

**PRODUCTION OF ORGANOLEPTICALLY AND NUTRITIONALLY ACCEPTABLE
COUSCOUS FROM LOCAL SORGHUM (*Sorghum bicolor*) AND WHEAT (*Triticum
durum*)**

BY

HABIBU SAFIYANU

**DEPARTMENT OF BIOCHEMISTRY,
AHMADU BELLO UNIVERSITY,
ZARIA, NIGERIA**

AUGUST, 2021

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HABIBU SAFIYANU

**BSc. BIOCHEMISTRY (ABU ZARIA 2014)
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**DEPARTMENT OF BIOCHEMISTRY,
FACULTY OF LIFE SCIENCES,
AHMADU BELLO UNIVERSITY,
ZARIA, NIGERIA**

AUGUST, 2021

Declaration

I declare that the work in this Dissertation entitled “**Production of Organoleptically and Nutritionally Acceptable Couscous from Local Sorghum (*Sorghum bicolor*) and Wheat (*Triticum durum*)**” has been carried out by me in the Department of Biochemistry faculty of life sciences, Ahmadu Bello University Zaria under the supervision of professor H.M. Inuwa and professor K.M. Anigo. The information derived from literature has been duly acknowledged in the text and a list of references provided. No part of this Dissertation was previously presented for another degree or diploma at this or any other Institution.

HABIBU SAFIYANU

(P14SCBC8066)

Signature

Date

Certification

This Dissertation entitled “**Production of Organoleptically and Nutritionally Acceptable Couscous from Local Sorghum (*Sorghum bicolor*) and Wheat (*Triticum durum*)**” by **Safiyanu HABIBU** meets the regulations governing the award of the degree of Master of Science in Nutrition of the Ahmadu Bello University Zaria, and is approved for its contribution to knowledge and literary presentation.

Prof. H. M. Inuwa

Chairman, Supervisory Committee

Signature

Date

Prof. K. M. Anigo

Member, Supervisory Committee

Signature

Date

Prof. A. B. Sallau

Head of Department

Signature

Date

Prof. Sani Abdullahi

Dean, School of Postgraduate Studies

Signature

Date

Dedication

This research work is dedicated to Almighty Allah for sparing my life up to this moment, my uncle Pharm Umar Garba Gumau, Alhaji Habibu Yusuf my Father and my caring Mother Hajiya Hajara Zubair.

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My sincere gratitude goes to Almighty Allah for being my only source of strength, hope, helper and provider in every phase of my studies. Alhamdulillah. I sincerely appreciate my Supervisor, guardian and confidant, Professor H.M. Inuwa, for all the support, advice and immense contribution towards the success of this work. Prof. thank you so much. May you live long and in total sound health Ma. My gratitude also goes to my Co-supervisor Professor K.M. Anigo for his immense contribution. I am forever grateful Sir.

I also want to thank my uncle Pharm Umar Garba Gumau, His wife Hajiya Amina Usman and his entire family for their immense financial and moral support toward my academic success. Sir, thank you very much.

To my mother and my everything, Hajiya Hajara Zubair, I can't thank you enough for your prayers day and night to see that I succeed in everything. I love you mother and may Almighty Allah continue to keep you for us, Ameen.

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My special regards also goes to my beloved wife Rabi'at Abubakar and my daughters Nusaibah Safiyanu, Mufida Safiyanu and my son Muhammad Habeeb Safiyanu. Rabi'at you have assisted me so much in my studies. Thank you so much may we live and succeed together forever. Thank you.

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List of Abbreviations

AOAC	Association of Official Analytical Chemists
BNF	British Nutrition Foundation
BSTID	Board of Science and Technology for International Development
DASH	Dietary Approaches to Stop Hypertension
CVD	Cardiovascular Diseases
CHD	Coronary Heart Diseases
FAO	Food and Agriculture Organization
GI	Glycemic Index
GL	Glycemic Load
NDNS	National Diet and Nutrition Survey
NSP	Non-Starch Polysaccharides
RDA	Recommended Dietary Allowance
RNI	Recommended Nutrient Intakes
TSM	Technicon Sequential Multi-Sample Amino Acid Analyzer
UNICEF	United Nations Children's Education Fund
USDA	United States Department of Agriculture
WHO	World Health Organization
WCRF	World Cancer Research Fund

Abstract

Cereals grains have been one of the most important raw materials for the production of a variety of local foods in Nigeria such as tuwo, kunu, pap, porridge and others. Couscous and many other baked products such as bread, biscuit, doughnut and others, are known to be produced using wheat flour both traditionally and at industrial level making it one of the most expensive and highly imported cereal grains. Also wheat gluten makes it difficult for people suffering from coeliac disease to safely consume food products from wheat. This work evaluated replacing gluten free sorghum with wheat in the production of couscous which could have more health benefits and be cost effective. Sorghum (*Sorghum bicolor*) and wheat (*Triticum durum*) grains were sourced locally and processed into flour locally for the production of couscous which were produced at three different levels: (1) using 100 % sorghum flour, (2) using 100% wheat flour (3) and using a mixture of 50 % wheat flour and 50% sorghum flour. The 100 % wheat flour couscous was produced to serve as a normal control. The following analyses were carried out on the sorghum and wheat couscous; proximate composition, dietary fibre, mineral elements contents and amino acid profile using standard methods. The results for the analysis carried out showed that sorghum couscous has significantly ($p<0.05$) high nutritional contents compared to wheat couscous. Using 9 point hedonic scale, sensory evaluation was carried out to compare the acceptability of the produced couscous. No significant difference was observed in overall acceptance between the 100 % sorghum flour couscous (test) at 5.80 ± 1.82 and 100 % wheat couscous (control) at 5.93 ± 2.02 . Sorghum 50 % and wheat 50 % couscous significantly ($p<0.05$) differ in overall acceptability from 100 % sorghum flour couscous and 100 % wheat flour couscous at 6.53 ± 1.32 . Based on the result of this study, it can be concluded that organoleptic and nutritionally acceptable couscous can be produced from local sorghum

(sorghum bicolor) due to its high nutritional contents compared to wheat and acceptability of the products by the panelist during sensory evaluation.

CHAPTER ONE

1.0

INTRODUCTION

1.1 Background

Cereals such as wheat sorghum, rice, maize, barley etc. are major staple foods for human nutrition and are considered as a major source of calories for human (Lafiandra *et al.*, 2014). The dietary components in cereals such as lipids, carbohydrates and proteins play an important role in processing and nutritional quality of food. Among cereals, wheat is one of the major staple crops across the world used in making a variety of food products (FAO, 2018). The estimated global production of wheat as at 2020 was 764.32 million metric tonnes (USDA, 2020). In Nigeria, traditional wheat cultivation is commonly done in the northeast region, particularly in Adamawa, Bauchi, Borno, Taraba, Yobe, Gombe, Jigawa and Kano states. However, the overall wheat production of these states has been reported to be too meagre to a population that is increasingly reliant on wheat flour based food products such as bread, semolina, pasta and other major staples in Nigeria's growing urban areas.

Data provided by the ministry of agriculture in 2019 indicated that Nigeria produces an annual volume of about 420,000 tonnes, while national consumption was recorded as 5.26 million metric tonnes, leaving a shortfall of about 4.5million tonnes. As a result, Nigeria is forced to import about 4 million tonnes of wheat per year and the country's import data shows that Russia, the United States, Canada, and Australia supply the bulk of Nigeria's wheat imports destined for milling and other productions. Data by International Trade Administration (ITA) also showed that Nigeria imported an estimated 1.8 billion dollar worth of wheat in 2019 and nearly 2 billion dollar in 2020 with the country's milling capacity of over eight million tonnes. According to USDA, Between January – November 2020, Nigeria imported a total of 1,292,029 tonnes of

wheat from the U.S, making it the fifth largest U.S. wheat importer in the world after Philippines, Mexico, Japan and China. Statistics from the central bank of Nigeria (CBN) also confirmed that Nigeria's high import expenditure when it disclosed that Nigeria spent about 6 billion dollars on importation of wheat from January 2016 to June 2020.

Though wheat has been used as important staple crop worldwide, in certain individuals and genetically predisposed people, it causes certain disorders and conditions arising mainly from the gluten fraction (Cabanillas *et al.*, 2019). Coeliac disease which is found in 1 in every 100 of the world population, is a chronic immune-mediated enteropathy of the small intestine that develops in genetically susceptible individuals by exposure to gluten protein found in wheat, rye and barley (Freeman *et al.*, 2011). Therapy for Coeliac disease patients are nonexistent and the only solution to the problem is stringent lifelong gluten free diet (Cohen *et al.*, 2019). Alternative cereals such as sorghum, maize, rice and millet are gluten free and can be substituted for wheat in the production variety of food products for people with coeliac disease. Currently, Nigeria is the second largest producer of sorghum in the world with annual production of about 6,665,000 metric tonnes (FAO STAT, 2019). Nigeria also account for about 65 – 70% of the total sorghum production in West Africa and it is mostly produced in northern states including Bauchi, Borno, Zamfara, Yobe, Gombe, Adamawa, Kaduna, Jigawa, Niger, Kebbi, Taraba, Plateau, Sokoto, Katsina and Nasara (Sanni *et al.*, 2003).

Sorghum grain has been shown to be a good source of carbohydrate, vitamins and to some extent protein and minerals for the poorest in Africa and Asia (FAO, 1995). It is used for the production of variety of local products in Nigeria which include; kunu, pap (akamu), tuwo, porridge and local alcoholic beverages and industrial products like breads, cakes, biscuits, sweeteners, malting and brewing (Momoh, 2012). Even though the grain sorghum is the most commonly cultivated in Nigeria, and second most

important grain in Africa (Berenji and Dahlberg, 2004), more research need to be done in utilization, improvement and marketing potential of the grain. This research therefore aimed at replacing wheat with local sorghum (*Sorghum bicolor*) to produce couscous thereby enhancing the utilization of the locally produced grain which can result into boosting up of the economy by making the products cheaply available and affordable. Production of couscous from sorghum can also pave way for a new staple food making it more available and can also serve as part of complementary food for children 12 to 24 months of age.

1.2 Sorghum

Sorghum, a tropical plant, belonging to the family of poaceae is one of the most important cereal crops in Africa, Asia and Latin America (Anglani, 1998). *Sorghum bicolor* is a warm season crop, intolerant of low temperature but fairly resistant to serious pest and diseases. It is known by a variety of names such as great millet and guinea corn in West Africa, Dawa (Kaura) in Hausa, Okababa in Yoruba, Gauri in Fulfulde, Kafircorn in South Africa, Jowarin India and Koaling in China. It is a staple food in many parts of Africa and Asia (FAO, 1995). Most of the sorghum produced in North and Central America, South America and Oceania is used for animal feed (FAO, 1995). The grain consists of a naked caryopsis, made up of a pericap, endosperm, and germ. Although there is a wide range of physical diversity, sorghums are classified into one of the four groups; the grain (sorghum), Forage sorghum, Grass sorghum, Sudan sorghum and broom corn (Macrae *et al.*, 1993). Sorghums are also grouped according to: the colour of the pericap (white, yellow and red), presence or absence of pigmented testa (with or without tannins) and pericap thickness.

The total production of sorghum in the world in 2020 was 61.86 million tonne (USDA, 2020). Sorghum is a good source of carbohydrate and contains protein, fat, vitamins,

minerals and some phytochemicals (Pins *et al.*, 2002). Sorghum has been used in the preparation of local products at home such as pap (Kunu), akamu, porridge, tuwo etc. and industrial products like noodles, cake, malting and brewing.

1.3 Wheat

Wheat (*Triticum* spp.) is a cereal grain, originally from the Levant region of the Near East but now cultivated worldwide. The estimated global production of wheat as at 2020 was 764.32 million metric tonnes (USDA, 2020). World trade in wheat is greater than that of all other crops combined (Curtis and Halford, 2002). Wheat provides a good source of protein in human food globally, having a higher protein content than other major cereals, maize (corn) or rice (World Wheat, 2015). Wheat gluten is found in thousands of normal food items, making life for those suffering from Coeliac disease very difficult (Shewry *et al.*, 1992). Fortunately, alternative grains and grasses, such as sorghum, can be eaten safely by those suffering from this increasingly common condition, without the painful inflammation, nausea, and gastrointestinal damage that gluten causes. Raw wheat can be ground into flour. Wheat is a major ingredient in many foods such as bread, crackers, biscuits, pancakes, pies, pastries, cakes, cookies, and doughnuts (Rooney and Waniska, 2000). In 100 grams, wheat provides 327 calories and is an excellent source (more than 19% of the Daily Value, DV) of multiple essential nutrients, such as protein, dietary fibre, manganese, phosphorus and niacin. Wheat is also rich in B vitamins and other dietary minerals. Wheat is 13% water, 71% carbohydrates, 1.5% fat and 13% protein (Sandberg *et al.*, 1986).

1.4 Couscous

The word “Couscous” is derived from the Arabic word “kaskasa” to mean pound small, but the word is also thought to be derived from Arabic name for the perforated earthen ware steamer pot, used in steaming the couscous called keskes in Arabic and

couscousirre in French (Bender and Bender, 2005).It is the crushed and steamed semolina of hard wheat (*Triticum durum*). Couscous could be made with sorghum, millet, barley or corn (Bender and Bender, 2005). Couscous is an icon food in North Africa for dietary and cultural reasons. Similar to rice, pasta, bread, couscous is a highly nutritive product, made from durum wheat or other cereals such as pearl millet, corn, soft wheat semolina and rice flour (Caballeroet al.,2003). With a modern cooking system, it is possible to prepare an everyday meal or a luxury feast, a main course or a desert. A versatile dish couscous can be mixed with vegetables, legumes, meat or fish or can be eaten with butter or fresh fruit (Caballeroet al.,2003).

The origin of couscous is uncertain. Lociebolens affirms that Berbers were preparing couscous as early as 238 to 149 B.C.E (Bolens, 1989). Nevertheless, Charles perryin (1990) stated that couscous originated between the end of the Zirid dynasty, and the rise of the Almohadiandynasty between the eleventh and thirteenth centuries.

1.5 Statement of Research Problem

There is over dependence on imported expensive wheat by a large number of Nigerian citizens for the production and processing of couscous as a staple food. There was no available cereals that may be alternative raw materials for couscous production that were explored in order to reduce the over dependency on imported wheat and to meet the demand of the people who consume couscous as a staple food. Wheat products contain gluten, a non common protein to cereals which is intolerable by coeliac patients(Kasarda, 2001).

1.6Justification

Production of acceptable couscous from sorghum, could reduce the over dependence on imported wheat, and to some extent rice grain and can pave way for a new staple food in

the country. Sorghum is often recommended as safe food for coeliac patients (Kasrada, 2001). Therefore couscous from sorghum could have more health benefits. Sorghum, one of the major food crops that farmers in northern Nigeria cultivate was used in this study to serve as an alternative raw material for couscous production so as to reduce the cost, as well as permit couscous production in many parts of Nigeria which could result into boosting up of the economy and social wellbeing.

1.7 Aim and Objectives

1.7.1 Aim

The main aim of this work was to produce an organoleptically and nutritionally acceptable couscous from sorghum other than wheat couscous.

1.7.2 Specific Objectives

The following are the objectives of the work;

1. To determine the proximate composition, dietary fibre content and amino acid composition of the locally prepared couscous from sorghum (*Sorghum bicolor*) and wheat (*Triticum durum*) grains.
2. To determine the antinutrient and mineral elements composition of the locally prepared couscous from sorghum (*Sorghum bicolor*) and wheat (*Triticum durum*) grains
3. To carry out a comparative sensory and organoleptic evaluation of the locally prepared couscous from sorghum (*Sorghum bicolor*) and wheat (*Triticum durum*) grains.

