

**Effect of partial mutton meat substitution with Bambara groundnut (*Vigna subterranea* (L.) Verdc.) flour on physicochemical properties, lipid oxidation and sensory acceptability of low-fat patties**

by

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## Abstract

Health concerns regarding fat consumption, as well as shifts in customer preference, have prompted substantial studies into low-fat products. This study examined the nutritional, colour, functional, and antioxidants properties of Bambara groundnut (BGN) flour varieties (cream, brown, and red-coated) grains as well as their influence on the physicochemical properties, lipid oxidation, and sensory acceptability of reduced-fat mutton patties. The patties were formulated with 2.5%, 5%, 7.5%, and 10% of BGN flours for each variety, and 100% mutton patties were used as a control. The BGN flours showed significant ( $p < 0.05$ ) differences in their nutritional composition (except for ash content), colour, functional (excluding the emulsion stability), antioxidant properties. The inclusion of BGN flours significantly increased the fibre (0.00 to 0.79%) and ash (1.16 to 1.99%) contents of the formulated mutton patties. However, moisture, protein contents, and carbohydrates decreased. The cooking yield of the formulated patties significantly increased with values ranging from 76.39 to 86.80% but the diameter decreased. The inclusion of BGN flours significantly increased the lightness, hue angle, colour difference and whiteness of patties. Nevertheless, the redness, yellowness, chroma, and yellowness index of the patties decreased. The hardness and resilience of formulated patties significantly increased with values varying from 16.41 to 17.66 N, and from 0.35 to 0.48 J/J, respectively, whereas the springiness, cohesiveness, and chewiness decreased. The lipid oxidation of formulated mutton patties significantly increased from day 7 to 21 but was still less than that of the control sample within storage days. The sensory properties of formulated patties were not significantly different from the control sample and were above the acceptable score of five. Different types of BGN flour varieties can each be utilised as additives in mutton patties without detrimental effects on the quality parameters of the patties.

**Key words:** Bambara groundnut flour, mutton patties, physicochemical properties, lipid oxidation, and sensory acceptability.

## Declaration

I, **Ramatsetse Kgaogelo Edwin**, student number 16014230, hereby declare that this research project for Master of Food Science and Technology under the supervision of Dr. S.E. Ramashia and Mr. M.E. Mashau has been submitted to the Department of Food Science and Technology, Faculty of Science, Engineering and Agriculture at the University of Venda and has not been previously submitted to any other institution for any degree. All citations and sources of information have been duly acknowledged in the text.



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Student signature

15 February 2023

Date

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## List of Abbreviations

a*	Redness of meat
AlCl <sub>3</sub> .6H <sub>2</sub> O	Aluminium chloride hexahydrate
AOAC	Association of Agricultural Chemists
b*	Yellowness of the meat
BGN	Bambara groundnut
BHA	Butylated hydroxyanisole
BHT	Butylated hydroxytoluene
C*	Chroma
CHO	Carbohydrates
CVDs	Cardiovascular diseases
DPPH	1,1-diphenyl-2-picryl hydrazyl radical
Fe <sup>2+</sup>	Ferrous ions
Fe <sup>3+</sup>	Ferric ions
FRAP	Ferric reducing antioxidant power
H <sub>2</sub> SO <sub>4</sub>	Sulphuric acid
L*	Lightness of the meat
NaNO <sub>2</sub>	Sodium nitrite
NaNO <sub>2</sub>	Sodium nitrite
NaOH	Sodium hydroxide
O <sub>2</sub>	Oxygen molecule
OAC	Oil absorption capacity
R•, ROO•	Free radicals
ROOH•, RH, ROOR	Hydroperoxides
TBARS	Thiobarbituric acid reactive substances
TBHQ	Tertbutyl hydroquinone
TFC	Total flavonoids content
TPC	Total phenolic content
WAC	Water absorption capacity
WI	Whiteness index
YI	Yellowness index
ΔE	Colour difference

## 1 CHAPTER 1: INTRODUCTION

### 2 1.1 Background

3 Meat is a nutrient-dense food made up of water, proteins, lipids, vitamins, and minerals, as well  
4 as little amount of carbohydrates (Aminzare *et al.*, 2019). Mutton meat is from an adult sheep,  
5 and it is less tender. It is India and South Africa's most common meat, owing to its lack of social  
6 and religious restrictions (Mendiratta *et al.*, 2013; Mazhangara *et al.*, 2022). This is the primary  
7 motive why mutton meat is still more expensive than other meat. Nevertheless, consuming a lot  
8 of processed meat has negative effects such as causing cardiovascular disease or colon cancer  
9 (Willett *et al.*, 2019; Grosso *et al.*, 2022). However, adding non-meat additives such as soybeans,  
10 eggs, durum wheat powder, potato mash, rice, and milk products are widely utilised as extenders,  
11 binders, and fillers in minced meat products may be beneficial in preventing nutrition-related  
12 disorders and for the mental-physical wellbeing, as well as increase quality features and minimise  
13 production losses (Yang, 2019; Possidonio *et al.*, 2021; Owusu-Ansah *et al.*, 2022).

14 Consumers are currently very concerned with their well-being and the food products they  
15 consume. Therefore, the meat industry must process meat that is less fat, and rich in protein to  
16 meet the claim for nourishing and balancing the diet. Health concerns regarding fat consumption,  
17 as well as shifts in customer liking, have prompted substantial studies into low-fat products  
18 (Yogesh, 2021; Di Vita *et al.*, 2022). The addition of legumes as non-meat additives in  
19 manufactured meat products is a potential answer to the current market trend for low fat and great  
20 amount of fibre in meat products (Ponnampalam *et al.*, 2019). For example, Bambara groundnut  
21 (*Vigna subterranea* (L.) Verdc.) contains a 5.8% level of crude fat, and it can be categorised as a  
22 source of healthy fat that can be employed in less-fat diet formulations such as lean meat  
23 (Oyeleke *et al.*, 2012). Beside their intrinsic functional properties, incorporation of legumes, fruits  
24 and vegetables into comminuted meat products increases yield, oxidative stability, nutritional  
25 composition, and fibre content and reduces production costs (Kumar *et al.*, 2016; Beriain *et al.*,  
26 2018).

