

# ASSESSMENT OF THE ANTI-NUTRITIONAL FACTORS, MICROBIAL AND FUNCTIONAL PROPERTIES OF FORMULA PRODUCED FROM MILLET, SOYA BEAN AND BAOBAB LEAF

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# ABSTRACT

The development of nutrient rich food is essential to combating the global issues of food insecurity and malnutrition, especially in developing nations. However, anti-nutritional factors (ANFs) and microbial contamination that might impair nutrient absorption and bioavailability frequently restrict the use of these basic materials. This study aims to evaluate the anti-nutritional characteristics, microbiological safety, and functional gualities of a formula produced from millet and soy beans and baobab leaf. The samples; millet (M), soya bean (SB), and baobab leaf (BL) were obtained from Gamboru Market, Maiduguri, Borno State, Nigeria. The samples were grouped as Blend 1(40% M: 40% SB: 20% BL), Blend 2 (20% M: 60% SB: 30% BL) Blend 3 (10% M: 30% SB: 60% BL), Blend 4 (20% M: 20% SB: 60% BL) and Blend 5 (40% M: 30% SB: 30% BL). The samples were assayed for functional properties and anti-nutritional factors using standard methods. Results obtained were subjected to statistical analysis using SPSS version 23.0. Duncan multiple range was used to compare the means The results were considered significant at ( $p \le 0.05$ ). The results of the bulk density of the samples revealed that Blend 2  $(1.25\pm0.01)$ , Blend 3  $(1.25\pm0.01)$ , unprocessed baobab leaves (1.25±0.01), processed baobab leaves (1.25±0.01) had highest bulk density than rest of the food samples. Based on the result obtained, for the Blend 1, Blend 2, Blend 3, Blend 4 and Blend 5, the viscosity measurements were 96.00±0.58, 96.00±0.58, 492.00±0.95, 528.00±0.76 and 288.00±0.84 respectively. While for the unprocessed millet, processed millet, unprocessed soya bean, processed soya bean, unprocessed baobab leaves and processed baobab leaves the viscosity measurements were 72.00±0.58, 84.00±0.58, 114.00±0.73, 60.00±0.58, 3624.00±2.77 and 2022.00±1.56 respectively. Processing

drastically reduced the antinutritional factors (Tannin, Phytate and oxalate) of the entire samples, while for the blends, low antinutritional factors were observed. Microbial analysis did not significantly alter the microbial load of the sample. However, all the microbial isolated from these samples were below the WHO limits. The study underscore the potentials of the formulated food blends based on millet, soybean, and baobab leaves as nutrient-rich, functional foods with acceptable microbiological safety and less anti-nutritional elements. This result demonstrates that the formulated blends could serves as viable strategy for combating food insecurity and malnutrition.

**Keywords:** Anti-Nutritional Factors; Millet, Soya Bean; Baobab Leaf; Microbiological Safety.

# INTRODUCTION

Traditional crops with great potential as sustainable and reasonably priced food sources include millet, soy beans, and baobab leaves. Soya beans are a great source of protein and bioactive substances, while millet is a hardy grain high in carbs and vital minerals. Because of their rich vitamin, mineral, and antioxidant content, baobab leaves have long been employed for their health-promoting gualities. Legumes and cereals have been crucial in lowering the global malnutrition issue. They are employed as a main element in the creation of starch- and protein-based food formulations because they are abundant sources of carbs, fats, and proteins. Different cereals and legume flours have different molecular structures, biochemical compositions, food component conformations, and functional characteristics.