1.8 Null hypothesis

H₀: Organoleptically and nutritionally acceptable couscous cannot be produced from sorghum (*Sorghum bicolor*).

H₀: Organoleptically and nutritionally acceptable couscous cannot be produced from wheat (*Triticum durum*).

1.9 Alternative hypothesis

H₁: Organoleptically and nutritionally acceptable couscous can be produced from sorghum (*Sorghum bicolor*).

H₁: Organoleptically and nutritionally acceptable couscous can be produced from wheat (*Triticum durum*).

CHAPTER TWO

2.0

LITERATURE REVIEW

2.1 Cereals

Cereals are the edible seeds of the grass family (Bender and Bender,2005). A number of cereals grown in different countries include rye, oats, barley, maize, sorghum, and millet. Cereals are grown for their highly nutritious edible seeds which are often referred to as “grains”. Some cereals have been staple foods both directly for human consumption, and indirectly for livestock feed since the beginning of civilization (BNF, 1994). Cereals are the most important sources of food (FAO, 2002) and cereals based foods are a major source of energy, protein, B vitamins, and minerals for the world population.

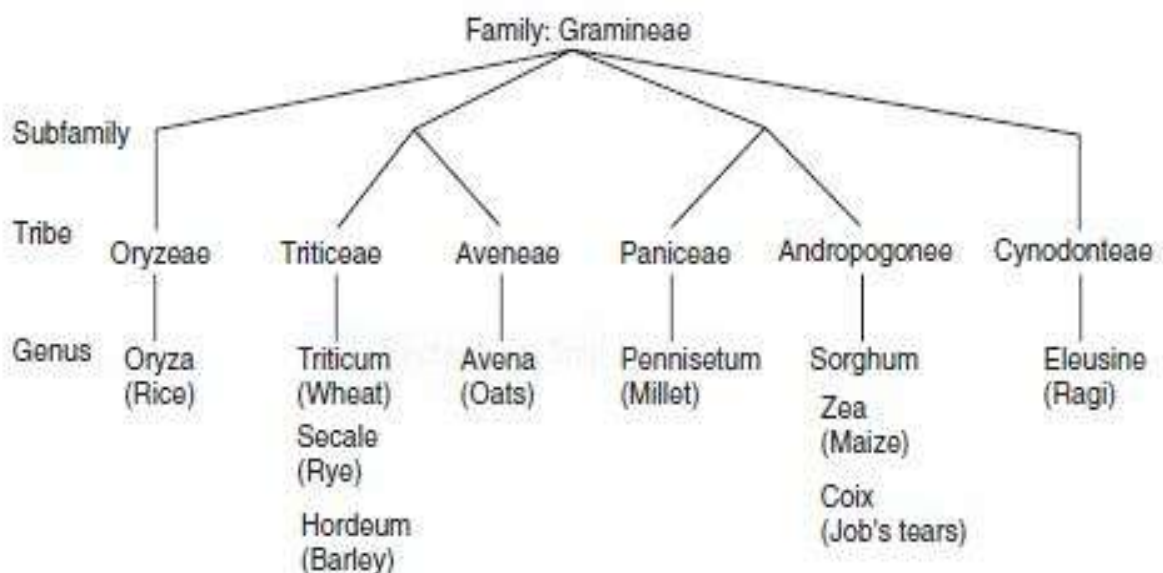


Figure 2.1 Taxonomy of the gramineae family
(Source: *Shewry et al.*, 1992)

2.1.1 General structure of cereal grains

Grains develop from flowers and florets although the structures of the various cereal grains are different. There are some typical features, the embryo or germ is a thin walled structure containing the new plant, it is separated by the scutellum (which is involved in mobilization of food reserves for the grain during germination) from the main part of the grain, the endosperm (Montonenet *al.*, 2003).

The endosperm consists of thin walled cells, packed with starch grains, if the cereal grain germinates; the seedling uses the nutrients provided by the endosperm until the development of green leaves that allow photosynthesis to begin (FAO, 1991: Kent and Evers, 1994). The endosperm is surrounded by the aleurone consisting of one or three cell layers (wheat, rye, oats, maize and sorghum have one; rice and barley have three each). The other layers of the grain are the pericarp (derived from the ovary of the flower) which surround the seed coat (the *testa*). The thick walled structures form the bran (BNF, 2004).

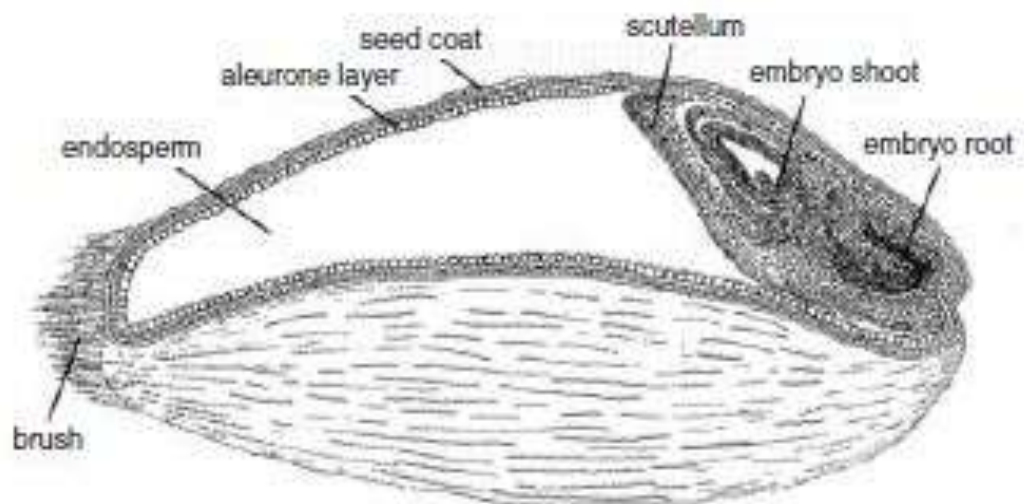


Figure 2.2 General structure of a grain

[Source: *The Big Picture* (Dr. Gary Barker, webmaster. Gary.Barker@Bristol.ac.uk
Adapted from: McKeivith, (2004)].

2.1.2 Cereals production

Cereals are grown in a number of countries. The forecast for the world cereals production in 2021 is 2821 million tonnes. More than 50% of world's cereals are produced in developing countries (FAO, 2020).

2.1.3 Processing

Cereals typically undergo a range of processes to produce a variety of different products, including non-food products. Milling is the main process associated with cereals especially the bread cereals; wheat and rye. The range for processing of cereals may also include extrusion and fermentation, most especially in the production of cereals products, as well as having technical consequence. Processing also changes the nutritional contents of cereals (BNF, 2004).

2.1.4 Milling

The process of milling can basically be described as grinding, sifting, separation and regrinding. These steps are repeated to extract the endosperm. Before milling begins, the cereal grains are cleaned. Most modern equipments used differ in size, shape, colour, specific weight, and response to magnetic force to separate foreign materials from the grain. Prior to grinding, water may be added to the cereal which is allowed to rest before milling (tempering), this allows absorption of water by the grains toughening the pericarp and germ so that they do not splinter during milling. If heat is also applied during tempering then the process is referred to as conditioning (Hoseney, 1994).

2.1.5 Nutritional Value of Cereals

Cereals are staple foods providing a major source of carbohydrates, protein, B vitamins and minerals for the world population, as well as containing a range of phytochemicals,

which may provide some health benefits seen among populations consuming diets based on plants foods. Cereals also contain a number of anti-nutrients (Goldberg, 2003).

2.1.5.1 Carbohydrate

Cereals are often classed as carbohydrate-rich foods, as they are composed of approximately 75% carbohydrate. Starches, the major component of the cereal, occur in starch granules in the endosperm. Starch granules differ in size (e.g. in rice they have a diameter of only 5 mm, while in wheat they may be 25– 40 mm) and shape (either large, lens-shaped granules or small, spherical granules). The ratio of amylose to amylopectin within the starch granules varies, depending on the cereal and its variety. Within common varieties of cereals, 25–27% of starch is present as amylose, while in waxy varieties (e.g. rice and corn) most of the starch is amylopectin. However, in cereal products, a proportion of this starch is not digested and absorbed in the small intestine. This is referred to as resistant starch (RS) and it appears to act in a similar way to dietary fibre. Four categories of resistance have been defined (Baghurst *et al.*, 1996):

- i. RS1 refers to starch that is physically inaccessible for digestion as it is ‘trapped’ (e.g. intact whole grains and partially milled grains).
 - ii. RS2 refers to native resistance starch granules (e.g. found in high amylose maize starch).
 - iii. RS3 refers to retrograded starch (e.g. found in cooked and cooled potatoes, bread and some types of corn- flakes).
 - iv. RS4 refers to chemically modified starch (e.g. commercially manufactured starches).
- A small amount of free sugars is also present (~1–2%), mainly as sucrose but low concentrations of maltose and very low concentrations of fructose and glucose occur.

Glycaemic index (GI)

The GI is used for classifying carbohydrate containing foods. It can be defined as ‘the incremental area under the blood glucose curve after consumption of 50g carbohydrate from a test food, divided by the area under the curve after eating a similar amount of control food (generally white bread or glucose) (Ludwig and Eckel, 2002). The glycaemic load (GL) assesses the total glycaemic effect of the diet and is the product of dietary GI and total amount of dietary carbohydrate (Jenkins *et al.*, 2002). The rate of digestion and absorption of carbohydrates is influenced by a range of factors (Pi-Sunyer, 2002), including: The nature of the monosaccharide components, the nature of the starch (e.g. the amylose to amylopectin ratio), cooking or food processing; (e.g. milling increases the GI of cereals), other food components (e.g. fat, protein and fibre). Cereals traditional starchy foods such as steamed bread and noodle, and food processed by modern technology such as breakfast cereals and some instant foods are rich in carbohydrates. The significant difference of GI of different cereal foods suggested that food procession and food type were factors influencing the value of GI (Jenkins *et al.*, 2002).

Sorghum foods have demonstrated slow starch digestibility *in vitro* and in animal feeding trials, suggesting favourable effects on post-prandial glycemic and insulinemic responses in humans. Studies have shown that sorghum in the diet effectively improves glucose metabolism compared to sorghum-free diets (Chung *et al.*, 2011). The apparent ileal digestibility of sorghum starch was lower than for corn (Cervantes-Pahm *et al.*, 2014) which was attributed to the high level of resistant starch in sorghum that appeared to be fully fermented isince ~100% starch disappearance was reported. The low apparent ileal digestibility of its starch in the study suggests that sorghum may be of value for reducing the glycemic index of human foods (Cervantes-Pahm *et al.*,

2014).Dixit *et al.* (2011) have even recommended sorghum grain to be regularly consumed in the modern Indian diet to assist in the reduction of Type 2 diabetes and cardiovascular disease in the population.

Several metabolic effects may be related to the reduced rate of glucose absorption after a low GI food, e.g. a lower blood glucose concentration and a reduced post prandial rise in gut hormones and insulin, maintaining suppression of free fatty acids. The GI concept suggests a possible role for the rate of carbohydrate digestion in the prevention and treatment of chronic diseases. There may also be a role for low GI foods in weight management, as they promote satiety. Although one study observed that a similar amount of weight loss occurred with a high GI diet as with a low-GI diet (Wolever *et al.*,1992), several intervention studies have found that energy-restricted diets based on low GI foods produced greater weight loss than those based on high GI foods (Brand-Miller *et al.*, 2002). A systematic review highlighted inconsistent results in short-term studies measuring appetite sensations following low GI vs. high GI foods. In terms of weight loss, the 20 longer- term intervention studies found no advantage in using a low-GI diet compared to a high GI diet (1.5 kg loss on a low GI diet vs. 1.6 kg on a high GI diet) (Raben, 2002). Several epidemiological studies have also discovered a relationship between a high-GI diet and chronic diseases, e.g. coronary heart disease (CHD), type 2 diabetes and cancer (Jenkins *et al.*, 2002). A high fibre wheat or high fibre rye diet has been shown to decrease post prandial plasma insulin by 46– 49% and post prandial plasma glucose by 16–19% in overweight, middle-aged men compared to a low fibre diet but it is unclear if subjects were healthy, or had impaired glucose tolerance or type 2 diabetes (McIntosh *et al.*, 2003). Although the authors suggested that more comprehensive testing should be undertaken, they concluded that it was promising that

even in the short term, wholegrain foods were capable of decreasing the glycemic response.

The recent WHO/FAO report on nutrition and chronic disease associated low-GI foods with an overall improvement in glycemic control in people with diabetes, and several countries educate people with diabetes about GI. The WHO/FAO report also listed low GI foods as a possible factor in decreasing the risk of developing type 2 diabetes and reducing the risk of weight gain (WHO/FAO, 2003).

2.1.5.2 Protein

Cereals contain about 6–15% protein (Goldberg, 2003). The major storage proteins in wheat are gliadins and glutenins, while in rice it is glutelin (oryzenin), in maize it is prolamin (zein); barley has hordeins and glutelins, and in oats there are albumins and globulins (Kulp and Ponte, 2000). Although cereals provide a good range of amino acids, the building blocks of proteins, some are present in relatively low amounts. Essential amino acids must be supplied by the diet, and from these the human body is able to make other (termed non-essential) amino acids for itself. The essential amino acid that is in shortest supply in relation to need is termed the limiting amino acid. For cereals, the limiting amino acid is lysine, except for rye, where tryptophan is the first limiting amino acid (Macrae *et al.*, 1993). More favourable essential amino acid compositions can be found in rice, rye, barley and high-lysine cultivars (e.g. maize, sorghum and barley) (Macrae *et al.*, 1993). Combining cereals with other plant foods (e.g. rice and beans) can compensate for these limiting amino acids.

2.1.5.3 Lipids

Although the germ is the richest source of lipids, overall, lipids are only a minor component of cereals, with the amount varying from a lipid content of 1–3% in barley, rice, rye and wheat, to 5–9% in corn and 5–10% in oats, on a dry-matter basis

(Southgate and Johnson, 1993), this lipid fraction is rich in the essential fatty acid linoleic acid.

2.1.5.4 Micronutrients Composition of Cereals

Cereals can contribute to vitamins and minerals intake, although the micronutrient content will depend on the proportion of germ, bran and endosperm present (Jood, and Kapoor, 1994). The pericarp, germ and aleuronic layer are rich in vitamins and minerals so refined cereal products lose some of these nutrients.

2.1.5.5 Vitamins

Cereals contain no vitamin C or vitamin B12, no vitamin A and, apart from yellow corn, no beta- carotene (Courdain, 1999). However, cereals are an important source of most B vitamins, especially thiamin, riboflavin and niacin (Kulp and Ponte, 2000). Cereals also contain appreciable amounts of vitamin E.

2.1.5.6 Minerals

Cereals are low in sodium and are a good source of potassium, in common with most plant foods. Wholegrain cereals also contain considerable amounts of iron, magnesium and zinc, as well as lower levels of many trace elements, e.g. selenium (Kivisto *et al.*, 1986). Rice contains the highest level of selenium among the cereal grains, providing between 10 and 13 mg per 100g (Koh-Banerjee and Rimm, 2003). The selenium content of a cereal will vary depending on the selenium content of the soil; for example, the selenium content of wheat-grain can range from 0.001 mg per 100 g to 30 mg per 100 g (Lyons *et al.*, 2003). Wheat grown in North America generally has a higher selenium content compared to that grown in Europe and the switch to European wheat in recent years is suggested as the main explanation of falling selenium intake in the UK, although the effect (if any) of this decrease on health is not currently known (Goldberg, 2003).