27 There is significant information that the consumption of legumes including Bambara  
28 groundnut (BGN) and other crop products is good for human well-being and decreases the  
29 chances of being exposed to cardiovascular diseases (Schwingshackl *et al.*, 2018; Ramatsetse  
30 *et al.*, 2023). The antioxidant properties of BGN can prevent non-transmissible diseases such as  
31 diabetes, malignancy, Alzheimer, and arthritis (Ramatsetse *et al.*, 2023). Moreover, rising  
32 consumers' desire for healthier food products is spurring the production of novel products that are  
33 more sustainable such as incorporating plant sources such as BGN (Rout *et al.*, 2022). Mubaiwa

34 *et al.* (2018) state that the high amount of protein in BGN provides the spotlight more on creating  
35 new updated processes for its production and extending its utilisation. According to Jideani and  
36 Diedericks (2014), the varied nutritional component of BGN grains implies that they have the  
37 capacity to fulfil the nutritional demands of a significant number of people.

38 Meat fat serves as a flavour compound reservoir and contributes to food texture. As a  
39 result, moisture drip and fat may affect or change product quality (Alves *et al.*, 2016). The loss of  
40 liquid decreases the sensory result, the heaviness of the commodity, and possibly its selling value.  
41 Shrinkage may also be caused by a lack of fluid in the product. Utilising plant-based protein  
42 derivatives can help to lessen the moisture loss that takes place when cooking (Aslinah *et al.*,  
43 2018; Ravani and Sharma, 2022). Different non-meat additives have been used in processed  
44 meat products. For instance, Bagdatli (2018) added quinoa flour to beef meatballs, Amadi (2020)  
45 added soybean to buffalo meat patties, and Ayandipe *et al.* (2020) added cassava and coconut  
46 composite powder to chicken sausages. Introduction of non-meat ingredients such as quinoa and  
47 BGN flours in processed meat products improves the sensory properties such as chewiness,  
48 juiciness, and taste (Alakali *et al.*, 2010; Muchekeza *et al.*, 2021).

49 There is little data regarding the utilisation of BGN flour in processed meat products.  
50 Alakali *et al.* (2010) investigated the impact of BGN flour on the nutritional qualities and sensory  
51 properties of beef patties stored at 4C for 21 days. The physicochemical, microbiological, and  
52 sensory properties of the formulated patties were acceptable over 21 days of refrigeration.  
53 Nevertheless, consumers preferred formulated beef patties added with up to 5% BGN flour.  
54 Dzudie *et al.* (2002) state that BGN flour was used because it is a cheaper source of protein and  
55 improves water retention and the production of structure in processed meat products. The limited  
56 use of BGN, like with other pulses, is associated with several problems, including the hard-to-  
57 cook and hard-to-mill characteristics of the grains, which also affect the nutritional quality (Gwala  
58 *et al.*, 2019; Diedericks *et al.*, 2020). To overcome these problems, the BGN grains are processed  
59 into flour. The incorporation of BGN flour in different foods could improve its utilisation, especially  
60 in developing countries. To fully explore the use of BGN flours in meat products, it is crucial to  
61 assess their effect on physicochemical, lipid oxidation and sensory acceptability of low-fat meat  
62 products. Therefore, this study assessed the nutritional, colour, functional and antioxidants  
63 properties of BGN flour varieties (cream, brown, and red coated grains) as well as their influence  
64 on the physicochemical properties, lipid oxidation, and sensory acceptability of mutton patties.  
65 The novelty of this research arises from the utilisation of cream, brown and red BGN flours in the  
66 development of low-fat mutton patties and reporting the results for the very first time. The results

67 obtained from this study may promote the utilisation of BGN flours in food products, improve the  
68 nutritional value, technological properties, and sensory characteristics of mutton patties.

## 69 **1.2 Problem statement**

70 The desire for less-fat meat products has risen dramatically in recent years, since consumption  
71 of great amount of fat, particularly saturated fats, has been linked to an elevated risk of  
72 cardiovascular disorders and some various malignancies. (Price *et al.*, 2013; Paglarini *et al.*,  
73 2020). However, reduction of fat content can affect product quality by making it stiffer, rubberier,  
74 less juicy, and dark in colour (Alves *et al.*, 2016; Gómez *et al.*, 2020). As results, the mouthfeel of  
75 low-fat meat products sought by customers may be improved by adding various alternative fat  
76 replacers (Paglarini *et al.*, 2020; Kumar, 2021). Fat substitutes in meat products are additives that  
77 add a small number of calories to treated meat and do not drastically change organoleptic and  
78 processing qualities. They change viscosity, softness, flavour and mouth feel, and other  
79 processing and sensory qualities (Purohit *et al.*, 2016). Modified starches, proteins, gums,  
80 cereals, and legume flours are added to decrease the negative effects of low fat (Al-Juhaimi *et*  
81 *al.*, 2016), thereby increase the retention of moisture and fat in meat products, enhancing juiciness  
82 and lowering brittleness.

83 Artificial antioxidants including propyl gallate, butylated hydroxyanisole (BHA), butylated  
84 hydroxytoluene (BHT), and tertbutyl hydroquinone (TBHQ) are usually utilised to delay oxidation  
85 processes or reactions as well as preservative to extend the durability of minced meat products.  
86 Nevertheless, these artificial antioxidants have been assessed as potential cause of cancer (Choi  
87 *et al.*, 2019). Therefore, the utilisation of natural antioxidant such as tocopherols, ascorbic acid,  
88 flavonoids, and phenolic compounds from plants extracts has piqued customer interest because  
89 of their possible health advantages, safety (Mishra *et al.*, 2021; Bag *et al.*, 2022) and ability to  
90 increase meat products shelf life (Falowo *et al.*, 2017). Additionally, the utilisation of natural  
91 preservatives has rendered high nutritional value foods to be safe from microorganisms and free  
92 from chemical preservatives (Olatunde and Benjakul, 2018; Mashau *et al.*, 2021). Due to  
93 increased consumers' intervention in the utilisation of artificial antioxidants, the utilisation of  
94 natural antioxidants in muscle foods is becoming extremely relevant and important in the meat  
95 industry.

## 96 **1.3 Rationale of the study**

97 The study is important to meat industry, consumers, and researchers. It may provide the meat  
98 industry with healthier and safer alternatives for meat preservation increasing the healthy  
99 consumer base and serving. The research may potentially improve consumer health and

100 customer satisfaction as they request the use of natural preservatives to replace chemical  
101 compounds. The study may widen the researcher's knowledge on the application of natural  
102 preservatives.