Millet (*Pennisetum glaucum*) is group of cereal that belongs to the *poaceae* family which is widely consumed and contains major and minor nutrients in remarkable amount (Himanshu *et al.*, 2018). Millets provide more essential amino acids than most other cereal. Millets are a group a small, round whole grains grown in India, Nigeria, and other Asian and African countries. Considered an ancient grain, they are used both for human consumption and livestock and bird feed. They have multiple advantages over other crops, including drought and pest resistance. They're also able to survive in harsh environments and less fertile soil. These benefits stem from their genetic composition and physical structure for example, its small size and hardness (Rathore *et al.*, 2019). Although all millet varieties belong to the Poaceae family, they differ in colour, appearance, and species. This

crop is also divided into two categories – large and small millets, with major millets being the most popular or commonly cultivated varieties (Chandra et al., 2020). Soya bean constitute an important component of the traditional diets of many people particularly in Asia and Africa (Ali *et al.*, 2020). Soya bean are valued because of their high protein and fat contents (Asgar *et al.*, 2010). Soya bean seeds contain on average 40 – 41% protein on a dry matter basis although a range of 33.2 to 51.3% protein in some genotypes has been reported. Soya bean proteins contain all amino acids essential to human nutrition; hence soya bean's protein quality is regarded as almost equivalent to animal sources. The deciduous baobab tree (Adansonia digitata) is indigenous to the Middle East, Australia, and Africa. Baobab leaves are very nutrient-dense and have been used traditionally to cure a variety of illnesses, including malaria, TB, anemia, diarrhea, and toothaches. The bark, seed, and fruits are also medicinal (Kornei et al., 2021). Even in urban regions, the impoverished cannot afford the high cost of iron-rich supplements, which are particularly scarce in rural areas. The iron content of locally accessible raw materials, such as cereal, legumes, and vegetables, is particularly high when combined into a single recipe. This can be used to treat anemia in children and even in elderly people who are susceptible to the condition due to ignorance of the readily available, reasonably priced, and locally accessible iron-rich sources. Thus, the creation of an iron-rich formula is the goal of this effort in order to manage nutritional anemia, specifically iron-deficiency anemia (Fransceshi et al., 2019). Antinutritional factors (ANFs) are biological compounds found in food that can decrease the uptake or use of nutrients, resulting in decreased metabolic and gastrointestinal functioning. Edible crops contain important anti-nutritional components such as goitrogens, lectins, protease inhibitors, amylase inhibitors, phytic acid, and gossypol. When combined with nutrients, anti-nutritional factors become the main cause for worry due to decreased nutrient bioavailability. Other variables that decrease protein digestibility and mineral absorption include trypsin inhibitors and phytates, which are typically found in cereals and legumes. One important element that lowers the bioavailability of different components of cereals and legumes is anti-nutrients. These substances may result in mineral deficits and micronutrient malnutrition. Developing safe and nutrient-balanced food substitutes requires an understanding of these constituents' functional characteristics and how they work together in prepared goods. Cereals and legumes are rich in macronutrients and micronutrients, but they also contain anti-nutritional elements. The study was conducted to determine

the microbial, functional properties, and anti-nutritional factors of formulas produce from millet, soya bean and baobab leaf.

# MATERIALS AND METHODS

#### **Materials**

All chemicals and reagents used in the study are of analytical grades.

## Sample Collection

The Millet, soya bean, and baobab leaf were bought at Gamboru Market in Maiduguri, Borno state.

## Methods

## Sample Preparation

The samples were sorted to get rid of foreign matters, stored in an air tight container at ambient temperature, until analysis.

# Sample Preparation

## Millet:

Ten kilogram (10 kg) of millet sample was sorted for stones, dirt, insects and defects. The grains were cleaned. The millet was soaked in two litres (2L) of distilled water for 72 hours for fermentation to take place. The fermented grains were then transferred to a grease-free tray and sun-dried for 2 days. The grains were milled and a sieve of 5 mm pore was used to remove the larger bits which was obtained was stored in an airtight container until use (Oladeji, 2014).

## Soya Bean

About 300g of soya bean seeds was cleaned, roasted, and grinded and stored in an air tight container at room temperature for further use (Eboagu, *et al.*, 2020).

