant difference in water absorption capacity in the various flour samples. Sample A (100 % white flour) had the least water absorption capacity (106.65±3.11) while sample E (50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour) had the highest water absorption capacity (168.93±1.74). Oil absorption capacity ranged from 91.70±3.19 % in sample A (100 % white flour) to 99.75±2.02 % in sample E (50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour). Composite flour viscosity ranged from 1072.65±4.11 to 1851.50±5.34 (RVU) at a temperature of 49.7 ^{o}C in 4.3 minutes. There was a significant difference ($p<0.05$) in viscosity of the various flour samples.

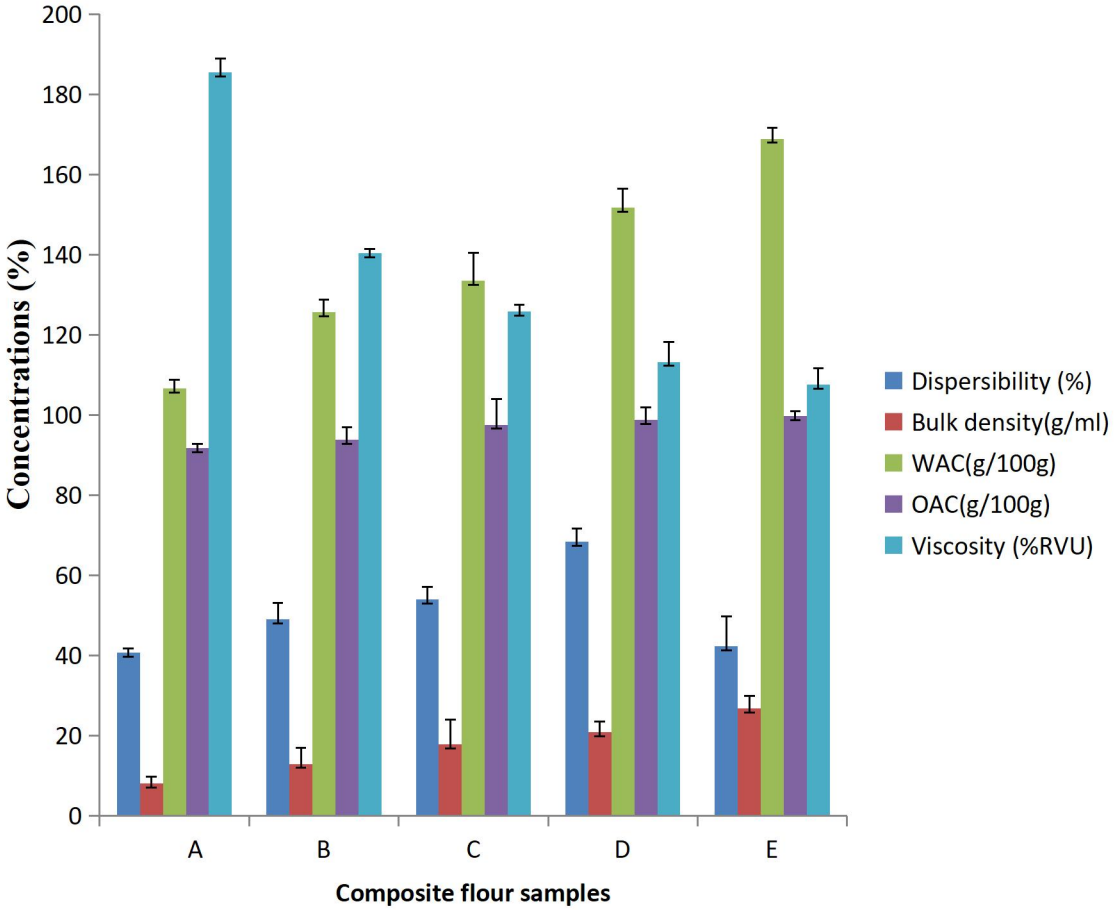

Figure 4.2: Functional Properties of Composite Flour Mixes

Key: WAC= Water absorption capacity, OAC= Oil absorption capacity

A: 100 % White flour, B: 80 % white flour, 20 % sprouted finger millet flour, C:70 % White flour, 20 % sprouted finger millet flour, 10% sprouted water melon seed flour, D: 60 % White flour, 30 % sprouted finger millet flour, 10 % sprouted water melon seed flour, E: 50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour.

4.1.8 Pictorial representation of snacks produced from composite flour mixes

The pictorial representation of snacks produced from composite flour mixes of white flour and substituted sprouted flours are presented in Plate IV and V. The mixes are represented in percent as follows; A: 100 % White flour, B: 80 % white flour, 20 % sprouted finger millet flour, C: 70 % White flour, 20 % sprouted finger millet flour, 10 % sprouted water melon seed flour, D: 60 % White flour, 30 % sprouted finger millet

flour, 10 % sprouted water melon seed flour, E: 50 % White flour, 30% sprouted finger millets flour, 20 % sprouted water melon seed flour respectively.

4.1.9 Results of physical properties of bread

The results of physical properties of bread produced from composite flour mixes is presented in Table 4.6. The following parameters; length, breadth, height, thickness and temperature were evaluated in bread samples. No Significant difference ($p>0.05$) was observed among the length and breadth of the bread samples. The height of bread samples ranged from 3.01±1.06 to 4.34±2.05 cm. There was a significant difference in the height of bread samples. Increase in substitution of sprouted seed flours increased the thickness of bread sample increased ranging from 2.95±0.03 to 3.88±0.18 respectively. The temperature at which the bread was fully cooked reduced with increased substitution of sprouted seed flours with sample A (100 % white flour) having the highest temperature range of 180 ^{o}C while sample E (50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour) having the least temperature range of 150 ^{o}C.

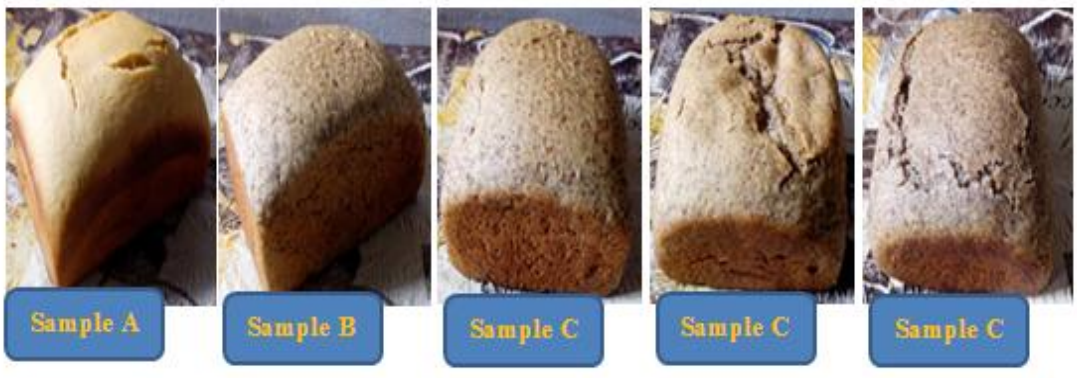

Plate IV: Bread Samples Produced from Composite Flour Mix

Key; A: 100 % White flour, B: 80 % white flour, 20 % sprouted finger millet flour, C:70 % White flour, 20 % sprouted finger millet flour, 10 % sprouted water melon seed flour, D: 60 % White flour, 30 % sprouted finger millet flour, 10 % sprouted water melon seed flour, E: 50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour.

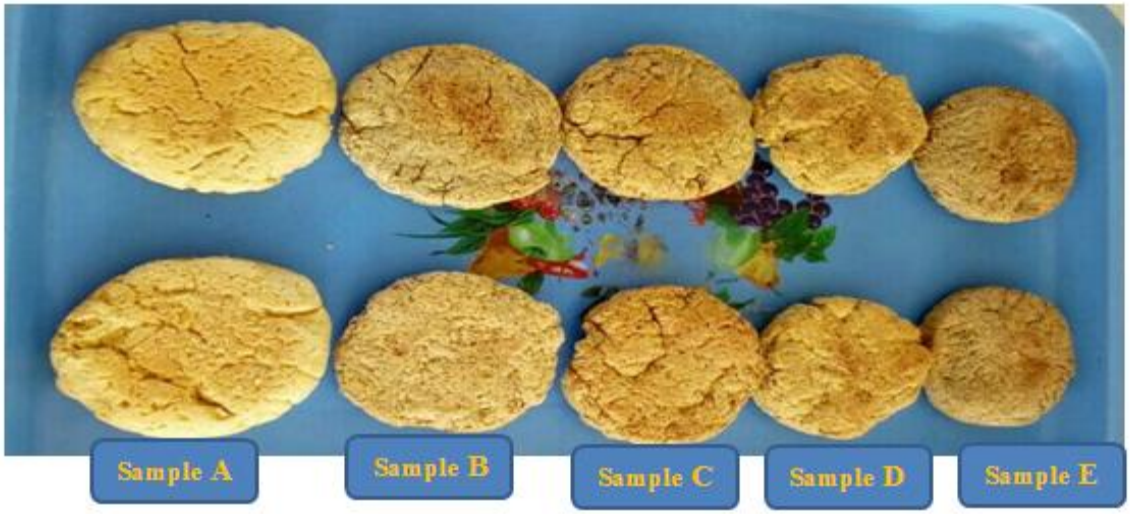

Plate V: Cookie Samples Produced from Composite Flour Mix

Key; A: 100 % White flour, B: 80 % white flour, 20 % sprouted finger millet flour, C:70 % White flour, 20 % sprouted finger millet flour, 10 % sprouted water melon seed flour, D: 60 % White flour, 30 % sprouted finger millet flour, 10 % sprouted water melon seed flour, E: 50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour.

Table 4.6: Impact of Baking on Physical Properties of Bread Samples

Parameters/Sample	A	B (%)	C	D	E
Length (cm)	4.35±1.37^{b}	4.32±1.12^{b}	4.20±1.10^{a}	4.22±0.97^{a}	4.30±1.25^{b}
Breadth (cm)	2.50±1.90^{b}	2.25±1.25^{b}	2.13±1.27^{a}	2.08±1.17^{a}	2.03±0.97^{a}
Height (cm)	4.34±2.05^{e}	4.11±0.57^{d}	3.90±1.71^{c}	3.23±1.50^{b}	3.01±1.06^{a}
Thickness (cm)	2.95±0.03^{a}	3.07±0.34^{b}	3.55±0.66^{c}	3.67±0.55^{d}	3.88±0.18^{e}
Temperature (^{0}C)	180 ^{0}C	172 ^{0}C	160 ^{0}C	157 ^{0}C	150 ^{0}C

Values are mean of triplicate determinations ± SEM

Values along rows with different superscript are significantly different ($p<0.05$)

Key: A: 100 % White flour, B: 80 % white flour, 20 % sprouted finger millet flour, C:70 % White flour, 20 % sprouted finger millet flour, 10 % sprouted water melon seed flour, D: 60 % White flour, 30 % sprouted finger millet flour, 10 % sprouted water melon seed flour, E: 50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour.

4.1.10 Results of proximate composition of bread samples

The result of the proximate composition of bread samples is presented in Table 4.7. There was a significant difference ($p<0.05$) in the crude protein content of bread samples. Bread produced from composite flour of sample E (50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour) had the highest amount of protein (11.90±1.03) while Sample A (100 % white flour) had the least amount of protein (5.93±1.03). Bread produced from Sample A (100 % white flour) had the least moisture contents (6.11±3.27 %) while sample E (50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour) had the highest amount of moisture contents (7.21±2.69 %). The Crude fat contents of the various bread samples ranged from 1.35±0.13 to 3.51±0.82 % while the crude fibre contents ranged from 1.13±0.14 to 3.69±1.07 %. There was a significant difference ($p<0.05$) in the fat and fibre contents of bread samples. The ash contents ranged from 1.82±1.40 to 5.48±1.72 %. Bread produced from sample A (100 % white flour) had the highest total carbohydrates (83.82±2.10 %) while bread produced from sample E (50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour) had the least total carbohydrates (68.73±0.09 %) contents.