## 103 **1.4 Aim and objectives**

### 104 **1.4.1 Aim**

105 Evaluation of the effect of partial mutton meat substitution with Bambara groundnut flour on  
106 physicochemical properties, lipid oxidation and sensory acceptability of low-fat patties

### 107 **1.4.2 Specific objectives:**

- 108 ➤ To determine the effect of Bambara groundnut flour on proximate composition (fat, protein,  
109 ash, moisture, crude fibre, carbohydrate, & energy content) of low-fat mutton patties
- 110 ➤ To examine the effect of Bambara groundnut flour on physical properties (colour  
111 properties, cooking yield, & diameter reduction) of low-fat mutton patties.
- 112 ➤ To analyse the effect of Bambara groundnut flour on lipid oxidation of low-fat mutton  
113 patties
- 114 ➤ To assess the effect of Bambara groundnut flour on sensory attributes of low-fat mutton  
115 patties.

## 116 **1.5 Hypotheses**

- 117 ➤ Alternative hypothesis: The addition of Bambara groundnuts flour may have effect on  
118 physicochemical properties, lipid oxidation and sensory acceptability of low-fat mutton  
119 patties.
- 120 ➤ Null hypothesis: The addition of Bambara groundnuts flour may not have effect on  
121 physicochemical properties, lipid oxidation and sensory acceptability of low-fat mutton  
122 patties.

123

124

125

126

## 127 CHAPTER 2: LITERATURE REVIEW

### 128 2.1 Introduction

129 Bambara groundnut (*Vigna subterranean*) is a legume belonging to the family and subfamily of  
130 *Fabaceae* and *Faboidea*, respectively (Ibny *et al.*, 2019). Bambara groundnut is a legume  
131 indigenous to Africa and it is believed that its origin is between west and central Africa (Temegne  
132 *et al.*, 2018). It grows in the wild in northern Nigeria and eastwards to southern Sudan and is  
133 currently grown across tropical Africa and to a smaller range in tropical portions of America, Asia,  
134 and Australia (Adeleke *et al.*, 2018). Bambara groundnut (BGN) is the third most significant  
135 legume after groundnut (*Arachis hypogaea*) and cowpea (*Vigna hypogaea*) (Arise *et al.*, 2015;  
136 Oyeyinka and Oyeyinka, 2018). It plays a crucial socio-economic impact in the semi-arid regions  
137 globally and contains sufficient protein. Therefore, if combined with some native protein sources,  
138 it could help improve the nutritional issues globally (Massawe *et al.*, 2005; Mbosso *et al.*, 2020).

139 Bambara groundnut grain is a proper healthy food due to its high iron (4.9–48 mg/100 g)  
140 and protein contents (18.0–24.0%) when compared to other legume products, with great amount  
141 of amino acids content, fibre (5.0–12%), fat (5.0–7.0%), carbohydrate (57.43–63.09%), calcium  
142 (95.8–99 mg/100 g), potassium (1144–1935 mg/100 g), and sodium (2.9–12.0 mg/100 g) (Mbosso  
143 *et al.*, 2020; Tan *et al.*, 2020). Bambara groundnuts diverse nutritional composition suggests that  
144 it can meet dietary needs of people globally. Its high protein content maintains a greater focus on  
145 improving procedures for processing its grains and expanding food applications (Jideani and  
146 Diedericks *et al.*, 2014). Bambara groundnut also contains several polyphenols such as flavonoids  
147 (medioresinol, catechin, catechin dimer anthocyanin, and epicatechin) and phenolic acids (quinic  
148 acid, chlorogenic acid, caffeic acid, and ellagic acid) that have positive health benefits and  
149 strengthen the immune system against infectious diseases (Onyilagha *et al.*, 2009; Nyau *et al.*,  
150 2015; Harris *et al.*, 2018; Okafor *et al.*, 2021). Furthermore, antioxidants properties from plant  
151 extracts including BGN have also been reported to inhibit lipid oxidation in food products, thereby  
152 preventing transition metals and oxygen from reacting with food (Oyeyinka *et al.*, 2017; Manassis  
153 *et al.*, 2020; Oyeyinka *et al.*, 2021).

154 Bambara groundnut thrives at temperatures ranging from 20 to 28 °C and grows to  
155 maturity in three to five months. The plant is extremely versatile and can withstand extreme  
156 weather conditions than other crops. It thrives in well-drained soil and may even thrive in low-  
157 nutrient soil, with a pH range of 5.0 to 6.5. It is not susceptible to total crop failure resulting from  
158 rainfall scarcity or other unpredictable weather conditions (Bamshaiye *et al.*, 2011; Hillocks *et al.*,  
159 2012). Given these factors, BGN can improve malnutrition and poverty in this period of global

160 warming and food security threats globally, particularly in places with severe water problems (Chai  
161 *et al.*, 2016; Tan *et al.*, 2020).

162 Bambara groundnut variants include the red seed with huge kernels that mature late and  
163 has a high production rate. The seven types of BGN seeds from various landraces are depicted  
164 in Figure 1. The black-coloured seed matures quicker than other cultivars and often has a one  
165 seed in a tiny to medium sized kernel. The cream/brown eye produces sizable kernel. The  
166 cream/no eye has a relatively little kernels and pods, produces mostly one seed, and has low  
167 productivity. The speckled or spotted variety has a strong purple colour, small kernels, and  
168 primarily one-seeded pods. Finally, the brown variety contains medium to big kernels that varies  
169 in colour from pale to dark brown (Department of Agriculture, Forestry and Forestry and Fisheries,  
170 2011).

171



172

173

174 **Figure 1:** Seven different colour types of BGN seeds: black, red, cream/black eye, cream/brown eye, cream  
175 with no eye, purple, and brown colour (A to G), respectively (Kone *et al.*, 2007).

176 **2.2 Utilisations of Bambara groundnut grains**

177 Legumes have long been a staple food of low-cost meals around the globe. Thus, it is important  
 178 that their low levels of intake be elevated (Temba *et al.*, 2016; Maphosa and Jideani., 2017). Plant  
 179 protein contributes about 64% of the world’s protein supply for human beings, with cereals  
 180 accounting for 44% – 50% and legumes for 10% – 15%. Legumes are rich in unprocessed protein,  
 181 valuable for urban and rural inhabitants, particularly in developing countries (Bamshaiye *et al.*,  
 182 2011). After being picked and dried, BGN is processed and consumed in a variety of ways (Table  
 183 1).

184 **Table 1.** Various foods products of Bambara groundnut produced in selected African countries.