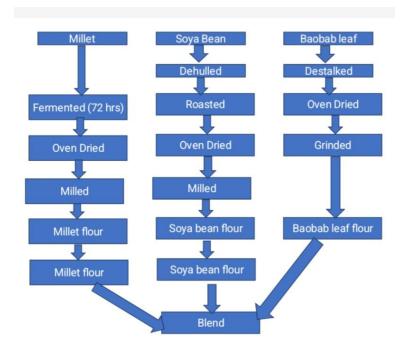
## Baobab Leaf

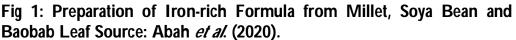
The leaves were soaked, washed, sun-dried, de-stalked, and grinded and sieved into a fine powder using a 3mm sieve. (Gebauer *et al.*, 2002).

I able 1.							
Samples (%)	Formula 1	Formula 2	Formula 3	Formula 4	Formula 5	Control	
Millet	40	20	10	20	40	-	
Soya Bean	40	60	30	20	30	-	
Baobab Leaf	20	20	60	60	30	-	
Total	100	100	100	100	100	100	

## Table 1: Production of the Formula

Key: Formula 1= 40% Millet: 40 Soya Bean: 20% Baobab Leaf Formula 2= 20% Millet: 60 Soya Bean: 30% Baobab Leaf Formula 3= 10% Millet: 30 Soya Bean: 60% Baobab Leaf Formula 4= 20% Millet: 20 Soya Bean: 60% Baobab Leaf Formula 5= 40% Millet: 30 Soya Bean: 30% Baobab Leaf





## Determination of Functional Properties (AOAC 2017) Determination of Viscosity

Viscosity was determined by placing twenty grams of the sample in a measuring cylinder of 100 ml of water in boiling water bath of 75-80°C. The

slurry was constantly stirred until boiling and was continued for five minutes. The slurry was cooled to room temperature 23°C and the viscosity was measured using canon viscometer.

# Bulk Density (BD)

Fifty grams (50 g) of the sample was introduced into a 100 ml graduated cylinder. The cylinder was tapped 40 to 50 times and the bulk density was calculated as weight per unit volume of sample.

wt. of flour  $BD = \frac{1}{Volume of the bulk flour}$ 

# Determination of Water Absorption Capacity

Eighty (80ml) of tap water at 28°C and 50°C were added to Ten grams (10g) of the sample. Pulverized extrudates allowed standing for one hour in a One Hundred and Fifty (150ml) beaker.

The hydrated extrudates was collected by inverting the beaker over a 20 mesh screen (Bs) for 60sec. The percentage hydration was defined as %hydration =  $\frac{wet \ sample - initial \ weight}{x \ 100}$ 

initial weight

#### **Determination of Antinutritional Factors: Determination of Tannins Content**

The tannin content was determined using Vanillin Hydrochloric Acid quantitative method. The sample (0.25g) was introduced into Erlenmeyer flask and 10ml of 4% HCl in methanol was also added to the mixture; the flask was closed with paraffin wax. The flask was shaken for 20 minutes on a wrist action shaker centrifuged for ten minutes at 4500 rpm. One gram (1q) of the extract was taken and 1.0 ml of 1.0 % vanillin was added, and 0.5 ml of concentrated HCl phenol standard was prepared and five different test tubes were labelled as 1, 2, 3, 4 and 5; 0.1, 0.3, 0.5, 0.7 and 1.0 ml of phenol reagent was introduced into the tubes respectively. The volume of the test tubes was made up to 1.0 ml with 8 % HCl in methanol and 0.1ml of 1% vanillin and 0.5 ml of concentrate and made up to volume to 5.5 ml with 4 % HCl in methanol. Blank sample was prepared using five mills of 4% HCI in methanol instead of the extract. The absorbance of standard solutions, sample extracts and sample blank were taken in the spectrophotometer at five hundred (500 nm) exactly for twenty minutes into incubation.