Table 4.7: Proximate Compositions of Bread Samples

Parameters/ Sample	A	B (%)	C	D	E
Moisture	6.11±3.27[a]	6.24±3.09[a]	6.82±2.26[a]	7.15±2.43[b]	7.21±2.69[b]
Ash	1.53±0.72[a]	1.97±1.03[a]	2.11±1.17[b]	3.05±1.09[c]	4.96±0.33[d]
Crude fibre	1.13±0.14[a]	1.18±0.53[a]	2.71±0.77[b]	3.44±2.01[c]	3.69±1.07[c]
Crude fat	1.35±0.13[a]	1.68±0.72[a]	2.69±1.03[b]	2.95±1.42[b]	3.51±0.82[c]
Crude protein	5.93±1.03[a]	6.37±2.04[b]	8.59±1.14[c]	9.89±2.03[c]	11.90±1.03[d]
Carbohydrates	83.82±2.10[e]	82.69±2.06[d]	77.08±2.51[c]	73.52±1.89[b]	68.73±0.09[a]

Values are mean of triplicate determinations ± SEM Values along rows with different superscript are significantly different ($p<0.05$)

Key: A: 100 % White flour, B: 80 % white flour, 20 % sprouted finger millet flour, C:70 % White flour, 20 % sprouted finger millet flour, 10 % sprouted water melon seed flour, D: 60 % White flour, 30 % sprouted finger millet flour, 10 % sprouted water melon seed flour, E: 50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour.

4.1.11 Results of proximate composition of cookie samples

The result of the proximate composition of cookie samples is presented in Table 4.8. There was a significant difference ($p<0.05$) in the crude protein content of cookie samples. Cookies produced from composite flour of sample E (50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour) had the highest amount of protein (11.98±2.03 %) while Sample A (100 % white flour) had the least amount of protein (5.73±1.03 %). Cookies produced from Sample A had the least moisture contents (6.01±1.27 %) while sample E had the highest amount of moisture contents (7.00±1.03 %). The Crude fat contents of the various cookie samples ranged from 2.35±0.13 to 4.51±0.82 %. And crude fibre contents ranged from 1.03±0.14 to 3.35±1.01. There was a significant difference ($p<0.05$) in the fat and fibre contents of cookie samples. The ash contents ranged from 1.48±0.52 to 4.15±0.32 %. Cookies

produced from 100 % white flour (Sample A) had the highest amount of total carbohydrates (82.82±2.10 %) while cookie produced from sample E (50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour) had the least total carbohydrates (67.83±0.09 %) contents.

Table 4.8: Proximate Compositions of Cookie Samples

Parameters/ Sample	A	B (%)	C	D	E
Moisture	6.01±1.27^{a}	6.21±0.09^{a}	6.34±1.26^{a}	6.82±1.03^{b}	7.00±1.69^{b}
Ash	1.48±0.52^{a}	1.67±1.03^{a}	2.01±1.07^{b}	3.01±0.09^{c}	4.15±0.32^{d}
Crude fibre	1.03±0.14^{a}	1.09±0.53^{a}	2.21±0.77^{b}	3.35±1.01^{c}	3.08±1.07^{c}
Crude fat	2.35±0.13^{a}	2.68±0.72^{a}	3.69±1.03^{b}	3.95±1.42^{b}	4.51±0.82^{c}
Crude protein	5.73±1.03^{a}	7.17±2.04^{b}	9.01±1.14^{c}	10.49±2.03^{c}	11.98±2.03^{d}
Carbohydrates	82.82±2.10^{e}	81.69±2.06^{d}	75.08±2.51^{c}	73.02±1.89^{b}	67.83±0.09^{a}

Values are mean of triplicate determinations ± SEM
Values along rows with different superscript are significantly different ($p<0.05$)

Key: A: 100 % White flour, B: 80 % white flour, 20 % sprouted finger millet flour, C:70 % White flour, 20 % sprouted finger millet flour, 10 % sprouted water melon seed flour, D: 60 % White flour, 30 % sprouted finger millet flour, 10 % sprouted water melon seed flour, E: 50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour.

4.1.12 Shelf life stability and microbial counts of bread and cookie samples

The results of the shelf life stability and microbial counts of bread and cookie samples are presented in Table 4.9a and 4.9b. Shelf life of bread ranged from 4-7 days while cookies lasted from 6-10 days before spoilage occurred. Spoilage on bread and cookie samples was indicated by some yellow and green colouration which later turned to black colouration and this was suspected to be mold growth which consisted of *Aspergillus flavus, Rhizopus stolonifer, and Mucor spp* when stained and examined under the light microscope.

The total bacterial counts of the bread and cookie samples ranged from 1.0×10^2 cfu/g to 1.8×10^6 cfu/g and 3.7×10^3 cfu/g to 1.6×10^6 cfu/g with the highest being recorded for bread and cookie made from sample E (50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour) while the lowest counts were obtained in bread produced from 100 % white flour (sample A) and cookies produced from 80 % White flour, 20 % sprouted finger millet flour (sample B). The fungi counts ranged from 2.5×10^1 cfu/g to 1.2×10^3 cfu/g for bread and 3.0×10^1 cfu/g to 1.4×10^3 cfu/g for cookies. With the highest being recorded for bread and cookie produced from 50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour (sample E) while the lowest counts were obtained in bread produced from 100 % white flour (sample A) and cookies produced from 70 % White flour, 20 % sprouted finger millet flour, 10 % sprouted water melon seed flour (sample C). The species of bacterial isolated were *Bacillus subtilis*, *Micrococcus sp*, and *Staphylococcus aureus,* while fungi were *Rhizopus nigricans, Aspergillus niger,* and *Mucor sp.* There were no detectable values for both bacterial and fungal count in some cookie samples.

Table 4.9a: Microbial Counts and Shelf Life of Freshly Baked Bread

Sample/ treatment	Bacterial (cfu/g)	Fungi (cfu/g)	Spoilage of bread started after (days)
A	1.0×10^2	2.5×10^1	7
B	2.1×10^3	4.2×10^1	7
C	3.1×10^5	5.0×10^1	5
D	4.6×10^5	3.2×10^2	4
E	1.8×10^6	1.2×10^3	4

Key: Cfu/g: colony forming units per gram; A: 100 % White flour, B: 80 % white flour, 20 % sprouted finger millet flour, C: 70 % White flour, 20 % sprouted finger millet flour, 10 % sprouted water melon seed flour, D: 60 % White flour, 30 % sprouted finger millet flour, 10 % sprouted water melon seed flour, E: 50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour.

Table 4.9b: Microbial counts and shelf life of freshly baked cookies

Sample/ treatment	Bacterial (cfu/g)	Fungi (cfu/g)	Spoilage of cookies started after (days)
A	NG	NG	10
B	3.7×10^{3}	NG	10
C	4.7×10^{3}	3.0×10^{1}	7
D	6.4×10^{5}	4.7×10^{2}	6
E	1.6×10^{6}	1.4×10^{3}	6

Key: Cfu/g: colony forming units per gram; A: 100 % White flour, B: 80 % white flour, 20 % sprouted finger millet flour, C: 70 % White flour, 20 % sprouted finger millet flour, 10 % sprouted water melon seed flour, D: 60 % White flour, 30 % sprouted finger millet flour, 10% sprouted water melon seed flour, E: 50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour.

4.1.13 Results of mean sensory scores of breads

The result of the mean sensory scores of bread products is presented in Figure 4.3a. There was a significant difference ($p<0.05$) in bread sample produced from composite flours. The colour of bread ranged from 4.48±1.23 to 8.43±1.28. The aroma and taste of bread ranged from 6.03±1.09 to 8.21±1.44 and 4.25±1.11 to 8.80±1.70. There was a significant difference ($p<0.05$) in the aroma and taste of bread samples. The crumb texture of bread samples ranged from 4.42±1.08 to 8.05±1.30 while appearance ranged from 5.93±1.43 to 8.71±1.64. The overall acceptance of bread samples ranged from 5.02±1.18 to 8.44±1.47 with 100 % white flour bread having the highest acceptability range.

4.1.14 Results of sensory scores of cookies

The result of the sensory scores of cookie produced from composite flour is presented in Figure 4.3b. There was a significant difference ($p<0.05$) in cookie sample produced from composite flours. The colour of cookies ranged from 4.81±0.58 to 8.30±1.67. The

aroma and taste of cookies ranged from 4.55±1.59 to 8.75±1.73 and 5.15±0.94 to 7.90±1.19. There was a significant difference ($p<0.05$) in the aroma and taste of cookie samples. The texture of bread samples ranged from 5.00±0.59 to 8.00±1.40. Cookie crispiness ranged from 5.11±1.04 to7.10±1.70, while appearance ranged from 5.72±0.26 to8.12±0.15. The overall acceptance of cookie samples ranged from 5.19±0.89 to 8.02±1.56 with 100 % white flour cookie having the highest acceptability range.

4.1.15 Result of cost effectiveness

The result of cost effectiveness of each sample is presented in Figure 4.4. There was a significant difference ($p<0.05$) in cost effectiveness of each samples. However, bread and cookie produced from sample A (100 % white flour) had same production and selling price (₦100)

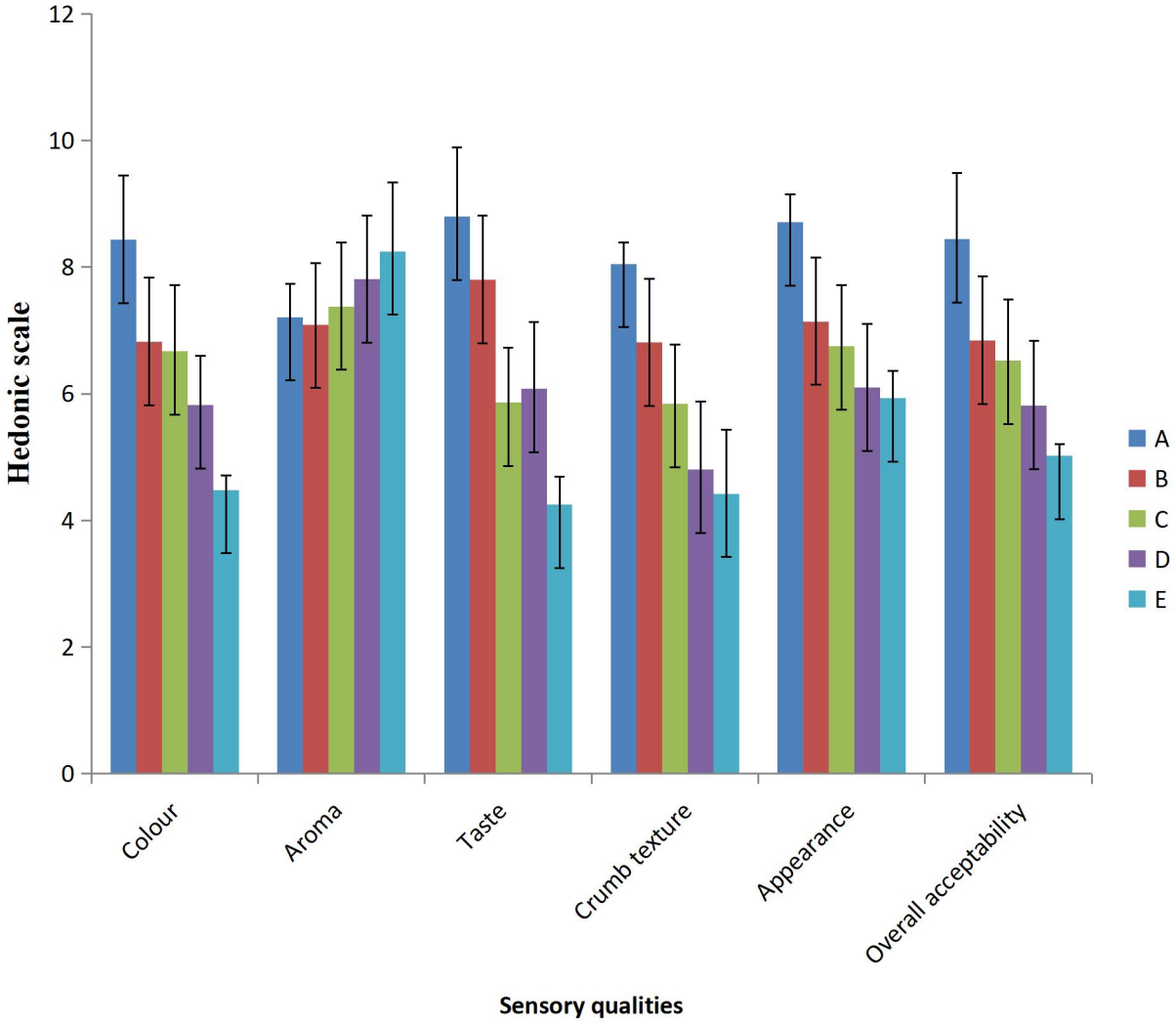

Figure 4.3a: Sensory Scores of Bread Samples

Key: A: 100 % White flour, B: 80 % white flour, 20 % sprouted finger millet flour, C: 70 % White flour, 20 % sprouted finger millet flour, 10 % sprouted water melon seed flour, D: 60 % White flour, 30 % sprouted finger millet flour, 10 % sprouted water melon seed flour, E: 50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour.