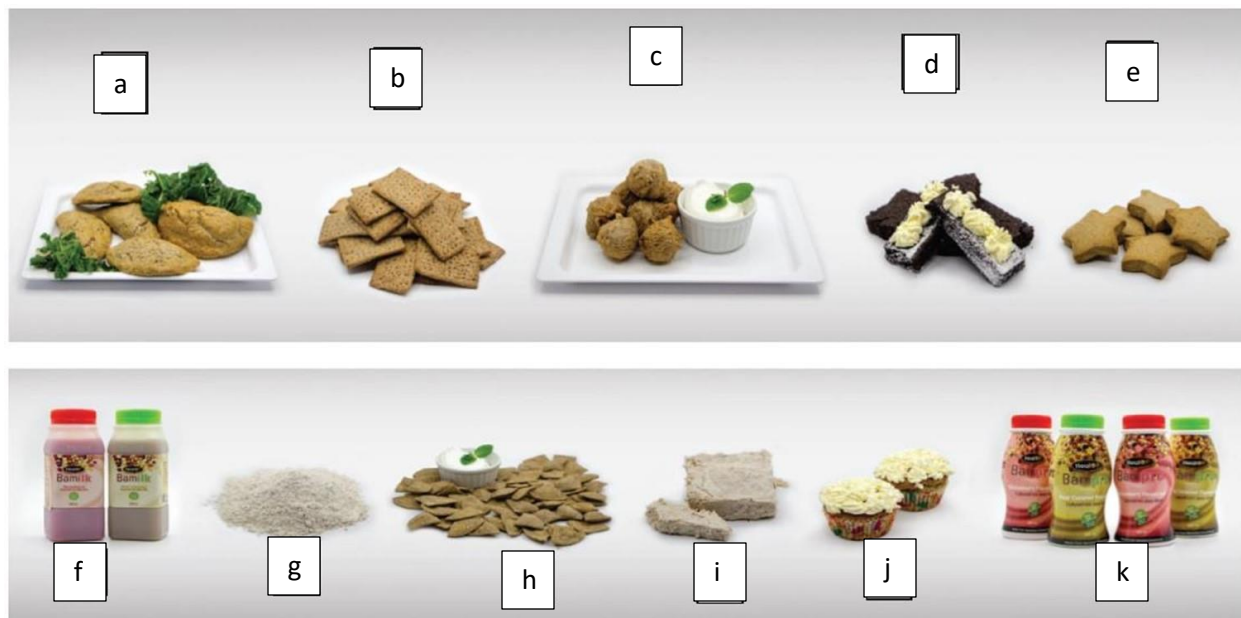
Country	Product name and processing of the product	References
South Africa	“ <i>Tshipupu</i> ” in Tshivenda- (Soup or porridge is made by boiling and stirring BGN).	(Harris, 2017)
Nigeria	“ <i>Okpa</i> ”- BGN is steamed into pudding.	(Okwunodulu <i>et al.</i> , 2019)
Kenya	“ <i>Njugumawe</i> ”- BGN grains are soaked, then cooked with maize, crushed, and fried into a stew maize.	(Harris, 2017)
Zimbabwe	“ <i>Matakura</i> ”- BGN grains are boiled to prepare <i>Matakura</i> .	(Mubaiwa <i>et al.</i> , 2018)
Zambia	“ <i>Ntoyo</i> or <i>Katoy</i> ”- BGN is ground into flour to bake bread.	(Cook, 2017; Harris, 2017)
Ghana	“ <i>Koose</i> ”- A paste of fried BGN grains. “ <i>Tuban</i> ”- BGN powder is mixed with water to make a stiff paste.	(Nti, 2009; Tan <i>et al.</i> , 2020)
Botswana	“ <i>Ditloo</i> ”- BGN is boiled and salted, and then eaten as a snack. “ <i>Dikgobe</i> ”- A porridge prepared by boiled BGN grains mixed with dry maize.	(Nti, 2009; Tan <i>et al.</i> , 2020)
Cameroon	“ <i>Akara</i> ”- It is made by rolling BGN grain paste into balls and fried immediately in oil. BGN grains are milled into powder, other times following roasting to prepare a porridge. BGN flour is also mixed with maize flour to enrich traditional meals.	(Temegne <i>et al.</i> , 2018; Olatunde <i>et al.</i> , 2021)
Sudan	Dried grains powder is utilised to prepare a stiff porridge.	(Tan <i>et al.</i> , 2020)
Mali	BGN grain powder is incorporated with sorghum or millet powder to prepare croquettes and grilled bread.	(Tan <i>et al.</i> , 2020)
Tanzania	Ground roasted BGN grains are roasted to prepare a soup or stiff porridge.	(Tan <i>et al.</i> , 2020)

185

186 After being grilled or boiled for almost an hour, picked fresh grains are eaten as snacks  
187 (Azam-Ali *et al.*, 2001; Jideani and Diedericks., 2014). Boiling dry grains makes them a functional  
188 food. Furthermore, after scorching or roasting, BGN can be eaten matured and consumed as a  
189 snack or as porridge (Stephens, 2012; Jideani and Diedericks, 2014). Dry grains are hard to  
190 ground because of their stiff and closely fitting grain coats. The grains are ground into powder,  
191 which is utilized to bake little flat cakes, make biscuits, bread and as additives to yogurt and  
192 vegetable milk (Murevanhema and Jideani *et al.*, 2013; Okafor *et al.*, 2015; Adu-Dapaah *et al.*,  
193 2016). Bambara groundnut is roasted and ground in Eastern Africa, and the powder is utilized to  
194 produce relish, soup, and coffee. The powder is also utilized to make a thin or hard porridge (De  
195 Kock, 2013; Esho *et al.*, 2018). Figure 2 indicates various foods products produced from BGN.

196 Interestingly, several tribes in Africa utilise BGN as traditional remedy for various ailments.  
197 For instance, in Senegal, the grains are crushed and mixed with water to treat vision problems  
198 and nausea and vomiting in pregnant women in South Africa (Murevanhema and Jideani, 2013)  
199 as well as nausea and diarrhea in Kenya (Khan *et al.*, 2021). In Ghana, white BGN grains are  
200 blended with Guinea fowl meat to treat diarrhea, whereas children with diarrhea are treated by  
201 mixing black grains with water (Damfami and Namo, 2020). Joint pains, bone decalcification,  
202 stomach pains, sore throats and amoebic dysentery are treated by BGN in Cameroon (Damfami  
203 and Namo, 2020). Bambara groundnut is utilized to treat sexually transmitted infections by the  
204 Ibo people of Nigeria (Brink *et al.*, 2006). It aids in digestion due to its laxative qualities. It helps  
205 to breastfeed mothers to produce more milk and is given to new mothers to cure their wounds  
206 (Temegne *et al.*, 2018; Mbosso *et al.*, 2020). Kaempferol, an antioxidant found in BGN, lowers  
207 the risk of numerous long-lasting diseases, including malignancy (Jideani and Diedericks, 2014;  
208 Yao *et al.*, 2015).

