



Effects of Combination of Cereals and Legumes on the Proximate and Microbial Contents of Ndaleyi Production: A Fermented Nigerian Food in Borno State

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Abstract: The study aimed to investigate the effects of dehulling and fermentation on a combination of cereals and legumes in the production of Ndaleyi from blends of millet, sorghum, and soybean flour, while also determining the proximate composition of the raw materials and finished products and evaluating the bacterial content of the finished products. The raw materials—millet, sorghum, and soybean—were purchased from Maiduguri Monday Market in Borno State, Nigeria, on October 18, 2023, and transported to the Department of Food Science and Technology at Ramat Polytechnic, Maiduguri for processing. They were stored in 25 kg polyethylene bags at room temperature. Millet, sorghum, and soybean samples (2 kg each) were soaked in water for one week, except for soybean which was soaked for three days. The soaking water was replaced regularly to prevent odor caused by microbial action during fermentation. Following dehulling and fermentation, the grains were dried in sunlight. The dehulled and unde-hulled flours were then blended in ratios of 75:25 and 90:10 with soybean flour. AOAC methods were used for laboratory analysis, revealing that the proximate composition of millet decreased with dehulling—protein content fell from 12.84% to 11.63%, fats from 2.77% to 2.36%, and carbohydrates from 73.73% to 70.115%, while ash content increased from 2.66% to 3.37%. Similar changes were observed in dehulled sorghum and soybean flour. Bacterial analysis indicated the presence of coliforms, *Escherichia coli*, *Salmonella*, and *Staphylococcus aureus* in both raw materials and finished products.

Keywords: Effects, Combination, Cereals, Legumes Proximate, Microbial

Introduction

Nutrition plays a vital role in prevention of diseases. The richest source of proteins and nutrients are those that can be derived from cereal grains presented abundant in developing countries (Verma *et al.* 2016). Some soybean seed may have been sent from China by missionaries as early as 1740 and planted in France. Soybean in U.S. literature was in 1804, however, it is through that soybean was first introduced to Nigeria in 1908 and was cultivated as an export crop in a small area in Benue State where the introduced variety 'Malayan' was adopted. The crop is usually grown in small holdings in mixed cropping with sorghum or maize or as an intercrop in citrus orchards.

Mutual supplementation of cereals with legume/oil seed has been advocated by several workers because of amino acid complementation, hence further improvement of nutritive value of the products can be done by the addition of defatted soy flour. Defatted soy flour is rich in high quality protein and an excellent source of iron, calcium and B vitamins.

Soy foods are also rich in non-nutrient functional component such as flavones. Soybean provides both soluble and insoluble fibre, which may help lower serum cholesterol level, control blood sugar, increase stool bulk, prevent colon cancer and relief symptoms associated with some digestive disorders. Other health benefits of soybeans in diet include cancer prevention, cholesterol lowering, combating osteoporosis and menopause regulation (Carroll, 1991).

Pearl millet is an important source of energy in the form of starch, but it can also contribute significant amount of fiber, minerals and other nutrients to diet. The nutritional composition of the grain shows that it is equal to wheat in its protein and superior to wheat in fats and minerals particularly iron and calcium (Asha, 1999). But information regarding the use of pearl millet in product development is scanty. The major constraints for widespread utilization of pearl millet are its coarse fibrous seed coat, coloured pigment, characteristics astringent flavor and poor keeping quality of the processed products (Archana *et al.*, 2001). Some of the constraints associated with pearl millet use for product development may be overcome by malting the grain. Malting has been reported to be beneficial in improving nutritive value and sensory quality of grains (Asha, 1999).

Statement of the Problem

Protein energy malnutrition (PEM) has been identified as one of the most endemic notational problems in sub-Saharan Africa including Nigeria. Attempts have been made to devise certain strategies for combating this menace. Highly nutritious food rich in protein and high calorie value promoted this research. Cereal grain and legume complementation has been suggested. In Africa, traditional food with adequate nutritional value like *Ndaleyi* is recommended. Nutritional deficiency disease like kwashiorkor and marasmus can be equally controlled.

A healthy diet incorporates all nutrients in moderation. Low protein intake has several health consequences and a severe lack of protein in the diet causes death (UNICEF, 2008).

The common legumes such as soybean, mung bean, black bean, lentils and chickpea increase the protein content and improving protein quality of cereal-based complementary foods. Both legumes and cereals however are rich in phytic acid which is a potent inhibitor of mineral and trace element absorption. However, a combination of cereals, millet and oil seeds provides most of the amino acids which complement each other to provide better quality proteins.

Objectives of the Study

The objectives of the study are to:

- (i) Produce *Ndaleyi* from cereals and defatted soybean
- (ii) Evaluate the proximate composition of the raw materials and finished products.
- (iii) Evaluate the bacterial content of the finished products.

LITERATURE REVIEW

The study was based on the research carried out by Nkama *et al.*, (1994) who determined the traditional method of *Ndaleyi* production from millet as a fermented sun-dried agglomerated powder made from millet or sorghum. It is a major traditional food of the Kanuri people of Borno State. The research was carried out in Maiduguri, Borno State. The research was to conduct and

examine the efficacy of the traditional method and suggesting the best approach and highly acceptable hygienic method for the production. Chemical analysis involving protein, ash moisture content, fats, crude fibre and carbohydrate by difference will be examined (AOAC, 1994). This research was different, because of the introduction of soybean as a compliment to enhance the nutritional amino acid i.e. methionine and lysine are both essential amino acid to the finish product. Relevant books, journals, conference papers, etc were consulted on the topic.

Cereal and Grain Legumes

The major cereals cultivated throughout the world are wheat, rice, barley, maize, rye, sorghum, oat and millet. The total cereal production in the world in 1991 was about 1.883,888,000 metric tonnes. Africa accounted for about 5.2% (99,397,000 metric tonnes) of the total cereal production in Africa during the same period (FAO, 1991).

Some of the important cereals grown in Nigeria are sorghum, millet, rice and maize. The country was the largest producer of sorghum and millet in Africa in 1991 and it produced about 14% and 8% of the world total production of millet and sorghum, respectively. (FAO 1991) Cereals are food from the cultivated grasses, member of the monocotyledonous family Gramineae. Cereals have been an important crop for thousands of years. The successful production, storage and use of cereals have contributed in no small measure to the development of modern cultivation (Kent, 1994).

A cereal is any of the edible components of the grain. Botanically, a type of fruit called caryopsis of cultivated grass, composed of endosperm, germ and bran. Cereal grains are grown in greater quantities and provide more food energy worldwide than any other type of crop (IDRC, 2016) and therefore, are staple crops for significant number of people. In their natural, unprocessed, whole grain form, cereals are rich source of vitamins, minerals, carbohydrates, fats, oils and proteins. When processed by the removal of the bran and germ, the remaining endosperm is mostly carbohydrate. In developing countries, production and consumption of grains such as rice, wheat, millet, sorghum or maize constitute majority of daily sustenance. In developed countries, cereal consumption is moderated and varied but still substantial (IDRC, 2016).

Production of Cereal Grains

Cereal remains the most important single food in many rural areas of Asia and African providing more than 70% of the common diets (Stanley, et al. 1975). Cereal grains are relatively low in proteins and lack some of the essential amino acids. Considerable quantities of cereal grains such as maize, sorghum, rice among others are produced in various part of the world (Kent, 1983). Global production statistics (FAOSTAT, 2018) indicated that China is the highest producer of cereals in the world. As of 2016, cereal production in China was 580 million metric tons which accounts for 20.40% of the world's cereal production, followed by the United States of America, India, Russian Federation and Indonesia collectively accounting for 55.04% of the world's total cereal production which was estimated at 2,846 million metric tonnes .(FAOSTAT,2015).

Table 2.1: World cereal production (tons) from 1961 to 2013

Cereal crops	Year				
	1961	2010	2011	2012	2013
Maize	205	851	888	872	1016
Rice (paddy)	285	703	725	720	745
Wheat	222	650	699	671	713
Barley	72	124	133	133	144
Sorghum	41	60	58	57	61
Millet	26	33	27	30	30
Oat	50	20	22	21	23
Rye	12	12	13	15	16
Triticale	35	14	13	14	14.5
Total	948	2467	2578	2533	2762.5

Source (FAO 2013)

Close examination of the production figures between 1961 and 2013 (Fig. 2.1) indicated that significant annual increase is recorded in developing countries than developed countries indicating huge dependence of developing countries on cereal grains for sustenance. The Food and Agricultural Organization (FAO) recent projected global cereal production in 2018 to be 2.591 million tons which is 2.4% less than the 2017 production figure. Corn is currently the leading cereal in terms of quantity produced followed by wheat and rice (Fig. 2.1).

World wheat production in 2018 is forecast at 722.4 million tons representing the lowest level since 2013. The current reduction may mostly be as a result of lower yields because of reduced rainfall and flood in many parts of the world. Global rice production in 2018 therefore is anticipated to amount to 513 million metric tonnes up 1.3 percent from 2018 all-time high. These increased production in 2018 follows evidence of greater Asian land area planted with rice, particularly in India even though short water availabilities for irrigation might have dampened the expected harvest in Mali, Pakistan and the Philippines. World production of coarse grain (cereal grains other than wheat and rice) in 2018 is forecast at 1,356 million tons which is 35.6 million tons (2.6 percent) below the 2017 recorded production.