Calculations:

AU/CU Astd/Cstd =

Cu

= Au ×C<sub>std</sub> = mg/g Astd

Where AU = Absorbance of unknown

- Astd = Absorbance of standard
- CU = Concentration of unknown
- Cstd = Concentration of standard

#### Determination of Phytate Content

Four grams (4 g) of the sample was soaked in 100 cm<sup>3</sup> of 2 % HCl for 3 hours and then filtered through two layers of filter paper. The filtrate (25 cm<sup>3</sup>) was placed in 250 cm<sup>3</sup> conical flask and 5cm<sup>3</sup> of 0.3 % NH<sub>4</sub>SCN solution was added as an indicator. Distilled water (25.0 cm<sup>3</sup>) was added to reach the proper acidity. This mixture was titrated against FeCl<sub>3</sub> solution, which contains about 0.00195 g of Fe per cm<sup>3</sup> of FeCl<sub>3</sub> solution. The result was multiplied by factor 1.95 to obtain Phytate P. Phytate P result was multiplied by factor 3.55 to convert to Phytate.

#### **Determination of Oxalate Content**

Total Oxalate was extracted by three sequential extractions from each sample with 5% HCI acid according to the method of Oxalate in the pooled supernatants was then determined by high-pressure liquid chromatography (HPLC) (25). Intra-assay variability of the Ox assay was 4.7%. Purified NaOx (Sigma) was the standard (Ibis *et al.*, 2020).

#### Microbial Analysis

The pour plate method was used to determine the load count. One gram of each of the formulations was suspended into 9ml of sterile distilled water in a Mac Cartney bottles to give 10–1 dilution. Serial dilutions were made up to 10–3. Each diluents of the samples were plated out in duplicate using the pour plating technique by transferring 1ml from each Mac Cartney bottle into 2 different Petri dish and pouring 15ml of the nutrient agar media on each sample. Incubation of microorganism was done in an aerobic incubator for 48 h at 37°C. After incubation period the colonies appearing on the agar plates were counted using a colony counter (Gallenkamp colony counter, U.K). The average colony obtained from the

countable duplicate plates, were expressed as colony forming unit per gram (Naibaho *et al.*, 2023).

## Data Analysis.

During analysis, all the data measurements and recordings were carried out in triplicates, and the results were expressed as mean ± SEM. The SPSS 23.0 for Windows Computer Software Package was used for the Analysis of Variance (ANOVA) (Desai *et al.*,2023), followed by Duncan's multiple ranges to compare means.

# RESULTS

## **Physical properties**

The results of viscosity of the food samples are shown in Table 3, based on the result obtained, for the Formula 1, Formula 2, Formula 3, Formula 4 and Formula 5 were 96.00±0.58, 96.00±0.58, 492.00±0.95, 528.00±0.76 and 288.00±0.84 respectively. While for the unprocessed millet, processed millet, unprocessed soya bean, processed soya bean, unprocessed baobab leaves and processed baobab leaves the viscosity measurements were 72.00, 84.00±0.58, 114.00±0.73, 60.00±0.58, 3624.00±2.77 and 2022.00±1.56 respectively. However, from the results, the viscosity of Formula 3 and Formula 4 are higher than that of Formula 1, Formula 2 and Formula 5, hence Formula 3 and Formula 4 are more concentrated than Formula 1, Formula 2 and Formula 5. A significant (P<0.05) increase in the viscosity of unprocessed baobab leaves and processed baobab leaves compared to soya bean and millet both in the raw and processed forms. Furthermore, significant (P<0.05) difference existed among the food samples. A food sample or fluid with large viscosity resists motion because its molecular makeup gives it a lot of internal friction whereas a fluid with low viscosity flows easily because its molecular makeup results in very little friction when it is in motion. The results of the bulk density of the samples as shown revealed that Formula 2 (1.25±0.01), Formula 3 (1.25±0.01), unprocessed baobab Leaves (1.25±0.01), processed baobab leaves (1.25±0.01) had highest bulk density than rest of the food samples. However, according to the results listed in Table 2 there was significant difference (P > 0.05) between bulk densities of the samples. The general low bulk density of the samples could be an advantage in formulation of baby foods, where a high nutrient density to low bulk density is desired. The results of water absorption capacity of the samples are shown in Table 2. The results obtained showed that the water absorption capacity for Formula 1, Formula 2, Formula 3, Formula 4 and Formula 5, were