2.1.6 Non-starch polysaccharides in cereals

All cereals are a rich source of non-starch polysaccharides (NSP). There are two types of NSP, insoluble and soluble and, although both may help in weight control (by delaying food leaving the stomach); they have different effects in the body. The insoluble NSP content of most cereals is similar, while the composition of the water-soluble NSP varies (Liu *et al.*, 2000). Arabinoxylans are the main water-soluble NSP in wheat, rye and barley, while in oats it is the beta- glucans. The amounts of beta-glucans and arabinoxylans are higher in barley, oats and rye compared to wheat (on a dry weight basis, 3–11%, 3–7%, 1–2% and <1%, respectively) (Wood, 1997).

2.1.7 Phytochemicals composition of cereals

Cereals contain a range of substances, which may have health-promoting effects that are often referred to as phytochemicals or plant bioactive substances (Goldberg, 2003). Although flavonoids are only present in cereals in small quantities, a number of other antioxidants are present, including small amounts of tocotrienols, tocopherols and carotenoids (McNulty *et al.*, 1996). In laboratory studies, wholegrain breakfast cereals have been found to have antioxidant content similar to those of fruits and vegetables (Miller *et al.*, 2000) and one study suggests that the major contributors of overall antioxidant activity are bound phytochemicals (Adom and Li, 2002). Lignans are a type of phytoestrogen found in cereals, and although the amount may be low (e.g. compared to that in linseed), cereals may be an important source because of the large quantities eaten daily.

2.1.8 Contribution of Cereals and Cereals' Products in diet

Cereal products play a central role in most countries and are staple foods for most of the world's population. In UK's Balance of Good Health food model, cereals and cereal products are grouped with bread and potatoes and this group of foods should form a

main part of meals. The dietary guidelines accompanying the Balance of Good Health encourage ‘plenty of foods rich in starch and fibre. Many of the foods in this group could be described as wholegrain foods, although no legal definition currently exists in the UK (although the American definition of a minimum of 51% wholegrain ingredient has been used by the Joint Health Claims Initiative). Currently America is the only nation to make specific recommendations regarding wholegrain foods (three servings a day) (Lang and Jebb, 2003).

2.1.8.1 Bread

Bread making goes back to prehistoric times, when a mixture of grass seeds was ground into a crude form of flour, to which water was added to form dough (Patent and Ainsworth, 1994). Bread is made from four ingredients namely, flour, water, yeast and salt and in the UK most bread is produced using wheat flour, although other flours, e.g. rye, are sometimes used. Different types of bread are produced from wheat flour depending on the proportion of the grain used, with brown breads being made from flour of an intermediate extraction rate (about 80–85%) (Wood, 1997). The traditional method of bread making involves the mixing of the four ingredients to form dough, which is then kneaded to develop the gluten, before being left to stand (during which time fermentation occurs). As this method is quite time-consuming and labour-intensive, a mechanical method, the Chorleywood process, was developed which uses a mechanical mixer, a fast-acting dough improver and a small amount of fat (Kent and Evers, 1994). White bread may be the most commonly eaten food in the UK; data from the recent adult National Diet and Nutrition Survey (NDNS) indicated that 93% of men and 89% of women ate it during the 7 day recording period (Henderson *et al.*, 2002). In the UK, weekly household consumption of bread has decreased by almost 1 kg since the 1940s. However, over the last 10 years there has been an increase in breads such as

French bread, naan bread, pitta bread and bagels (DEFRA and National Statistics, 2001). The current average adult intake of bread (including wholemeal, soft grain and other bread) is about 91g a day, roughly three slices (Henderson *et al.*, 2002).

2.1.8.2 Breakfast cereals

Developed in the late 19th century in America and introduced to the UK in the early 20th century, RTE breakfast cereals are an important source of nutrients. For example, in a sample of schoolchildren in Northern Ireland, fortified RTE breakfast cereals were associated with higher daily intakes of most micronutrients and fibre, and with a macronutrient profile consistent with current nutritional recommendations. Inadequate intakes of riboflavin, niacin, folate and vitamin B₁₂ (and iron in girls) were more likely in those children not consuming fortified breakfast cereals (McNulty *et al.*, 1996). Although vitamin D is not usually associated with breakfast cereals, because of fortification of RTE breakfast cereals, the recent NDNS report indicated that they now account for 13% of the average daily vitamin D intake in UK men and women (Henderson *et al.*, 2003). Similarly, in children, breakfast cereals account for 20% of the average daily vitamin D intake in girls and 24% in boys (Gregory *et al.*, 2000). Some recent work in school children has suggested that breakfast cereals may help maintain mental performance over the morning compared to no breakfast or a glucose drink (Wesnes *et al.*, 2003). A small study in adults also found that a high fibre carbohydrate rich breakfast was associated with the highest post breakfast alertness rating and the greatest alertness between breakfast and lunch (Holt *et al.*, 1999). A larger study found an association between breakfast cereal consumption and subjective reports of health, with those adults who ate breakfast cereal every day reporting better mental and physical health, compared to those who consumed it less frequently (Smith, 1999).

2.1.8.3 Pasta

Pasta is traditionally made from very hard (*durum*) wheat, which is high in protein, and water. The mixture is kneaded to produce a very stiff dough which is then extruded, cut and dried. Pasta was brought to Britain in the 18th century and in 2000/2001 in Britain, among those men and women who ate pasta (52% men and 53% of women), the mean consumption was 406 g and 330g a week, respectively (Henderson *et al.*, 2002). Biscuits, buns, cakes and pastries: Although cereal foods are generally low in fat, this sub- group contributes 7% to the average daily intake of total fat in adults and 5% in children (Gregory *et al.*, 2000; Henderson *et al.*, 2003 a, b). Within the balance of good health, therefore, they do not fall into the same category as the cereal products discussed above. Biscuits and buns, cakes and pastries form part of the group of foods containing fat; foods containing sugar.

2.1.8.4 Other foods containing cereal

Cereals and cereal products are used in a wide range of other foods. Cereal-based porridges are traditionally used as weaning foods in many parts of the world and in Nigeria baby cereals are often used at the first stage of weaning (Onofiako and Nnanyelugo, 2008).

2.1.9 Biomedical importance of cereals

There is much interest in understanding the role of particular foods, such as cereals, in the diet and their effect on health. Some of the work on cereals has focused specifically on wholegrain cereals and work suggests that people who eat wholegrains may have better nutrient intake profiles. For example, people in the USA who ate wholegrains had higher intakes of vitamins and minerals, and lower intakes of total fat, saturates and added sugars compared to those who did not eat whole grains (Cleaveland *et al.*, 2000). Cereals may also have a range of health benefits as discussed below.

2.1.9.1 Energy balance

Cereal foods have a relatively low energy density, and foods rich in wholegrain cereals may help reduce hunger as they are relatively bulky (Holt *et al.*, 1999; Saltzman *et al.*, 2001). Cereals may also affect body weight regulation through effects on hormonal factors (Koh-Banerjee and Rimm, 2003). By focusing on increasing cereal intake, it is possible to achieve a reduction in consumption of other foods and a reduction in fat intake. For example, a study using free living subjects found that including 60g of breakfast cereal with semi skimmed milk every day decreased the average intake of energy from fat by 5.4%, with a similar increase in energy contribution from carbohydrate (Kirk *et al.*, 1997). In another study, 14 subjects consumed four different breakfasts of the same energy content but with differing macronutrient content two fat rich and two carbohydrate rich (low or high fibre). The high fibre, carbohydrate rich breakfast was the most filling meal and was associated with less food intake during the morning and at lunch. Hunger returned at a slower rate after this meal than after the low fibre, carbohydrate- rich meal. Both fat rich breakfasts were more palatable but less satiating than the carbohydrate-rich meals (Holt *et al.*, 1999). The recent WHO/FAO expert committee report on nutrition and chronic diseases suggested that a high intake of NSP may be a protective factor against overweight and obesity (WHO/FAO, 2003).

2.1.9.2 Diabetes

A potential role for fibre in the prevention of diabetes was put forward over 30 years ago, and a high intake of cereal fibre has consistently been associated with a lower risk of diabetes (Willett *et al.*, 2002). For example, in a large prospective study of more than 42,000 men followed for about 12 years, an inverse association was found between wholegrain intake and type 2 diabetes. After adjustment for confounding factors, men in the highest quintile of intake compared to those in the lowest had an RR of 0.58 (Fung

et al., 2002). Similar results have been seen in women. Montonen *et al.* (2003) studied the intake of wholegrain and fibre of over 4,000 Finnish men and women, and the subsequent incidence of type 2 diabetes during a 10year follow up. An inverse association was found between wholegrain intake and risk of type 2 diabetes, with an RR between the highest and lowest quartiles of wholegrain consumption of 0.65, i.e. a 42% reduction in risk (Liu *et al.*, 2000; Meyer *et al.*, 2000). A reduced risk of type 2 diabetes was also associated with cereal fibre (RR 0.39). Data pooled from seven prospective cohort studies (including the Montonen study) provided a summary estimate of a 30% reduction in risk (RR 0.70) (Liu, 2003). The Nurses' Health Study found for comparing highest GI with lowest GI, the RR of developing diabetes was 1.37. The GL was also associated with diabetes (RR 1.47), and a high GL combined with a low cereal fibre intake (< 2.5 g/day) increased the risk of diabetes further (RR 2.50) (Salmeron *et al.*, 1997a).

Similarly, in the Health Professionals' Follow up Study, men on a high-GI diet had an increased risk of diabetes. Those men with a high GL diet and a low cereal fibre intake (< 2.5 g/day) had a further increased risk compared to men with a low GL diet and high cereal fibre intake) (Salmeron *et al.*, 1997b). In contrast, The Iowa Women's Health Study, while demonstrating a negative association between cereal fibre intake and risk of diabetes, found no significant association between GI and GL and diabetes incidence (Jenkins *et al.*, 2002). However, an elderly cohort was used in this study, which could have introduced selection bias (Augustin *et al.*, 2002).

There is strong evidence for the role of a high carbohydrate high fibre diet in improving glycaemic control for people with type 1 or 2 diabetes (Mann, 2001) and a higher fibre intake has been associated with better glycaemic control in people with type 1 diabetes (Buyken *et al.*, 1998). A recent RCT demonstrated that, in people with type 2

diabetes, a high fibre diet (containing 25g soluble fibre and 25 g insoluble fibre) could decrease blood glucose and insulin more than a diet of equivalent macronutrient and energy content, containing moderate amounts of fibre (Chandalia *et al.*, 2000). It is worth noting that the amount of fibre used in this study (50g) is high. The current UK recommendation for adults is 18 g a day and the average UK intake of fibre in 2000/2001 was 15.2g for men and 12.6 g for women (Henderson *et al.*, 2003a). Additionally, the method used for measuring fibre in the USA differs from that used in the UK, making comparisons difficult. There is also a role for low GI foods in the management of diabetes. Medium term studies have shown that improvements in glycemic control can be seen when people with diabetes replace high GI foods with low GI foods, such as wholegrain, minimally refined cereal products (Willett *et al.*, 2002). For example, a randomized, crossover study by Jarviett *al.* (1999) demonstrated that a low-GI diet improved glycemic control as well as decreasing LDL-cholesterol and normalizing fibrinolytic activity compared to a high-GI diet, identical for macro-nutrient composition and amount of dietary fibre. Although long term studies are required to establish the long term consequences, the GI concept is often used when counseling people with diabetes.

2.1.9.3 Digestive health

Insoluble fibre, which is found in a range of foods including cereals, may be important for gut health. Insoluble fibre absorbs fluid, increasing stool weight. It also promotes the growth and activity of the gut bacteria, which could also be beneficial for gut health. Recently, a study demonstrated that moderate intakes of high fibre wheat and high fibre rye foods could improve other markers of gut health, such as decrease faecal beta glucuronidase, secondary bile acids and paracresol concentrations, and decrease faecal pH, compared with a low-fibre diet (McIntosh *et al.*, 2003). From their work in Africa,

Burkitt and Walker suggested the importance of dietary fibre for digestive health, and a role in particular for preventing colorectal cancer (Vossenaar *et al.*, 1997). The World Cancer Research Fund currently lists NSP/fibre as a possible factor in decreasing the risk of colorectal cancer (Vossenaar *et al.*, 1997), although a UK report concluded that there was moderate evidence that diets rich in fibre would reduce colorectal cancer (Department of Health, 1998). Since this report several other studies have been published. A study of 455 000 older women with relatively low fibre intake followed for a mean of 8.5 years found little evidence that dietary fibre intake lowered the risk of colorectal cancer. However, this study was set up to investigate breast cancer and not colorectal cancer, and the highest quintile only had an average fibre intake of 17g a day (Mai *et al.*, 2003).

Two more recent studies have investigated the intake of dietary fibre and the incidence of colorectal cancer. The European Prospective Investigation into Cancer and Nutrition (EPIC) study followed more than 50,000 subjects aged 25–70 years for almost 2 million person per years. An inverse relationship between dietary fibre in foods and incidence of large bowel cancer was found, with an adjusted RR in the highest vs. lowest quintile of fibre from food of 0.58 (0.41–0.85). No food source of fibre was found to be significantly more protective but the results suggested that in populations with a low average intake of dietary fibre, doubling of total fibre intake from foods could reduce the risk of colorectal cancer by 40% (Bingham *et al.*, 2003). Another study performed within the Prostate, Lung, Colorectal, and Ovarian Cancer Screening Trial found that high intakes of dietary fibre were associated with a lower risk of colorectal adenoma, those in the highest quintile having a 27% decrease in risk compared to those in the lowest quintile. Fibre from cereals and from fruits showed the strongest inverse association (Peters *et al.*, 2003). Intakes in the highest quintiles of these two studies

were more than 30g of fibre a day, at least double the current average UK intake. The results from these last two studies are in contrast to those of a review of RCTs in this area which found no evidence to suggest that increased dietary fibre would reduce the incidence or recurrence of adenomatous polyps within a 2 - 4year period (Asano and McLeod, 2002). The role of fibre in the treatment of other bowel problems has been investigated. The faecal bulking action of insoluble fibre makes it useful in the treatment of constipation and diverticular disease (Thomas, 1994). In the past, a highfibre diet was the normal treatment for irritable bowel syndrome, but a recent review by Burden (2001) revealed a move-away from this approach, towards manipulation of the fibre fractions in the diet, dependent on the individual's symptoms.