Table 2.2: The Proximate composition of some cereal grains

Components	Rice	Wheat	Maize	Sorghum	Millet	Barley	Oat	Rye
Moisture (%)	12	12.5	13.8	11	11.8	11.1	8.3	11.0
Calorie (kcal/100g)	360	330	348	332	327	349	390	334
Protein (%)	7.5	12.3	8.9	11	9.9	8.2	14.2	12.1
Fat (%)	1.9	1.8	3.9	3.3	2.9	1.0	7.4	1.7
NFE (%)	77.4	71.7	7.2	73	72.9	78.8	68.3	73.4
Fibre (%)	0.9	2.3	2.0	1.7	3.2	0.5	1.2	2.0
Ash (%)	1.2	1.4	1.2	1.7	2.5	0.9	1.9	1.8
Thiamine (g/100g)	0.34	0.52	0.37	0.38	0.73	0.12	0.60	0.43
Riboflavin (g/100g)	0.05	0.12	0.12	0.15	0.38	0.05	0.14	0.22
Niacin (g/100g)	4.7	4.3	4.2	3.9	2.3	3.1	1.0	1.6

Source: Ajayi (2006)

Physical, Chemical and Nutritive Characteristics of Millet

Millet is the smallest of the cultivated cereal, in size, 2mm length by 1-2.5m in width with average grain weight of 7g per width, with average grain weight per 1000 grains. The grain comprised of

the seed coat, germ and endosperm. Each main part of the grain is further divided in various layers, tissues or regions. The pericarp and test are hard and indigestive. The aleurone layer contains a high proportion of protein than the flavor. The germ and scutellum are rich in protein and fat. The endosperm comprising about 70-80% of the whole grain consist of mainly starch and protein (Kent, 1994).

Chemical Composition of Millet

The mature millet grain consists of carbohydrates, proteins, lipids, minerals matter and water in addition to small amounts of vitamin and enzymes (Table 2). The carbohydrates are the most important constituent forming about 77-87% of the total dry matter, carbohydrates present in millet are starch, cellulose, hemicellulose, pentosans, dextrans and sugars. The protein content of pearl millet is from 8.6 to 17.4% depending in part on variety and class but more largely on environmental factors during growth. The removal of some of the outer layers of millet grain by dehulling lowers protein content but increases the digestibility of the dehulled product (Nkama and Ikwelle, 1997).

The protein content of pearl millet and foxtail millet is comparable with other cereal. Fearly, millet has higher protein content than sorghum or maize. Finger millet appears to be adequate in all the essential amino acids compared to sorghum, which is deficient in lysine, tryptophan and sulfur containing amino acids. Millet is rich in the B-complex vitamins with levels of thiamine, niacin, pantothenic acid and pyridoxine familiar to that of wheat. Finger millet is high in calcium (344mg/100g fresh basis). Millet has been shown to possess anti-pellagra properties. It has been observed that incidence of pellagra was lower in areas where millet is consumed than where maize is the sole cereal food (Kent, 1983, Nkama, 1994, Ahmed and Dominguez, *et al.* 1993).

Millet Legume Combination

The nutritional complementarity of cereals and legumes proteins seems well known in Africa. Millet or sorghum with cowpea or groundnut is used in the Sahelian countries. Chemical analysis has shown that amino acids deficient in the millet are generally adequately compensated by the proteins of legumes and vice versa. The mutual compensation is closer to deal when the ratio by mass of cereal to legume is about 70:30 in which proportion each provides about equal part by mass of protein (Nkama, 1993; Amleida Dominguez, *et al.*, 1993; Merero *et al.* 1988).

Processing and Food Uses of Millet

Nigeria uses million tonnes of pearl millet as staple food in many homes, especially among the poor, predominantly in Northern Nigeria (FAO, 2007). It is also used in making a popular fried cake known as "masa". Its flour is also used in preparing "Tuwo", a thick binding paste, also referred to as "Toh" in northern Africa. It contains 18% protein, rich in Vitamin B especially niacin, B6 and folic acid. It is fitted for flat bread especially because it lacks gluten. It is an important food across the Sahel. It is particularly the main staple in a large region of northern Nigeria, Niger, Mali and Burkina Faso. It is often ground into flour, rolled into large balls, parboiled, liquefied into watery paste using fermented milk and then consumed as a beverage. This beverage called "Fura" in Hausa or "Tukura" in Marghi language is a popular drink in northern Nigeria and southern Niger. Pearl millet is an excellent forage crop because of its low hydrocyanide content (Green, 2012).

Millets are also used to prepare alcoholic beverages. Millets are traditionally important grains used in brewing millet beer in some cultures, for instance by the Tao people of Orchid Island and the Amisr Atayal of Taiwan. It is also the base ingredient for the distilled liquor rakshi in Nepal and the indigenous alcoholic drink of the Sherpa, Tamang, Ri and Limbu people, Tongba, in Eastern Nepal (Amadou, 2019).

Millet is processed for food in several ways depending upon need and local habits. The main objective of processing is to improve appearance, texture, culinary properties and palatability and alter the bioavailability of nutrients.

Millet grains, before consumption and for preparing of food are usually processed by commonly used traditional processing techniques include decorticating, malting, fermentation, roasting, flaking and grinding to improve their edible, nutritional and sensory properties. Millet can be used to make bread, beer, cereal and other dishes. Even today, millet is a staple food around the world in fact, millet is gaining renewed population because of how versatile and easy it is to grow (Merero *et al.* 1988).

MATERIALS AND METHODS

The samples were purchased at the Maiduguri Monday Market and transported in polythene bags to the Department of Hospitality Management Technology Ramat Polytechnic, Maiduguri, Borno State, for the processing of 25kg millet, 25kg sorghum and 15kg soybean grains obtained and kept in the kitchen at room temperature.

Production of Flours

The cereal grains (millet) flours were processed according to the method demonstrated by Ihekoronye and (Ngoddy 1985). Essentially, the grains were clean to remove extraneous matters such as stones, chaffs, sands and broken grains conditioned to moisture content of 14% and mill with a hammer mill (Meadows Model 35). The flour was sieved using sieves of 315 microns to separate the bran from the endosperm to produce fine flour ready for use in composite blending and the soybean flour was steeped in water for some time.

The flours produce millet, sorghum and soybeans are packaged and stored at room temperature for both processing and laboratory analysis in the demonstration kitchen of the Department of Hospitality Management and Department of Food Science and technology, Ramat Polytechnic Maiduguri respectively.

Determination of Physical and Functional Properties of Millet

A portion of the millet sample (2kg) was placed in an earthen ware pot and about 2.5 litres of step water (fresh borehole water or kadal) added the sample was steeped for 6 days at room temperature ($30\pm 2^{\circ}\text{C}$), then removed, washed with fresh water and briefly sun-dried for 1 hour to remove surface moisture.

Clean soybean grains were placed in an earthen ware pot measuring 500g of step water (fresh borehole water) was fermented for one week by steeping at room temperature then removed, washed with fresh water and briefly dried for 2 hours to remove surface moisture. The steeped millet and steep soybean were mixed in ratio of 4:1 and millet homogenous using a No.2A premier grinding mill (Christy Hunt Engineering Ltd, Atlas Wors Earls Colne, U.K.). A very thin slurry was prepared from the mixed millet and soybean by adding excess water (1:10w/v). This was then sieved using a very fine cheese cloth. The bran fraction (over tail or biria) was removed, sun-dried and reserved for analysis. The filtrate was transferred into a larger earthen ware pot. Vigorously

stirred by hand and allowed to settle for two or three days. Excess water was removed and a portion of it reserved for analysis.

The top-lighter layer Chir was carefully removed and placed on a mater in thin sheets and the bottom denser layer *Ndaleyi* was then recovered and placed on another mat. Both samples (*Ndaleyi* and Chir) were sun-dried for 6 to 8 hours.

Determination of Ndaleyi and by-product (Chir and bran). Each of these was expressed as a percentage of dry matter content of cleaned dried mixed of millet and soybean grain (Nkama *et al.* 1994).

Physical Analysis of Millet, sorghum and Soybeans Flours

The moisture content, crude protein (m x 6.25), crude fat and ash content of millet and soybean grain, *Ndaleyi*, Chir, millet and soybean mixed ogi and bran was determined using relevant AOAC methods. Titratable acidity (TA) and pH of millet grain and soybean grain during steeping, the steep liquor, *Ndaleyi*, Chir and millet and soybean ogi mix was determined according to AOAC methods. Reducing sugars in raw millet grain, soybean *Ndaleyi*, Chir and millet ogi were determined according to Lane and Erynon method as described by (Pearson and Nkama, 1994).

Functional Properties of the Flour Blends and the *Ndaleyi*

Determination of Bulk Density

The method described by (Onwuka 2005) is adopted. Ten (10ml) capacity graduated measuring cylinder will be pre-weighted. The cylinder was filled gently with the sample. The cylinder was tapped gently several times on the laboratory beach until no further reduction of the sample level after filling to the 10ml mark, it was weighted and calculated as follows:

$$\text{Bulk density (g/ml)} = \frac{\text{weight of sample (g)}}{\text{volume of samples}}$$

The bulk density (g/cm³) will be determined as weight of the sample (g) divided by the sample volume (cm³) as reported by (Adejuyitan *et al.* 2009). Fifty-gram flour of the complementary foods will be placed into a 100ml measuring cylinder and tapped to a constant volume. The bulk density (g/cm³) was determined as weight of developed complementary food (g) divided by developed complementary food (g) divided by developed complementary food volume (cm³) as reported by (Adejuyitan *et al.*, 2009).