209



210

211 **Figure 2.** Various products produced from Bambara groundnut flours/ grains: a - crackers, b - chili-bite, c -  
 212 brownie, d- tofu, e - cake, f - biscuits, g - milk, h - baking flour, i - chips, j - tofu, and k - pro beverages,  
 213 <https://www.innovationbridge.info/ibportal/sites/default/files/bambara-tia-brochure-2019.pdf>  
 214 (innovationbridge.info). (Accessed 19 March 2022).

### 215 **2.3 Nutritional composition and health benefits of Bambara groundnut**

216 The BGN grains have proteins, fat, carbohydrates, and ash (Table 2), which serve as a  
 217 major source of nutrients in developing countries. Bambara groundnut is a balance food product  
 218 for a nutritionally balanced diet because it has enough crude fiber, proteins, carbohydrates, fats,  
 219 and minerals (Halimi *et al.*, 2019). Compared to other legumes, they contain high protein quality  
 220 and has an excellent balance of essential amino acids such as tryptophan, lysine, valine,  
 221 threonine, isoleucine, and phenylalanine, with relatively high lysine (8.5%) and methionine (6.4%)  
 222 (Olaleke *et al.*, 2006; Baptista *et al.*, 2016). Olaleke *et al.* (2006) found 2.84% of total crude protein  
 223 in cream testa BGN. Therefore, due to ongoing studies on therapeutic benefits of the grains and  
 224 its high protein content, its consumption is expected to rise in future. Polyunsaturated fatty acid  
 225 makes up most of the oil (Adeleke *et al.*, 2017). The grains of BGN contain 5.8% level of crude  
 226 fat and is a source of healthy fat for low-fat diet food formulations (Oyeleke *et al.*, 2012). Okpuzor  
 227 *et al.* (2010) stated that BGN is composed of polyunsaturated fatty acids (linolenic & linoleic acids)  
 228 and saturated fatty acids (stearic & palmitic).

229

**Table 2.** The nutritional profile of various types of Bambara groundnut grains, flour and grain coat

Cultivars	Crude protein (%)	Fat (%)	Moisture (%)	CHO (%)	Ash (%)	References
<b>Seeds</b>						
Red	19.5- 20.57	5.2- 6.5	8.0- 8.7	54.4- 63.9	2.4- 3.4	(Bamshaiye <i>et al.</i> , 2011; Oyeyinka <i>et al.</i> , 2021)
Black	21.7- 26.3	5.2- 8.5	8.9- 9.0	52.8- 57.4	2.2- 3.5	(Bamshaiye <i>et al.</i> , 2011; Oyeyinka <i>et al.</i> , 2021)
Cream	19.5- 23.51	6.0- 7.8	8.9- 9.7	56.0- 58.6	2.5- 2.9	(Bamshaiye <i>et al.</i> , 2011; Kaptso <i>et al.</i> , 2015; Hlanga <i>et al.</i> , 2021)
Brown	19.0- 24.3	5.3- 8.0	8.7- 10.3	54.4- 59.4	2.4- 3.0	(Jideani and Diedericks, 2014; Hardy, 2016; Oyeyinka <i>et al.</i> , 2021)
<b>Flour</b>						
Red	17.1- 20.9	3.0- 7.0	7.7- 9.3	48.0- 57.43	2.0- 3.90	(Bamshaiye <i>et al.</i> , 2011; Jideani and Diedericks, 2014; Shanono and Muhammad, 2015)
Black	22.6- 25.3	4.0- 5.9	9.0- 11.3	32.0- 61.7	2.0- 3.8	(Bamshaiye <i>et al.</i> , 2011; Jideani and Diedericks, 2014; Kaptso <i>et al.</i> , 2015)
Cream	22.3	3.0	9.0	49.6	1.5	(Bamshaiye <i>et al.</i> , 2011)
Brown	19.4- 20.9	3.5- 8.5	6.41- 10.0	48.0- 56.87	2.0- 4.12	(Bamshaiye <i>et al.</i> , 2011; Shanono and Muhammad, 2015)
<b>Grain coat</b>						
Red	5.7	0.5	3.0	8.4	1.0	(Bamshaiye <i>et al.</i> , 2011)
Black	6.1	2.0	2.0	6.0	1.5	(Bamshaiye <i>et al.</i> , 2011)
Cream	6.8	1.0	1.0	9.2	1.0	(Bamshaiye <i>et al.</i> , 2011)
Brown	6.3	2.0	3.0	9.1	1.0	(Bamshaiye <i>et al.</i> , 2011)

Note: CHO = Carbohydrate

1 Dietary fiber in BGN decreases blood pressure and serum cholesterol and protects against  
 2 cardiovascular diseases (Tan et al., 2021). They contain oligosaccharides carbohydrates such as  
 3 raffinose (0.4 g/mg) and stachyose (1.6 g/mg), which cause flatulence (Hardy and Jideani., 2016,  
 4 Halimi et al., 2019). It also contains reducing sugars (3.6 g/mg). Its chemical makeup of legume  
 5 starch is characterized by high amylopectin concentration (Schuster-Gajzago, 2004; Iwe et al.,  
 6 2006), indicating that BGN is rich in energy. The grains are abundant in minerals such as iron,  
 7 phosphorus, potassium, magnesium, sodium, and calcium as shown in Table 3.

8 Iron contributes to the synthesis of proteins myoglobin and haemoglobin by functioning as  
 9 an oxygen transporter, thereby reducing chances of getting diseases such as anaemia, physical  
 10 weakness, and fatigue (Fasoyiro et al., 2012; Atoyebi et al., 2017). Phosphorus assists in the  
 11 development of bones, proper functioning of kidneys and formation of cells. Potassium helps to  
 12 adjust the human's body acid-alkaline balance (Ndamitso et al., 2017). Calcium is significant for  
 13 blood coagulation, contraction of muscles, and the metabolic activity of certain enzymes.  
 14 Magnesium activates many enzyme systems. Water-soluble vitamins, including niacin, riboflavin  
 15 and thiamine is essential for carbohydrate, lipid, and protein metabolism (Okpuzor et al., 2010).

16  
 17 **Table 3.** Mineral composition of Bambara groundnut grains.