2.1.9.4 Hypertension

Hypertension or high blood pressure (defined in the guidelines of the European Society of Hypertension as >140/90 mmHg) is a major risk factor for CVD and renal disease (Hermansen, 2000). Changes in sodiumintake have been shown to affect blood pressure in older people and those with hypertension and diabetes, but more recently a food based approach has been investigated. The Dietary Approaches to Stop Hypertension (DASH) studies focused on increasing consumption of a range of foods including wholegrain cereals, but with particular emphasis on fruits and vegetables, and low-fat dairy products. The DASH diet demonstrated a strong effect on hypertension, with a decrease in systolic blood pressure (SBP) of 11.4 mmHg and in diastolic blood pressure (DBP) of 5.5 mmHg among those hypertensive subjects (n = 133) (Appel *et al.*, 1997). The benefits seen with the DASH diet may in part be due to its high fibre content, and several studies have specifically looked at the effect of fibre on blood pressure, for example:

- i. In a double blind RCT, dietary fibre given as a supplement (7 g/day) was found to significantly reduce DBP among hypertensive patients (n = 32) compared to those receiving a placebo (n = 31) (Eliasson *et al.*, 1992).
- ii. A pilot RCT involving 18 hypertensive patients found the addition of 5.52 g beta glucan/day decreased SBP by 7.5 mmHg and DBP by 5.5mmHG. Virtually no change was seen in the control group (Keenan *et al.*, 2002).
- iii. Another small RCT study of hypertensive patients (n = 88) found that by including a wholegrain oat cereal, more patients in the oats group could stop or reduce their anti-hypertensive medication (77% vs. 42%). Those in the oat group whose medication was not reduced had substantial decreases in blood pressure, suggesting that wholegrain oats can have a beneficial effect on blood pressure (Pins *et al.*, 2002).

2.1.9.5 Heart health

Several large cohort studies in America, Finland and Norway have found that people eating relatively large amounts of wholegrain cereals have significantly lower rates of CHD and stroke. A review by Hu (2003), identified several prospective cohort studies showing an inverse association between wholegrain consumption and risk of cardiovascular disease (CVD). In addition, the prospective Physicians' Health Study in the USA following 86,000 men for over 5 years found men in the highest category for wholegrain breakfast cereal intake (≥ 1 serving/day) had a 20% decreased risk of dying from CVD, compared to those in the lowest category (relative risk (RR) of 0.80) (Liu *et al.*, 2003).

2.1.9.6 Cancers

Fibre may also decrease the risk of pancreatic and breast cancer (WCRF, 1997). A series of case control studies in Italy found an inverse association between wholegrain

food intake and the risk of a range of cancers, including those of the upper gastrointestinal tract, the bladder and the kidney (La Vecchia *et al.*, 2003). Cereals may have a protective effect on hormone-related cancers because of their lignan content (Goldberg, 2003). Lignans, a type of phytoestrogen, are modified by gut bacteria to be more similar in structure to mammalian lignans (Truswell, 2002).

2.2 Sorghum

Sorghum bicolor is a warm season crop, intolerant of low temperature but fairly resistant to serious pest and diseases (Chelowski, 1991). It is known by a variety of names such as great millet and guinea corn in West Africa, Kafir corn in South Africa, Jowar in India, and Koaling in China. It is a staple food in many parts of Africa and Asia (FAO, 1995). Most of the sorghum produced in North and Central America, South America and Oceania is used for animal feed (FAO, 1995). The grain consists of naked caryopsis, made up of a pericarp, endosperm and germ. Although there is a wide range of physical diversity, sorghums are classified into one of the four groups.

- i. The grain (grain sorghum)
- ii. Forage sorghum
- iii. Grass sorghum
- iv. Sudan Sorghum and broom corn (Macrae *et al.*, 1993).

Sorghums are grouped, using the following characteristics

- a. The colour of the pericarp (white, yellow and red)
- b. Presence or absence of pigmented testa (with or without tannins)
- c. Pericarp thickness

2.2.1 Botanical description of sorghum

Sorghum belongs to the tribe Andropogonea, and family poacea. Like most angiosperm (flowering plants) lineages sorghum is thought to be 200million years old (Paterson *et al.*, 2003). The main races of cultivated sorghum are: *S. bicolor*, vulgar, caudatum, kair, guinea and durra (Dev *et al.*, 1994 and BASTID-NRC, 1996). Sorghum is also found in temperate regions, and at altitude of up to 2300 meters in the tropics. It is well suited to heavy soils commonly found in the tropics, where tolerance to water logging is often required (African Journal of Biotechnology, 2006). *S. bicolor* is the fifth most important cereal crop after Wheat, rice, maize and barley in terms of production (FAO, 2005). The total production of sorghum in the world in 2014 is 26.4 million metric tons. United States of America (USA) is the world leading sorghum producer with annual production of 11.5 million metric tons. India ranked the second produces an average of 7.5 million metric tons. Nigeria is the top sorghum producing country in Africa and third in the world with an annual production of 7.4 million metric tons (FAO, 2014).

2.2.2 Sorghum grain as human food

Sorghum is Africa's contribution to the small number of elite grains that supply about 85 percent of the world's food energy. Only four other foods; rice, wheat, maize, and potatoes are consumed in greater amounts. Sorghum is the dietary staple of more than 500million people in more than 30 countries of the semi-arid tropics, thus being one of the most familiar foods in the world (Cleaveland *et al.*, 2000). Sorghum is indeed an ancient grain. Dahlberg and Wasylikowa., (1996) report on sorghum remains found in the Nabta Playa archaeological site in the Western Desert, in southern Egypt, dating back to 8000 B.C.E.

Sorghum is valued for its grain, stalks and leaves. Many people in the U. S. are familiar with sorghum for the syrup made from the sweet juice in stalks of certain sorghum

varieties or for the use of sorghum in silage or for pastures. The U.S. produces “food grade sorghum” a white coloured grain grown on a “tan” plant that produces light coloured glumes used to produce gluten free, bland flour suitable for incorporation into many cereal containing food products. Specialty sorghums are under study for their health promoting properties.

Sorghum is used extensively worldwide in food production system (Dordrecht and Ainsworth, 1994). Around the world, many types and colours of sorghum are used to produce various types of traditional foods and beverages. Unfermented bread, such as chapatti and roti are common in India, while tortillas are made from sorghum in Central America and Mexico. Fermented breads such as kisra and dosa are found in Africa, Sudan, and India, while injera is popular in Ethiopia. Stiff porridges called ugali, tuwo, karo, and mato are found throughout Africa, India and Central America, while thin porridges such as ogi, koko, and akasa can be found in Nigeria and Ghana.

2.2.3 Worldwide utilization of sorghum

Sorghum is grown in the United States, Australia, and other developed nations essentially for animal feed. However, in Africa and Asia the grain is used both for human nutrition and animal feed. It is estimated that more than 300 million people from developing countries essentially rely on sorghum as source of energy (Godwin and Gray, 2000). The main foods prepared with sorghum are: tortillas (Latino America), thin porridge, e.g. “bouillie” (Africa and Asia), stiff porridge, e.g. tuwo (West Africa), injera (Ethiopia), nasha and kisra (Sudan), traditional beers, e.g. dolo, tchapallo, pito, burukutu, etc. (Africa), ogi (Nigeria), baked products (USA, Japan, Africa), etc.

2.2.4 Gluten-Free whole grain sorghum

Whole grain gluten free sorghum flour is an excellent source of dietary fibre with 6.6g/100g flour (USDA, 2011). Most Americans of all ages under consume dietary fibre with an average intake of about 15 grams versus the 25 grams that are recommended (CDC, 2006). Concentrated sources of fibre include whole grains, dried peas and beans, vegetables, nuts and fruits. Fibre is intrinsic and intact only in plants. Dietary fibres are non-digestible forms of carbohydrates and lignin. Dietary fibre can be classified as fermentable or non-fermentable-both forms are needed for good health. Along with helping to provide satiety, a moderate amount of evidence suggests that dietary fibres from whole food sources protect against cardiovascular disease, obesity, and type 2 diabetes and is essential for optimal digestive health (Ness *et al.*, 2002). The 2010 Dietary Guidelines for Americans was careful to point out that increasing total grains was not recommended. However increased consumption of the proportion of whole grains to total grains was recommended because of the need to increase whole grain fibre (Ness *et al.*, 2002). In particular, individuals with coeliac disease may not consume enough dietary fibre and need gluten free whole grains such as sorghum in their diets.

A study with adolescents (Montonen *et al.*, 2003) found that higher dietary fibre intakes, but not low saturated fat or cholesterol intakes, were associated with lower incidence of metabolic syndrome. Since metabolic syndrome in adolescence leads to a higher incidence of metabolic syndrome, type 2 diabetes, and cardiovascular disease in adulthood, teens are a major group in need of education about whole grains and other sources of dietary fibre. The Nutrition Facts panel on food labels requires that the amount of dietary fibre per serving be shown. If a food product contains at least 2.5 g/serving, the label may state the food is a good source of fibre. Based upon an

extensive review of the evidence, the FDA recently approved a health claim for whole grains that permits manufacturers to state on foods: "Diets rich in whole grain foods and other plant foods and low in total fat, saturated fat, and cholesterol may reduce the risk of heart disease and some cancers" (Peters *et al.*, 2003) In order to use the claim, the serving size must contain a specified amount of dietary fibre per serving. Four serving sizes are specified ranging from a 35 gram serving with 1.7 grams of fibre to a 55 gram serving with 3.0 grams of dietary fibre (Peters *et al.*, 2003). Thus sorghum grain is an excellent source of dietary fibre.

2.2.5 Sorghum a safe diet for coeliac patients

Coeliac disease is a syndrome characterized by damage to the mucosa of the small intestine caused by ingestion of certain wheat proteins and related proteins in rye and barley (Fasano and Cattasi, 2001). The gliadins (Kagnoff *et al.*, 1982) and glutenins (Vande *et al.*, 1999) of wheat gluten have shown to contain protein sequence that is not tolerable by coeliacs. Sorghum is inherently gluten free and was demonstrated to be safe for people with coeliac disease (Neel, 1999).

2.2.6. Nutritional Composition of Sorghum

Like other cereal grains, sorghum is composed of three main parts, seed coat (pericarp), grain (embryo) and endosperm (storage tissue). The relative proportion varies, but most kernels are made up of 6 percent seed coat, 10 percent grain, and 84 percent endosperm. In its chemical composition the kernel (in its whole grain form) are about 70 percent carbohydrate, 12 percent protein, 3 percent fat, 2 percent fibre and 1.5 percent ash. When the seed coat and the grain are separated to leave stableflour (from the starchy endosperm) the chemical composition is about 83 percent carbohydrate, 12 percent protein, 0.6 percent fat, 1 percent fibre and 0.4 percent ash (BSTID-NCR, 1996).

Sorghum is an excellent source of energy, containing about 75% complex carbohydrate. Complex carbohydrates (fibres and starches) are usually digested slowly and therefore provide satiety and delayed hunger. Sorghum flour is whole grain and thus aligns well with the 2010 Dietary Guidelines to increase dietary whole grains while keeping the suggested total dietary carbohydrate intake the same (USDA, 2010). A Nutrient comparison of the commodity grains sorghum, wheat, corn, and rice shows that with about 3.3% fat content, grain sorghum contains more fat than wheat and rice, but slightly less than corn. Sorghum and wheat grain contain similar amounts of protein (~11- 12%) while both contain more protein than rice and corn (~6-9%) (USDA, 2011).

Table 2.1 Amino Acid Score of sorghum and wheat couscous based on FAO/WHO/UNU (2007)

Amino acid	FAO/WHO/UNU (2007) Maintenance amino acid pattern (mg/g protein)	Couscous	
		Sorghum	Wheat
Leucine	59	173.73	105.76
Lysine	45	56.00	81.78
Isoleucine	30	140.33	122.67
Phenylalanine	38	131.84	118.16
Valine	39	107.95	125.64
Tryptophan	6	205.00	253.33
Methionine	16	128.13	123.13
Histidine	15	156.00	165.33
Threonine	23	146.96	143.04

2.2.6.1 Carbohydrate

Carbohydrate is the grain major components, with starch making upto 32 to 79 percent of its weight. The remaining carbohydrates are largely sugars, which can be quite high in certain race of varieties of sorghum grain. The starches in most sorghum occur in both polygonal and spherical granules, ranging in diameter from about 5 to 28 microgram. Chemically, the starch is normally made up of 70 – 80 percent amylopectin (a non-gelling type) and 20 – 30 percent amylose (a gelling type). However, some sorghum starches contain as much as 62 percent amylose (BSTID-NCR, 1996).

2.2.6.2 Protein

Sorghum protein contents are more variable than that in maize and can range from 7 to 18 percent. The protein amino acid composition is much like that of maize protein. Lysine is the first limiting amino acid, followed by Threonine. The proteins contain no gluten. A large portion of it is polyamine, a cross linked form that humans cannot easily digest. In fact, prolamin makes about 59% of the total protein in normal sorghum. This is higher than in other major cereals and it lowers the food value considerably (BSTID-NCR, 1996).

2.2.6.3 Fat

Generally, sorghum contains about 1 percent less fat than maize. Free lipids make up to 2 - 4 percent of the grain and bond lipids, 0.1 - 0.5 percent. The oil properties are similar to those of maize oil. The fatty acids are highly unsaturated oleic and linoleic acid(BSTID-NCR, 1996).

A comparison of 100g of commodity sorghum to the World Health Organization (WHO) Recommended Nutrient Intakes (RNI) (35) for children ages 13 years shows the following micronutrients are met: Magnesium = 366% RNI; Iron based on 10% bioavailability = 73% RNI; Zinc based on moderate bioavailability = 38% RNI;

Thiamin = 47% RNI; Riboflavin = 28% RNI; Niacin = 49% RNI; Pantothenate = 63% RNI; Vitamin B 6 = 118% RNI. WHO does not have an established RNI for copper and manganese, thus using the United States Recommended Dietary Allowance (RDA) (FNB,2001) criteria, 100g sorghum meets the RDA for children ages 4-8 years as follows: Copper = 245% RDA; Manganese = 92% RDA. Iron and zinc are two of the four micronutrients (iron, zinc, iodine, and vitamin A) identified (Holland *et al.*, 1988) as at risk in populations of developing countries. Sorghum is a good to excellent source of iron and zinc. Sorghum is rich in B complex vitamins that play a major role in energy metabolism. Sorghum's high energy content and ready supply of B-complex vitamins are a perfect combination for energy utilization.

2.2.6.4 Vitamins

Compared to maize, sorghum contains higher levels of B vitamin pantothenic acid, niacin, folate and biotin. Similar levels of riboflavin, pyridoxine and lower levels of vitamin A (carotene). Most B vitamins are located in the germ (BSTID-NCR, 1996).

2.2.6.5 Minerals

The grain ash content ranges from about 1 to 2 percent. As in most cereals, potassium and phosphorus are the major minerals found. The calcium and zinc levels tend to be low. Sorghum has been reported to be a good source of more than 20 micronutrients (BSTID-NCR, 1996).

2.2.7 Phytochemicals

Sorghum provides good sources of phytochemicals such as phenolic acids, anthocyanins, phytosterols and policosanols. These compounds are familiar to the public as a result of health claims around sterols and stanols (heart health) and the publicity attributed to the antioxidant properties of anthocyanins (pigmented berries-

blueberries, strawberries, etc). Awika and Rooney, (2004) provide an excellent review of the potential health benefits of phytochemicals in sorghum. One category of phytochemicals, condensed tannins regardless of grain colour is not found in U.S. sorghum varieties and most sorghum produced elsewhere (Miller *et al.*, 2000).

2.2.8. Food products from Sorghum

2.2.8.1 *Cake*

Preparation of cakes is similar to that of cakes prepared from wheat Maida. Fine sorghum grain flour is used for the preparation of cakes. Sorghum flour is comparatively superior to the wheat flour for cake preparation. Fine sorghum grain flour is mixed with required quantities of sugar, egg, emulsifiers and fat. The dough is made little soft and nuts are decorated on the top and kept in oven for baking after keeping the dough in a mould (Rooney and Waniska, 2000).