Foaming Capacity and Stability of Millet, Sorghum and Soybeans Flours

The procedure of (Lawhom *et al.* 1971) in determining forming capacity and stability of millet, sorghum and soybean flour was adopted. Two grams of flour blends sample and 50ml distilled water were mixed in a Braun blender at room temperature. The suspension will be mix and shake for 5 minutes at 1600rpm. The content along with the foam was poured into a 100ml graduated measuring cylinder. The total volume was recorded after 30 seconds. Then the content was allowed to stand at room temperature for 30 minutes and the volume of foam only was recorded.

$$\text{Foaming capacity (Fc) (\%)} = \frac{\text{volume of foam} - \text{volume of BW}}{\text{Volume of foam AW}}$$

Where: AW = After whipping

BW = Before whipping

P_5 = The volume of foam only (total volume – liquid volume) after the 30 min standing is taken as foam stability

Oil Absorption Capacity of Millet, Sorghum and Soybeans Flours

Oil absorption capacities of the sample of millet sorghum and soybean were determined by the centrifugal method elicited by (Beuchat 1977) in the slight modifications. One gram of sample was mixed with 10ml of pure canola oil for 60 sec, the mixture then was allowed to stand for 10 min at room temperature, centrifugal at 400g for 30 min and the oil that separated was carefully decanted and the tubes will be allowed to drain at a 45° angle for 10 min and then weighed. Oil absorption is expressed as percentage increase of the sample weight.

Water Absorption Capacity (WAC) Millet, Sorghum and Soybeans Flours

Water absorption capacity of millet, sorghum and soybean flour was determined using (AOAC 1990) official methods of analysis. About 2g each of the flour blends of ingredients and soybean and millet blends was weighed into a centrifuge tube. Five milliliters of water were added and mixed well. The mixture was allowed to stand for 30 minutes and centrifugal at 600rpm for 15 minutes. The supernatant was decanted, and the new weight of the sample was taken as water absorbed and the result is expressed on weight (g) of water per 100g dry samples. The experiment was repeated, and triplicate determination was made for each *Ndalayi* blend.

$$\% \text{ WAC} = \frac{\text{weight of sample after centrifuge}}{\text{weight of the original samples}}$$

Swelling Power and Solubility Index Determination of Millet, Sorghum and Soybean Flours

The swelling power and solubility index method as described by (Hirsch and Kokini 2002) One gram of the flours of ingredients of soybean and millet blends was poured into pre-weighed graduated centrifuge tube appropriately labelled. Then 10ml of distilled water was added to the weighed sample. In the centrifuge tube and the solution was stirred and placed in a water bath heated at different temperature of 85oC for one hour while shaking the sample gently to ensure that the starch granules remained in suspension until gelatinization occurred. The samples were cooled to room temperature under running water and centrifugal for 15 min at 3000rpm. After centrifuging, the supernatant was decanted from the sediment into a pre-weighed petri-dish; the supernatant in the petri-dish was weighed and dried at 105°C for 1h. The sediment in the tube was weighed and the reading recorded. The starch swelling power and solubility were determined according to the equations below:

$$\begin{aligned} \text{Swelling power} &= \frac{\text{Weigh of swollen sediment}}{\text{Weight of star sample}} \\ \text{Solubility} &= \frac{\text{Weigh of dry supernatant}}{\text{Weigh of starc sample}} \times 100 (\%) \end{aligned}$$

Determination of viscosity in Millet, Sorghum and Soybean Flours

The hot paste viscosity of the samples was determined with Brookfield Viscometer RV Model (Brookfield Engineering Laboratories Stoughton, U.S.A) as described by (Badau *et al.* 2006). The viscosity of the developed complementary foods was determined by first preparing the gruel. The slurry of the complementary *Ndaleyi* was prepared by dissolving 40g in 200ml and 20g in 200ml of distilled water to give 20% and 10% w/v concentration respectively. The slurry was heated in water bath timed to reach a cooking temperature of 95% for 7 minutes. The viscosity of the flours and complementary *Ndaleyi* foods measured appropriate ambient temperature of 32.1%, revolution per minutes (RPM) of 2.5 for 10% w/v concentration using spindle "L" and 28.9°C RPM of 6 for 20% w/v concentration using spindle (Mosha and Svanberg, 1983; Badau, *et al.* 2006).

Chemical Methods

Crude Protein Determination

Principle

In Kjeldahl procedure, proteins and other organic food components in a sample are digested with sulphuric acid in the presence of catalysts. The total organic nitrogen is converted to ammonium sulphate. The digest was neutralized with alkali and distilled into a boric acid solution. The borate anions formed are titrated with standardized acid which was converted to nitrogen in the sample. The result of analysis represents the crude proteins content of the food since nitrogen also come from non-protein components.

Digestion

After preparation, 1g of the sample was introduced into 100ml digestion flask. The following was added into flask: 2g anhydrous sodium sulphate again hydrated cupric sulphate, a pinch of selenium powder and 10ml of concentrated sulphuric acid. This was digested at 42°C for 45 minutes until clear digest was obtained. After digestion, distilled water was added into the flask up to 100ml mark and then mixed thoroughly.

Distillation

Five mills of boric acid after digestion were introduced into 250ml conical flask and 2 drops of indicator was added. Then 5ml of the diluted was introduced into the distillation apparatus. The inlet was closed and blocked with distilled water to prevent escape of ammonia after 20ml of 40°C NaOH introduced into it. It was distilled into the 250ml conical flask containing the boric acid and indicator (bromocresol and methyl red) until about 50-75ml of distilled was collected (Egan *et al.*, 1988; Nielsen, 2002).

Titration

The distillate in the conical flask was titrated with standard solution of hydrochloric acid (HCl) to the end point. The titre volume was recorded, that is, volume of HCl that exchanged with the indicator and percentage crude protein was calculated.

$$\% \text{ Crude Protein} = \frac{A \times C \times 100 \times 1 \times 6.25}{B \times D \times 100 \times E}$$

Where :

- A = Volume of solution of standard HCL (titre value)
B = Volume of Sample solution taken from distillation
C = Volume of sample made after digestion
D = weight of sample taken for distillation
E = acid factor

Amino Acid Determination

The amino acid profile in the known samples was determined using methods described by Benitez (1989). The known samples were dried to a constant weight, defatted, hydrolyzed, evaporated in a rotary evaporator and later loaded into Technicon Sequential Multi-sample Amino Acid Analyser (TSM).

Oven Drying Methods

In oven drying methods, the sample was heated under specified conditions and the loss of weight was used to calculate the moisture content of the sample. The moisture content value obtained was highly dependent on the type of oven used, conditions in the oven and the time and temperature of drying. Various drying oven methods are AOAC approved for many food products. The methods are simple, and many ovens allow for simultaneous analysis of large numbers of samples. The time required may be from a few minutes to 24 hours.

Calculation of moisture and total solid contents of foods

Moisture and total solids contents of foods can be calculated as follows using oven drying procedure:

$$\% \text{ moisture} = \frac{w_t H_2O \text{ in sample}}{wt. of \text{ wet sample}} \times 100$$

$$\% \text{ moisture} = \frac{wt. of \text{ wet sample} - wt. of \text{ dry sample}}{wt. of \text{ wet sample}} \times 100$$

$$\% \text{ total solids (wt/wt)} = \frac{wt. of \text{ dry sample}}{wt. of \text{ wet sample}} \times 100$$

(ii) proein content analysis (kjeldahl meth) (AOAC 1991).

Principle

In the Kjeldahl procedure, proteins and other organic food components in a sample are digested with sulphuric acid in the presence of catalysts. The total organic nitrogen was converted to ammonium sulphate. The digest was neutralized with alkali and distilled into a boric acid solution. The borate anions formed were titrated with standardized acid which is converted to nitrogen in the sample. The result of the analysis represents the crude protein content of the sample of the food since nitrogen also comes from non-protein components.

Defatted Sample of Soybean

The samples were defatted using chloroform and methanol (solvents) mixed in the ratio of 2;1 and 4g of each of the samples were placed in extraction thimble for 15 hours defatting in soxhlet extraction apparatus (AOAC, 2006).

Ash Content Analysis

The Association of Official Analytical Chemists (AOAC) international has several dry ashing procedures (e.g. AOAC methods) for certain individual foodstuffs

The general procedure includes the following steps:

- i. The sample were weighed in a 5g sample into a tarred crucible pre-dry if the sample is very moist and crucibles was placed in cool muffle furnace use tongs gloves and protective eye wear if the muffle furnace is warm.
- ii. The muffle furnace was turned off and waits to open it until the temperature has dropped to at least 250°C preferably lower. Open door carefully to avoid losing ash that may be fluty.
- iii. A safely tongs was used to quickly transfer crucibles to a desiccator with a porcelain plate and desiccant. Cover crucibles, close desiccator and allow crucibles to cool prior to weighing.