Mineral content (mg/100g)	Bambara groundnut	References
Calcium	0.3- 76.01	(Atoyebi <i>et al.</i> , 2017; Halimi <i>et al.</i> , 2019, Hussin, <i>et al.</i> , 2020)
Potassium	31.50- 2200.00	(Halimi <i>et al.</i> , 2019; Musah, <i>et al.</i> , 2021)
Magnesium	1.30- 555.10	(Halimi <i>et al.</i> , 2019)
Sodium	0.10- 25.20	(Halimi <i>et al.</i> , 2019; Musah, <i>et al.</i> , 2021).
Phosphorus	17.40- 563.00	(Halimi <i>et al.</i> , 2019; Musah, <i>et al.</i> , 2021).
Iron	0.10- 180.2	(Atoyebi <i>et al.</i> , 2017; Musah, <i>et al.</i> , 2021)
Sulphur	0.34	(Hussin <i>et al.</i> , 2020)
Zinc	2.14- 139.00	(Atoyebi <i>et al.</i> , 2017; Halimi <i>et al.</i> , 2019)
Copper	0.26- 30.00	(Atoyebi <i>et al.</i> , 2017; Musah, <i>et al.</i> , 2021)

18  
 19 Bambara groundnut also has vitamin C (1.170.20 mg/100 g), vitamin E (3. 180.15 mg/100  
 20 g) and vitamin A (26.050.14 mg/100 g) (Brink *et al.*, 2006; Jideani and Diedericks, 2014).  
 21 However, the mineral composition of BGN grains might vary depending on the variety of the grain.  
 22 For example, Kaptso *et al.* (2015) found significant difference in phosphorus content of black and  
 23 white BGN flours, with the white variety having lower content (216.8 mg/100 g) than the black

24 variety (231.9 mg/100 g). Furthermore, several studies reported significant difference of mineral  
 25 composition in different BGN accessions from wide geographical areas (Amarteifio *et al.*, 2006;  
 26 Alake and Alake, 2016; Atoyebi *et al.*, 2017). These variations might be due to environmental  
 27 factors and crop management.

28 The grains have a lot of amino acids, which are the building blocks of protein and are  
 29 molecules with amino (-NH<sub>2</sub>) and carboxyl (COOH) group (Kamei *et al.*, 2020). Amino acids are  
 30 essential for metabolism, anabolism, and neuron transmission in human body. They assist in  
 31 curing injury, tissue regeneration, nutrients transportation and storage, cell structuring, body  
 32 strengthening and the creation of arteries (Adebiyi *et al.*, 2019; Kamei *et al.*, 2020). Amino acids  
 33 are divided into two groups, essential and non-essential amino acids. Bambara groundnut has a  
 34 lot of essential amino acids (Table 4). Human cells cannot synthesize essential amino acids,  
 35 which is why their consumption via food is critical. However, non-essential amino acids can be  
 36 produced by human bodies (Bujang and Taib., 2014).

37  
 38 **Table 4.** Amino acids content of Bambara groundnut grains

Amino acids (g/100g)	Bambara groundnut	References
<b>Essential amino acids</b>		
Valine	0.70- 0.98	(Adeyeye <i>et al.</i> , 2012; Adebiyi <i>et al.</i> , 2019)
Lysine	1.15- 5.91	(Adeyeye <i>et al.</i> , 2012; Adebiyi <i>et al.</i> , 2019)
Histidine	0.50- 2.44	(Adeyeye <i>et al.</i> , 2012; Adebiyi <i>et al.</i> , 2019)
Phenylalanine	0.80- 4.13	(Adeyeye <i>et al.</i> , 2012; Adebiyi <i>et al.</i> , 2019)
Threonine	0.53- 3.04	(Adeyeye <i>et al.</i> , 2012; Adebiyi <i>et al.</i> , 2019)
Methionine	0.24- 1.22	(Adeyeye <i>et al.</i> , 2012; Adebiyi <i>et al.</i> , 2019)
Leucine	1.16- 6.92	(Adeyeye <i>et al.</i> , 2012; Adebiyi <i>et al.</i> , 2019)
<b>Non-essential amino acids</b>		
Proline	0.61- 2.86	(Adeyeye <i>et al.</i> , 2012; Adebiyi <i>et al.</i> , 2019)
Alanine	0.65- 4.04	(Adeyeye <i>et al.</i> , 2012; Adebiyi <i>et al.</i> , 2019)
Serine	0.79- 3.33	(Adeyeye <i>et al.</i> , 2012; Adebiyi <i>et al.</i> , 2019)
Glycine	0.58- 2.38	(Adeyeye <i>et al.</i> , 2012; Adebiyi <i>et al.</i> , 2019)
Arginine	1.18- 5.68	(Adeyeye <i>et al.</i> , 2012; Adebiyi <i>et al.</i> , 2019)
Glutamic acid	2.56- 10.01	(Adeyeye <i>et al.</i> , 2012; Adebiyi <i>et al.</i> , 2019)
Aspartic acid	1.63- 10.03	(Adeyeye <i>et al.</i> , 2012; Adebiyi <i>et al.</i> , 2019)
Tyrosine	0.56- 3.21	(Adeyeye <i>et al.</i> , 2012; Adebiyi <i>et al.</i> , 2019)

39  
 40 Amino acids are commonly used to complement the nutritional content of some food  
 41 products. For example, lysine and methionine are not available in some grains but are present in

42 BGN (Arise *et al.*, 2017; Arise *et al.*, 2017). Therefore, they can be utilized to complement the  
 43 amino acids content of such products when introduced. Amino acids are also used as flavouring  
 44 agent in food because they are sweet, salty, sour, and bitter or possess umami taste (Ninomiya,  
 45 2016). For example, they contain great amounts of glutamic acid (2.56 g/100 g) that can be utilized  
 46 as a flavouring substance in food products (Adebisi *et al.*, 2019; Kamei *et al.*, 2020; Tan *et al.*,  
 47 2020). Bambara groundnut is rich in lysine and glutamic acid and contains considerable quantities  
 48 of phenylalanine, aspartic acid, leucine, valine, arginine, and isoleucine (Adebisi *et al.*, 2019).

#### 49 2.4. Antimicrobial activities of Bambara groundnut

50 This section examines the most current research on raw legume extracts and compounds  
 51 to evaluate their anti-microbiological properties (Klompong and Benjakul, 2015; Pina-Pérez *et al.*,  
 52 2017; Adebisi *et al.*, 2019). The section also addresses unanswered questions about bioactivity  
 53 and sustainability of legume additives and compounds as anti-microbes, bioavailability, influence  
 54 on microbiological cells and metabolic process and pathogenicity production (Adebisi *et al.*, 2019).  
 55 Table 5 shows the ability of BGN to fight several foodborne pathogens.