2.2.8.2 *Noodles*

Noodles of acceptable quality could be made from hard endosperm sorghums while pasta could be made from sorghum and wheat composite flours. Sorghums with soft texture, yellow endosperm, and white pericarp and without pigmented testa produce best pasta products (Rooney and Waniska, 2000). The technology for making noodles and pasta from sorghum makes the product cheaper and healthier as sorghum products are known for high B vitamin and dietary fibre.

2.2.8.3 *Biscuits*

Biscuits are prepared from fine sorghum flour in combination with Maida to an extent of 15%. Biscuits are prepared as follows. Sorghum flour is mixed with Maida, vegetable fat, sugar, baking powder (15%) and essences required. The mixed dough is then compressed in a mould and baked at the required temperature. Studies on the parameters

like compressibility, breaking strength etc. showed that sorghum biscuits are having lesser breaking strength. Biscuits are good in taste and taste panel acceptability score is high for cakes and biscuits at NRCS, pilot studies are in progress in collaboration with a food Industry in Hyderabad. It is possible to produce biscuits of acceptable quality using 50:50 wheat and sorghum composite flour (Cashel *et al.*, 2001).

2.2.8.4 Bread

Bread can be made with sorghum flour by blending with Maida up to 40-60%. Bread can be made using the modified starches like carboxymethyl. Very fine flour of sorghum mixed with salt, sugar, fat and bread improvers and is made into dough. Baker's yeast is added to the dough and allowed for fermentation for longer period compared to the normal wheat bread and after the fermentation the dough is ready for baking. The dough is then kept in the mould and baked for one hour. To improve the leavening and softness comparable to wheat bread, more yeast and external gluten are added. 30- 40% of Maida along with sorghum may result in a tasty product. Studies on the shelf life are very important, as it is a key factor that is directly related to economy of bakery industry. Bread can be made using the modified starches like carboxy methyl starch. The shelf life varies from 24 to 72 hrs. Bread made with a composite flour containing sorghum is nutritionally valuable. However, sorghum alone lacks the functional proteins for bread making. Cashel *et al.* (2001) investigated the rheological properties and bread baking potential of a sorghum-based composite flour system containing various amounts of vital wheat gluten (exogenous gluten protein). Mixograph and Kieffer test results showed that exogenous gluten protein significantly enhanced the composite dough's strength. At fixed gluten protein levels, as sorghum flour increased, water absorption decreased slightly, dough strength and extensibility decreased, and mixing time increased significantly.

2.2.8.5 Malting and Brewing

Malting and brewing with sorghum to produce stout has been conducted on a large commercial scale since the late 1980's notably in Nigeria (Oloriet *et al.*, 1996). Brewing with sorghum is now taking place in east Africa southern Africa and the USA (Mackintosh and Hoggins, 2004). There have been extension research and development work and several excellent reviews published covering enzymes in sorghum malting and brewing technology (Tayloret *et al.*, 2006).

2.3 Couscous

Couscous is the Husked and crushed, but underground semolina of hard wheat (*Triticum durum*). Couscous could be made with sorghum, rice, millet, barley or corn. Semolina is the hard part of the grain of hard wheat, which resists the grinding of the mills ton. The word "couscous" is derived from the Arabic word "Kaskasa" meaning to pound small, but the word is also thought to be derived from Arabic name for the perforated earthen ware steamer pot, use in steaming the couscous called keskes in Arabic and couscousirre in French (Bender and Bender, 2005).

Couscous is an icon food in North Africa for dietary and cultural reasons. Similar to rice, pasta, bread, couscous is an expensive and highly nutritive product, made from wheat or other cereals (Caballero *et al.*, 2003). With a basic cooking system, it is possible to prepare an everyday meal or a luxury feast, a main course or a dessert. A versatile dish, couscous, can be mixed with vegetables, legumes, meat or fish or can be eaten with butter or fresh fruit (Caballero *et al.*, 2003).

Couscous is prepared by mixing flour water mixture into small particles, steaming, and drying. The cooked granules are a combination of gelatinized particles of different sizes

which must then be separated into desirable sizes (1-2mm) called couscous (Aboubakaret *al.*, 1999).

2.3.1 Storage

Sealed containers or bags are best to store couscous in to keep the moisture out. It will keep well at room temperature or cooler storage. It is not a whole grain product, but optimal flavour will be achieved if heat in storage is avoided. The origin of couscous is uncertain. Lociebolens affirms that Berbers were preparing couscous as early as 238 to 149 B.C.E (Bolens, 1989). Nevertheless, Charles Perry, states that couscous originated between the end of the Zirid dynasty, and the rise of the Almohadian dynasty between the eleventh and the thirteenth centuries (Perry, 1990).

2.3.2 Couscous Dish

Couscous is a major food of North African cuisine and it can also be used as side dish. Its popularity is due to its good nutritional benefits when served with a rich sauce. Couscous food contains complex carbohydrates, Vitamin B, and minerals (Laveen and Wilmington, 2000). Couscous is cooked in a special pot called *couscousirre* or couscous steamer, which has two components; a bottom perforated pan which contains the grain, and a globular pot that stands underneath it and contains boiling water whose steam cooks the granules (Caballero *et al.*, 2003).

CHAPTER THREE

3.0

MATERIALS AND METHODS

3.1 Materials

3.1.1 Sorghum and wheat sample collection

The samples used for this research work are; Sorghum and Wheat. The sorghum and wheat grains were purchased from Samaru market, put in polythene bags and identified at the herbarium unit of the department of Botany, Ahmadu Bello University Zaria and voucher numbers (Wheat 1365, Sorghum 2118) were allocated.

3.1.2 Equipment

- PTH analyzer: 120A PTH Amino Acid Analyzer
- Desiccator: Duran desiccator - Z317438
- Oven: Royal Premium 220 – Volt Italy
- AAS: BUCK Scientific VGP 210
- Furnace: Viny Trading Company, India TC - 334
- Kjeldahl Soxhlet apparatus: Ogawa Seik Company Ltd, Japan

3.2 Methods

3.2.1 Production of sorghum and wheat Flour

Measurements;

- i. Sorghum grains 8kg
- ii. Wheat grains 8kg
- iii. Water 4liters

The sorghum and wheat grains were processed into flour by traditional method. The grains were measured as indicated above and processed by winnowing and air aspiration to remove foreign and light materials present in the grains. Stones and other metallic materials were removed by hand picking. Exactly 8kg each of the grains were measured and steeped for one and half hours, winnowed and then coarse milled using the hammer

mill and subsequent milling to make the flour finer. The flour was then packaged in polythene bags and stored for production of couscous.

Sorghum and wheat Grain



Cleaned



Steeped for 1.5 hours



Winnowed



Coarse milled



Fine milled



Sieved



Flour

Figure 3.1: Flow chart for sorghum and wheat flour production

3.2.2 Production of Couscous from Sorghum and Wheat Flour

Measurements;

- | | | |
|------|---------------|--------|
| i. | Sorghum flour | 500g |
| ii. | Wheat flour | 500g |
| iii. | Water | 1liter |

Two different sieve sizes were used during the couscous production; Sieve with 1.5mm mesh size used for producing desirable couscous particles before steaming and a sieve with 2.5mm mesh size used in producing desirable couscous particles after steaming. The difference in the mesh sizes used before and after steaming was because the couscous particles tend to swell after steaming therefore 2.5mm mesh size was used instead of 1.5mm mesh size. Also during steaming the couscous particles tend to stick together making a large chunk which was broken intermittently so that the couscous particles can be steamed uniformly.

Exactly 500g each of the finely ground flour was measured, moistened with cold water (250 ml) and kneaded until the flour particles agglomerate. Then, the mixture was forced through a fine screen of 1.5mm mesh size to make desirable couscous particles before steaming. The mixture was put in a perforated pot fitted over another pot containing boiling water. The lower pot was heated for 15 minutes to steam the grains above until a single large chunk is formed. The chunk was then taken out, broken into small aggregates and transferred back to the steam for another 15 minutes. It was removed, broken into small aggregates again and then sifted out through sieve (2.5mm). The product was then finally dried and packaged.

3.2.3 Production of sorghum – wheat flour blend couscous

Exactly 2 cups (250g) of both sorghum and wheat flour were measured, mixed very well and used in producing the sorghum – wheat flour couscous blend. All the

procedures for the production are the same as explained above and in the flow chart below;

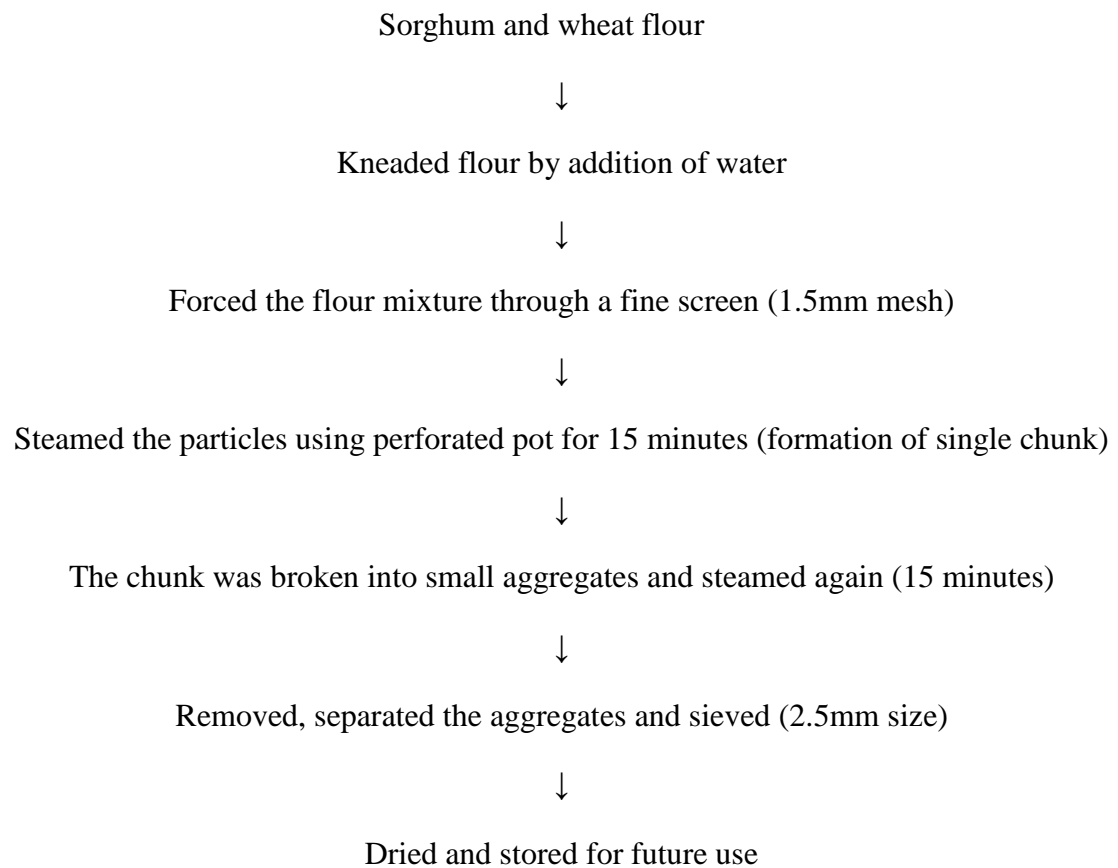


Figure 3.2: Flow chart for couscous production

3.2.4 Proximate analysis of couscous from sorghum and wheat flour

The samples were analyzed for moisture, crude protein, crude fat, ash, crude fibre and available carbohydrate according to the method recommended by Association of Official Analytical Chemists (AOAC).

3.2.4.1 Determination of moisture content

Principle:

The method employed for the determination of moisture content of the sample was based on the measurement of the loss in weight due to drying at a temperature of 105°C as described by AOAC (1985). It also involves the measurement of the weight lost due to the evaporation of water at near boiling point. The proportion of water lost increases as the temperature is raised.

The moisture content of each of the sample (Wheat and sorghum couscous) was determined according to AOAC (1985) method as follows: Five grams (5.0g) of each of the samples was weighed into a tarred clean dried dish and then weighed again. The dish containing the sample was placed in an oven maintaining the heat at 105°C for 2 hours to a constant weight. The sample was then cooled in a desiccator and then reweighed. The difference between the two weights gives the moisture content of the sample.

The percentage moisture content of each sample was calculated using the expression;

$$\text{Moisture(\%)} = \frac{\text{loss of weight on drying}}{\text{Initial weight of the sample}} \times 100$$

3.2.4.2 Determination of ash content

Principle:

When organic substances are burned in the presence of oxygen to CO₂, H₂O and N₂ between 500 and 600°C, the residue is ash which consists of the organic component in

the form of oxides. The ash obtained is not necessarily exactly the same as the mineral matter present in the original sample as there may be losses due to volatilization or some interactions between constituents. The term ash refers to the residue left after the combustion of the oven dried sample. It is a measure of the total mineral content.

The ash content of the sample (couscous) was determined according to the method described by AOAC (1985) as follows: A silica dish (7cm) was ignited, cooled and weighed. Five grams each of the couscous was measured into the silica dish and ignited in the muffle furnace at 500°C for 46 hours. It was then cooled in a desiccator and weighed again to get the total ash.

Percentage ash was calculated using the expression:

$$\text{Ash (\%)} = \frac{\text{Weight of Ash}}{\text{Weight of dry sample}} \times 100$$

3.2.4.3 Determination of crude protein

Principle:

This involves digestion of the organic matter with H₂SO₄ in the presence of catalyst. The digest was reacted with alkali (NaOH) and distilled to liberate ammonia followed by titrating the distillate with acid.

The Kjeldahl method of AOAC (2000) was used for the crude protein determination.

Exactly 2.0g of each sample was weighed into 100ml Kjeldahl flask and a few antibumping granules were added. One gram of the mixed catalyst (CuSO₄ and K₂SO₄ in the ratio 8:1 respectively) and 15ml of concentrated sulphuric acid was added. The flask was placed on a Kjeldahl digestion rack and heated until a clear solution was obtained. At the end of the digestion, the flask was cooled and the sample was quantitatively transferred into a 100ml volumetric flask and made up to the mark with

distilled water. Ten millilitres of the digest was pipetted into a semi micro nitrogen steel tube, 10ml of 40% NaOH solution was then added cautiously. The sample was then steam distilled liberating ammonia into a 100ml conical flask containing 10ml of 4% boric acid using methyl blue indicator until the colour changed from pink to green. Exactly 30ml of sample volume was collected. The content of the conical flask was then titrated with 0.1M HCl. The end point was indicated by a colour change from green to pink and the volume (v) of the acid for each distillate was noted. Percentage nitrogen per sample was calculated using the expression below;

$$\% \text{Nitrogen} = \frac{(M \times 14 \times 100 \times 100)}{(\text{Weight of sample} \times 1000 \times 10)}$$

Where; M = Molarity of HCl

14 = Atomic weight of Nitrogen.

100 = Total volume of digest.

100 = % conversion.

10 = Volume of the digest taken.

1000 = Conversion to litre.

The crude protein was calculated as: % Crude Protein = 6.25 x % nitrogen.

3.2.4.4 Determination of crude fat

Principle;

This was based on continuous extraction of fat content from the sample using petroleum ether in a soxhlet extractor. In this principle, non-polar components of the samples are easily extracted into the organic solvent.