The ash content is calculated as follows:

$$\% \text{ ash} = \frac{\text{wt. after ashing} - \text{tare wt of crucible}}{\text{Original sample wt} \times \text{dry matter coefficient}}$$

Where: dry matter coefficient = % solids/100

Crude Fat Analysis (Continuous Solvent Extract Methods)

For continuous extraction, sample was put in an extraction ceramic thimble and the solvent is added into the boiling flask. The continuous method gives faster more efficient extraction than semi-continuous extraction methods. However, they may cause channeling which results in incomplete extraction. The Wiley under writers and goldfish tests are examples of continuous lipid extraction methods.

Soxhlet method – procedures – preparation of sample: If the sample contains more than 10% H₂O dry the sample to constant weight at 95-100% under pressure ≤ 1000mmHg for about 5 hours (AOAC method).

Soxhlet method procedure

- i. The samples were weighed to the nearest mg, about 2g. Pre-dried sample into a pre-dried extraction thimble, with porosity permitting a rapid flow of ethyl ether, cover sample in thimble with glass wool; it was weighed pre-dried in a boiling flask.
- ii. The samples were put anhydrous ether in boiling flask. Note: the anhydrous ether was prepared by washing commercial ethyl ether with two or three portions of H₂O adding NaOH or KOH and letting stand until most of H₂O is absorbed from the ether. Add small pieces of metallic Na and let hydrogen evolution cease (AOAC Method). Petroleum ether may be used instead of anhydrous ether (AOAC method).

- iii. The samples were assembled in boiling flask Soxhlet flask and condenser. The sample was extracted in a Soxhlet extractor at a rate of 5 or 6 drops per second condensation for about 4 hours or for 16 hrs. at a rate of 2 or 3 drops per second by heating solvent in boiling flask.
- iv. The sample were dry boiled in a flask with the extracted fat in an air oven at 100°C for 30 mins, cool in desiccator and weigh.
Calculation
$$\frac{\text{fat on dry weigh basis}}{(\text{dried sample}) \times 100} = \frac{\% \text{fats in sample}}{100}$$

Dietary Fibre Analysis

Dietary fibre was estimated by two basic approaches i.e. gravimetrically or chemically. In the first approach, digestible carbohydrate, lipids and proteins are selectively solubilised by chemicals and/or enzymes. Indigestible materials are then collected by filtration and the fibre residue is quantitated gravimetrically. In the second approach, digestible carbohydrates are removed by enzymatic digestion, fibre components are hydrolysed by acid and monosaccharide are measured, sum of monosaccharide in the acid hydrolysed represents fibre.

Carbohydrate Content Determination

The available percentage of carbohydrates in the sample was determined by difference i.e. (100% -moisture + protein + ash + fibre) as described by Chibuzo and Ali (1994; 1995) and Asma et al. (2006).

Total Caloric Value

The total caloric value or energy value was estimated by using the A+ water factor as reported by Chibuzo and Ali (1994-95).

Determination of Mineral in samples

The minerals analyses of the samples were determined with the Atomic Absorption spectrophotometer (AAS) as described by (AOAC 2000). The minerals that were analysed are calcium, iron, zinc, sodium, magnesium, potassium, phosphorus, copper and manganese.

Procedure

About five grams of the formulated flours and fortificant was weighed into a clean dry porcelain crucible. It was shed in muffle furnace for two to. Hours at 475 to 500°C, cooled and dissolved in five mills of 20% Hcl. It was filtered into fifty mills volumetric flask after thorough acid wash. It was diluted with distilled water. Iron was determined by wavelength of 248.3nm. Zinc at 213.9nm, sodium and potassium was determined with flame photometer calorimetric method.

Determination of Vitamin Content in sample

Vitamin content was determined by high power liquid, chromatography (HPLC) method as described by (Angelica et al. 1996). One gram of sample will be dissolved in water in a beaker placed in an ultrasonic water bath for extraction for thirty minutes (30 minutes), it was centrifuged at 1200rpm for ten (10) min and then it was filtered with disc filter membrane. The filtrate was inserted into the mobile phase containing 0.1% trifluoro acetic acid (TFA) and acetonitrile (standard solutions) in the high-power liquid chromatography for separation. The data generated were displayed as peaks on the chromatography. Peaks were identified based on

retention time of reference standard, the vitamins determined are Thiamine, Riboflavin and Niacin.

Microbial Analysis

The pour plate technique (Hanigan and McCane, 1976) was used for the inoculation. Ten grams (10g) of the samples of dehulled, unde-hulled fermented and fermented millet, sorghum, soybean and its fortificants were weighed and dissolved in 10ml of sterile peptone water. It was allowed to stand for 10-15 minutes after shaking vigorously. Media such as Eosine methylene blue agar, monnisol salt agar, McConkey Agar, Deoxycholate citrate Agar, salmonella, Shigella Agar and Selenite "E" were all prepared and used. The suspension was poured to each of these media and allowed to cover it completely for 10-15 minutes, the excess water discarded. The plates were incubated at 37°C aerobically and anaerobically for 48 hours. Observations were made for the growth of colonies appearing on the plates with digital colony counts. The mean values of the triplicate plates counts, were expressed as colony forming units per grams (cfu/g). Microorganism associated with the complementary food formulations were determined by Harrigan and McCance (1976) and AOAC (2000).

Preparation of the Nutrient Agar

Nutrient agar should be prepared and kept at pH 5.8. The colony forming unit (cfu/g) of the millet samples were determined using plate count agar (PCA) or nutrient and potato dextrose agar (PDA) for bacteria and fungi respectively via pour plate method after serial dilution ($10^{-1} - 10^{-5}$).

100µl of serially diluted *Ndeleye* sample was dispensed into petri-dishes with plate count agar (PCA) or nutrient agar and potato dextrose agar (PDA) and incubated at ambient temperature (20-29°C) for 24-36h and 48-72h for bacteria and fungi respectively (Eruteva and Odunfa, 2017). After incubation, the colonies were enumerated, and distinct colonies will be selected and sub-cultured successively to obtain pure cultures.

Identification of Bacterial Isolates

Pure cultures of bacteria isolated was characterized and identified on the basis of their cultural (size, pigment, opacity, edge, elevation, shape, consistency) morphological (gram staining and spores staining) and bio-chemical properties (catalase, citrate, urease, coagulase, gelatinase, starch, hydrolysis, sugar fermentation oxidate test). Bergey's manual of Determinative Bacteriology (Holt *et al.* 1994). Pure cultures were preserved in slant and stored in 20% glycerol broth at 70°C for further analysis.

Identification of Fungal Isolates

The fungi isolated was characterized based on their macroscopic appearance on the culture medium microscopic morphology using lactophenol cotton blue and type of asexual spores produced and identified by reference to the compendium of Soil Fungi.

Preparation of Samples for Microbial Analysis

Samples of the complementary food formulations was sealed in plastic bags and kept in dry plastic containers for analysis.

Culture Media Preparation

Mannitol Salt Agar (MSA): This is a medium for the isolation and enumeration of staphylococcus.

Procedure:

Fifty-six (56g) of the sample was weighed into 500ml distilled water. It was allowed to soak for 10 minutes, swirled to dissolve completely and then sterilized by auto claving for 15 minutes at 12°C, it was cooked to 47°C before pouring into petri dishes.

Deoxycholate Agar

This is a selective medium for the growth of salmonella and shigella.

Procedure

Fifty-two grams (52g) of the sample was weighed and dispersed in one litre of distilled water. It was heated to dissolve the medium completely. It was not autoclaved but cooled to 47°C before pouring onto sterilized petri dishes.

Eosine Methylene Blue Agar (EMBA)

This is a differential medium for the isolation and enumeration of E-coli form organisms.

Procedure

Exactly 37.5g of the sample was weighed and dispersed in litre of distilled water. It was allowed to be soaked for 10 minutes and gently agitated to ensure complete dissolution of the precipitated and then autoclave at 121°C for 15 minutes cooled to about 47°C and then poured into petri dishes

Nutrient Agar (NA)

This is a general-purpose medium that supports the growth of a wide range of micro-organisms and contains sufficient nutrients for the organisms.

Procedure

Twenty-eight grams (28g) of the nutrient agar was weighed and dispensed in a 250ml conical flask containing one litre of distilled water which was allowed to be soaked for 10 minutes. The mixture was swirled and sterilized in autoclave at 121°C for the 15 minutes; it was allowed to cool to 47°C and then aseptically poured into sterile petri dishes to set.

Potato Dextrose Agar (PDA)

This is a medium for the growth of mould and yeasts.

Procedure:

Some Irish potatoes was peeled and sliced into pieces. It was cooled in one litre of distilled water until very soft extract was then be put into one litre conical flask and 15g agar powder and 10g of dextrose sugar was added. The mixture was dissolved and made up to one litre mark with distilled water. The medium was supplemented with 2ml of a broad-spectrum antibiotic which was dissolved in warm alcohol over a flame to dissolve completely. It was sterilized using autoclave for 15 seconds at 121°C.

McConkey Agar

This is a different medium for the isolation of coliforms and intestinal pathogens in water, dairy products and biological specimens.

Determination of Total Viable Counts

The pour plate technique (Harrigan and McCane, 1976) was used for the inoculation. After inoculation the plates was incubated at 37°C for twenty-four hours. The colonies were obtained and counted with an electric colony counter (Gallen Kemp Colony Counter).

Isolation and Identification

One gram of the sample was smeared over one corner of the solidified medium which was sufficiently dried. A nichrome wire 100p was sterilized over spirit lamp and allowed to cool and

was made parallel streaks from the main inoculums. The plates were incubated at 37°C for twenty-four hours.