56 **Table 5.** Antimicrobial activities of Bambara groundnut grains and against crucial foodborne  
 57 pathogens

Legume	Extracts	Microorganisms	Effect	References
Bambara groundnut	BGN seed	<i>Escherichia coli</i> <i>Candida albicans</i> <i>Staphylococcus aureus</i>	Growth inhibition	Oyeyinka <i>et al.</i> , 2021
Bambara groundnut	BGN seed coat, 99.9% ethanol extracts	<i>Escherichia coli</i> <i>Staphylococcus aureus</i> <i>Bacillus cereus</i>	Growth inhibition	Klompong and Benjakul, 2015
Bambara groundnut	BGN red and brown hulls, 70% methanol and 70% ethanol extracts	<i>Staphylococcus aureus</i> <i>Candida albicans</i> <i>Pseudomonas aeruginosa</i>	Growth inhibition	Harris, 2017

58  
 59 Different authors have reported an increase in inhibition of gram-negative bacteria such  
 60 as *Klebsiella pneumonia*, *Klebsiella aerogenes*, and *Pseudomonas aeruginosa*, as well as gram-  
 61 positive bacteria such as *Escherichia coli*, *Candida albicans*, *Staphylococcus aureus*, *Bacillus*  
 62 *aureus*, and *Aspergillus Niger* with an increase in BGN grain extracts (Klompong and Benjakul,  
 63 2015; Harris, 2017; Wanyama *et al.*, 2017; Wanyama, *et al.*, 2018; Oyeyinka *et al.*, 2021).

64 Moreover, Udeh *et al.* (2020) states that the anti-microbiological properties of BGN are also  
65 dependent on the concentration of the extract and the variety of the legume. The ability of BGN  
66 grains extracts to inhibit microbial growth is due to polyphenols such as flavonoids, anthocyanins,  
67 and tannins in the hulls of BGN grains that exhibit antimicrobiological and antioxidant potentials  
68 (Klompong and Benjakul, 2015; Tsamo *et al.*, 2018).

69 Harris (2017) found antibacterial action of raw BGN extract and no antibacterial action in  
70 BGN milk and yogurt extracts; possibly indicating that phytochemical concentration was lower  
71 than in a raw BGN grains. The author further concluded that incorporating food products with high  
72 BGN hulls can improve their antioxidant and anti-microbiological properties. Moreover, Alakali *et*  
73 *al.* (2010) found an elevation of total plate count in beef patties incorporated with BGN powder for  
74 the first 14 storages days of the samples. However, after 14 days of storage to 21 days, the  
75 elevation of total plate in untreated beef patties was substantially greater than of treated patties  
76 with BGN powder.

77 Significant polyphenolic compounds in legumes can be found in the roots and seeds,  
78 depending on a particular product (Chon, 2013). Many polyphenolic compounds in legumes have  
79 been thoroughly investigated and linked with antimicrobial activity against extensive range of  
80 microbes. Chelation of crucial micro minerals such as iron and zinc, suppression of cell surface  
81 microbiological enzymes and straight forward intervention in microbiological activity are  
82 processes that are widely recognized as liable for polyphenols' antimicrobial capability (Heinonen,  
83 2007; Daglia, 2012). Moreover, polyphenol compounds present in BGN such as tannins and  
84 flavonoid have been reported to possess antimicrobial activities (Klompong and Benjakul, 2015;  
85 Ajiboye and Oyejobi, 2017).

86 Antimicrobial activity has also been discovered in proteinase inhibitors found in plants,  
87 especially legumes. Plant-derived substances that protect from pests and pathogens have great  
88 prospects for application as anti-microbiological agents in products. These substances work by  
89 inhibiting enzyme activity in response to proteinases generated by phytopathogenic microbes  
90 attacking them (Arulpandi and Sangeetha, 2012; Lopes and Brandelli, 2018). Sitohy *et al.* (2013)  
91 determined the impact of chickpea, broad bean, and soybean proteins extracted and esterified  
92 with methanol that they inhibited microbial development. By utilizing the agar, a concentrated-  
93 dependent suppression area was noted against Gram-positive (*Bacillus subtilis* and  
94 *Staphylococcus aureus*) and Gram-negative (*Pseudomonas aeruginosa*, and *Escherichia coli*)  
95 bacterial upon adding methylated proteins at concentrations between 0.1 and 10 mg/ml (Sitohy  
96 *et al.*, 2013). Similarly, Kanatt *et al.* (2011) found growth inhibition against *Bacillus cereus*,

97 *Staphylococcus aureus*, *Escherichia coli*, and *Pseudomonas fluorescens* by chickpea, mung, and  
98 pigeon pea extracts at concentration of 0.05%.

## 99 2.5 Natural antioxidants of Bambara groundnut

100 Natural antioxidants are substances that are utilized to prevent fat oxidation from  
101 occurring, as well as to prolong shelf-life of food (Glodde *et al.*, 2018). Moreover, natural  
102 antioxidants protect food from spoilage due to oxidation, which may lead to lipid rancidity or  
103 alterations of colour in products (Aminzare *et al.*, 2019). Despite their lower efficiency, natural  
104 antioxidants are well tolerated by consumers and are safe for consumption. Moreover, other  
105 natural substances possess greater antioxidant potential compared to artificial compounds and  
106 others have beneficial impacts on processed meat sensory attributes (de Florio Almeida *et al.*,  
107 2017; Oyeyinka *et al.*, 2017).

108 Bambara groundnut has natural antioxidants such as flavonoids and phenolic, as well as  
109 DPPH and ferric reducing antioxidant power (Table 6). Thus, the shelf life of products containing  
110 fats can be extended due to availability of such substances. Natural antioxidants from BGN grains  
111 also help to maintain health. Antioxidant qualities of BGN are influenced by different ways of  
112 processing, crop variant and assay methodologies. The availability of phenolic substances in the  
113 grains is responsible for these qualities. Oyeyinka *et al.* (2017) found that antioxidant activities of  
114 BGN decrease significantly after the removal of hulls. Furthermore, Xu and Chang (2008) reported  
115 that most phenolic substances which are contained in hulls are mostly accountable for antioxidant  
116 activities. The reason for this is presumable due to the uneven distribution of phenolic compounds  
117 in plants. For instance, the seed coats of grains have greater concentrations of phenolic  
118 compounds than those present in the inner portions (Laura *et al.*, 2019; Oyedeji *et al.*, 2021).

119 **Table 6.** Phytochemicals of Bambara groundnut grains and their health benefits.