1.20 $\pm$ 0.06, 0.85 $\pm$ 0.06, 5.10 $\pm$ 0.06, 3.40 $\pm$ 0.76 and 2.60 $\pm$ 0.06 respectively. While for the unprocessed millet, processed millet, unprocessed soya bean, processed soya bean, unprocessed baobab leaves and processed baobab leaves the viscosity measurements were 3.30 $\pm$ 0.06, 2.10 $\pm$ 0.06, 3.10 $\pm$ 0.06, 2.40 $\pm$ 0.06, 2.08 $\pm$ 0.06 and 2.20 $\pm$ 0.06 respectively. However, significant (P<0.05) difference existed among the samples. The functional properties of the formulas are presented in Table 3. No significant (P<0.05) difference was observed in the viscosity of Formula 1 and 2. Formula 4 fad the highest viscosity level, followed by formula 3, while die 1 ad 2 recorded the lowest viscosity level. Bulk density of formulas 3, 4 and 5 did not show any significant (P>0.05). A significant (P<0.05) decrease was observed in the water absorption capacity of Formulas 1, 3, 4 and 5 when compared to the WAC of formula 2. No significant (P>0.05) difference was observed in the viscosity of Formulas 1, 3, 4 and 5 when compared to the WAC of formulas 1, 3, 4 and 5.

				Components				
	Millet		Soya Beans		Baobab Leaf			
parameters	Raw	Processed	Raw	Processed	Raw	Processed		
Viscosity (Pi)	72.00±0.58°	84.00±0.58°	114.00±0.73 <sup>b</sup>	60.00±0.58°	3624.00±2.77 <sup>d</sup>	2022.00±1.56°		
Bulk Density (g/ml)	1.16±0.01°	1.22±0.01°	1.22±0.01°	1.22±0.01 <sup>ª</sup>	1.25±0.01°	1.25±0.01°		
Water Absorption Capacity (g/cm <sup>3</sup> )	3.30±0.06 <sup>b</sup>	2.10±0.06°	3.10±0.06°	2.40±0.06ª	2.08±0.06°	2.20±0.06°		

 Table 2: Functional Properties of Unprocessed and Processed Millet, Soya

 Bean and Baobab Leaf.

Data are expressed as mean  $\pm$  SEM, of three determinations. Values with different superscripts horizontally along the row are significantly different. (P < 0.05).

parameters	Formula 1	Formula 2	Formula 3	Formula 4	Formula 5
Viscosity (Pi)	96.00±0.30°	96±0.23°	492±0.12 <sup>b</sup>	528±0.23 <sup>b</sup>	288±0.15°
Bulk Density (g/ml )	1.90±0.03ª	1.90 ±0.01°	1.25±0.02 <sup>b</sup>	1.22±0.01 <sup>₅</sup>	1.20±0.01 <sup>b</sup>
Water Absorption Capacity (g/cm <sup>3</sup> )	2.1±0.01°	3.0±0.02 <sup>b</sup>	2.4±0.01°	2.0±0.01°	2.2±0.01°

Table 3: Functional properties of formulated formulas

Data are expressed as mean  $\pm$  SEM, of three determinations. Values with different superscripts horizontally along the row are significantly different. (P < 0.05).