The fat content of each sample was determined by the procedure described by AOAC (2000). A clean dry round bottom flask containing antibumping granules was used. Exactly 210ml of petroleum ether (40 – 60°C) was put in to a flask fitted with soxhlet extraction unit. The weighed sample was transferred into a thimble already fixed into the Soxhlet extraction unit. Cold water was put into circulation. The heating mantle was then switched on and the heating rate adjusted until the solvent is refluxed at a steady rate. Extraction was carried out for 2½ hours.

The sample was removed and dried to a constant weight in an oven. It was then cooled in a dessicator, reweighed and the percentage crude fat content was determined;

$$\text{Crude fat(\%)} = \frac{\text{Weight of lipid extracted}}{\text{Weight of dried sample}} \times 100$$

Where the weight of lipid extracted was the loss in weight of the sample after extraction dried in an oven and cooled in a desiccator.

3.2.4.5 Determination of crude fibre

Principle:

This involves sequential digestion of the sample with dilute acid and alkali solutions. The residue obtained was ignited at 550°C to obtain crude fibre.

Crude fibre was determined by the method of AOAC (2000). Two grams of the ground sample was placed in a round bottom flask. 100ml of 0.25M H₂SO₄ was added and mixture was boiled under reflux for 30 minutes. The insoluble matter was washed several times with hot water until it was acid free (C₁). It was then transferred into a flask containing 100ml of 0.25M NaOH solution. The mixture was boiled again under reflux for 30 minutes and filtered under suction, then washed with hot water until it is alkaline free (C₂). It was then dried and ashed in a furnace at 550°C for 2 hours. The

furnace was then switched off and allowed to cool down. The sample was removed and cooled in a desiccator and weighed (C_3). The crude fibre content was calculated as loss of weight in ashing. Weight of original sample was used as W .

$$\text{Crude Fibre(\%)} = \frac{(C_2 - C_3)}{W} \times 100$$

3.2.4.6 Determination of total carbohydrate

Percentage carbohydrate was obtained by the method adopted by Ihekoronye and Ngoddy (1985) and it was carried out as follows: The sum of the measured moisture content (%MC), Ash (%A), Crude protein (%P), Crude fat (% F), Fibre (%F) was subtracted from the initial weight of the couscous sample (100%).

3.2.5 Determination of dietary fibre

Principle:

This involves sequential enzymatic digestion of the sample by heat-stable – α -amylase, protease and amyloglucosidase according to AOAC, (1992).

Exactly 1.0g of the sample was weighed inside a 400ml beaker; 40ml of the buffer solution pH 6.9 was added and stirred with a magnetic stirrer to prevent lump formation. The solution was then incubated with 50 μ l of heat stable α - amylase solution while stirring at low speed and incubated in water bath at 95-100°C for 35min with continuous stirring. After then, the solution was allowed to cool before adding 100ml protease solution and incubated with shaking in a water bath at 60°C for 30min. The mixture was further incubated with 200ml amyloglucosidase solution, while stirring and also incubated at the same temperature of 60°C for 30min with constant agitation.

The solution was then filtered, the residue was washed twice with 10ml of distilled water, 95% ethanol and acetone, and then dried in the crucible for protein/ash

determination for insoluble fibre analysis. The filtrate preserved was used for soluble fibre analysis, by precipitation using 4 volume of 95% extract preheated to 60°C and allowed at room temperature for 60 min. The precipitate was filtered. The residue was washed successively with two volumes of 15ml of 78% ethanol, 95% ethanol, and acetone. The residue was later dried, weighed, and then ashed and protein content was determined.

$$\text{Dietary fibre(\%)} = \frac{(\text{Weight of residue} - \text{Protein} - \text{ash} - \text{blank})}{\text{Weight of sample}} \times 100$$

3.2.6 Determination of amino acid profile of sorghum and wheat couscous

The Amino Acid profile of the known samples was determined using the method described by Benitez (1989). A known weight of the sample was dried to constant weight, defatted, hydrolyzed, evaporated in a rotary evaporator and loaded into Applied Biosystems Phenyl-thiohydantoin (PTH) model 120A (Applied Biosystem, Inc. Foster city, CA USA) Amino Acid Analyzer.

i. Defatting Sample:

The sample was defatted using chloroform/methanol mixture of 2:1 ratio. Exactly 4.0g of the sample was put in extraction thimble and extracted for 15 hours in soxhlet extraction apparatus (AOAC, 2006).

ii. Nitrogen determination:

A small amount (200mg) of ground sample was weighed, wrapped in Whatman No.1 filter paper and put in the Kjeldahl digestion flask. Concentrated sulphuric acid (10ml) was added. Catalyst mixture (0.5g) containing sodium sulphate (Na_2SO_4), copper sulphate (CuSO_4) and selenium oxide (SeO_2) in the ratio of 10:5:1 was added into the flask to facilitate digestion. Four pieces of anti-bumping granules were added.

The flask was then put in Kjeldahl digestion apparatus for 3 hours until the liquid turned light green. The digested sample was cooled and diluted with distilled water to 100ml in standard volumetric flask. Aliquot (10ml) of the diluted solution with 10ml of 45% sodium hydroxide was put into the Markham distillation apparatus and distilled into 10ml of 2% boric acid containing 4 drops of bromocresol green/methyl red indicator until about 70ml of distillate was collected. The distillate was then titrated with standard solution of 0.01 N hydrochloric acid to grey coloured end point.

$$\text{Percentage Nitrogen} = \frac{[(a - b) \times 0.01 \times 14 \times V \times 100]}{W \times C}$$

Where:

- a = Titre value of the digest
- b = Titre value of the blank sample
- V = Volume after dilution (100ml)
- W = Weight of the dried sample (mg)
- C = Aliquot of the sample used (10ml)
- 14 = Nitrogen constant in mg

iii. Hydrolysis of the sample

A known weight of the defatted sample was weighed into glass ampoule. Seven (7) ml of 6N HCl was added and oxygen was expelled by passing nitrogen into the ampoule (this is to avoid possible oxidation of some amino acids during hydrolysis e.g. methionine and cysteine). The glass ampoule was then sealed with Bunsen burner flame and put in an oven pre-set at $105^{\circ}\text{C} \pm 5^{\circ}\text{C}$ for 22 hours. The ampoule was allowed to cool before broken open at the tip and the content was filtered to remove the humins. However, tryptophan decomposes during acid hydrolysis, as such alkaline hydrolysis was adopted by using sodium hydroxide (NaOH) instead of barium

hydroxide to prevent problems with both precipitation and adsorption of tryptophan. In this study, the tryptophan in the known sample was hydrolyzed with 4.2 M Sodium hydroxide (Robel, 1984).

The filtrate was then evaporated to dryness using rotary evaporator. The residue was dissolved with 5ml to acetate buffer (pH 2.0) and stored in plastic specimen bottles, which were kept in the freezer.

iv. Loading of the Hydrolysate into TSM analyzer

The amount loaded was 60 microlitre; this was dispensed into the cartridge of the analyzer. The analyzer is designed to separate and analyze free acidic, neutral and basic amino acids of the hydrolysate.

v. Method of Calculating Amino Acid Values from the Chromatogram Peaks.

An integrator attached to the Analyzer calculates the peak area proportional to the concentration of each of the amino acids. Alternatively, the net height of each peak produced by the chart recorder of Phenylthiohydantoin (each representing an amino) was measured. The half-height of the peak on the chart was found and width of the peak on the half height was accurately measured and recorded. Approximate area of each peak was obtained by multiplying the height with the width at half-height.

The norleucine equivalent (NE) for each amino acid in the standard mixture was calculated using the formula:

$$NE = \text{Area of Norleucine} \frac{\text{Peak}}{\text{Area}} \text{ of each } \textit{amino acid}$$

A constant was calculated for each amino acid in the standard mixture:

Where $S_{std} = NE_{std} \times \text{Molecular weight} \times \mu MAA_{std}$

Finally, the amount of each amino acid present in the sample was calculated in g/100g protein using the following formula:

$$\text{Concentration (g/100g protein)} = \text{NH} \times \text{W at NH/2} \times \text{S}_{\text{std}} \times \text{C}$$

$$\text{C} = ((\text{Dilution} \times 16)/(\text{Sample weight (g)} \times 10 \times \text{Volume loaded})) \div (\text{NH} \times \text{W(nleu)})$$

Where: NH = Net height

 W = Width at half height

 nleu = Norleucine

3.2.6.1 Determination of tryptophan

The tryptophan in the known sample was hydrolyzed with 4.2M sodium hydroxide (Robel, 1984). A known weight of the sample was dried to constant weight, defatted, hydrolyzed, evaporated in a rotary evaporator and loaded into the Applied Biosystems PTH Amino Acid Analyzer. All other procedures are the same as above except for hydrolysis.

Hydrolysis of the sample for tryptophan determination

A known weight of the defatted sample was weighed into the glass ampoule. Then 10ml of 4.2M NaOH was added and oxygen was expelled by passing nitrogen into the ampoule. The glass ampoule was then sealed with Bunsen burner flame and put in an oven pre-set at $105^{\circ}\text{C} \pm 5^{\circ}\text{C}$ for 22 hours. The ampoule was allowed to cool before been broken open at the tip and the content filtered. The filtrate was neutralized with 6 N HCl and evaporated to dryness at 40°C under vacuum in a rotary evaporator. The residue was dissolved with 5ml of acetate buffer (pH 7.0) and stored in plastic specimen bottles, which were kept in the freezer.

3.2.7 Determination of antinutritional factors

3.2.7.1 Determination of phytic acid

Principle:

When phytate react with hydrochloric acid in the presence of chloride ion and ammonium thiocyanide, the chloride ion bind to the phosphate ester, making it unavailable to react with sulfosalicylic acid resulting in a brownish colour formation.

The phytic acid was determined using the procedure described by Lucas and Markakas (1975). Exactly 2.0g of each couscous powder was weighed into 250ml conical flask; 100ml of 2 % HCl was added to each couscous sample in the conical flask and allowed to stand for 3hours. it was then filtered through a double layer of harden filter paper, 50ml of each filtrate in 250ml beaker and 107ml of distilled water were added in each. Ten milliliters of 0.3% ammonium thiocyanide solution was added into each and titrated with standard iron (III) chloride solution which contains 1.95mg of iron/ml. The end point was brownish colour persisting for 5minutes. The percentage of phytic acid was calculated using the formula:

$$\% \text{ phytic acid} = (Y \times 1.19 \times 100)/(\text{couscous weight})$$

Where;

Y = titre value x 1.95mg.

3.2.7.2 Determination of oxalates

Principle:

Oxalate react with ammonia and potassium permanganate at 100°C in the presence of hydrochloric acid and methyl red indicator resulting in a faint yellow colouration which later turn to pink.

Oxalate was determined using method described by Oke (1969). The total oxalic acid of the powdered couscous sample was determined by weighing 2g of the couscous sample into 250ml of conical flask. 190ml of distilled water and 10ml of 6M HCl acid were then added. The mixture was incubated for 1hour on a boiling water bath, cooled, transferred into a 250ml volumetric flask, diluted with distilled water to volume and filtered. Four drops of methyl red indicator was added, followed by concentrated ammonia till the solution turned faint yellow. It was then heated to 100°C, allowed to cool and filtered to remove precipitate containing ferrous irons. The filtrate then boiled and 10ml of 5% CaCl₂ added with a constant stirring. It was then being allowed to stand overnight.

The mixture was filtered through Whatman No. 4 filter paper. The precipitate was then washed several times with distilled water and transferred to a beaker and 5ml of 25% sulphuric acid was added to dissolve the precipitate. The resultant solution was maintained at 80°C and titrated against 0.5% potassium permanganate until the pink colour persists for approximately one minute.

A blank was run. From the amount of potassium permanganate to be used, the oxalate content of the unknown couscous sample was calculated using the equation below:

1ml potassium permanganate = 2.24mg oxalate.

3.2.7.3 Determination of tannins

Tannin was estimated according to the method described by (Makkar *et al.*, 1993).

Principle:

The tannin and tanninlike compounds reduce phosphomolybdic acid in alkaline solution to produce a highly coloured blue solution, the intensity of which is proportional to the

amount of tannin and can be estimated against standard tannic acid solution at wavelength of 725nm.

Couscous sample Preparation and Extraction of Tannin

Tannin extraction was done using 400mg of ground couscous sample in a conical flask with 40ml of diethyl ether containing 1% acetic acid (v/v) to remove the pigment material. The supernatant was carefully discarded after 5minutes and 20ml of 70% aqueous acetone was added and the flask sealed with cotton plug covered with aluminium foil and kept in an electrical shaker for 2hours for extraction. It was then filtered through Whatmann No. 1 filter paper and the sample was kept in a refrigerator at 4°C until analysis.

Reagents

Folin Ciocalteu reagent (1N) commercially available Folin Ciocalteu reagent (2N) was diluted with equal volume of distilled water, sodium carbonate and standard tannic acid solution (0.5mg/ml).

Standard Calibration Curve Preparation

From the stock solution of tannic acid (0.5mg/ml) 0, 10, 20, 30, 40 and 50µL each was put in a test tube and their volumes made up to 1ml with distilled water. After which 0.5ml of folin reagent and 2.5ml of 1 N sodium carbonate was added and the whole content mixed properly and after 40minutes, absorbance reading was taken at 725nm in a spectrophotometer.

Method

50µL of tannin extracts for each of the couscous sample was taken in a test tube and the volume was made up to 1ml with distilled water. Then 0.5ml of Folin Ciocalteu reagent was added and mixed properly. Then 2.5ml of 20% sodium carbonate solution was

added, mixed and kept for 40 minutes at room temperature. Optical density was then taken at 725nm on the spectrophotometer and the concentrations was obtained from the standard curve. Percentage concentration of tannin was determined using the expression:

$$\% \text{ Tannin (mg/100g)} = (A_n \times C \times D_f) / (A_s \times W) \times 100$$

Where;

A_n = Absorbance of test couscous sample

A_s = Absorbance of standard tannic acid

C = Concentration of standard tannic acid (mg/ml)

D_f = Dilution factor V_{ex}/V_a

W = Weight of test couscous sample (mg)

V_{ex} = Volume of extract

V_a = Volume of extract analysed

3.2.7.4 Quantitative determination of saponin

Principle:

The gravimetric method of saponin determination is based on double extraction of saponin using organic solvent which can be quantified in the presence of a basic medium.

The saponin content of the couscous was determined by double extraction gravimetric method described by Harborne (1973). A known weight (5.0g) of the powdered couscous sample was mixed with 50 ml of 20% aqueous ethanol solution in a flask. The

mixture was heated with periodic agitation in water bath for 90 minutes at 55°C; it was then filtered through Whatman filter paper (No. 42). The residue was extracted with 50 ml of 20% ethanol and both extract were poured together and the combined extract was reduced to about 40 ml at 90°C and transferred to a separating funnel where 40 ml of diethyl ether was added and shaken vigorously. Re extraction by partitioning was done repeatedly until the aqueous layer become clear in colour. The saponins were extracted, with 60 ml of normal butanol. The combined extracts were washed with 5% aqueous sodium chloride (NaCl) solution and evaporated to dryness in a pre-weighed evaporation dish. This process was repeated two more times to get an average. Saponin content was determined by difference and calculated as a percentage of the original sample thus:

$$\text{Percentage (\%) Saponin} = \frac{W_2 - W_1}{\text{Weight of couscous}} \times \frac{100}{1}$$

Where;

W_1 = Weight of evaporating dish

W_2 = Weight of evaporating dish + sample

3.2.7.5 Determination of Cyanogenic glycoside

Principle:

Free cyanide is completely ionized in alkaline solution to form a stable metal – cyanide complexes which is indicated by faint permanent turbidity.