The colonies were separated from one another based on the difference of colony monopoly. One of the separated colonies were taken using a sterilized wire loop and inoculated in another media then was inoculated for twenty-four hours at 37°C colonies will be obtained on the medium after twenty-four hours.

Statistical Analysis

One-way analysis of variance of each parameter was used to produce least significant differences (LSD) i.e. the value such that a difference between two samples of that amount or greater at 5% level of significance. The analysis was done using the SAS/STAT Computer programmes SAS Institute Carry, NC) on an IBM 808 personal computer.

Table 3.1: Formulation and Fortification of *Ndaleyi* Powder with Defatted Soybean Flour

MILLET	<i>Ndaleyi</i> (%)	Defatted Soybean (%)	Total Fortificant (%)
Dehulled	90	10	100
Undehulled	75	25	100
SORGHUM			
Dehulled	90	10	100
Undehulled	75	25	100

The above table shows the formulation and fortification of millet, sorghum and soybean in ratio of 10%, 25% of soybean to 90 % and 75% of dehulled and unde-hulled millet and sorghum flour respectively.

Preparation of Millet Fermented to Flour

Millet was cleaned from dirt, washed and soaked for at least 12hour then washed with clean water, sun-dried for 3-4 hours. The millet was milled into flour and sieve to obtain fine flour and the fine flour were packaged.

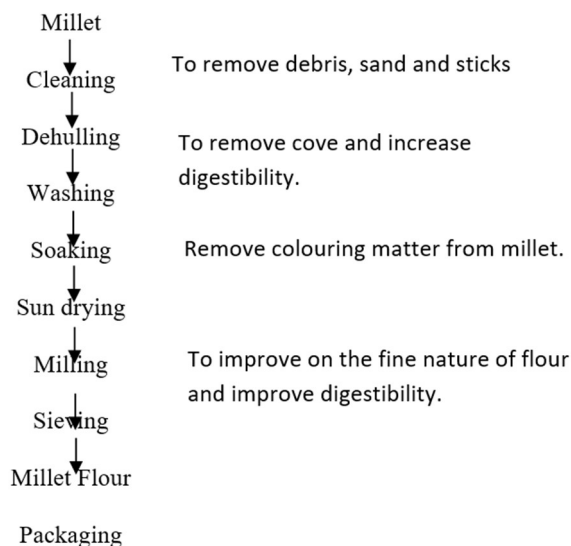


Fig. 3.1: Flow chart to produce millet flour (Nkama *et al.*, 1994)

Method of Preparation of Soybeans Fermented to Flour

Soybeans were cleaned from dirt, washed and soaked for at least 12 hours then washed with clean water, sun-dried (for at least 3-4 hours). The soybean was milled into flour and sieved to obtain fine flour were packaged.

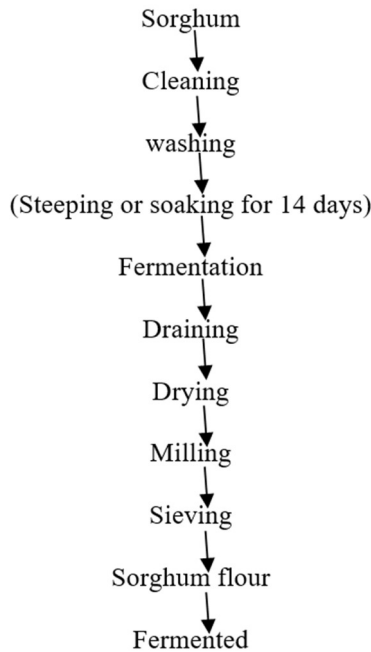


Fig.3.2: The flowchart for the preparation of soybean flour (Ngoddy *et al.*, 1985).

Production of Fermented Sorghum Flour

The sorghum seeds are fermented after cleaning, dehulling and undehulled seeds are grinded into fine particles by the attrition and pass through a 0.8mm mesh size sieve and packaged in transparent plastic buckets and store until when needed for use (Badau *et al.* 2006).

Flow chart to produce Fermented Sorghum.



Steps Involved in Production of Ndaleyi from Millet

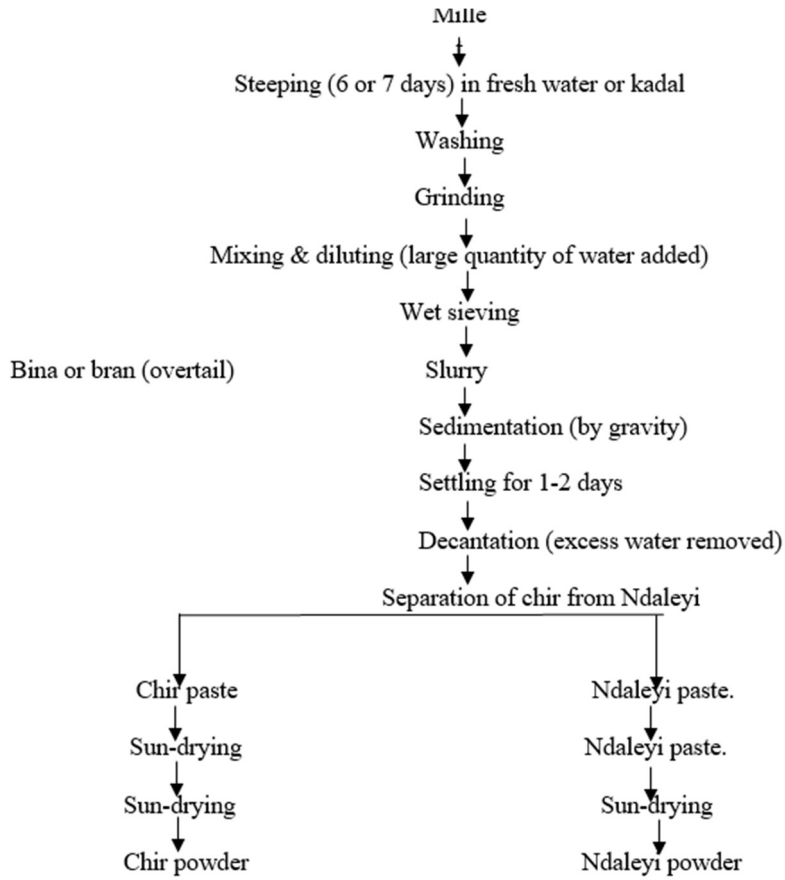


Fig. 3.4 *Ndaleyi* Production Flow Chart
Nkama et al 1994

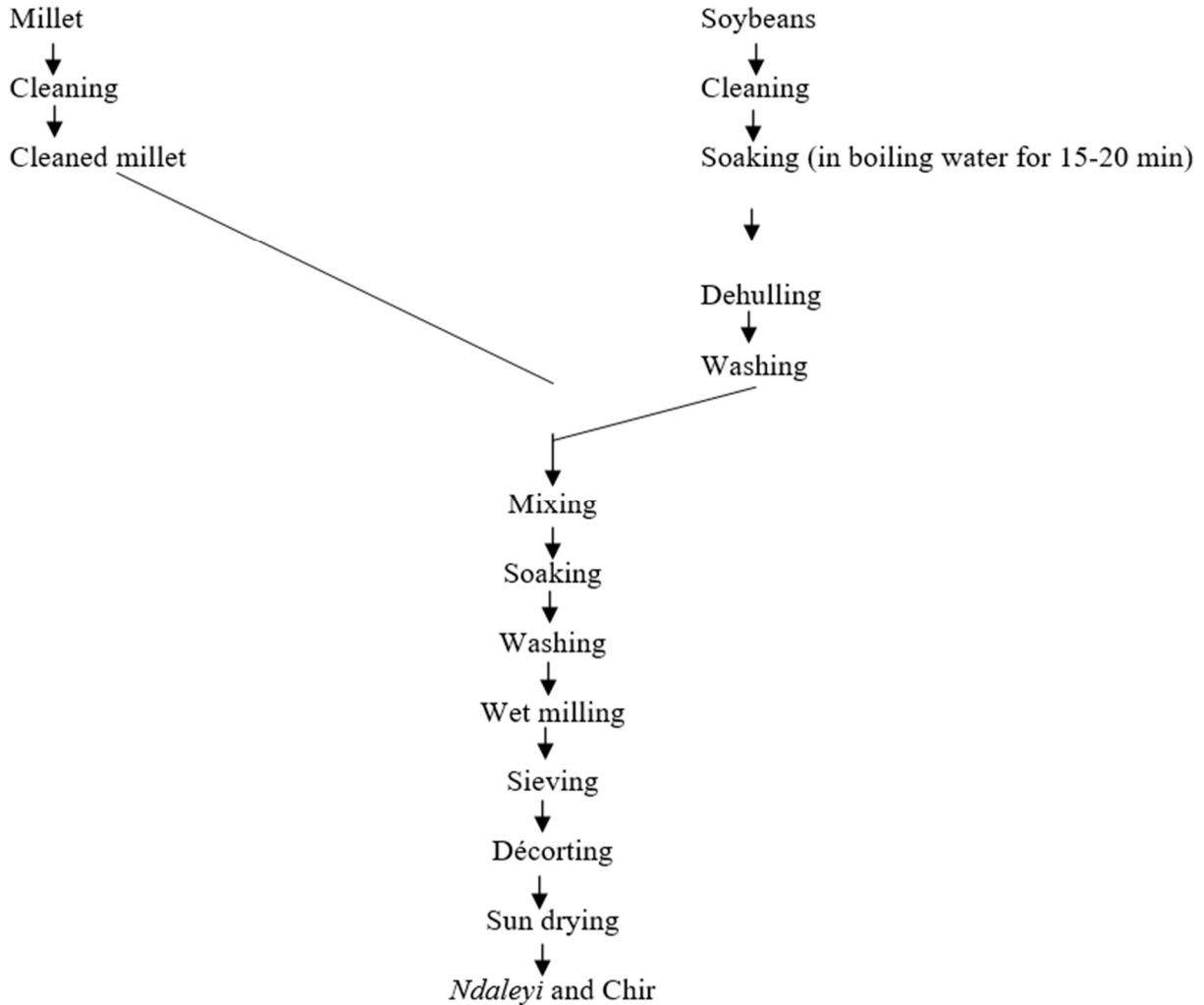


Fig. 3.5: *Ndaleyi* and Chir Production Flow Chart

Source: Nkama (1994)