## **Anti Nutrient Content Of The Samples**

The result of the anti-nutritional content of the raw and process samples were presented in Table 4, the Table shows the significant reduction in oxalate tannin and phytate content of all the sample after processing. A percentage reduction of 50.97, 64.50, and 57.66% in oxalate as observed for processed samples of millet, soya beans and baobab leaf respectively. While for tannin, the percentage reduction of 64.05%, 66.79%, and 57.39% for the processed samples of millet, soya beans and baobab leaf respectively. A percentage reduction of 65.60% was observed in millet affect processing while processed soya beans and Baobab leaf recorded a percentage reduction of 64.94% and 62.32% respectively. For the formulated Formula, significant difference was observed in the oxalate content of Formula, 1, 2, and 3, so also in formula 4 and 5. Tannin content of formula 1-4 shows no significant difference. The Phytate content of the formulated formula range between 44.73-68.94mg/100g with the formula 5 having the highest Phytate content while formula 1 has the lowest Phytate content.

Table 4: Antinutrients baobab leaf and its resp		ocessed millet, soya bear uction (mg/100g)	n and
Millet	Soya Beans	Baobab Leaf	

	Ivimet		Soya Beans		Baobab Leal	
parameters	Raw	Processed	Raw	Processed	Raw	Processed
Oxalate	27.7	13.62	461.71	163.71	617.76	261.59
% Decrease	50.97%		64.5%		57.66%	
Tannin	32.93	11.84	24.87	8.26	672.32	286.45
% Decrease	64.05%		66.79%		57.39%	
Phytate % Decrease	185.84 65.60%	63.93	324.16	113.65	47.84 52.32%	22.81
10 Declease	03.00%		64.94%		JZ.JZ /0	

Table 5: Antinutrients of formulated formula (mg/100g)

					<u> </u>	
Parameters	Formula 1	Formula 2	Formula 3	Formula 4	Formula 5	Limit
Oxalates	32.99±0.01°	37.19±0.01°	42.67±0.00°	27.56±0.01 <sup>b</sup>	29.64±0.01°	50
Tannins	20.17±0.01°	21.32±0.01°	23.66±0.01°	24.04±0.01°	28.93±0.00 <sup>b</sup>	150
Phytates	44.73±0.51°	51.48±0.01°	54.67±0.01°	64.04±0.01 <sup>b</sup>	68.94±0.01 <sup>b</sup>	100-400

Data are expressed as mean ± SEM, of three determinations. Values with different superscripts horizontally along the row are significantly different. (P < 0.05).

Key;

Formula 1 = 40% Millet: 40% Soya bean: 20% Baobab leaf Formula 2 = 20% Millet: 60% Soya bean: 30% Baobab leaf Formula 3 = 10% Millet: 30% Soya bean: 60% Baobab leaf Formula 4 = 20% Millet: 20% Soya bean: 60% Baobab leaf Formula 5 = 40% Millet: 30% Soya bean: 30% Baobab leaf

## **Microbial Analysis**

The microbial analysis of the food samples is presented in Table 6. A significant (P<0.05) difference was observed in the microorganisms grown on nutrient agar of the formulations. With highest microorganisms were detected in processed soya, millet and Baobab leaf have the lowest non-fastidious organism. Coliform bacteria were not detected in all the samples while yeast and mould were detected only in processed millet and unprocessed soya.