Cyanide contents of the samples were measured by alkaline titration method as described by (AOAC, 1990). Exactly 15 g of the samples was measured into 800ml Kjeldahl flask containing 200ml of distilled water and allowed to stand for 3 hours at

25°C. Autolysis was carried out with the apparatus connected to a distiller. A 150ml of distillate was collected in 20ml, 25% of NaOH solution and further diluted to 250ml with distilled water. Next, 100ml of the diluted distillate was mixed with 8.0ml of 6.0N NH_4OH and 2.0ml of 5% KI indicator solution and titrated against 0.02N AgNO_3 . The end point was indicated by a faint permanent turbidity appearance. The cyanide content mg/100g wet weight of the sample was evaluated from the expression; 1ml 0.02N $\text{AgNO}_3 = 1.08\text{mg HCN}$.

3.2.7.6 Alkaloids determination

Principle:

Alkaloids react with hydrogen ions under acidic conditions in the presence of organic solvent to form a coloured complex.

The alkaloids content of the sample was determined using gravimetric method as described by Harbone (1973). Exactly 5.0 g of each of the sample was weighed using a weighing balance and dispersed into 50 ml of 10% acetic acid solution in ethanol. The mixture was well shaken and allowed to stand for about 4 hours before it was filtered. The filtered was then evaporated to one quarter of its original volume on hot plate. Concentrated ammonium hydroxide was added drop wise to precipitate the alkaloids. A pre-weight filter paper was used to filter off the precipitate and it was then washed with 1% ammonium hydroxide solution. The filter paper containing the precipitate was dried on an oven at 60°C for 30 min, transferred into desiccators to cool and then reweighed until a constant weight was obtained. The constant weight was recorded. The weight of the alkaloids was determined by weight difference of the filter paper and expressed as a percentage of the sample weight analyzed.

$$\% \text{ Alkaloids} = \frac{w_2 - w_1}{W} \times 100$$

Where;

w = weight of sample

w_1 = weight of empty filter paper

w_2 = weight of paper plus precipitate

3.2.8 Mineral analysis

Zinc (Zn), Iron (Fe), Calcium (Ca), and Magnesium (Mg) were determined by Atomic Absorption Spectrometry (AAS); while Sodium (Na) and Potassium (K) were determined by flame photometry according to the method of AOAC (2003).

Principle:

Atomic absorption spectroscopy is based on the ability of an excited atom of an element to absorb energy from wavelengths of the same frequency as the element. This creates a decrease in the initial signal energy and this difference is proportional to the concentration of that element.

3.2.8.1 Wet digestion of couscous sample

For wet digestion of couscous sample, exactly 1.0g of the powdered couscous was taken into digesting glass tube. Twelve milliliters (12ml) of 1 N HNO_3 was added to the couscous samples and mixtures was kept overnight at room temperature. Then 4.0ml 2 N perchloric acid (HClO_4) was added to this mixture and was kept in the fume hood for digestion. The temperature was increased gradually starting from 50°C and increasing up to $250 - 300^\circ\text{C}$. The digestion was completed in about 70- 85 minutes as indicated by the appearance of white fumes. The mixture was left to cool down and the content of

the tubes was transferred to 100ml volumetric flasks and the volumes of the contents were made up to 100ml with distilled water. The wet digested solution was transferred to plastic bottles labelled accurately and stored for mineral determination.

3.2.8.2 Procedure for analysis of mineral contents

Digested couscous samples were analyzed for mineral contents. The absorption measurement of the elements for the samples was read out. A different electrode lamp was used for each mineral. The equipment was run for standard solutions of each of the mineral, before and during determination to check that it is working properly. The dilution factor for all minerals was 100, further dilution of the original solution was done using 0.5ml original solution and enough distilled water was added to it to make the volume up to 100ml. Also the determination of calcium (Ca), 1.0ml lithium oxide solution was added to the original solution to unmask Ca from Mg. The concentration of minerals was determined using the formula below:

$$M_w = \text{absorbency (ppm)} \times \text{dry wt.} \times D / \text{wt. of couscous sample} \times 100$$

Where M_w = Conc. of minerals

D = Dilution factor

Determination of Sodium (Na) and Potassium (K) was done by the method of flame photometry. The same wet digested couscous sample solutions used in AAS was used for the determination of Na and K. Standard solutions of 20, 40, 60, 80 and 100milli equivalent/L was used both for Na and K. The calculations for the total mineral intake involved the same procedure for determination of Zn, Fe and Ca using AAS as shown above.

3.2.9 Sensory evaluation

3.2.9.1 Preparation of couscous

Measurements:

- | | | |
|------|--------------------|-----------------|
| i. | Precooked couscous | 500g |
| ii. | Water | 750ml |
| iii. | Vegetable Oil | 1 serving spoon |
| iv. | Tomatoes | 100g |
| v. | Onions | 100g |
| vi. | Iodized Salt | to taste |

Number of servings: 5 (1 serving of a small couscous = 100g)

Preparation Time: 22 minutes

Based on the measurements above, the precooked couscous was prepared with a sauce and presented to a panel of 15 judges who were asked to score the three produced couscous (100% SF, 100 % WF and 50%/50% WSF) samples for aroma, texture, colour, taste and overall acceptability using 9 points hedonic scale where analyzed (9) represent extremely like and (1) represent extremely dislike. The results were analyzed using Analysis of variance (ANOVA).

3.3 Statistical analysis

Data analysis was carried out using statistical package for social science (SPSS version 20). All values were expressed as mean \pm SD of triplicate determinations. Values with P value ≤ 0.05 were considered significant. Independent sample T-test was used for the statistical analysis to compare the results for the two samples (Wheat and sorghum couscous) results. One way Analysis of variance (ANOVA) was used to compare the means of the sensory evaluation results.

CHAPTER FOUR

4.0

RESULTS

4.1 Proximate Composition of the locally prepared couscous from sorghum (*S. bicolor*) and wheat (*T. durum*) flour

The proximate composition of sorghum and wheat couscous is presented in Table 4.1. The result shows that there is no significant difference ($p>0.05$) between sorghum ($8.18\pm0.46\%$) and wheat ($8.19\pm0.18\%$) couscous in protein contents, but there is a significant ($p<0.05$) difference between sorghum ($3.91\pm0.13\%$) and wheat ($2.42\pm0.11\%$) couscous in crude fibre content, sorghum ($3.40\pm0.15\%$) and wheat ($2.47\pm0.23\%$) couscous in lipid contents, sorghum ($1.40\pm0.06\%$) and wheat ($1.81\pm0.06\%$) couscous in ash contents, sorghum ($74.07\pm1.32\%$) and wheat ($78.44\pm0.01\%$) couscous in carbohydrate contents.

4.2 Dietary fibre Content of the locally prepared couscous from sorghum (*S. bicolor*) and wheat (*T. durum*) flour

Results of dietary fibre revealed a significant ($p<0.05$) difference in soluble, insoluble and total dietary fibre contents of sorghum and wheat flour couscous with values of soluble fibre; sorghum couscous ($1.51\pm0.02\%$), wheat couscous ($2.30\pm0.03\%$), insoluble fibre; sorghum couscous ($8.11\pm0.11\%$), wheat couscous ($10.55\pm0.15\%$) and total dietary fibre; sorghum couscous ($9.61\pm0.21\%$) and wheat couscous ($12.85\pm0.12\%$) as shown in Table 4.2.

Table 4.1: Proximate Composition of the Locally Prepared Couscous from Sorghum (*S. bicolor*) and wheat (*T. durum*) Flour

Parameters	Couscous	
	Sorghum (%)	Wheat (%)
Moisture	9.06±0.64 ^b	6.68±0.23 ^a
Crude protein	8.18±0.46 ^a	8.19±0.18 ^a
Crude fibre	3.91±0.13 ^b	2.42±0.11 ^a
Crude fat	3.40±0.15 ^b	2.47±0.23 ^a
Ash	1.40±0.06 ^a	1.81±0.06 ^b
Total Carbohydrate	74.07±1.32 ^a	78.44±0.91 ^b

Values are mean ± standard deviation of triplicate determination. Values with different superscripts across the row are significantly different (p<0.05).

Table 4.2 Dietary Fibre Content of the locally prepared couscous from Sorghum (*S. bicolor*) and Wheat (*T. durum*) flour

Dietary fibre	Couscous	
	Sorghum(%)	Wheat(%)
Soluble	1.50± 0.02 ^a	2.30± 0.03 ^b
Insoluble	8.11 ± 0.11 ^a	10.55 ± 0.15 ^b
Total	9.61±0.21 ^a	12.85±0.12 ^b

Values are mean ± standard deviation of triplicate determination. Values with different superscripts across row are significantly different (p<0.05).

4.3 Amino Acid Composition of Locally prepared Couscous from Sorghum (*S. bicolor*) and Wheat (*T. durum*) Flour

The amino acid profile results of couscous from both grains are presented in Table 4.3.

The results showed that wheat couscous has significantly ($p < 0.05$) higher content of some of the amino acids (lysine, valine, arginine, glutamate and glycine) when compared to sorghum couscous, while sorghum couscous has significantly higher content of the amino acids (leucine, isoleucine, proline, tyrosine, cysteine, alanine and aspartate) compared to wheat couscous. There is no significant difference between sorghum and wheat couscous in the contents of the amino acids; phenylalanine, tryptophan, methionine, histidine, threonine and serine.

4.4 Antinutrient Content of the locally prepared couscous from Sorghum (*S. bicolor*) and Wheat (*T. durum*) Flour

The result of antinutrients composition of couscous from both grains is presented in Table 4.4. The result showed that wheat couscous has significantly ($p < 0.05$) higher content of Phytic acid, oxalate and saponin and alkaloids when compared to that obtained from sorghum couscous, whereas, higher content of tannin and cyanide was found in sorghum couscous when compared to that obtained from wheat couscous.

Table 4.3 Amino Acid Composition of Locally Prepared Couscous Sorghum (*S. bicolor*) and Wheat (*T. durum*) Flour (g/100g protein)

Amino acid	Couscous	
	Sorghum	Wheat
Leucine (Leu)	10.25±0.95 ^b	6.24±0.11 ^a
Lysine (Lys)	2.52±0.18 ^a	3.39±0.31 ^b
Isoleucine (Ile)	4.21±0.06 ^b	3.68±0.25 ^a
Phenylalanine (Phe)	5.01±0.07 ^a	4.49±0.57 ^a
Tryptophan (Trp)	1.23±0.03 ^a	1.52±0.33 ^a
Valine (Val)	4.21±0.04 ^a	4.90±0.15 ^b
Methionine (Met)	2.05±0.06 ^a	1.97±0.05 ^a
Arginine (Arg)	3.89±0.16 ^a	5.44±0.17 ^b
Histidine (His)	2.34±0.14 ^a	2.48±0.11 ^a
Threonine (Thr)	3.38±0.18 ^a	3.29±0.06 ^a
Proline (Pro)	9.63±0.31 ^b	7.48±0.53 ^a
Tyrosine (Tyr)	3.91±0.08 ^b	2.81±0.11 ^a
Cysteine (Cys)	1.97±0.11 ^b	1.32±0.07 ^a
Alanine (Ala)	9.08±0.07 ^b	4.34±0.28 ^a
Glutamate (Glu)	19.94±0.16 ^a	24.72±0.69 ^b
Glycine (Gly)	3.45±0.23 ^a	4.52±0.13 ^b
Serine (Ser)	4.45±0.35 ^a	4.46±0.19 ^a
Aspartate (Asp)	7.50±0.35 ^b	6.35±0.21 ^a

Values are mean ± standard deviation of triplicate determination. Values with different superscripts across row are significantly different.

Table 4.4 Antinutrient Content of Locally Prepared Couscous from Sorghum (*S. bicolor*) and Wheat (*T. durum*) flour

Antinutrients	Couscous		Permissible limits (AOAC, 2000)
	Sorghum	Wheat	
Phytic acid (mg/g)	3.48±0.07 ^a	4.65±0.09 ^b	250 – 500mg
Oxalate (mg/kg)	0.43±0.01 ^a	0.55±0.07 ^b	3–5 mg/kg
Tannin (mg/g)	0.18±0.02 ^b	0.13±0.002 ^a	20 mg/g
Saponin (%)	1.55±0.07 ^a	1.98±0.04 ^b	2.3%
Alkaloids (%)	1.55±0.05 ^b	1.38±0.03 ^a	5%
Cyanide (mg/kg)	8.6±0.01 ^b	6.6±0.001 ^a	5 - 6 mg/kg

Values are mean ± standard deviation of triplicate determination. Values with different superscripts across row are significantly different.

4.5 Mineral Composition of the locally prepared couscous from Sorghum (*S. bicolor*) and Wheat (*T. durum*)

The result for mineral analysis of couscous from both grains is presented in Table 4.5.

The result showed that wheat couscous contains significantly ($p<0.05$) higher amount of Na, K and Fe when compared to that obtained from sorghum couscous, while sorghum couscous contains significantly ($p<0.05$) higher amount of calcium and zinc (Ca and Zn) when compared to that obtained from wheat couscous.

4.6 Sensory and Organoleptic Evaluation of Locally prepared Couscous from Sorghum (*S. bicolor*) and Wheat (*T. durum*) flour

The result of sensory and organoleptic evaluation of the different couscous from both grains is presented in Table 4.6. The result showed that 100% sorghum flour couscous significantly ($p<0.05$) differ in texture when compared to the remaining couscous whereas, couscous produced from 100% wheat flour has significantly ($p<0.05$) higher preference in colour and aroma when compared to sorghum flour couscous. Couscous produced from 50% wheat flour/50% sorghum flour has higher preference in terms of taste and overall acceptability when compared to 100% sorghum flour and 100% wheat flour couscous.

Table 4.5 Mineral Composition of Locally Prepared Couscous from Sorghum (*S. bicolor*) and Wheat (*T. durum*) flour

Element (mg/Kg)	Couscous	
	Sorghum	Wheat
Na	0.9489±0.0008 ^a	1.0286±0.0007 ^b
K	0.0460±0.0011 ^a	0.8782±0.0014 ^b
Ca	0.0632±0.0008 ^b	0.0456±0.0007 ^a
Fe	0.2940±0.0071 ^a	0.5353±0.0007 ^b
Zn	0.0158±0.0072 ^b	0.0017±0.0001 ^a

Values are mean ± standard deviation of triplicate determination. Values with different superscripts across row are significantly different (p<0.05).

Table 4.6 Sensory and Organoleptic Evaluation of Couscous from Sorghum (*S. bicolor*) and Wheat (*T. durum*)

Quality attributes	Products (couscous)		
	SFC	WFC	SWFC
Colour	5.40±0.99 ^a	7.53±0.92 ^b	5.93±1.87 ^{ab}
Texture	7.67±0.49 ^b	6.33±1.35 ^a	6.73±0.22 ^a
Aroma	6.93±0.88 ^a	7.60±0.74 ^b	7.73±1.03 ^b
Taste	7.27±0.80 ^a	6.97±0.70 ^a	8.13±0.74 ^b
Overall acceptability	5.80±1.82 ^a	5.93±2.02 ^a	6.53±1.69 ^b

Values are mean ± standard deviation of triplicate determination. Values with different superscripts across row are significantly different(p<0.05).