RESULTS AND DISCUSSION

Table 1: Effects of Dehulling on the Proximate Composition of Millet, sorghum and Soybean Flours

The result of the proximate composition of the dehulled and undeulled samples of millet, sorghum and soybean flour were determined. The result shows that the protein content of undeulled millet, sorghum and soybean shows (12.84%, 11.88 and 38.92%) respectively which is very high in protein composition than the dehulled samples of millet, sorghum and soybean (11.63%, 10.80% and 36.46%) respectively which are low in its protein composition (Oghbaei and Prakash, 2016). The carbohydrate content of millet and sorghum at 73.75% and 72.28% are higher than that of the soybean in which is very low for both dehulled and undeulled samples.

The ash content of both dehulled and undeulled millet and sorghum samples are lower than that of soybean because of its high mineral content. This study shows that ash content of soybean for both dehulled and undeulled sample are 4.31% and 4.63%.

The fibre content for all millet and sorghum flours for both dehulled and undeulled samples are less than 1% when compared to the soybean flours under investigation. The lost in fibre content is due to the removal of bran, pericarp, and endosperm, amongst others during dehulling. (Nkama and Ikwelle 1997)

The fat content of the samples of millet and sorghum for both dehulled and unde-hulled flours at (2.36%, 2.77%), (2.86%, 3.18%) for millet and sorghum is very low when compared to (16.21%, 18.27%) for dehulled and unde-hulled soybean flour. The high fat content of soybean is because of 20% saturated fat, 3% monosaturated fat, 4% polyunsaturated fat mainly as linoleic acid (Merrit-Russe et al. 2004). The germ and scutelum are rich in protein and fats, the endosperm comprising about 70%-80% of the whole grain consists of mainly starch and protein (Kent 1993).

Table 4.2: Effect of Dehulling on the Proximate Composition of Millet, Sorghum and Soybean Flours

Samples (CODE) ⁽²⁾	Parameters (%) ⁽¹⁾						
	Crude Protein	Crude Fat	Crude Fibre	Ash	Moisture	Carbohydrates	Energy value*
DMF	11.63 ^e ±0.01	2.37 ^c ±0.03	0.61a±0.10	2.65 ^f ±0.02	9.03 ^f ±0.0252	73.70 ^a ±0.07	363.87 ^c ±0.70
UMF	12.84 ^c ±1.0E.02	2.78 ^c ±0.02	0.84a±0.02	3.38 ^e ±0.02	9.96 ^c ±0.02	70.11 ^d ±0.07	352.32 ^f ±0.69
DSF	10.82 ^d ±0.04	2.85 ^c ±0.01	0.93a±5.84E.03	3.78 ^d ±0.01	9.34 ^e ±1.0E.02	72.27 ^b ±0.04	366.30 ^b ±0.70
USF	11.85 ^d ±0.10	3.18 ^c ±0.01	0.97a±0.01	3.89 ^c ±5.774E.03	9.48 ^d ±1.0E.02	70.54 ^c ±0.02	360.06 ^d ±0.73
DSOF	36.64 ^b ±1.0E.02	11.87 ^b ±7.50	2.79a±2.87	4.31 ^b ±5.4E.03	11.53 ^b ±0.02	25.20 ^e ±0.04	359.77 ^e ±0.74
USOF	38.91 ^a ±0.02	18.23 ^a ±0.02	3.29a±2.75	4.62 ^a ±0.02	11.65 ^a ±0.03	20.69 ^e ±0.08	406.96 ^a ±0.97

(1) Values are mean ± standard deviation of triplicate determinations. In any column, mens bearing similar superscripts are not significantly different (P≥0.05).

(2) DMF = Dehulled Millet Flour; UMF = Undehulled Millet Flour; DSF = Dehulled Sorghum Flour; USF = Undehulled Sorghum Flour
DSOF = Dehulled Soybean Flour; USOF = Undehulled Soybean Flour

Table 2: Effects of Fermentation on the proximate composition of Millet, Sorghum and Soybean

The results of the proximate composition of the fermented samples, for all the dehulled and unde-hulled millet, sorghum and soybean flour were determined. Fermentation had been reported to improve sensory and nutritional quality of cereals, generally, natural lactic acid fermentation of cereals, results in a decrease in carbohydrate content together with increase in some amino acids and B-group vitamins. Furthermore, there is improved bioavailability and starch digestibility. (Merero etal 1988.,)

The result of the proximate composition is attested to that literature, there is increase in protein content from 11% to 12% in dehulled and unde-hulled samples to about 15% to 16% in fermented millet. The sorghum cereal sample also exhibit same increase in protein content as a result of fermentation, most studies regarding natural fermentation of cereals and legumes with bacteria or yeasts have described changes in proteins, amino acids and B-group vitamins (Chavan et al. 1988). Sorghum grain has a lot of nutritional benefits due to its rich in antioxidant properties, high in protein (11.8% to 16.5%) and high in calories than several other grains (Jacobs etal., 2013). Sorghum grain has high carbohydrates with 10% protein and 3.4% fats, because of the fermentation process the amount of protein in dehulled and unde-hulled samples has increased to (14.40% to 15.94%) as revealed by the results in table 4.7 and in conformity with the findings obtained by (Jacobs etal 2013). Among the legumes, groundnuts and soybeans have exceptional high oil contents, about 45% and 20% respectively (Ngoddy etal 1985). Soybeans have high fibre and potassium and low saturarated fats and sodium content.They are good sources of protein, folate and magnesium as indicated on results table 4.7 in the conformity with results obtained by (Ngoddy etal., 1985).

Table 2: Effects of Fermentation on the Proximate Composition of Millet, Sorghum and Soybean Flour

Sample (Code)	Parameter (%)						
	Crude Protein	Crude Fats	Crude fibre	Ash	Moisture	Carbohydrate	Energy value
FDMF	15.95 ^d ±5.03	3.62 ^e ±0.02	0.60 ^d ±5.03	4.36 ^d ±0.02	9.00 ^d ±5.03	66.47 ^b ±0.03	443.03 ^a ±0.97
FUMF	16.21 ^c ±0.01	3.81 ^d ±0.02	0.62 ^d ±0.02	4.71 ^d ±0.02	9.51 ^c ±1.02	65.13 ^d ±0.06	360.85 ^e ±0.69
FDSF	14.40 ^e ±5.03	3.81 ^d ±0.01	0.62 ^d ±0.02	4.21 ^f ±0.02	8.01 ^f ±0.02	68.90 ^a ±0.01	368.71 ^c ±0.70
FUSF	15.95 ^d ±0.03	3.93 ^c ±0.03	0.94 ^c ±0.01	4.77 ^c ±0.02	8.23 ^e ±0.02	66.16 ^c ±0.06	365.65 ^d ±0.71
FDSOF	42.61 ^b ±0.02	18.57 ^a ±0.02	3.37 ^a ±0.02	7.49 ^b ±0.02	12.36 ^b ±0.04	15.59 ^e ±0.07	350.43 ^f ±0.69
FUSOF	46.95 ^a ±0.02	20.65 ^b ±0.02	3.31 ^b ±0.02	7.72 ^a ±0.01	12.56 ^a ±1.02	8.77 ^f ±0.07	415.07 ^b ±0.96

1. Values are means ± standard deviations of triplicate determinations. In any column, means bearing similar superscripts are not significantly different (P≥0.05)
2. FDMF = Fermented Dehulled Millet Flour; FUMF = Fermented Undehulled Millet Flour
FDSF = Fermented Dehulled Sorghum Flour; FDSOF = Fermented Dehulled Soybean Flour
FUSOF = Fermented Undehulled Soybean Flour
3. Energy value = kcal

Proximate composition of sample of *Ndaleyi*, Chir and Bran for both millet and sorghum flour

The proximate composition of millet and sorghum flours—specifically *Ndaleyi*, Chir, and Bran—was analyzed, and the results showed that the primary components such as protein, ash, and carbohydrate content for these samples were consistent with the values reported by Nkama et al. (1994). Among the different types, millet Chir exhibited the highest protein content at 1.71%, surpassing millet *Ndaleyi*, which contained 1.6% protein. Conversely, sorghum *Ndaleyi* had the lowest protein content at approximately 1.21%. Sorghum bran flour from millet was found to be richer in protein compared to its *Ndaleyi* counterpart. The carbohydrate content was notably higher in both millet and sorghum *Ndaleyi* compared to other nutrients like ash, fat, and fiber. Additionally, a significant loss of nutrients, particularly protein, fat, and ash, was observed during the production of *Ndaleyi*.