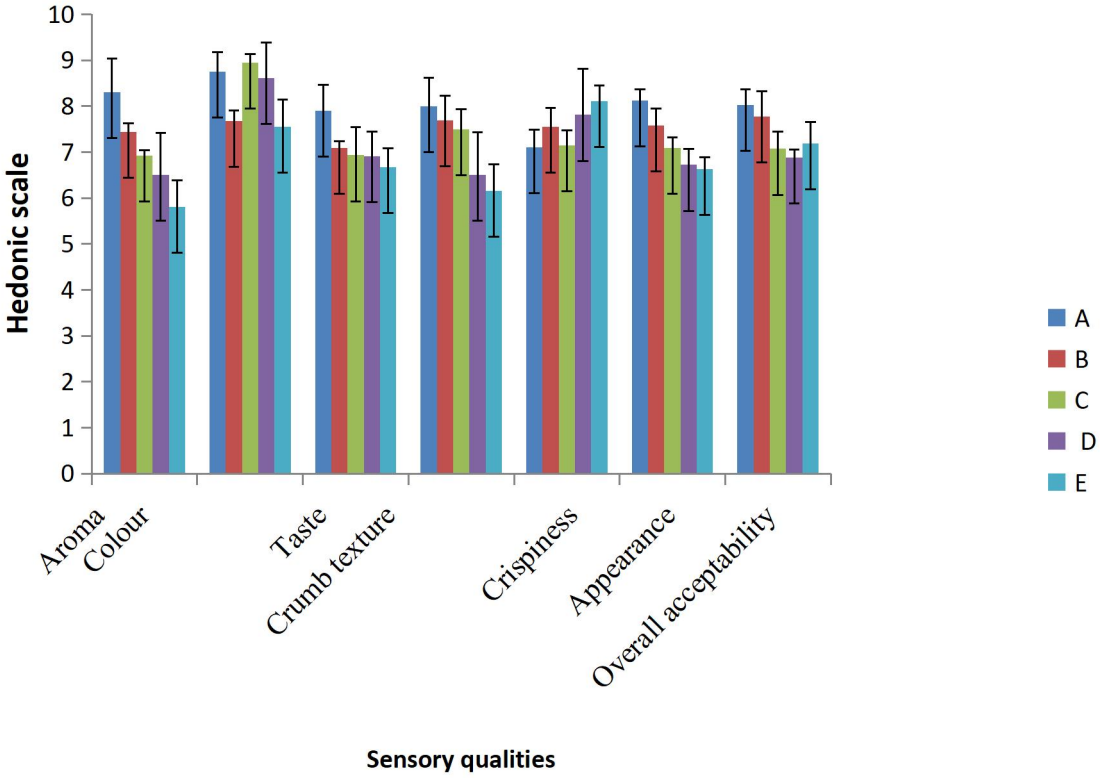

Figure 4.3b: Sensory Scores of Cookie Samples

Key: A: 100 % White flour, B: 80 % white flour, 20 % sprouted finger millet flour, C: 70 % White flour, 20 % sprouted finger millet flour, 10 % sprouted water melon seed flour, D: 60 % White flour, 30 % sprouted finger millet flour, 10 % sprouted water melon seed flour, E: 50 % White flour, 30 % sprouted finger millets flour, 20% sprouted water melon seed flour.

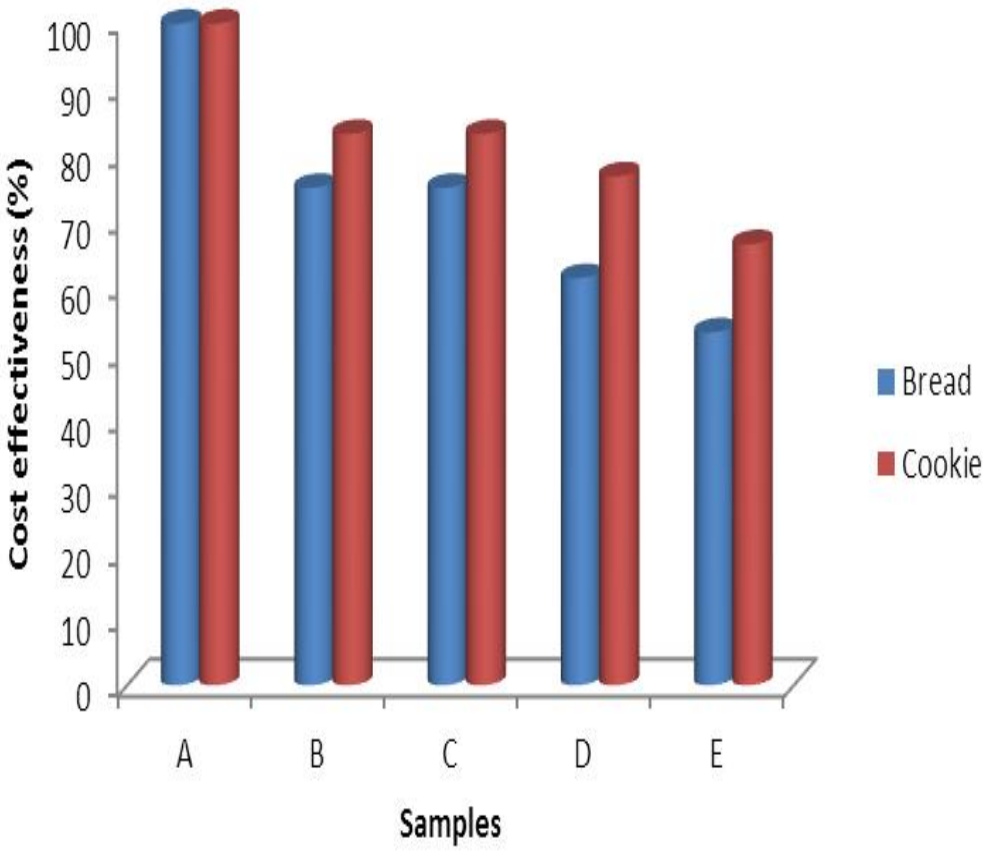

Figure 4.4: Cost Values for Bread and Cookie

Key: A: 100 % White flour, B: 80 % white flour, 20 % sprouted finger millet flour, C: 70 % White flour, 20 % sprouted finger millet flour, 10 % sprouted water melon seed flour, D: 60 % White flour, 30 % sprouted finger millet flour, 10 % sprouted water melon seed flour, E: 50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour.

4.2 Discussion

As drying time increases, moisture contents decreased in sprouted finger millets and water melon seeds as shown in (Figure 4.1a and 4.1b), this may be due to the hydroscopic nature of grains. The drying curve demonstrated falling rate pattern which is common to all agricultural products as reported by Ajala *et al*. (2012). However, seeds sprouted for six (6) days had better drying rate, hence, may have good storage potentials and suggested for formulations into composite flour.

Result from this study (Table 4.1a and 4.1b) has shown that finger millets and water melon seeds are good source of proteins. The protein contents increased upon sprouting and were significant at day six, this may be attributed to activation of metabolic enzymes such as proteinases, leading to the release and synthesis of some amino acids and peptides as a consequence, nutritional quality of proteins are enhanced. This is in line with the study of Faltermaier *et al*. (2015), who reported that protein contents increased in wheat and rice when sprouted for 3 to 7 days at 27 ^{0}C. Proteins are macronutrients, essential for growth and repair of body tissues (ward law, 2004). The decrease in protein contents at longer sprouting times may be attributed to leaching of water-soluble peptides in the steeping water. Sprouting seeds for six days may be suggested for improving protein quality in food crops. High protein content in sprouted finger millets and water melon seeds suggest that they may be desirable for fortification in confectionaries. The significantly low moisture content in sprouted seeds shows that sprouted finger millets and water melon seeds may have good storage potentials. This is in line with the work of Adeleke and Odedeji (2010) who reported that moisture content less than 14 % is recommended for long storage periods. Moisture content is an important parameter that indicates the self-life stability of food products (Fellows, 2000).

Increase in moisture contents, increases microbial contamination and chemical reaction that may lead to reduction in food quality and stability.

The decline in fats content especially at longer sprouting days may be attributed to denovo synthesis occurring during sprouting where by lipids are used up during respiration process. This is unique with the studies of Peterson, (1999), who reported that matured oat grains have high levels of lipase activity that remain unchanged or even decline during sprouting. Low fat contents in sprouted seeds suggest that they may be suitable in food formulations for children and the aged.

The ash contents that slightly increased during sprouting but decreased with long sprouting days this may be attributed to the loss of starch compounds especially when long sprouted times are applied. The presence of ash in food represents the total mineral contents of a food material (Nielson, 2002). Sprouting decreases percentage Carbohydrate contents in finger millets and water melon seeds. This is because during sprouting, metabolic enzymes such as amylases are involved in hydrolysis of starch into simple sugars. As a result, starch compounds are lost. This finding agrees with the research of (Xu *et al.*, 2012; Chungcharoen *et al.,* 2015) who reported 5 to 15 % decrease in starch contents of brown rice sprouted for 1 to 3 days at 30 ^{o}C and barley sprouted for 4 days at 17 ^{o}C.

The variation in vitamin contents of sprouted finger millets and water melon seeds as shown in (Table 4.2a and 4.2b) may depend on the type of seed and conditions of steeping or sprouting. (EFSA, 2016). Sprouting improved vitamins in finger millets and water melon seeds particularly B vitamins such as riboflavin (vitamin B_2), niacin (vitamin B_3) and folic acids (vitamin B_9), ascorbic acids (vitamin C) and Tocopherols (vitamin E a fat soluble vitamin), these increases in vitamins may have been driven by

enzymatic hydrolysis of starch by amylases and diastasis which increases the availability of glucose a precursor for the synthesis of vitamins. This finding is in line with the studies of Kariluoto *et al.* (2006) and Koehler *et al*. (2007), who reported a 4 to 6 fold increase in B vitamins in wheat sprouted for 4 to 6 days at 25 ^{0}C. Moongngarm and Setung (2010) observe no significant changes in vitamin E content in brown rice sprouted for a day at 28 ^{0}C. While Esa *et al*. (2011) found that vitamin E contents increased from undetectable levels to 60 mg/100 g in brown rice when sprouted for 3 to 6 days at 28 ^{o}C. This may be an indication that long sprouting times are required for denovo synthesis of vitamin E. Vitamin E is an important antioxidant that protects cellular membranes from oxidative damages. Thus, seeds sprouted for six (6) days may be suggested as good supply of antioxidants to the human body.

Niacin (vitamin B_3) was greatly increased especially at the sixth day of sprouting. The high amounts of niacin is of great importance to the human body as it aids in the conversion of carbohydrates into glucose for release of energy, promotes normal functioning of the nervous and digestive system, sex hormone and stress-related hormones and in metabolism of fat and proteins. Increase in folates (vitamin B_9) may be due to the increased demand for methyl groups as metabolic activities of the developing seeds increases. Folates are essential vitamins required by the body to produce and maintain new cells especially in pregnancy. Folic acids are also used as medication for treatment of folic acid deficiencies and certain types of anemia (lack of red blood cells). The loses in thiamine contents in sprouted seeds may be due to leaching during sprouting or drying process. This is in line with the reports of Moongngam and Saetung (2010) who reported that thiamin is a heat labile B vitamin and may be degraded or leached during sprouting. The decrease in most B vitamins during longer sprouting times may be attributed to some of the vitamins especially (vitamin B_3) are coenzymes,

and may be reduced after starch are mobilized for energy. Hence, appropriate process conditions are required to carefully preserve these vitamins.

However, high levels of these vitamins in sprouted cereals when consumed may significantly contribute to recommended daily intake (RDA). EFSA (2006) reported that sprouted cereals may account for about 20 to 35 % RDA of Niacin and Folate, 5 to 30 % RDA of Riboflavin and Pyridoxal, and 3 to 36 % RDA of vitamin E.