Parameter	N0. factor	of NO. colony	Of N0. O colony	of CFU/g	microorganism
Processed Soya Bean	10 <sup>7</sup> 10 <sup>7</sup> 10 <sup>7</sup>	10 <sup>7</sup> 10 <sup>7</sup> 10 <sup>7</sup>	41 Absent Absent	4.1×10 <sup>®</sup> Absent Absent	Lactobacillus Yeast and mould Coliform bacteria
Raw Soya Bean	10 <sup>7</sup>	10'	101	1.01×10°a	Lactobacillus
	10 <sup>7</sup>	10'	TFTC	<10	Yeast and mould
	10 <sup>7</sup>	10'	Absent	Absent	Coliform bacteria
Processed Millet	10 <sup>7</sup>	10 <sup>7</sup>	78	7.8×10⁵c	Lactobacillus
	10 <sup>7</sup>	10 <sup>7</sup>	TFTC	<10	Yeast and mould
	10 <sup>7</sup>	10 <sup>7</sup>	Absent	Absent	Coliform bacteria
Raw Millet					
	10 <sup>7</sup>	10'	107	1.07×10°a	Lactobacillus
	10 <sup>7</sup>	10'	Absent	Absent	Yeast and mould
	10 <sup>7</sup>	10'	Absent	Absent	Coliform bacteria
Raw Baobab Leaf	10 <sup>7</sup>	10'	92	92	Lactobacillus
	10 <sup>7</sup>	10'	Absent	Absent	Yeast and mould
	10 <sup>7</sup>	10'	Absent	Absent	Coliform bacteria
Processed Baobab Leaf	10 <sup>7</sup> 10 <sup>7</sup> 10 <sup>7</sup>	10 <sup>7</sup> 10 <sup>7</sup> 10 <sup>7</sup>	106 Absent Absent	1.06×10°a Absent Absent	Lactobacillus Yeast and mould Coliform bacteria

# Table 6: Mean microbial population of unprocessed and processed soya bean, millet and baobab leaf (CFU/g)

abc= Values with same alphabets for same foods and counts across the rows and same counts down the column are not significantly different. Key:

CFU/g = Colony-forming unit per gram, TFTC= Too few to count

# DISCUSSION

# **Functional properties**

# Viscosity

Viscosity of a food sample is a product of attractive forces between the particle that make up the fluid Increase in viscosity due to fermentation of millet could be due starch degradation caused by the action of hydrolytic enzymes ( $\alpha$  and  $\beta$  amylase) that developed during the fermentation process which hydrolyses some of the starch into limit dextrin and maltose, which do not swell when cooked. Fermentation also decreases the total amount of carbohydrate and other nutrients. This reduction could be attributed to possible hydrolysis of complex carbohydrate to simple sugars, which were used for metabolic process. Decrease in viscosity observed in the roasted samples of soya bean and dried sample of baobab leaves might be as a result of the processing method which generated heat that denatured the protein, thereby leading to decrease in the viscosity of the flour. All the food formulas exhibited low viscosity including the control formula. Flour from fermented and sprouted grains can therefore be used in greater amount to give the same viscosity as flour from unfermented grains, thereby giving higher nutrient and energy density. This is in line with earlier work reported by (Dhakal et al., 2023).

# **Bulk Density**

The values for bulk density obtained in this study for formulas including the were higher than those who obtained from bulk density ranging from 0.80 to 0.82 g/ml in durum wheat formulas. Bulk density gives an indication of the relative volume of packaging material required and high bulk density is a good physical attribute when determining mixing quality of a particular matter. The density of processed products dictates the characteristics of its container or package. Product density limits the caloric and nutrient per feed of a child which can result in growth faltering (De *et al.*,2022). However, low bulk density would be an advantage in the formulation of complementary formula. This is because lower bulk density values lead to higher amount of flour particles which can stay together and thus increasing the energy content of such. Therefore, more of the samples could be prepared using a small amount of water yet giving the desired energy nutrient density and semi-solid consistency which can easily be fed to an infant (Dhakal *et al.*, 2023).