Millet Chir flour had a higher protein content than sorghum Chir flour, registering at 1.71% compared to the latter's lower value. The ash content in millet Chir was also higher, recorded at 0.67%. Millet Chir flour's moisture content was lower than that of sorghum Chir, due to a continuous three-day drying process. Millet bran, or the overtail, had a lower protein content at 1.55% compared to sorghum bran flour's 3.64%. However, millet bran flour had a higher moisture content at 3.98% compared to sorghum bran flour's 3.34%. The carbohydrate content in millet bran flour was significantly higher at 94.19%, attributed to the high starch content in cereals. Both millet Chir and sorghum bran flours showed higher nutrient content compared to *Ndaleyi*, which was affected by prolonged processing and fermentation. These processing techniques resulted in a reduction of pericarp, endosperm, bran, and other anti-nutritional factors like oxalates and tannins, as noted by Joseph et al. (2021).

Table 5: Effects of Fermentation on the Proximate Composition of *Ndaleyi* Samples

Sample Code (2)	Parameters (g/100mg) (1)						
	Crude Protein (%)	Crude Fats (%)	Crude Fibre (%)	Ash (%)	Moisture (%)	Carbohydrate (%)	Energy value (kcal)
MNF	1.60 ^d ±5.4E-03	0.48 ^b ±0.24	0.003 ^a ±3.0E-03	0.65 ^{ab} ±0.02	32.95 ^e ±0.01	94.08 ^a ±0.11	389.44 ^b ±0.87
MCF	1.71 ^c ±5.4E-03	0.31 ^c ±0.02	0.002 ^a ±1.E-03	0.67 ^a ±0.05	3.45 ^c ±5.4E-03	94.09 ^a ±0.05	394.54 ^a ±0.99
MBF	1.55 ^e ±5.4E-03	0.30 ^c ±0.02	0.002 ^a ±1.0E-03	0.67 ^a ±0.01	3.98 ^a ±0.01	94.19 ^a ±0.05	385.62 ^c ±0.96
SNF	1.22 ^e ±0.02	0.23 ^c ±0.01	0.003 ^a ±3.0E-03	0.62 ^{bc} ±0.01	3.82 ^b ±0.01	94.14 ^a ±0.03	383.42 ^f ±0.95
SCF	3.65 ^b ±0.02	0.77 ^a ±0.01	0.002 ^a ±1.0E-03	0.62 ^c ±0.02	3.14 ^f ±0.01	92.07 ^b ±0.07	388.42 ^c ±0.96
SBF	3.64 ^f ±0.01	0.56 ^b ±0.02	0.003 ^a ±3.0E-03	0.43 ^d ±0.01	3.34 ^d ±1.E-04	91.96 ^b ±0.05	387.44 ^d ±0.95

1. Values are means ± standard deviations of triplicate determinations in any column, means bearing similar superscripts are not significantly different (P>0.05)
2. MNF = Millet *Ndaleyi* Flour; MCF = Millet Chir Flour; MBF = Millet Bran Flour
SNF = Sorghum *Ndaleyi* Flour; SCF = Sorghum Chir Flour; SBF = Sorghum Bran Flour
3. Energy value in Kcal

Effects of Fortification on the Proximate Composition of *Ndaleyi* Samples

The results of the nutrient content in the proximate composition of fortified millet *Ndaleyi* and sorghum *Ndaleyi* flour, for both dehulled and unde-hulled samples with soybean added in specific ratios, demonstrated significant changes across all nutrients, including protein, fats, fibre, ash, and carbohydrate content, as outlined in the proximate composition table on April 14. Fortification of dehulled millet with 10% soybean flour led to an increase in protein content to 15.167%. Similar increases were observed in fats (1.57%), fibre (0.89%), and ash (2.41%), while carbohydrates decreased to 72.13% after dehulling, as reported by Oghaei and Prakash (2016).

Conversely, sorghum exhibited a notable increase in protein content to over 17.62% after fortification with 25% soybean flour. However, there was a reduction in fat (1.41%), fibre (1.22%), and ash (2.73%) content in the dehulled and fermented sorghum samples, despite the fortification. The unde-hulled sorghum, fortified with soybean flour, showed only minor variations in carbohydrate content. These findings are consistent with Nkama's (1994) research.

The fiber content in defatted soybean flour was notably higher at 4.05% compared to the fortified *Ndaleyi* samples of both dehulled and unde-hulled ratios (75:25), which had less than 2% fibre content. This difference is attributed to the non-removal of bran, germ, and pericarp in the samples (Prakash and Oghaei). Additionally, fortification with 10% soybean flour in 90% dehulled sorghum *Ndaleyi* flour resulted in a higher protein value of 17.62% compared to the unde-hulled sorghum *Ndaleyi* flour fortified with 25% soybean flour, which had a lower protein content of 16.19%. The ash content of 2.20% in dehulled millet *Ndaleyi* at a ratio of 75%:25% soybean fortification was lower than all other fortifications for both dehulled and unde-hulled millet and sorghum *Ndaleyi* flours, attributed to the high fibre content in millet as noted by Kent (1993).

Table 3: Effects of Fortification on the Proximate Composition of Ndaleyi samples

Sample Code (2)	Parameter (%) (1)						
	Crude Protein	Crude Fats	Crude Fibre	Ash	Moisture	Carbohydrate	Energy value
MNSO Dehulled (90:10)	16.17 ^c ±0.02	1.58 ^b ±0.04	0.89 ^d ±0.04	2.41 ^c ±0.03	6.24 ^c ±0.03	72.14 ^b ±0.08	369.19 ^b ±0.66
MNSO (undehulled (75:25)	15.16 ^d ±0.02	1.33 ^c ±0.02	0.82 ^e ±1.E-02	2.21 ^d ±0.02	6.67 ^b ±0.06	73.70 ^a ±0.14	369.07 ^b ±0.86
SNSO Dehulled (90:10)	16.19 ^c ±0.05	1.65 ^b ±0.11	1.15 ^c ±0.01	2.59 ^b ±0.06	6.79 ^b ±0.23	71.40 ^c ±0.07	367.51 ^d ±0.79
SNSO (undehulled (75:25)	17.63 ^b ±0.07	1.41 ^c ±0.02	1.23 ^b ±0.02	2.73 ^b ±0.16	6.14 ^c ±0.03	70.62 ^d ±0.16	368.09 ^c ±0.80
DSOF	34.85 ^a ±0.17	8.18 ^a ±0.05	4.05 ^a ±0.07	6.33 ^a ±0.05	7.12 ^a ±0.02	39.23 ^e ±0.12	411.96 ^a ±0.97

1. Values are means ± standard deviation of triplicates determination in any column, means bearing similar superscripts are not significantly different ($P \geq 0.05$)
2. MNSO = Millet Ndaleyi and Soybean (90:10) dehulled
MNSO = Millet Ndaleyi and Soybean (75:25) undehulled
SNSO = Sorghum Ndaleyi and Soybean (90:10) dehulled
SNSO = Sorghum Ndaleyi and soybean (75:25) undehulled
DSOF = Defatted Soybean Flour
3. Energy Value in kcal

Bacterial Count of Ndaleyi Chir and Bran from Fermented Millet Flour

The results displayed in Table 3 indicated the total bacterial, coliform, E. coli, Salmonella, and Staphylococcus aureus counts in cfu/g for samples of Ndaleyi, Chir, and bran from millet flour, respectively. The total bacterial counts ranged from 6.32×10^4 cfu/g for millet Ndaleyi, 7.41×10^4 cfu/g for Chir, and 7.94×10^4 cfu/g for bran, showing an increasing trend. Bran exhibited the highest bacterial counts compared to Chir and Ndaleyi, likely due to the outer covering of the grain, which harbors more dirt, germs, and other microorganisms.

Coliform counts, measured in cfu/g, were highest in Ndaleyi at 6.66×10^1 cfu/g and lowest in bran at 3.4×10^1 cfu/g. The increased coliform levels in Ndaleyi compared to Chir and bran were attributed to the activities of Lactococcus, Leuconostoc, and other bacteria during fermentation. E. coli was more abundant in bran (3.5×10^1 cfu/g) than in Ndaleyi (3.4×10^1 cfu/g) and Chir (2.26×10^1 cfu/g), though all values were below the WHO standard of 20 cfu/g. Salmonella was found in Chir (0.333×10^1 cfu/g) and bran (0.3×10^1 cfu/g) but was undetected in Ndaleyi. Staphylococcus aureus was not detected in Ndaleyi and Chir but was present in trace amounts in bran (0.5×10^1 cfu/g), possibly due to contamination from the outer layers of the grain during processing and fermentation.