The anti-nutrients levels in sprouted finger millets and water melon seeds as shown in (Table 4.3a and 4.3b) were reduced in minimal levels, this may be due to denovo synthesis occurring in sprouts. This finding is in line with the study of (H¨ubner and Arendt, 2013) who reported that sprouting improves bioavailability of micronutrients by reducing the levels of anti-nutrients in food crops. Anti-nutrients hinder the bioavailability of essential nutrients in foods. They interfere with absorption of nutrients, reduces nutrient digestion and utilization. Most of these anti nutrients elicit very harmful biological responses while some are used as pharmaceutically active agents.

Phytates, saponins and cyanogenic glycosides that were the most abundant anti-nutrients, suggests that they may be the most predominant in cereals and legumes. Phytates in food may reduce the bioavailability of mineral elements especially calcium. Umaru *et al*. (2006) reported that a phytate diet of 1-6 % over a long period may decrease the bioavailability of calcium elements in monogastric animals. Saponins and Tannins reduce protein digestibility of food by inhibiting trypsin, amylase, chymotrypsin and lipase activities (Salunkhe *et al*., 1990). The reduced level in saponins and tannins may not affect the digestibility of proteins obtained from these food materials. The amounts of tannins in sprouted seeds were below permissible limits (20.00±8.19 and 26.28±0.19 mg/100 g) for finger millets and water melon seeds

respectively. Cyanide is a known inhibitor of cytochrome oxidase and interferes with aerobic respiratory system (Onwuka, 2005). High levels of cyanides in food may be detrimental to health. The values of oxalates obtained in this work (0.19±0.01 to 0.33±0.02 mg/100 g) in finger millets and (0.10±0.01 to 0.52±0.03 mg/100 g) in water melon seeds were far below the lethal dose. High levels of oxalates in diets increase the risk of renal calcium absorption and this may virtually lead to kidney stones. Sprouting therefore decreased levels of acids in seeds within an acceptable range which may not be detrimental to health.

Results from this study (Table 4.4) have shown that amylose contents were decreased during sprouting; this may probably be as a result of biosynthesis of starch molecules in sprouted seeds during which metabolic enzymes (amylases) breaks down amylose chains into simples units. This is in line with the study of Li *et al*. (2017), who reported a decrease in amylose contents of germinated sorghum and millet. The major starch components of a food material are amylose and amylopectin (Wankhede *et al*., 1979). The presence of amylose content in flour determines its starch gelling and firmness property while amylopectin levels are primarily responsible for the thickening of paste (Wankhede *et al*., 1979). The reduced levels in amylose contents in sprouted seed flours may probably be responsible for decreased level of retrogradation in confectionaries.

High protein values as shown in (Table 4.5) were observed in sample E (50 % white flour, 30 % finger millets and 20 % water melon seed) compared to sample A (100 % white flour) which had the least protein contents, this may be attributed to protein synthesis during sprouting process which may have increased the protein availability in finger millets and water melon seeds (Ghavidel and Prakash, 2007). This may however suggests that substituting white flour with 20 to 30 % sprouted flour blends may be good sources of protein in confectionaries. The decreased moisture content of

composites flour blends suggests that the sprouted flour samples may have good storage potentials. The slight increase in fat contents on the addition of finger millets and water melon seed flour may be attributed to high fat contents in water melon seeds. Fats improve flavour and increase the mouth feel of foods and are reported to be a significant factor in bakery foods especially in cookies (Eke and Akobundu 1993; Iwe and Egwuekwe, 2010). High values of crude fibre in flour samples observed on the addition of sprouted finger millets and water melon seeds may have resulted from the hull of water melon seeds. The ash contents in flour samples may have increased due to increased substitution of sprouted seed flour. Total carbohydrate contents were decreased with increased substitution of sprouted seed flours, with higher values being recorded for sample A (100 % white flour) while least carbohydrate values were recorded for sample E (50 % white flour, 30 % sprouted finger millets flour and 20 % sprouted water melon seed flour). Suggesting that high fibre and low carbohydrate contents may enhance digestibility of food materials.

Percentage flour dispersibility as shown in (Figure 4.2) increased with increased substitution of sprouted finger millets and water melon seed flours, showing that the flour may be easily rehydrated with water and made into fine dough. Bulk density of composite flour increased on addition of sprouted finger millet and water melon seeds flour however, no significant difference ($p>0.05$) was observed in the values. This is a reflection of the load which the flour samples can carry when allowed to rest on each other. Higher levels of water absorption capacity and oil absorption capacity were recorded with increased substitution of sprouted finger millets and water melon seeds flours. These properties must have been influenced by the hydrophobic characters of seed macromolecules especially proteins. Similar trend was reported by Abegunde *et al*. (2014) for maize and cowpea blends. The high water absorption capacity of flour

composites makes them useful functional raw materials in fabrication of confectionaries and ready-to-eat foods. This is in line with the study of Iwe and Onadipe (2001), who reported that the ability of flour to absorb water improves its potentials in dough formation. High viscosity values were observed in flour samples, however, the viscosity values were reduced on percentage inclusion of sprouted seed flours. This may be due to high carbohydrates contents in the sprouted seed flours. Functional property of a food material is an indicator of its application and end use (Adeleke and Odedeji, 2010). Thus, sprouting improved functionality of composite flour which is an indication that composite flour of sprouted finger millets and water melon seed may perform useful function in baked products during dough making.

Results from this study (Table 4.6) have shown that height of bread decreased with increased substitution of sprouted seed flours, this may probably be due to absence of glutens in finger millets and water melon seeds. Gluten is the characteristic factor responsible for dough strength and elasticity. Presence of glutens in food causes inflammation in gut lining, which may however affect how the gut absorbs food. Suggesting that gluten free bread may be safe for consumption especially for patients with metabolic disorders. Increased inclusion of sprouted seed flours increased the weight of bread. The increase in weight of bread may be attributed to high bulk density of sprouted seeds. This signifies that bread samples may be allowed to rest directly on each other without any damage. The temperature at which snacks were baked decreased with increased inclusion of sprouted seed flours and ranged from (150 to 180 ^{0}C). This must have resulted from denovo synthesis which may have decreased the cooking time of sprouts. Suggesting that sprouted foods may cook faster than non-sprouted foods. No Significant difference ($p>0.05$) was observed among the length and breadth of the bread.

Although bread produced from sample A (100 % white flour) had the highest length and breadth values.

Protein contents as presented in (Table 4.7 and 4.8) were increased with increased substitution of sprouted flours and highest values were observed in bread and cookie produced from (50 % white flour, 30 % sprouted finger millets and 20 % sprouted water melon seed flours) while bread and cookie produced from sample A (100 % white flour) had the least protein values, this signifies that the snacks may be good sources of proteins. Bread and cookies produced from composite flour had moisture contents below 14 %, Suggesting that the snacks may be stored for a long time before spoilage. Fat contents in bread and cookie samples increased with substitution of sprouted flours and sample A had the least values, this may suggest that the snacks may supply part of energy required by the body. The ash contents increased with increased percentage inclusion of sprouted flours. Ash is an indication of presence of minerals in a food material (Alabi and Anuoye, 2007) and has been demonstrated in bread and cookies produced from substituted sprouted seed flours. Crude fibre contents also increased with increased substitution of sprouted flours. The high fibre contents in bread and cookies probably may have resulted from the hull of water melon seeds. The hull of seeds represents a variable fraction of dietary fibre and includes mostly the lignin and cellulose. High carbohydrate contents were observed with white flour bread and cookies while least values were recorded for bread and cookies produced from substituted sprouted seed flours. However, increased fibre and lower carbohydrate contents in bread and cookies suggest that the snacks may aid digestion and reduce constipation. Baking improves organoleptic properties of snacks due to heating conditions resulting from Millard reaction, but however has no significant effect on nutrient quality.

Bread and cookies samples as shown in (Table 4.9 a and 4.9 b), lasted for about 4-7 days and 6-10 days before spoilage occurred with effect to microbial loads, this must have been due to the low moisture contents observed in the samples. Thus high moisture can have an adverse effect on storage and stability of products as well as a reduced shelf life.

There were no detectable bacteria and fungi growth in some cookie samples. This may be correlated with the limits set by the Standard Organization of Nigeria, which states that the counts of aerobic bacteria must not exceed 100cfu/g and coliform growth must not be detected in food samples. This shows that such snack may be safe for consumption as there is no fecal contamination. The high bacteria growth in bread and cookie produced from substituted sprouted flour may have evolved during baking or from the raw ingredients used as these may provide favorable condition for bacteria growth. For example, *Bacillus sp.* form spores on flour, sugar, and yeast which enable the bacteria to survive heating conditions (Saranraj and Geetha, 2012). Microorganisms are found everywhere and the manner of food handles are usually the main source of food contamination (Talaro and Talaro, 1993). Higher fungi counts observed in bread and some cookie samples may probably be due to raw materials, processing and storage. The fungi isolated (*Rhizopus nigricans, Aspergillus niger,* and *Mucor sp)* may have been introduced at different stages of production. This finding is in line with the study of Daniyan and Nwokwu (2011), which identified similar organisms in bread.

Generally, panelists expressed preference for three bread samples produced from (100 % white flour, 80 % white flour, 20 % sprouted finger millets and 70 % white flour, 20 % sprouted finger millets and 10 % sprouted water melon seeds flour) and almost all cookie samples as shown in (Table 4.4 a and 4.4 b). Although, snacks produced from 100 % white flour had better acceptability probably because people are familiar with

consumption of white flour snack products. However, snacks produced from 20 % and 30 % substituted sprouted seed flours had closed level of acceptability. This may suggest that substituting white flour with 20-30 % sprouted finger millets and water melon seed flours may be desirable for making confectionaries in baking industries.

Fig 4.5 shows the cost effectiveness of bread and cookie samples. It was observed that apart from the 100 % white flour snacks, the price at which consumers were willing to purchase each bread and cookie products however had closed ratings with the cost of production, this may probably be due to individual's preference. This may however suggest that there is need to review product specificity and modification based on people's opinion.

CHAPTER FIVE

5.0 CONCLUSION AND RECOMMENDATIONS

5.1 Conclusion

The results obtained from this study proved that finger millets and water melon seeds sprouted for six (6) days had better drying rate as well as increased nutrient quality (particularly, proteins and B vitamins) and reduced anti-nutritional components. Therefore, sprouts are possibly suitable for storage and are desirable for formulations into composite flour.

Sprouting within six (6) days best improved the functional properties of composite flour blends, enhancing their use in various ready-to-eat products and bringing about a notable nutritional improvement in the quality of snacks with increased level of substituted sprouted finger millets and water melon seed (flours).

5.2 Recommendations

i. Further research is encouraged to determine the compositions of gluten contents in finger millets and impact of finger millet formulated diets in metabolic disorders

ii. Industries and house-hold should be encouraged to adopt the use of sprouting technique for improving nutrients quality in food materials.

REFERENCES

Abegunde, T. A., Bolaji, O.T., & Adeyemo, T. M. (2014). Quality evaluation of maize chips ("kokoro") fortified with cowpea flour. *Nigerian Food Journal*, 32, 97-104.

Abioye, V. F., Ogunlakin, G. O., & Taiwo, G. (2018). Effect of germination on anti-oxidant activity, total phenols, flavonoids and anti-nutritional content of finger millet flour. *Food Process Technology*, 6, 2-9

Adekunle, A. A. (2012). *Agricultural innovation in sub-Saharan Africa: experiences from multiple stake holder approaches*. Forum for Agricultural Research in Africa, Ghana. ISBN 978-99881.8373-2-4

Adeleke, R. O., & Odedeji, J. O. (2010). Functional properties of wheat and sweet potato flour blend. *Pakistan Journal of Nutrition,* 96, 535–538

Afify, A. E. M. R., El-Beltagi, H. S., El-Salam, S. M. A., & Omran, A. A. (2011). Bioavailability of iron, zinc, phytate and phytase activity during soaking and germination of white sorghum varieties. *Public Library of Science*, 6, 1–7.

Afify, A. E. M. R., El-Beltagi, H. S., El-Salam, S. M. A., & Omran, A. A. (2012). Protein solubility, digestibility and fractionation after germination of sorghum varieties. *Public Library of Science*, 7, 1–6.

Agu, R. C., & Palmer, G. H. (1997). The effect of temperature on the modification of sorghum and barley during malting. *Processes in Biochemistry*, 32, 501–507

Ahmed, S. B., Mahgoub, S. A., & Babiker, B. E. (1996). Changes in tannin and cyanide contents and diastic activity during germination and the effect of traditional processing on cyanide content of sorghum cultivars. *Food Chemistry*, 56, 159–162.