# Water Absorption Capacity

The water absorption capacity of food samples is the ability of a food material to take up water under specific conditions. This property is important in various food processing and culinary applications. Water absorption capacity is typically measured as the amount of water absorbed by a unit weight or volume of the food sample under controlled conditions (such as temperature and time). It affects the texture, cooking behaviour, and overall quality of food products. Foods with high water absorption capacity may require more water during cooking or processing (De et al.,2022). The results for the water absorption capacity of the samples as presented above showed that Formula 3 (10% Millet, 30% Soya bean, 60% Baobab leaf) shows the highest water absorption capacity  $(5.10 \text{ g/cm}^3)$ , significantly higher than other formulas. However, Formula 2 (20% Millet, 60% Soya bean, 30% Baobab leaf) has the lowest water absorption capacity among the formulas (0.85 g/cm<sup>3</sup>). Baobab leaves sample (both processed and unprocessed) generally show higher water absorption compared to millet and soya bean. While the processed Baobab leaves exhibit slightly higher absorption (2.20 g/cm<sup>3</sup>) compared to unprocessed (2.08 g/cm<sup>3</sup>). Processed Soya bean sample (2.40 g/cm<sup>3</sup>) absorb more water than unprocessed sample (3.10 g/cm<sup>3</sup>), which is unexpected but may be due to changes in structure or composition during processing as stated by Naibaho et al. (2023). Across formulas involving baobab leaves, there is a trend where higher baobab leaf content correlates with higher water absorption capacity. The millet sample generally absorbs less water compared to baobab leaves and soya bean across all formulations. Therefore, the results suggest that the water absorption capacity varies significantly depending on the composition of the formulas and the processing of the ingredients. Baobab leaves appear to be a major factor influencing water absorption capacity, with higher proportions leading to greater absorption. Processing also impacts absorption differently across ingredients (Mannar et al., 2022). These findings are crucial for understanding how different food components can affect hydration properties, which is essential for dietary formulation and product development. The water absorption capacity (WAC) observed in this study is probably related to the low viscosity patterns from starch granules as reported by S who worked on cookiemaking properties of corn and potato flours, respectively.

## Antinutrients

Antinutrients are natural or synthetic compounds that interfere with the absorption of nutrients, antinutrients may act by binding to vitamins and minerals, preventing their uptake, or inhibiting enzymes. Throughout history, humans have bred crops to reduce antinutrients, and cooking processes have developed to remove them from raw food materials and increase nutrient bioavailability. Phytate is an antinutrient that interferes with the absorption of minerals from the formula. has a strong binding affinity to minerals such as calcium, magnesium, iron, copper, and zinc. This results in precipitation, making the minerals unavailable for absorption in the intestines. Phytic acids are common in the hulls of nuts, seeds, and grains (Dolan et al., 2010). Oxalates are present in many plants and it bind to calcium, magnesium and iron, preventing their absorption in the human body, tanning on the other hand are a group of polyphenolic compounds that chelate metals such as iron and zinc and reduce the absorption of these nutrients. Overall processing appears to activate the reduction of antinutrients as shown in the above Table, making them potentially more nutritious compared to their unprocessed form, each shows variation in antinutritional content, suggesting that the choice of ingredients play a crucial role in the nutritional profile (Mannar et al., 2020). Ingredients high in antinutritional factor may impact nutrient absorption, emphasizing the importance of processing in enhancing the nutritional quality of food source.

# Microbial Analysis

The purpose of the microbiological analysis was to evaluate the items' safety for ingestion by concentrating on the existence of particular bacterial isolates and their biochemical traits. A rise in the number of bacteria in certain samples indicated that microbial activity was still present or that the samples were being stored in a way that allowed for growth. On the other hand, bacterial counts decreased in some samples, suggesting that there could be variables influencing the survivability of the bacteria or inhibiting their growth while they are being stored. The various treatments could have had an impact on the types and numbers of bacteria found in each sample. In food fermentation, Lactobacillus species are essential for the development of flavour, preservation, and the generation of desired metabolites such lactic acid. Their presence in fermented foods is typically linked to human health benefits and safety (Naibaho *et al.*, 2023). This research discusses the nutritional composition of various formula formulations designed to address iron deficiency anaemia. The findings

highlight the importance of ingredients like soya bean, millet, and baobab leaves in managing nutrient deficiencies, particularly iron.

# CONCLUSION

In conclusion, this study demonstrates the feasibility of creating a nutritious formula using baobab leaves notably the inclusion of baobab leaves has significantly enhanced the iron content of the formula, offering a promising alternative for the management of the iron deficiency anaemia.

## **Conflict of Interest**

None

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