SFC = 100 % Sorghum Flour Couscous

WFC = 100% Wheat Flour Couscous(Normal control)

SWFC= 50% Sorghum Flour and 50% Wheat Flour Couscous

CHAPTER FIVE

5.0 DISCUSSION

The result obtained from proximate composition shows that sorghum and wheat couscous have significant difference in almost all the parameters analyzed with the exception of protein contents. The crude protein, moisture contents, crude fat and ash contents for sorghum couscous were within the range reported by Abdullahi,(2017). The result of this research shows that sorghum couscous (3.91 ± 0.13 %) has higher crude fibre content as against wheat couscous (2.42 ± 0.11 %) which is similar to the study reported by Oppong *et al.*(2015) who reported that some level of crude fibre is lost during wheat refining process. Carbohydrate contents in both sorghum and wheat couscous which is 74.05% and 78.44% respectively were slightly above the research findings reported by Adeyeye and Adewale (1992) which can be due to the effect of processing methods. The higher crude fibre content of the sorghum couscous could also be of an advantage, since crude fibre in diet is known to enhance the digestibility, decrease cholesterol and reduce the risk of large bowel cancers. This finding is supported by the work of Anderson *et al.* (1995), who reported that crude fibre in diet improved digestibility and reduce the risk of chronic diseases. The higher moisture content of food serves as a measure of stability and susceptibility to endogenous and/or exogenous microbial and enzymatic activities. Thus longer shelf life could be linked to lower moisture content, this finding is supported by the work of Muhammad *et al.* (2018), who said higher moisture content of foods enhance microbial and enzymatic activities.

The total dietaryfibre (soluble and insoluble) result for wheat couscous (12.85 ± 0.18 %) is higher than that of sorghum couscous (9.61 ± 0.21 %). This research finding is lower than what was reported by Rakha *et al.*(2010) and Anderson *et al.*(2009)who studied

dietary fibre contents of cereals whole grain and benefits of dietary fibre. This shows that milling and other processing methods greatly reduce dietary fibre contents in cereals.

Dietary fibre is a measure of the quantity of indigestible cellulose, pentosans, lignin, pectin, mucilage and other components of this type present in food where as crude fibre refers to one type of dietary fibre primarily cellulose and lignin that remains as a residue after food receives a standardized laboratory treatment with dilute acid and alkali which dissolve all the soluble and some of the insoluble fibre. Crude fibre is less fermentable and may only have some benefits in peristaltic movement while dietary fibre is highly fermentable and may positively affect bowel movement and metabolism like lipid profile and glycemic control. Soluble fibre which dissolves in water forms a gel-like material that help lower blood cholesterol and glucose levels while insoluble fibre prevents the fermentation of foods in alimentary canal which subsequently may lead to colon cancer (Mayo, 2015).

The presence of eight essential amino acids (Leucine, Isoleucine, Lysine, Phenylalanine, Tryptophan, Valine, Methionine and Thereonine), two semi essential amino acids (Arginine and Histidine) and eight non-essential amino acids (Proline, Tyrosine, Cysteine, Alanine, Glutamate, Glycine, Serine and Aspartate) were observed in both sorghum and wheat flour couscous. However, the two non-essential amino acids (Asparagine and Glutamine) were absent, which may be due to conversion of amide group of these two amino acids to ammonia during hydrolysis, which tends to give high peak and overall conversion of the amide group of these amino acids to Aspartate and Glutamate. This is in agreement with the report of Muhammad *et al.* (2018). The higher percentage composition of leucine among the essential amino acids in both sorghum and wheat flour couscous respectively showed that leucine is an abundant essential

amino acid in plant products. The highest percentage composition of leucine in sorghum and wheat flour couscous was in agreement with the observations made by Aremu *et al.*(2010), who reported that leucine is the most concentrated essential amino acid in Nigerian plant products.

Sorghum couscous has higher concentrations (mg/100g crude protein) of Leucine and Isoleucine, among the essential amino acids, whereas higher concentration (mg/100g crude protein) of lysine and valine was found in wheat couscous). The major abundant non-essential amino acids found in both sorghum and wheat couscous were proline, glutamic acid + glutamine and aspartic acid + asparagines. Welch, (2005), reported higher content of the non-essential aspartic and glutamic in sorghum, wheat and other cereals such as rice oats and barley. Asparagine is important for proper functioning, maintenance and chemical balance in various tissues including the human brain this is supported by the report of Muhammad *et al.*(2018) that some amino acids are essential in proper functioning of human tissues. The presence of reasonable amount of alanine in sorghum couscous (9.08 ± 0.07 %) as against wheat couscous (4.34 ± 0.28 %) is very essential since alanine has an important metabolic function as reported by Gropper and Smith, (2013) that amino acid such as alanine helps to metabolize blood glucose and plays a role in muscle health. Also, according to Muhammad *et al.*(2018), Glutamate plays an important role as a component of glutathione and a precursor of γ -amino butyric acid (GABA), which is a neurotransmitter.

The nutritional quality of food protein depends on the types and amounts of amino acids it contains, and represents a measure of the efficiency with which the body can utilize the protein (Cheftel *et al.*, 1985). Thus a balanced or high quality protein is the one that contains essential amino acids in a ratio that commensurate with human needs. This was determined by comparing the amino acid content of the sorghum (*S. bicolor*) and wheat

(*T. durum*) couscous with FAO/WHO/UNU maintenance amino acid reference pattern. Of particular interest is the high contents of the essential amino acids which satisfy the values recommended by FAO/WHO/UNU (2007) with the exception of lysine, which is the limiting essential amino acid, having the lowest chemical score in both the sorghum and wheat couscous.

There were higher level of tannin (mg/g) and cyanide (mg/kg) in sorghum flour couscous compared to wheat flour couscous, whereas high level of phytatic acid (mg/g), oxalate (mg/kg) and saponin (%) were found in wheat couscous compared to sorghum couscous. The phytic acid contents for both sorghum and wheat couscous is similar to the research finding reported by Garcia *et al.* (1990). The tannin contents of the two couscous samples were slightly lower than that reported by Rao and Prabhavathi (1982) on tannin contents of cereal foods consumed in India. The Oxalate, Saponin, alkaloids and cyanide contents of the two couscous products were lower than what was reported by Salvan, (2004) on whole cereal grain and human health. The lower contents of these antinutrients can therefore be due to the effect of milling and steaming.

Higher concentration of calcium and zinc is found in sorghum couscous compared to wheat couscous. The result for Zinc, Calcium and Iron contents in both sorghum (0.0158mg/kg, 0.0632mg/kg, 0.2940mg/kg) respectively and wheat (0.0017mg/kg, 0.0456mg/kg, 0.5353mg/Kg) respectively couscous in this study are within the range reported by Gibson *et al.* (2010). Significant amount of sodium (1.0286mg/Kg), potassium (0.8782mg/Kg) and iron (0.5353mg/Kg) were found in wheat couscous compared to sorghum (Na 0.9489mg/Kg, K 0.0460mg/Kg and Fe 0.2940mg/Kg) couscous and these are slightly below what was reported by Abdulrahman and Omoniyi, (2016) who studied mineral compositions of different cereals variables in Gwagwalada market, Abuja. Sorghum couscous (*S. bicolor*) having higher amount of

calcium can be good for complementary feeding of children 12 to 24 months since children need to get enough calcium for their bones and teeth to develop normally. Calcium also combines with phosphorous to form bones and teeth making them hard and resistant to breaks and decay. Also getting enough calcium early in life help bones remain strong later in life (WHO, 2019). Higher amount of Zinc in sorghum couscous is also very important since zinc is an essential nutrient for all forms of life and its importance lies in the fact that many body functions are linked to zinc containing enzymes, also low levels of zinc in the body have been linked to the prevalence of diarrhoea in children (Fleet, 2006).

Parameters assessed during the sensory and organoleptic evaluation are; colour, texture, aroma, taste and overall acceptance. Sorghum flour (100% SFC) couscous (7.67 ± 0.49) has highest preference in terms of texture as against 100% WFC (6.33 ± 1.35) and 50%SWFC (6.73 ± 0.22). Samuel and Olalekan, (2016) also reported highest preference in terms of texture for cookies made from sorghum flour as against those made from wheat flour or composite flour from sorghum and wheat. The sensory evaluation result in this study showed that 100% WFC has highest preference for aroma (7.60 ± 0.74) and colour (7.53 ± 0.92) than 100% SFC. This is also similar to the research findings by Samuel and Olalekan (2016) which showed that 100%WF cookies have higher preference for aroma and colour than those produced from 100%SF. Couscous produced from 50% sorghum/wheat composite flour has the highest preference in terms of taste and overall acceptability than 100%SF and 100%WF. Adebowale *et al.* (2012) reported highest preference for taste in wheat/sorghum composite flour biscuit at the ratio of 75:25 however, the research findings by Gaines, (1994); Samuel and Olalekan, (2016) showed that 100%WF cookies and other baked products are more preferred in

terms of overall acceptance than those produced from 100% sorghum flour and this may be due to the role of wheat gluten in baked food products.

CHAPTER SIX

6.0 CONCLUSION AND RECOMMENDATION

6.1 Conclusion

From the result of the analysis carried out, sorghum (*Sorghum bicolor*) couscous has high nutrient composition compared to wheat couscous and therefore can be commercialized and produced at industrial level. The sensory evaluation result showed that couscous produced using Sorghum Flour is equally accepted as that made from wheat flour. Also blending sorghum with wheat flour to produce Couscous can give a good brand with more desirable taste and aroma.

6.2 Recommendations

From the finding of this study, it is recommended that;

- (1) Sorghum grains can be used as a substitute for wheat grains in couscous production due to its availability, low cost and high nutritional value.
- (2) Further research needs to be carried out on blending sorghum and wheat flour in production of variety of products so as to reduce cost associated with wheat as a raw material since sorghum is more cheaply available in Nigeria.
- (3) Further research to come up with improved techniques for traditional couscous production also needs to be carried out so that a standard couscous can be produced from a variety of the available local grains which will pave way for new staple foods for our people.
- (4) Additional research need to be carried out on blending sorghum with wheat flour at different ratio to produce couscous.

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List of Plates



Plate A: Wheat flour



Plate B: Sorghum flour



Plate C: Raw couscous

Desirable couscous particles are produced using a sieve with 1.5 mm mesh size (raw couscous) before steaming to produce couscous.

100% Wheat Flour couscous (A)

100% Sorghum flour couscous (B)

50% Wheat/50% Sorghum flour couscous (C)

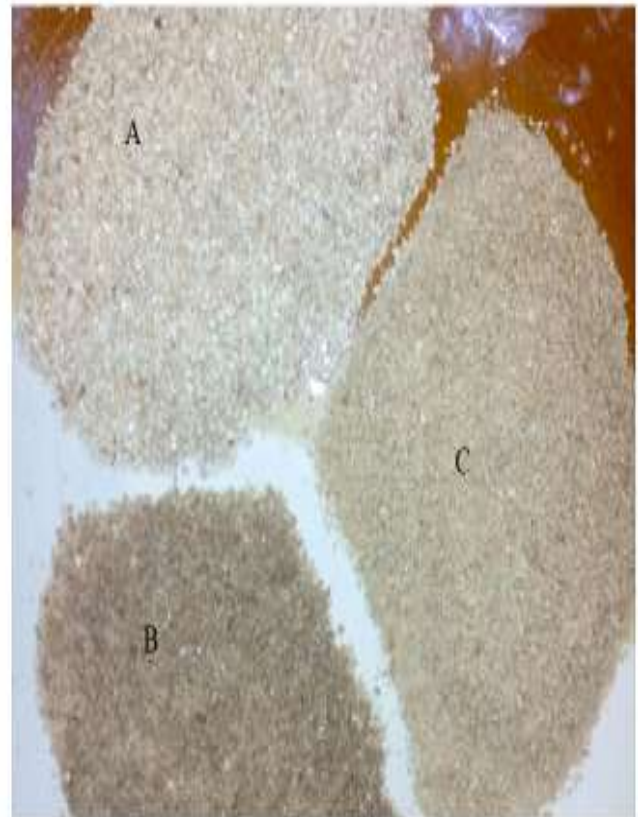


Plate: D The three produced couscous

List of Appendices

A nine point hedonic scale questionnaire to be completed by the panelist based on the preferences levels to the listed sensory and organoleptic quality attributes for the three produced couscous. Every panelist has to fill three questionnaires, one per couscous product.

Instructions:

Taste the given sample, then place an x mark on the point in the scale which best describes your feeling.

Panellist no:			Date:			
Product:						
Attributes:		Clour	Texture	Aroma	Taste	Overall acceptability
Scale:						
9	Like extremely					
8	Like Very much					
7	Like moderately					
6	Like slightly					
5	Neither like no dislike					
4	Dislike slightly					
3	Dislike moderately					
2	Dislike very much					
1	Dislike extremely					

Appendix 2

Hedonic scale results for the attribute “**Colour**” of the 100% sorghum flour, 100% wheat flour and 50/50 sorghum/wheat flour couscous for the 15 panelist.

JUGDES	SFC	WFC	SWFC
1	5	9	7
2	5	8	7
3	5	7	7
4	3	8	3
5	6	8	7
6	3	9	6
7	6	7	5
8	7	7	6
9	4	6	7
10	6	7	2
11	5	8	8
12	6	6	3
13	5	8	7
14	6	8	6
15	3	7	8
Total	81	113	87

Appendix 3

Hedonic scale results for the attribute “**Texture**” of the 100% sorghum flour, 100% wheat flour and 50/50 sorghum/wheat flour couscous for the 15 panelist.

JUGDES	SFC	WFC	SWFC
1	7	7	8
2	9	5	5
3	8	7	7
4	7	5	6
5	6	6	4
6	8	4	7
7	8	7	6
8	9	5	7
9	8	6	8
10	7	5	8
11	8	7	7
12	7	5	7
13	8	7	6
14	7	8	8
15	8	9	7
Total	115	95	101

Appendix 4

Hedonic scale results for the attribute “**Aroma**” of the 100% sorghum flour, 100% wheat flour and 50/50 sorghum/wheat flour couscous for the 15 panelist.

JUGDES	SFC	WFC	SWFC
1	8	9	9
2	6	7	8
3	6	8	7
4	9	8	9
5	7	7	8
6	7	8	7
7	8	8	6
8	7	8	9
9	7	7	8
10	7	8	9
11	6	6	6
12	7	7	7
13	6	8	8
14	6	8	7
15	7	7	8
Total	104	114	116

Appendix 5

Hedonic scale results for the attribute “**Taste**” of the 100% sorghum flour, 100% wheat flour and 50/50 sorghum/wheat flour couscous for the 15 panelist.

JUGDES	SFC	WFC	SWFC
1	9	6	7
2	8	7	8
3	7	8	7
4	7	8	7
5	9	7	7
6	9	6	7
7	8	7	6
8	8	7	9
9	8	7	7
10	8	8	8
11	8	7	8
12	7	6	7
13	8	6	7
14	9	7	6
15	9	7	8
Total	122	104	109

Appendix 6

Hedonic scale results for the attribute “**Overall acceptability**” of the 100% sorghum flour, 100% wheat flour and 50/50 sorghum/wheat flour couscous for the 15 panelist.

JUGDES	SFC	WFC	SWFC
1	8	6	8
2	7	7	8
3	6	6	6
4	8	8	7
5	6	6	7
6	2	5	3
7	3	2	5
8	4	7	5
9	5	5	6
10	7	8	7
11	8	9	7
12	7	5	8
13	7	6	9
14	6	5	8
15	3	4	4
Total	87	80	98