Ajala, A. S., Ajala, F.A., & Tunde-Akintunde, T. Y. (2012). Study on drying kinetics of fermented corn grains. *Food Science and Quality Management Journal*, 5, 10-17

Akbas, E., Kilercioglu, M., & Onder, O. N. (2017). Wheatgrass juice to wheat grass powder: Encapsulation, physical and chemical characterization. *Journal of Functional Foods*, 28, 19–27.

Akubor, P. I., & Ukwuru, M. U. (2005). Functional properties and biscuit making potential of soybean and cassava flour blends. *Plants Foods for Human Nutrition*, 58, 1-12.

Akwaowo, E. U., Ndon, B. A., & Etuk, E. U. (2000). Minerals and anti-nutrients in fluted pumpkin (*Telfairia occidentalis Hook f.*). *Food Chemistry*, 70(2), 35-40.

Alabi, M. O., & Anuoye, J. C. (2007). Nutritional sensory attributes of supplemented cereal meals. *Nigeria Food Journal*, 25, 100-110.

Aletor, V. A. (1993). Allelochemicals in plant foods and feeding stuffs: 1. Nutritional, biochemical and physiopathological aspects in animal production. *Veterinary and Human Toxicology,* 35, 57-67.

Alexopoulus, C. J., & Mims, C. W. (1979). *Introductory Mycology*. Wiley, New York, NY, USA, 3rd Edition.

Alok, B., Rajeev, K., Vivek, D., & Niyaz, A. (2011). Evaluation of Anti-Ulcer Activity of Citrullus lanatus Seed Extract in Wistar Albino Rats. *International Journal of Pharmacy and Pharmaceutical Sciences*, 4, 135-139.

American Association for Clinical Chemistry: AACC. (2008). International Board 2008. Available online: http://www.aaccnet.org/initiatives/definitions/Pages/ WholeGrain.aspx.

American Diabetes Association. (2005). Diagnosis and classification of diabetes mellitus. *Diabetes Care,* 28, 37-42.

Annison, G., & Topping, D. P. (1994). Nutritional role of resistant starch: chemical structure and physiological function. *Annual Review of Nutrition*, 14, 297–320.

Antony, U., & Chandra, T. S. (1998). Anti-nutrient reduction and enhancement in protein, starch and mineral availability in fermented flour of finger millet (*Eleusine coracana*). *Journal of Agriculture and Food Chemistry,* 46, 2578-2582

Association of Official Analytical Chemists (AOAC). (2011). *Official Methods of Analysis of AOAC International*. 18th Edition, AOAC International, Gaitherburg, 2590.

Aremu, C.O., & Ibirinde, D. B. (2012). Bio-diversity Studies on Accessions of African Yam Bean (Sphenostylis stenocarpa). *International Journal of Agricultural Research,* 7, 78-85.

Asensi-Fabado, M. A., & Munne´-Bosch, S. (2010). Vitamins in plants: occurrence, biosynthesis and antioxidant function. *Trends in Plant Science*, 15, 582-592.

Atkins, G. J., Welldon, K. J., Wijenayaka, A. R. (2009). Vitamin K promotesmineralization, osteoblast-to-osteocyte transition, and an anticatabolic phenotype by {gamma}-carboxylation-dependent and -independent mechanisms. *American Journal of Physiology and Cell Physiology*, 29, 58-67.

Atobatele, O. B., & Afolabi, M. O. (2016). Chemical composition and sensory evaluation of cookies baked from the blends from the blends of soya bean and maize flours. *Applied Tropical Agriculture*, 21, 8-13

Ayernor, G. S., & Ocloo, F. C. K. (2007). Physico-chemical changes and diastatic activity associated with germinating paddy rice. *African Journal of Food Science*, 1, 37–41.

Azeke, M. A., Egielewa, S. J., Eigbogbo, M. U., & Ihimire, I. G. (2011). Effect of germination on the phytase activity, phytate and total phosphorus contents of rice (Oryza sativa), maize (Zea mays), millet (Panicum miliaceum), sorghum (Sorghum bicolor) and wheat (Triticum aestivum*). Journal of Food Science and Technology,* 48, 724–729

Barky, A., Hussein, S., & Alm-Eldeen, Y. (2017). Saponins and their potential role in diabetes mellitus. *Diabetes Management (London*), 1, 48-58.

Bartnik, M., & Szafranska, I. (1987). Changes in phytate content and phytase activity during the germination of some cereals. *Journal of Cereal Science*, 5, 23–28.

Bhatt, A., Singh, V., Shrotria, P. K., & Baskheti, D. C. (2003). Coarse Grains of Uttaranchal: Ensuring sustainable Food and Nutritional Security. *Indian Farmer's Digest*, 4, 34-38.

Bouis, H. E., Hotz, C., McClafferty, B., Meenakshi, J. V., & Pfeiffer, W. H. (2011). Biofortification: A new tool to reduce micronutrient malnutrition. *Food and Nutrition Bulletin,* 32, 31–40.

Braide, W., Odiong, I. J., & Oranusi, S. (2012). Phytochemical and Antibacterial properties of the seed of watermelon (Citrullus lanatus). *Prime Journal of Microbiology Research*, 2, 99-104.

Bravo, A. B. (1998). Phenolic compounds in plants and agric-industrial by-products: Antioxidant activity, occurrence, and potential uses. *Food Chemistry*, 99, 191-203.

Bugusu, B. A., Campanella, O., & Hamakor, B. R. (2001), Imrovement of sorghum-wheat composite dough rheological properties and bread making quality through Zein addition. *Cereal Chemistry*, 78, 31-35.

C´aceres, P. J., Mart´ınez-Villaluenga, C., Amigo, L., & Frias, J. (2014). Assessment on proximate composition, dietary fibre, phytic acid and protein hydrolysis of germinated Ecuatorian brown rice. *Plant Foods for Human Nutrition*, 69, 261–267

Campos, F. A. P., & Richardson, M. (1983). The complete amino acid sequence of the bifunctional -amylase/trypsin inhibitor from seeds of ragi (Indian finger millet, *Eleusine coracana Gaertn*.), *Federation of European Biochemical Societies Letters*, 152, 300–304.

Centeno, C., Viveros, A., Brenes, A., Canales, R., & Lozano, A. (2001). Effect of several germination conditions on total P, phytate P, phytase, and acid phosphatase activities and inositol phosphate esters in rye and barley. *Journal of Agricultural and Food Chemistry,* 49, 3208–3215.

Ceserani, V., & Kinton, R. (2008). *Practical Cookery.* (10th Edition.). John Wiley and Sons, New York.

Chandrasekara, A., & Shahidi, F. (2010). Content of insoluble bound phenolics in millets and their contribution to antioxidant capacity. *Journal of Agricultural and Food Chemistry*, 58, 6706–6714

Chandrasekara, A., & Shahidi, F. (2011a). Determination of antioxidant activity in free and hydrolyzed fractions of millet grains and characterization of their phenolic profiles by HPLC-DAD-ESI-MSn. *Journal of Functional Foods,* 3, 144–58.

Chandrasekara, A., & Shahidi, F. (2011b). Antioxidant phenolics of millet control lipid peroxidation in human ldl cholesterol and food systems. *Journal of American Oil and Chemical Society,* 10, 1174-1918.

Cheeke, P. R., & Shull, L. R. (1985). *Natural toxicants in feeds and livestock*. West Port, Connecticut: AVI Publishing Inc.

Chrastil, J. (1987). Improved Colorimetric determination Amylose in Starches or flours. *Carbohydrate Research*, 159, 154-158.

Chu, X., Guo, Y., & Xu, B. (2015). Effects of tannic acid, green tea and red wine on hERG Channels Expressed in HEK293 Cells. *Public Library of Sciences*, 10, 143-797

Chung, T. Y., Nwokolo, E. N., & Sim, J. S. (1989). Compositional and digestibility changes in sprouted barley and canola seeds. *Plant Food for Human Nutrition*, 39, 267–278.

Chungcharoen, T., Prachayawarakorn, S., Tungtrakul, P., & Soponronnarit, S. (2015). Effects of germination time and drying temperature on dyring characteristics and quality of germinated paddy. *Food and Bioproducts Processing*, 94, 707–716.

Combs, G. F. (2007). Vitamin B6. In: The Vitamins. Academic Press, Waltham, MA.

Correia, I., Nunes, A., Barros, A. S., & Delgadillo, I. (2008). Protein profile and malt activity during sorghum germination. *Journal of the Science of Food and Agriculture,* 88, 2598–2605.

Coulibaly, A., Kouakou, B., & Chen, J. (2011). Phytic acid in cereal grains: structure, healthy or harmful ways to reduce phytic acid in cereal grains and their effects on nutritional quality. *American Journal of Plant Nutrition and Fertility Technology*, 1, 1-22

Crider, K. S., Bailey, L. B., & Berry, R. J. (2011). Folic acid food fortification; its history, effect, concerns, and future directions. *Nutrients,* 33, 70-84.

Dal-Bosco, A., Castellini, C., Martino, M., Mattioli, S., Marconi, O., & Benincasa, P. (2015). The effect of dietary alfalfa and flax sprouts on rabbit meat antioxidant content, lipid oxidation and fatty acid composition. *Meat Science*, 106, 31–37.

Daniyan, S. Y., & Nwokwu, O. E. (2011). Enumeration of microorganisms associated with the different stages of bread production in FUTMIN bakery, Nigeria*"*. *International Research Journal of Pharmacy,* 2, 88–91.

Day, R. A., & Underwood, A. L. (1986). *Quantitative Analysis*. 5th Edition, Prentice Hall Publication, Upper Saddle River, 701.

Devi, P. B.,Vijayabharathi, R., Sathyabama, S., Malleshi, N. G., & Priyadarisini, V. B. (2014). Health benefits of finger millet (Eleusine coracana L.) polyphenols and dietary fibre: a review. *Journal of Food Science Technology*, 51, 1021–1040.

Di Gioia, F., Renna, M., & Santamaria, P. (2017). *Sprouts, microgreens and "baby leaf" vegetables'. In Minimally Processed Refrigerated Fruits and Vegetables*. Springer: Boston, MA, USA, 27, 403–432.

Donkor, O. N., Stojanovska, L., Ginn, P., Ashton, J., & Vasiljevic, T. (2012). Germinated grains - Sources of bioactive compounds. *Food Chemistry*, 135, 950–959.

Duke, S. H. (2009). A comparison of barley malt amylolytic enzyme activitiesas indicators of malt sugar concentrations. *Journal of the American Society of Brewing Chemists*, 67, 99–111.

Eke, O. S., & Akobundu, E. H. T. (1993). Functional properties of African yam bean (Sphenostylis stenocarpa) seed flour as affected by processing. *Journal of Food Chemisrty*, 48, 337-340.

El-Adawy, T. A. (2001).Characteristics and composition of watermelon pumpkin and paprika seed oils and flours. *Journal of Agriculture and Food Chemistry,* 49, 1253 – 1259.

Elmaki, H. B., Babiker, E. E., & El Tinay, A. H. (1999). Changes in chemical composition, grain malting, starch and tannin contents and protein digestibility during germination of sorghum cultivars. *Food Chemistry*, 64, 331–336.

Elmoneim, A., Elkhalifa, O., & Bernhardt, R. (2010). Influence of grain germination on functional properties of sorghum flour. *Food Chemistry*, 121, 287–392

Esa, N. M., Kadir, K. A., Amom, Z., & Azlan, A. (2011). Improving the lipid profile in hypercholesterolemia-induced rabbit by supplementation of germinated brown rice. *Journal of Agricultural and Food Chemistry*, 59, 7985–7991

Essien, J. P., Akpan, E. J., & Essien, E. P. (2005). Studies on mold growth and biomass production using waste banana peel. *Bioresources Technology*, 96, 1451-1456.

European Sprouted Seeds Association (ESSA). (2006). Tolerable upper intake levels for vitamins and minerals. Scientific Committee on Food and Scientific Panel on Dietetic Products, Nutrition and Allergies. Parma, Italy.

European Sprouted Seeds Association (ESSA). (2016). ESSA Hygiene Guideline for the Production of Sprouts and Seeds for Sprouting; ESSA: Brussels, Belgium.