Table 4.: Bacterial Counts of Ndaleyi, Chir and Bran from Fermented Millet Flour (1)

Sample Code (2)	Parameter (cfu/g)				
	Total Bacterial Count	Coliform Count	E.coli count (fu/g)	Salmonella Count	S.aureus count
MNF	6.33x10 ^{4c} ±0.01	6.00x10 ^{1a} ±0.10	2.27x10 ^{1b} ±0.05	ND	ND*
MCF	7.42x10 ^{4b} ±0.16	4.67x10 ^{1b} ±0.15	3.40x10 ^{1a} ±0.10	0.33x10 ^{1a} ±0.06	ND*
MBF	7.94x10 ^{4a} ±0.03	3.40x10 ^{1c} ±0.10	3.50x10 ^{1a} ±0.10	0.30x10 ^{1a} ±0.10	0.50x10 ¹ ±0.10

1. Values are means ± standard deviations of triplicate determination in any column, means bearing similar superscripts are not significantly different (P≥0.05)
2. MF = millet flour; SF = sorghum flour; SOF = soybean flour
3. ND* = Not Detected

Bacterial Count of Ndaleyi, Chir, and Bran from Fermented Sorghum Flour

Table 5 presented the bacterial counts, including total coliform, E. coli, Salmonella, and Staphylococcus aureus, measured in cfu/g for Ndaleyi, Chir, and bran samples from fermented sorghum flour. The highest total bacterial count was observed in Chir, at 8.44x10⁴ cfu/g, followed by Ndaleyi at 7.63x10⁴ cfu/g, and bran at 5.90x10⁴ cfu/g. The elevated bacterial count in Chir was noted, while bran showed the lowest count, likely due to the extended fermentation period of 14 days used in Ndaleyi production.

The coliform count was highest in Ndaleyi, at 6.50x10¹ cfu/g, compared to bran, which had the lowest count at 0.4x10¹ cfu/g, though all counts were within acceptable tolerance levels. Chir had a coliform count of 4.66x10¹ cfu/g, falling between the levels found in Ndaleyi and bran. For E. coli, both bran and Ndaleyi exhibited similar counts of 2.466x10¹ cfu/g, while Chir had a lower count of 1.6x10¹ cfu/g. Salmonella and Staphylococcus aureus were minimally present in Ndaleyi, with counts of 0.233x10¹ cfu/g, while both were not detected in Chir and bran samples.

Table 5: Bacterial counts of Ndaleyi, Chir and bran from fermented sorghum flour

Sample Code (2)	Parameters (cfu/g) (1)				
	Total Bacterial Count	Coliform Count	E. coli count (fu/g)	Salmonella Count	S. aureus count
SNF	7.63x10 ^{4b} ±0.02	6.57x10 ^{1a} ±0.15	2.47x10 ¹ ±0.06	0.23x10 ¹ ±0.05	0.51x10 ¹ ±0.06
SCF	8.44x10 ^{4a} ±0.10	4.40x10 ^{1b} ±0.10	1.60x10 ¹ ±0.10	ND*	ND*
SBF	5.90x10 ^{4b} ±0.01	2.46x10 ^{1a} ±0.15	2.40x10 ¹ ±0.10	ND*	ND*

1. Values are means ± standard deviations of triplicate determination in any column, means bearing similar superscripts are not significantly different (P≥0.05)
2. SNF = sorghum Ndaleyi flour; SCF = Sorghum Chir flour; SBF = sorghum bran flour
3. ND* = Not Detected

Bacterial Counts of Fortified Ndaleyi with Soybean in Certain Ratios for Both Dehulled and Undehulled Sorghum Grain Flour

The results from the bacterial count analysis revealed that the total bacterial counts in the fortified millet samples varied depending on the fortification ratios. For the millet fortified with 90% millet and 10% soybean, the bacterial count was 6.33x10⁴ cfu/g. In comparison, the bacterial count for millet fortified with 75% millet and 25% soybean was slightly higher at 6.58x10⁴ cfu/g. The increased bacterial count in the undehulled millet was attributed to the

presence of bran, which can contribute to contamination. Dehulled sorghum flour showed a higher bacterial count of 6.95×10^4 cfu/g at the 90% sorghum and 10% soybean fortification level, which was greater than that of the dehulled sorghum at 75% sorghum and 25% soybean fortification. This increase was due to the reduced bran, husk, and pericarp resulting from the dehulling process. All recorded bacterial counts were within the acceptable contamination level, which should not exceed 25 cfu/g in 1000 liters.

Regarding coliform counts, the undehulled sorghum fortified Ndaleyí at a 75% sorghum and 25% soybean ratio exhibited a higher coliform count of 7.40×10^1 cfu/g compared to the dehulled sorghum Ndaleyí at a 90% sorghum and 10% soybean ratio, which had a coliform count of 5.70×10^1 cfu/g. Additionally, the fortified Ndaleyí with 75% millet and 25% soybean had a coliform count of 3.53×10^1 cfu/g, higher than the dehulled millet at a 90% fortification level, which had a count of 2.6×10^1 cfu/g. All these coliform counts remained within the acceptable contamination level, which is less than 100 cfu/g.

Escherichia coli counts showed that undehulled millet fortified at a 75% level had a higher count of 3.26×10^1 cfu/g compared to the dehulled millet at a 90% fortification level, which had a count of 2.65×10^1 cfu/g. Similarly, sorghum Ndaleyí at a 75% fortification level had an E. coli count of around 3.40×10^1 cfu/g, compared to the millet fortification at a 90% level, which had a total count of 2.36×10^1 cfu/g. Salmonella counts were very low in the dehulled millet fortified at 90% with 10% soybean, at 0.26×10^1 cfu/g. Salmonella was not detected in the undehulled millet at a 75% fortification level or in the undehulled sorghum Ndaleyí at 75% sorghum and 25% soybean fortification level. However, it was detected at 0.366×10^1 cfu/g in dehulled sorghum Ndaleyí at a 90% sorghum and 10% soybean fortification level. Staphylococcus aureus was detected in dehulled millet Ndaleyí at a 90% fortification level with a count of 0.2667×10^1 but was not detected in undehulled sorghum at a 75% sorghum and 25% soybean fortification level.

Table 6: Bacterial Count of Fortified Ndaleyí with the soybean in certain ratios for both dehulled and undehulled millet and sorghum grain flour

Sample code (2)	Parameter (cfu/g) (1)				
	Total Bacterial Count	Coliform Count	E. coli count	Salmonella Count	S.aureus count
DMSO (90:10)	$6.33 \times 10^{4b} \pm 0.01$	$2.60 \times 10^{1d} \pm 0.10$	$2.60 \times 10^{1b} \pm 0.10$	$0.27 \times 10^b \pm 0.55$	0.26×10
UMSO (75:25)	$6.56 \times 10^{4b} \pm 0.03$	$3.53 \times 10^{1c} \pm 0.06$	$3.27 \times 10^{1a} \pm 0.05$	ND	ND*
DSSO(90:10)	$6.95 \times 10^{4a} \pm 0.03$	$5.70 \times 10^{1c} \pm 0.10$	$2.36 \times 10^{1c} \pm 0.05$	$0.36 \times 10^a \pm 0.05$	$0.36 \times 10^1 \pm 0.05$
USSO (75:25)	$2.36 \times 10^{4c} \pm 0.01$	$7.40 \times 10^{1a} \pm 0.10$	$3.46 \times 10^{1a} \pm 0.05$	ND*	ND*

1. Values are means \pm standard deviations of triplicate determination in any column, means bearing similar superscripts are not significantly different ($P \geq 0.05$)
2. DMSO = dehulled millet sorghum flour; UMSO = undehulled millet sorghum flour; DSSO = dehulled sorghum soybean flour
USSO = undehulled sorghum soybean flour
3. ND* = Not Detected

CONCLUSION AND RECOMMENDATIONS

The proximate composition of Ndaleyí made from both millet and sorghum was found to be low in protein, fats, fiber, ash, and moisture content, as shown in Table 4.10, but high in carbohydrate

content. In contrast, Chir and bran (overtail) were richer in protein compared to Ndaleyí made from both types of flour. The functional properties, such as oil absorption capacity, bulk density, water absorption capacity, swelling capacity, gelatinization, and viscosity, were higher in Ndaleyí but lower in Chir, with bran exhibiting higher values. Traditional Ndaleyí was deficient in most B-vitamins, unlike Chir and bran. The fortified Ndaleyí, enriched with 10% and 25% soybean flour, was notably rich in phosphorus, calcium, and potassium, while magnesium, manganese, sodium, iron, and zinc were generally low, except for defatted soybean flour, which had high sodium and iron content. The proximate composition of fortified Ndaleyí showed increased protein levels but lower fats, fiber, and ash. Carbohydrate content remained very high. The fortified Ndaleyí exhibited high oil absorption capacity, water absorption capacity, and viscosity (all above 50%), while bulk density, swelling capacity, and gelatinization was relatively low.

Recommendations

The research work reveals the nutritional significance of Ndaleyí, fortified Ndaleyí and products: Dehulling of cereals and legumes reduces the number of micronutrients e.g., magnesium, phosphorus, calcium, potassium, sodium, iron and zinc. Dehulling of cereals and legumes should be discouraged in Ndaleyí processing. Undehulled cereals and legumes contains high amount of protein, fats, crude fibre, ash and carbohydrates than the dehulled samples, it is also recommended that dehulling of cereals and legumes should be discouraged to safeguard the availability of this essential nutrients in Ndaleyí processing.

Ndaleyí food product is highly recommended for a patient of celiac disease (protein intolerance) because of its lower protein content. Fermentation, had been reported to improve sensory and nutritional quality of cereals, because of the lactic acid bacteria that participated in the process which decreases carbohydrate content increase in amino acids and B-group vitamin contents, i.e. leucine, arginine, alanine, aspartic acid, glycine and lysine.

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