Falcinelli, B., Calzuola, I., Gigliarelli, L., Torricelli, R., Polegri, L., & Marsili, V. (2018). Phenolic content and antioxidant activity of wholegrain breads from modern and old wheat (Triticum aestivum L.) cultivars and ancestors enriched with wheat sprout powder. *Italian Journal of Agronomy,* 13, 297–302.

Faltermaier, A., Zarnkow, M., Becker, T., Gastl, M., & Arendt, E. K. (2015). Common wheat (Triticum aestivum L.): Evaluating microstructural changes during the malting process by using confocal laser scanning microscopy and scanning electron microscopy. *European Food Research and Technology,* 241, 239–252.

FAO, (2011). Facts sheet on watermelon production. Ghana.

Fardet, A., Rock, E., & Remesy, C. (2008). Is the in vitro antioxidant potential of whole-grain cereals and cereal products well reflected in vivo? *Journal of Cereal Science,* 48, 258–276.

Faurschou, A., Beyer, D. M., & Schmedes, A. (2012). The relation between sunscreen layer thickness and vitamin D production after ultraviolet B exposure: a randomized clinical trial. *British Journal of Dermatology,* 167, 391-5.

Fellows, P. J. (2000). "Food Processing Technology: Principles and Practice". *Ellis Horwood Series in Food Science and Technology*, 2, 234-280.

Fu, P. P., Xia, Q., Lin, G., & Chou, M. W. (2002). Genotoxic pyrrolizidine alkaloids mechanisms leading to DNA adduct formation and tumorigenicity. *International Journal of Molecular Sciences,* 3, 948-64.

Gan, R. Y., Chan, C. L., Yang, Q. Q., & Li, H. B. (2019). *Bioactive compounds and beneficial functions of sprouted grains. In Sprouted Grains*. AACC International Press: St. Paul, MN, USA, 191–246.

Gemede, H. F., & Ratta, N. (2014). Antinutritional factors in plant foods: potential health benefits and adverse effects. *International Journal of Nutrition and Food Science*, 3(28), 4-9.

Ghavidel, R., & Prakadh, J. (2007). The impact of germination and dehulling on nutrients, anti-nutrients, in vitro iron and calcium bioavailability and in vitro starch and protein digestibility of some legumes seeds. *Lebensmittel-Wissenschaft and Technologie*, 40, 1292-1299.

Gopalan, C., Ramashastri, B. V., & Balasubramanium, S. C. (2002). Nutritive value of Indian foods. *National Institute of Nutrition*, ICMR, Hyderabad.

Gregory, J. F., Park, Y., & Lamers, Y. (2013). Metabolomic analysis reveals extendedmetabolic consequences of marginal vitamin B6 deficiency in healthy human subjects. *Public Library of Sciences*, 8(6), 35-44.

Gull, A., Kmalesh, P., & Kumar, P. (2015). Optimisation and functionality of millet supplemented pasta. *Food Science and Technology*, 35, 626–632.

Haraldsson, A., Rimsten, L., Alminger, M. L., Andersson, R., Andlid, T., Aman, P. & Sandberg, A. (2004). Phytate content is reduced and β-glucanase activity suppressed in malted barley steeped with lactic acid at high temperature. *Journal of the Science of Food and Agriculture,* 84, 653–662.

Hassani, A., Procopio, S., & Becker, T. (2016). Influence of malting and lactic acid fermentation on functional bioactive components in cereal-based raw materials:

A review paper. *International Journal of Food Science and Technology,* 51, 14–22.

Hayden, M. R., & Tyagi, S. C. (2004). Homocysteine and reactive oxygen species in metabolicsyndrome, type 2 diabetes mellitus, and atheroscleropathy: the pleiotropic effects of folate supplementation. *Nutrition Journal*, 7, 3-4.

Hegde, P. S., Chandrakasan, G., & Chandraa, T. S. (2002). Inhibition of collagen glycation and crosslinking in vitro by methanolic extracts of Finger millet (Eleusine coracana) and Kodo millet (Paspalum scrobiculatum). *Journal of Nutritional Biochemistry*, 13(5), 17–21.

Hendek, M., & Bektas, M. (2018). Enhancement of bioavailable micronutrients and reduction of anti-nutrients in foods with some processes. *Food Health*, 4(1),5-9.

Hirota, Y., Sugawa, N., & Nakagawa, K. (2013). Menadione (vitamin K_3) is a catabolic product of oral phylloquinone (vitamin K1) in the intestine and a circulating precursor of tissue menaquinone-4 (vitamin K2) in rats. *Journal of Biololgy and Chemistry,* 288(3), 71-80.

Hithamani, G., & Srinivasan, K. (2014). Bioaccessibility of polyphenols from wheat (Triticum aestivum), sorghum (Sorghum bicolor), green gram (Vigna radiata), and chickpea (Cicer arietinum) as influenced by domestic food processing. *Journal of Agricultural and Food Chemistry,* 62, 11170–11179

Hübner, F., & Arendt, E. K. (2013). Germination of cereal grains as a way to improve the nutritional value: A review. *Critical Review of Food Science and Nutrition*, 53, 853–861.

Hulse, J. H., Laing, E. M., & Pearson, O. E. (1980). *Sorghum and the millets: their composition and nutritive value*. New York: Academic Press, 991–997.

Ikewuchi, C. C. (2012). Hypocholesterolemic effect of an aqueous extract of the leaves of Sansevieria senegambica Baker on plasma lipid profile and atherogenic indices of rats fed egg yolk supplemented diet. *Experimental and Clinical Sciences Journal*, 11(3), 46-56.

Iwe, M. O., & Onadipe, O. O. (2001). Effect of extruded full fat soy flour into sweet potato flour on functional properties of the mixture. *Journal of Environment and Sustainable Agriculture,* 3, 109-117.

Iwe, M.O., & Egwuekwe, E. I. (2010). Production and evaluationof cookies from *Xanthosoma sagitolium and Colocasia esculenta* blends. *Nigerian food journals*, 23, 145-153.

Jagerstad, M., Piironen, V., Walker, C., Ros, G., Carnovale, E., Holasova, M., & Nau, H. (2005). Increasing natural food folates through bioprocessing and biotechnology. *Trends in Food Science and Technology,* 16, 298–306.

Jabrin, S., Ravanel, S., Gambonnet, B., Douce, R., & Rebeille, F. (2003). One-carbon metabolism in plants. Regulation of tetrahydrofolate synthesis during germination and seedling development. *Plant Physiology*, 131, 1431–1439.

Jensen B. D., Toure, F. M., Hamattal, M. A., Toure, F. A., & Nantoumé D. A. (2011). Wtermelons in the Sand of Sahara: Cultivation and use of indigenous landraces in the Tombouctou Region of Mali. *Ethnobotany Research and Applications*, 9, 151-162.

Jideani, I. A., Takeda, Y., & Hizukuri, S. (1996). Structures and physiochemical properties of acha (*Digitaria exilis*), iburu (*D. iburua*) and tamba (*Eleusine coracana*). *Cereal Chemistry*, 73(7), 76-85.

Jideani, I. A. (2012). Digitaria exilis (acha/fonio), Digitaria iburua (iburu/fonio) and Eleusine coracana (tamba/finger millet) - Non-conventional cereal grains with potentials. *Scientific Review and Essays,* 7, 3834–3843

Johnson, I. T., Gee, J. M., Price, K., Curl, C., & Fenwick, G. R. (1986). Influence of saponins on gut permeability and active nutrient transport in vitro. *Journal of Nutrition*, 116, 2270-2277.

Joshi, H. C., & Katoch, K. K. (1990). Nutritive value of millets: A comparison with cereals and pseudo cereals. *Himalayan Research Development*, 9, 26-28

Kala, A., & Prakash, J. (2003). Vitamin B1e retention in cooked, stored and reheatedvegetables. *Journal of Food Science and Technology Mysore*, 40, 409-412.

Kariluoto, S., Liukkonen, K., Myllym¨aki, O., Vahteristo, L., Kaukovirta-Norja, A., & Piironen, V. (2006). Effect of germination and thermal treatments on folates in rye. *Journal of Agricultural and Food Chemistry,* 54, 9522–9528.

Kaur, K., Singh, N., & Singh, H. (2002). Studies on the effect of skim milk powder, sprouted wheat flour, and pH on rheological and baking properties of flour. *International Journal of Food Properties,* 5, 13–24.

Kennedy, M. M. O., Grootboom, A., & Shewry, P. R. (2006). Harnessing sorghum and millet biotechnology for food and health. *Journal of Cereal Science*, 44, 224–235.

Keogh, J. B., Cleanthous, X., & Wycherley, T. P. (2012). Increased vitamin B1e intake may berequired to maintain vitamin B1e status during weight loss in patients with type 2diabetes. *Diabetes Research and Clinical Practices*, 98, 40-42.

Kim, S. K., Choi, H. J., Kang, D. K., & Kim, H. Y. (2012). Starch properties of native proso millet (Panicum miliaceum L.). *Agronomy Research*, 10, 311-318

Klupšaitė, D., & Juodeikienė, G. (2011). Legume: composition, protein extraction and functional properties. A review. *Chemical Technology*, 66, 5-12.

Koehler, P., Hartmann, G., Wieser, H., & Rychlik, M. (2007). Changes of folates, dietary fibre, and proteins in wheat as affected by germination. *Journal of Agriculture and Food Chemistry*, 55, 4678–4683.

Kolodziejczyk, P., & Michniewicz, J. (2004). The changes of some enzymes activities during germination of rye kernels. *Electronic Journal of Polish Agricultural Universities*, 7, 1–9.

Krahl, M., Zarnkow, M., Back, W., & Becker, T. (2010). Determination of the influence of malting parameters on the water-extractable arabinoxylan content of wheat (Triticum aestivum), rye (Secale cereale), and spelt wheat (Triticum aestivum spp. spelta). *Journal of the American Society of Brewing Chemists,* 68, 34–40.

Krieg, N. R., & Holt, J. G. (1994). *Bergey's Manual of Systemic Bacteriology*, Williams and Wilkins, Baltimore, Md, USA.

Krishnan, P., Ramakrishnan, B., & Reddy, K. (2011). High temperature effects on rice growth, yield and grain quality. *Advances in Agronomy*, 111, 87-206.

Kubicka, E., Grabska, J., & Jedrychowski, L. C. B. (2011). Changes of specific activity of lipase and lipoxygenase during germination of wheat and barley. *International Journal of Food Sciences and Nutrition,* 51, 301–304.

Kulkarni, K. D., Kulkarni, D. N., & Ingle, W. M. (1991). Sorghum malted and soybean weaning food formulation: preparation, functional properties and nutritive value. *Food and Nutrition Bulletin*, 13, 322–329.

Kumar, A., Metwal, M., Kaur, S., Gupta, A. K., & Yadav, S. (2016). Nutraceutical Value of Finger Millet [Eleusine coracana (L.) Gaertn.], and Their Improvement Using Omics Approaches. *Front Plant Science,* 7, 9-34.

Kurosu, M., & Begari, E. (2010). Vitamin K_2 in electron transport system: are enzymes involved in vitamin K2 biosynthesis promising drug targets?. *Molecules,* 15, 1531-1553.

Lakshmi, K. P., & Sumathi, S. (2002). Effects of consumption of finger millets on hyperglycemia in non-insulin dependent diabetes mellitus (NIDDM) subjects. *Plant Food for Human Nutrition*, 57, 3-4

Lanska, D. J. (2010). Historical aspects of the major neurological vitamin deficiency disorders: the water-soluble B vitamins. *Clinical Neurology*, 95, 445-476.

Lee, S. H., Chung, I. M., Cha, Y. S., & Parka, Y. (2010). Millet consumption decreased serum concentration of triglyceride and C-reactive protein but not oxidative status in hyperlipidemic rats. *Nutrition Research,* 30, 290–296.

Lemmens, E., Moroni, A. V., & Pagand, J. (2018). Impact of Cereal Seed Sprouting on Its Nutritional and Technological Properties: A Critical Review. *Comprehensive Review in Food Science and Food Safety*, 18, 305–328.

Leskova, E., Kubikova, J., Kovacikova, E. (2006). Vitamin losses: retention during heattreatment and continual changes expressed by mathematical models. *Journal of Food and Comprehensive Analysis,* 19(2), 52-76.

Li, Q., Lauber, L., Gail, C., Pan, Y., & Hannah, S. (2017). Effects of Dietary Protein and Carbohydrate ratio on Gut Microbiomes in Dogs of Different Body Conditions. *Society for Microbiology*, 3, 16.

Lintschinger, J., Fuchs, N., Moser, H., J¨ager, R., Hlebeina, T., Markolin, G., & Gossler, W. (1997). Uptake of various trace elements during germination of wheat, buckwheat and quinoa. *Plant Foods for Human Nutrition*, 50, 223–237.

Lolas, G. M., & Markakis, P. (1975). Phytic acid and other phosphorus compounds of beans (*Phaseolus Vulgaris L.). Journal of Agriculture and Food Chemistry*, 23, 13-15.

Lu, L., Tian, S., Liao, H. Z., J., Yang, X., Labavitch, J. M., & Chen, W. (2013). Analysis of metal element distributions in rice (*Oryza sativa L.*) seeds and relocation during germination based on X-ray fluorescence imaging of Zn, Fe, K, Ca and Mn. *Public Library of Science* 5, 1–9.

Lupien, J. R. (1990). Sorghum and millets in human nutrition. FAO, ICRISAT. http://www.aaccnet.org/initiatives/definitions/Pages/WholeGrain.aspx.

Malleshi, N. G., & Klopfenstein, C. F. (1998). Nutrient composition, amino acid and vitamin contents of malted sorghum, pearl millet, finger millet and their rootlets. *International Journal of Food Sciences and Nutrition*, 49, 415–422

Mamboleo, T. (2015). Nutrients and antinutritional factors at different maturity stages of selected indigenous African green leafy vegetables. *International Journal of Food Sciences and Nutrition*, 25, 131-139.

Maskos, k., Huber, M., & Glockshuber, R. (1996). RBI, a one-domain α-amylase/trypsin inhibitor with completely independent binding sites. *Federation of European Biochemical Societies,* 397, 11-16

Masum, A. S. M. G., Crawford, H., & Berthold, J. (2011). Minerals (Zn, Fe, Ca and Mg) and anti-nutrient (Phytic acid) constituents in common bean. *American Journal of Food Technology*, 6(2), 35-43.

Mattioli, S., Dal Bosco, A., Martino, M., & Ruggeri, S. (2016). Alfalfa and flax sprouts supplementation enriches the content of bioactive compounds and lowers the cholesterol in hen egg. *Journal of Functional Foods*, 22, 454–462.

Maynard, D. N. (2001). *Watermelons: characteristics, production and marketing*. American Society for Horticultural Science (ASHS) Press. Horticulture Crop Production Series. Alexandria, VA, United States, 2-27.

Mbithi-Mwikya, S., Ooghe, W., Van Camp, J., & Huyghebaert, A. (2000). Amino acid profile after sprouting, Autoclaving and lactic acid fermentation of finger millet (*Elusine coracana*) and kidney beans (*Phaseolus vulgaris L.*). *Journal of Agriculture and Food Chemistry*, 48, 3081-3085.

McDowell, L. R. (2000). *Vitamins in Animal and Human Nutrition*. 2nd Edi. Iowa StateUniversity Press/Ames.

McKeown, N. M., Meigs, J. B., Liu, S., Wilson P. W., & Jacques, P. F. (2002). Whole-grain intake is favorably associated with metabolic risk factors for type 2 diabetes and cardiovascular disease in the Framingham Offspring Study. *American Journal of Clinical Nutrition*, 76, 390–398.

McWatters, K. H., Ouedraogo, J. B., Resurrection, A. V. A., Hung, Y. C., & Philips, R. D. (2003). Physical and sensory characteristics of sugar cookiescontaining mixtures of wheat, fonnio (*Digitaria exilis*) and cowpea (*Vigna unguiculata*) flours. *International Journal of Food Science and Technology*, 38, 403– 410

Meilgaard, M., Civile, G. V., & Carr, B. T. (2007). *Sensory Evaluation Techniques*. 4th Edition. Florida, USA: *CRC Press*, 1- 464.

Moongngarm, A., & Saetung, N. (2010). Comparison of chemical compositions and bioactive compounds of germinated rough rice and brown rice. *Food Chemistry*, 122, 782–788.

Mosolov, V. V., Grigor'eva, L. I., & Valueva, T. (2001). Plant protease inhibitors as multifunctional proteins (Review). *Applied Biochemistry and Microbiology*, 37, 545–551.

Nasr, S. I., & Abufoul, G. (2004). *Using free fat watermelon (Citrullus vulgaris) seed kemels in preparing high protein biscuits*. Al-Azhar University Gaza. Gaza Palestine, 7, 45-54

Natesh, N. H. (2018). An overview of nutritional and antinutritional factors in green leafy vegetables. *Horticulture International Journal*, 1, 58-65

Nelson, K., Stojanovska, L., Vasiljevic, T., & Mathai, M. (2013). Germinated grains: A superior whole grain functional food? Can. *Journal of Physiology and Pharmacology,* 91, 429–441.

Ngozi, O. O. P. (2014). Evaluation of tannin, phytate and mineral composition of different indigenous dishes based on pumpkin (*Cucurbita pepo). International Journal of Nutrition and Food Science*, 3, 493-496.

Nielson, S. S. (2002). *Introduction to the Chemical Analysis of Foods*, CBS Publishers and Distributors, New Delhi.

Nissar, J., Ahad, T., & Naik, H. R. (2017). A review phytic acid: As anti-nutrient or nutraceutical. *Journal of Pharmacognisis and Phytochemistry*, 6(15), 54-60.

Nonogaki, H., Bassel, G. W., & Bewley, J. D. (2010). Germination - Still a mystery. *Plant Science*, 179, 574–581.

Oakenfull, D., & Sidhu, G. S. (1989). *Saponins: In Toxicants of plant origin, Vol. II, Glycosides*. In: Florida: CRC. Press Inc, 97.

Obilana, A. B., & Manyasa, E. (2002). Pseudo cereals and less common génétique du fonio (Digitaria exilis stapf) en Afrique de l'Ouest, Conakry, *International Plant genetic Resourses Institute,* 73. 177-217.

Ogbonna, A. C., Abuajah, C. I., Ide, E. O., & Udofia, U. S. (2012). Effect of malting conditions on the nutritional and anti-nutritional factors of sorghum grist. *Food Technology*, 36, 64–72.

Oghbaei, M., & Prakash, J. (2012). Bioaccessible nutrients and bioactive components from fortified products prepared using finger millet (Eleusine coracana). *Journal of Science and Food Agriculture,* 92, 2281–2290.

Oghbaei, M., & Prakash, J. (2016). Effect of primary processing of cereals and legumes on its nutritional quality: A comprehensive review. *Cogent Food and Agriculture*, 2(1), 13-15.

Olagunju, A. I., & Ifesan, B. O. T. (2013). Nutritional composition and acceptability of cookies made from wheat flour and germinated sesame (Sesamumindicum) flour blends. *British Journal of Applied Science and Technology,* 3, 702-703.

Olawoye, B. T., & Gbadamosi, S. O. (2017). Effect of different treatments on in vitro protein digestibility, anti-nutrients, antioxidant properties and mineral composition of Amaranthus viridis seed. *Cogent Food and Agriculture*, 3, 1-7.

Olorode, O., Idowu, M., Bamgbose, A., & Animashaun, O. (2017). Evaluation of Potentials of some selected seeds' flours as partial Substitute for Wheat in Cookies production. *International Journal of Scientific and Engineering Research,* 8, 2229-5518

Oloyede, O. I. (2005). Chemical profile of unripe Pulp of *Carica papaya. Pakistan Journal of Nutrition*, 4, 379-381.

Onwuka, G. I. (2005). *Food Analysis and Instrumentation: Theory and Practice*. Lagos, Nigeria: Naphtali Prints.

Ou, K., Cheng, Y., Xing, Y., Lin, L., Nout, R., & Liang, J. (2011). Phytase activity in brown rice during steeping and sprouting. *Journal of Food Science and Technology*, 48, 598–603.

Oyeleke, G. O. I., Olagunju, E. O., & Ojo, A. (2012). Functional and Physicochemical Properties of Watermelon (Citrullus Lanatus) Seed and Seed-Oil. *Journal of Applied Chemistry*, 2, 29-31.

Oyeyinka, S., Oyeyinka, A., Karim, O., Toyeeb, K., Olatunde, S., & Arise, A. (2014). Biscuit making potentials of flour from wheat and plantain at different stages of ripeness. *Croation Journal of Food science Technology*. 6, 36-42.

Pandey, R., Patil, N., & Rao, M. (2007). Proteases and protease inhibitors: implications in antitumorigenesis and drug development. *International Journal of Human Genetics*, 7, 67–82.

Park, S. S., & Obha, H. (2004). Suppressive activity of protease inhibitors from buck-wheat seeds against humanT-acutelymphoblasticleukemiace lllines, *Applied Biochemistry and Biotechnology,* 117, 65–74.

Peterson, D. M. (1999). Lipase activity and lipid metabolism during oat malting. *Cereal Chemistry*, 76, 159–163

Plaza, L., de Ancos, B., & Cano, M. P. (2003). Nutritional and health-related compounds in sprouts and seeds of soybean (Glycine max), wheat (Triticum aestivum.L) and alfalfa (Medicago sativa) treated by a new drying method. *European Food Research and Technology*, 216, 138–144.

Pore, M. S., & Magar, N. G. (1976). Effect of ragi feeding on serum cholesterol level, Indian. *Journal of Medical Research,* 64, 909–914.

Raboy, V. (2003). myo-Inositol-1,2,3,4,5,6-hexakisphosphate. *Phytochemistry,* 64, 1033–1043.

Saleh, S. M., Zhang, Q., Chen, J., & Shen, Q. (2013). Millet grains, Nutritional quality, processing and potential health benefits. *Comprehensive Reviews in Food Science and Technology*, 12, 281–295.

Salunkhe, D. K., Chavan, J. K., & Kadam, S. S., (1990). *Dietary tannins: Consequences and remedies*. Boca Raton, Florida*: CRC Press,* 1, 50-73.

Saman, P., Vazquez, J. A., & Pandiella, S. S. (2008). Controlled germination to enhance the functional properties of rice. *Process in Biochemistry,* 43, 1377–1382.

Saranraj, P., & Geetha, M. (2012). "Microbial spoilage of bakery products and its control by preservatives," *International Journal of Pharmaceutical and Biological Archives*, 3, 38– 48.

Schlemmer, U., Frolich, W., Prieto, R. M., & Grases, F. (2009). Phytate in foods and significance for humans: Food sources, intake, processing, bioavailability, protective role and analysis. *Molecular Nutrition and Food Research,* 53, 330–375.

Sen, S., & Dutta, S. K. (2012). Cloning, expression and characterization of biotic stress inducible Ragi bifunctional inhibitor (RBI)gene from *Eleusine coracana Gaertn*. *Journal of Plant Biochemistry and Biotechnology,* 21, 66–76.

Sen, S., Dutta, S. K., & Ghosh-Dastidar, S. (2011). Development of a highly potent therapeutic regimenforchronicmyeloid leukemia using the extract of *Eleusine coracana* seeds. *International Journal of Biomedicine and Pharmaceutical Science,* 5, 7–11.

Shearer, M. J., & Newman, P. (2014). Recent trends in the metabolism and cell biology ofvitamin K with special reference to vitamin K cycling and MK-4 biosynthesis. *Journal of Lipid Research*, 55(3), 45-62.

Shearer, M. J., Fu, X., & Booth, S. L. (2012). Vitamin K nutrition, metabolism, andrequirements: current concepts and future research. *Advanced Nutrition*, 3(1), 82-95.

Shiihii, S. U., Musa, H., Bhati, P. G., & Martins, E. (2011). Evaluation of physicochemical properties of Eleusine coracana starch. *Nigerian Journal of Pharmaceutical Sciences,* 10, 91–102.

Shingare, S. P., & Thorat, B. N. (2013). Fluidized bed drying of sprouted wheat (Triticum aestivum). *International Journal of Food Engineering*, 10, 29–37.

Shivaraj, B., & Pattabiraman, T. N. (1981). Natural plant enzyme inhibitors.Characterization of anunusualalpha-amylase/trypsin inhibitor from ragi (*Eleusine coracana Geartn.), Biochemistry Journal,* 193, 29–36.

Shobana, S. (2009). Investigations on the carbohydrate digestibility of finger millet (*Eleusine Coracana*) with special reference to the influence of its seed coat constituents. (Doctoral dissertation,University of My sore), 1-221.

Shobana, S., Sreerama, Y. N., & Malleshi N. G. (2009). Compositionandenzyme inhibitory properties of finger millet (Eleusine coracana L.) seed coat phenolics: mode of inhibition of -glucosidase and -amylase, *Food Chemistry*, 115, 1268–1273

Shokoohi, M., Razavi, S. H., Labbafi, M., Vahidinia, A., & Gharibzahedi, S. T. (2014). Wheat sprout flour as an attractive substrate for the producing probiotic fermented beverages: Process development and product characterisation. *Quality Assurance and Safety of Crops and Foods,* 7, 469–475.

Singh, A., & Sharma, S. (2017). Bioactive components and functional properties of biologically activated cereal grains: A bibliographic review. *Critical Reviews in Food Science and Nutrition*, 57, 3051–3071

Singh, J., Singh, N., Sharma, T. R., & Saxena, S. K. (2003). Physicochemical, rheological and cookie making properties of corn and potato flours. *Food Chemistry,* 83, 387-393.

Soetan, K. O. (2008). Pharmacological and other beneficial effects of antinutriional factors in plants. –A review. *African Journal of Biotechnology*, 7, 4713-4721.

Srivastava, S., Singh, N. S., & Shamim, M. Z. (2015). Nutritional composition of weaning food using malted cereal and pulses flour for infants. *International Journal of Pure & Applied Bioscience*, 3, 171–185.

Sung, H. G., Shin, H. T., Ha, J. K., Lai, H. L., Cheng, K. J., & Lee, J. H. (2005). Effect of germination temperature on characteristics of phytase production from barley. *Bioresource Technology*, 96, 1297–1303.

Swieca, M., & Dziki, D. (2015). Improvement in sprouted wheat flour functionality: Effect of time, temperature and elicitation. *International Journal of Food Science and Technology,* 50, 2135–2142.

Taiwo, E. O., Sekinat, A. A., Adegbola, D. O., Arise, A. K., & Sotunde, A. J. (2017). Chemical composition and sensory qualities of wheat-sorghum date cookies. *Croatian Journal of Food Technology, Biotechnology and Nutrition,* 12, 71-76.

Takhellambam, R. D., Chimmand, B. V., & Prkasam, J. N. (2016). Ready-to-cook millet flakes based on minor millets for modern consumer. *Journal of Food Science and Technology*, 53, 1312–1318.

Talaro, K., & Talaro, A. (1993). *Foundations in Microbiology*, Wm.C Brown Publishers,Dubuque, La, USA., 34-67.

Taylor, J. R. N., & Emmambux, M. N. (2008). Gluten-free foods and beverages from millets. In: Gallagher E, editor. Gluten-free cereal products and beverages. Burlington, MA: *Elsevier*, 1–27.

Taylor, J. R. N., Schober, T.J., & Bean, S. R. (2006). Novel food and non-food uses for sorghum and millets. *Journal of Cereal Science,* 44, 252–71.

Teixeira, C., Nyman, M., Andersson, R., & Alminger, M. (2016). Effects of variety and steeping conditions on some barley components associated with colonic health. *Journal of the Science of Food and Agriculture*, 96, 4821– 4827.

Tharanathan, R. N., & Mahadevamma, S. (2003). Grain legumes—a boon to human nutrition. *Trends in Food Science and Technology,* 14, 507–518.

Thompson, I. U. (1993). Potential health benefits and problems associated with anti-nutrients in foods. *Food Research International Journal,* 26, 131–149.

Tian, B., Xie, B., Shi, J., & Wua, J. (2010). Physicochemical changes of oat seeds during germination. *Food Chemistry*, 119, 1195–1200.

Tibe, O., Amarteifio, J. O., & Njogu, R. M. J. (2007). Trypsin inhibitor activity and condensed tannin content in bambara groundnut (Vigna subterranean (L.) Verdc) grown in Southern Africa. *Applied Science and Environmental Management,* 11(1), 5-94.

Tizazu, S., Urga, K., Abuye, C., & Retta, N. (2010). Improvement of energy and nutrient density of sorghum-based complementary foods using germination. *African Journal of Food Agriculture Nutrition and Development*, 10, 2927–2942.

Ubbor, S. C., & Akobundu, E. N. T. (2009). Quality characteristics of cookies from composite flours of Water melon seed, cassava and wheat. *Pakistan Journal of. Nutrition*, 8,1097–1102.

Udeme, J. J. I., Helen, S. A., Mercy, O. A., & Sesan, A. A. (2014). Microbiological, Nutritional, and Sensory Quality of Bread Produced from Wheat and potato Flour Blends. *International Journal of Food Science*, 6, 12-20.

Umaru, H. A., Daniel, D., & Nadro, M. (2006). Levels of antinutritional factors in some wild edible fruits of Northern Nigeria. *African Journal of Biotechnology,* 6, 10-16.

Van Hung, P., Maeda, T., Yamamoto, S., & Morita, N. (2011). Effects of germination on nutritional composition of waxy wheat. *Journal of the Science of Food and Agriculture,* 92, 667–672.

Vinje, M. A., Duke, S. H., & Henson, C. A. (2015). Comparison of factors involved in starch degradation in barley germination under laboratory and malting conditions. *Journal of the American Society of Brewing Chemists,* 73, 195–205.

Vinoth, A., & Ravindhran, R. (2017). Biofortification in Millets: A Sustainable Approach for Nutritional Security. *Front Plant Science*, 8, 29-37.

Vis, R. B., & Lorenz, K. (1998). Malting and brewing with a high β-glucan barley. *Lebensmittel-Wissenschaft und -Technologie,* 31, 20–26.

Vishwanathan, V., Urooj, A., & Malleshi, N. G. (2009). Evaluation of anti-oxidant and anti-microbial properties of finger millet polyphenols (*Eleusine coracana). Journal of Food Chemistry*, 114, 340-346.

Wankhede, D. B., Shehnaz, M. R., & Raghavendra, R. (1979). Preparation and Physicochemical properties of Starches and their fraction from finger millet (*Eleusine coracana*) and foxtail millet (*Setaria italica*). http://doi.org/10.1002/star.19790310505 (31).

Wardlaw, G. M., Hampl, J. S., & DiSilvestro, R. A. (2004). *Perspective in Nutrition*. 6th Edition, Mcgraw-Hill College, 1, 10-56.

Watanabe, M., Maeda, T., Tsukahara, K., Kayahara, H., & Morita, N. (2004). Application of pregerminated brown rice for breadmaking. *Cereal Chemistry*, 81, 450–455.

Wcislo, G., & Szarlej-Wcislo, K. (2014). Colorectal cancer prevention by wheat consumption: A three-valued logic - true, false, or otherwise? In: Wheat and Rice. *Disease Prevention and Health*, 1, 91-111.

WHFoods, 2017. World's Healthiest Foods. http://whfoods.org/.

Xu, J., Zhang, H., Guo, X., & Qian, H. (2012). The impact of germination on the characteristics of brown rice flour and starch. *Journal of the Science of Food and Agriculture*, 92, 380–387.

Yang, F., Basu, T. K., & Ooraikul, B. (2001). Studies on germination conditions and antioxidant contents of wheat grain. *International Journal of Food Sciences and Nutrition*, 52, 319–330.

Yang, R., Wang, P., Elbaloula, M. F., & Zhenxin, G. U. (2016). Effect of germination on main physiology and biochemistry metabolism of sorghum seeds. *Bioscience Journal*, 32, 378–383.

APPENDICES

APPENDIX A

Functional Properties of Composite Flours Mix

Sample/parameters	A	B	C	D	E
Dispersibility (%)	35.78±1.03[a]	45.05±1.78[b]	47.05±2.18[c]	47.44±1.12[c]	48.37±3.41[d]
Bulk density(g/mL)	0.84±4.07[a]	0.85±4.33[a]	0.86±3.12[a]	0.86±3.10[a]	0.86±1.12[a]
WAC(g/100g)	106.65±3.11[a]	125.65±6.12[b]	133.50±6.92[c]	151.75±6.48[d]	168.93±1.74[e]
OAC(g/100g)	91.70±3.19[a]	93.80±2.66[b]	97.60±4.79[c]	98.85±3.13[d]	99.75±5.02[e]
Viscosity (%RVU)	1851.50±7.34[e]	1401.35±3.13[d]	1251.79±2.70[c]	1131.27±1.16[b]	1072.65±4.11[a]

Values are mean of triplicate determinations ± SEM

Values along rows with different superscript are significantly different ($p<0.05$)

Key: A: 100 % White flour, B: 80 % white flour, 20 % sprouted finger millet flour, C: 70 % White flour, 20 % sprouted finger millet flour, 10 % sprouted water melon seed flour, D: 60 % White flour, 30 % sprouted finger millet flour, 10 % sprouted water melon seed flour, E: 50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour.

APPENDIX B

Sensory Scores of Bread Products (%)

Sample/ Parameters	A	B	C	D	E
Colour	8.43±1.02^{d}	6.82±1.03^{c}	6.67±1.05^{c}	5.82±0.78^{b}	4.48±0.23^{a}
Aroma	8.21±0.53^{c}	6.63±0.97^{b}	6.09±1.01^{a}	6.25±1.00^{a}	6.03±1.09^{a}
Taste	8.80±1.09^{d}	6.80±1.01^{c}	5.86±0.87^{b}	6.08±1.05^{c}	4.25±0.44^{a}
Crumb texture	8.05±0.34^{d}	6.81±1.01^{c}	5.84±0.94^{b}	4.80±1.08^{a}	4.42±1.01^{a}
Appearance	8.71±0.44^{d}	7.14±1.01^{c}	7.14±0.97^{c}	6.10±1.00^{b}	5.93±0.43^{a}
Overall acceptability	8.44±1.05^{c}	6.84±1.02^{b}	6.32±0.97^{b}	5.81±1.03^{a}	5.02±0.18^{a}

Values are mean of triplicate determinations ± SEM

Values along rows with different superscript are significantly different ($p<0.05$)

Key: A: 100 % White flour, B: 80 % white flour, 20 % sprouted finger millet flour, C: 70 % White flour, 20 % sprouted finger millet flour, 10 % sprouted water melon seed flour, D: 60 % White flour, 30 % sprouted finger millet flour, 10 % sprouted water melon seed flour, E: 50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour.

APPENDIX C

Sensory Scores of Cookie Products (%)

Sample/ Parameters	A	B	C	D	E
Colour	8.30±0.94^{d}	6.10±0.19^{b}	7.25±0.12^{c}	6.90±0.91^{b}	4.81±0.58^{a}
Aroma	8.75±1.04^{d}	6.68±0.23^{b}	7.95±0.19^{c}	6.60±0.77^{b}	4.55±0.59^{a}
Taste	7.90±0.56^{c}	6.58±0.15^{b}	6.85±0.61^{b}	5.15±0.54^{a}	6.00±0.41^{b}
Crumb texture	8.00±0.72^{c}	6.69±0.53^{b}	6.50±0.44^{b}	6.50±0.93^{b}	5.00±0.59^{a}
Crispiness	7.10±1.00^{c}	7.00±0.49^{c}	6.81±0.32^{b}	6.15±1.00^{b}	5.11±0.34^{a}
Appearance	8.12±0.15^{c}	6.58±0.37^{a}	7.09±0.23^{b}	6.63±0.35^{a}	5.72±0.26^{a}
Overall acceptability	8.02±0.34^{d}	6.77±0.55^{b}	7.07±0.38^{c}	6.32±0.18^{b}	5.19±0.89^{a}

Values are mean of triplicate determinations ± SEM

Values along rows with different superscript are significantly different ($p<0.05$)

Key: A: 100 % White flour, B: 80 % white flour, 20 % sprouted finger millet flour, C: 70 % White flour, 20 % sprouted finger millet flour, 10 % sprouted water melon seed flour, D: 60 % White flour, 30 % sprouted finger millet flour, 10 % sprouted water melon seed flour, E: 50 % White flour, 30 % sprouted finger millets flour, 20 % sprouted water melon seed flour.

APPENDIX E

Sprouted Finger Millets (*Eleusin coracana*)

Key: Day 0 = non-sprouted seeds, day 3 = seeds sprouted for three days, Day 6 = seeds sprouted for six days, Day 9 = seeds sprouted for nine days.

APPENDIX F

Sprouted water melon seeds (*Citrullus lanatus)*

Key: Day 0 = non-sprouted seeds, day 3 = seeds sprouted for three days, Day 6 = seeds sprouted for six days, Day 9 = seeds sprouted for nine days.