Phytochemicals	Quantity (mg/g)	Health benefit	References
<b>Flavonoids</b>			
Kaempferol	0.05- 2.18	Boost the body's antioxidant defence over free radicals, which lowers the chances of developing chronic diseases, particularly cancer and stroke.	(Chen and Chen, 2013; Salawu, 2016; Harris <i>et al.</i> , 2018; Alam <i>et al.</i> , 2020; Silva Dos Santos <i>et al.</i> , 2021)
Anthocyanin	0.038	Regulate signalling pathways, aid in improving vision, controls weight, lowers the chances of developing cardiovascular	(Adedayo <i>et al.</i> , 2021; Gonçalves <i>et al.</i> , 2021)

		diseases, malignancy, and neurodegenerative disorders.	
Medioresinol	0.0008	Prevents CVDs and metabolic syndrome by lowering glucose and cholesterol levels.	(Hwang <i>et al.</i> , 2018; Adebisi <i>et al.</i> , 2021)
Myricetin	0.062-1.80	Protect liver injuries, lowers hepatic triglycerides, inhibits hyperglycemia, reduces oxidative stress.	Salawu, 2016; Semwal <i>et al.</i> , 2016; Harris <i>et al.</i> , 2018; Imran <i>et al.</i> , 2021
Catechin	0.01- 2.34	Prevents cancer, obesity, diabetes, liver injuries, CVDs, and lowers cholesterol level.	Salawu, 2016; Isemura, 2021; Kim and Heo, 2022
Quercetin	0.007-6.39	Lowers high glucose level, prevent kidney failure, heal wounds, prevents Alzheimer's disease, and eases arthritis-related pain and inflammation.	Salawu, 2016; Thomas <i>et al.</i> , 2017; Borghi <i>et al.</i> , 2018; Harris <i>et al.</i> , 2018; Salehi <i>et al.</i> , 2020
Epicatechin	1.15	Decreases chances of CVDs and diabetes.	Salawu, 2016; Aina <i>et al.</i> , 2017
<b>Phytic acids</b>			
Ellagic acid	0.0005-1.09	Heal wounds, reduce exhaustion and insomnia. prevents liver damage, heart disease, skin damage, atherosclerosis, fibrosis, and nociceptive.	Salawu, 2016; Harris <i>et al.</i> , 2018
Chlorogenic acid	0.03- 2.37	Prevents diabetes, cancer, stroke, Alzheimer disease, blood pressure, and obesity.	Salawu, 2016; Tajik <i>et al.</i> , 2017; Harris <i>et al.</i> , 2018; Yan <i>et al.</i> , 2020
Caffeic acid	3.65	Prevents cancer, neurological disorders, diabetes, inflammation, and premature aging and improves collagen synthesis.	Magnani <i>et al.</i> , 2014; Salawu, 2016; Harris <i>et al.</i> , 2018; Cizmarova <i>et al.</i> , 2020
Gallic acid	0.05- 1.03	Ability to treat cancer, neoplastic, diabetes, Alzheimer disease, hepatitis C virus, Parkinson's disease, and CVDs.	Salawu, 2016; Harris <i>et al.</i> , 2018; Samad and Javed, 2018; Kahkeshani <i>et al.</i> , 2019
Quinic acid	0.00309	Inhibit Human Immunodeficiency Virus, Hepatitis B virus, and Herpes Simplex virus loads. Improves immunological functioning and improves DNA repair.	Bhatia <i>et al.</i> , 2015; Zanello <i>et al.</i> , 2015; Adebisi <i>et al.</i> , 2021

120 Note: CVDs = cardiovascular diseases.

121 Most research on antioxidant activities of BGN utilised entire grain or grains without hulls  
 122 which probably lowered the levels of antioxidants tested. Therefore, extracting phenolic  
 123 compounds from the grains and determining antioxidant activities is essential.

124 Bambara groundnut contains variety of phytochemicals with different physiological active  
125 constituents; therefore, BGN constituents should be utilised as an additive in the production of  
126 nutraceuticals and functional foods (Okafor *et al.*, 2022). Dried grains are an excellent source of  
127 phenolic compounds, and they can contribute to antioxidants consumption in other foods.  
128 Phenolic compounds in legumes can attach to proteins, minerals and amino acids that lack some  
129 of their electrons, therefore lowering the digestibility of crucial minerals and possibly improving  
130 the uptake of the nutrients (Thakur *et al.*, 2019; Ahmmed *et al.*, 2020). Phenolic compounds  
131 possess health benefits due to several biological processes by acting as free radical scavenger,  
132 reducing potential, chain breaking agent and changing of signal transduction pathways (Moyo *et*  
133 *al.*, 2018). Flavonoids are a type of aromatic secondary metabolites from plants. Plants primarily  
134 utilise them to make essential pigments that are vital in forming colour (Karak *et al.*, 2019; Khatun,  
135 2021). Anthocyanidins, flavones, flavanols, and flavan-3-ols are among flavonoids found in BGN  
136 grains (Oyedeki *et al.*, 2021).

137 Ferric reducing antioxidant power (FRAP) of legumes such as BGN grains demonstrates  
138 their potential to decrease ferric ions ( $\text{Fe}^{3+}$ ) to ferrous ions ( $\text{Fe}^{2+}$ ) while changing the colour from  
139 yellow to blue green (Oluwatoyin, 2014; Masek *et al.*, 2018). In general, the existence of  
140 components that disrupt the free radical chains by acting as hydrogen-donor indicates FRAP  
141 (Ahmmed *et al.*, 2020). The 1,1-diphenyl-2-picryl hydrazyl radical (DPPH) of legumes is a steady  
142 nitrogen-centered free radical donor that is mostly utilised to determine the scavenging activity of  
143 plant extracts. Extracts that possess antioxidant potential to alter DPPH colour from violet to  
144 yellow, due to their ability of donating electrons to a steady DPPH radical (Vijayaraghavan *et al.*,  
145 2018; Ahmmed *et al.*, 2020). Oyeyinka *et al.* (2021) found comparatively greater DPPH activity in  
146 maroon and black BGN grains aqueous extract, due to possibly greater amount of protic  
147 flavonoids in them than in brown BGN grains (Ahmed *et al.*, 2015). In addition, the differences in  
148 composition of the BGN grains may affect how differently the hydrogen atoms are donated to  
149 DPPH, which could be possibly account for variations in antioxidant content across varied BGN  
150 grains extract (Klompong and Benjakul, 2015). Chinnapun and Sakorn (2022) discovered a new  
151 substance in BGN grains that was classified as luteolin glycoside ( $\text{C}_{20}\text{H}_{18}\text{O}_9$ ), which inhibited  
152 DPPH radical in them.

153 However, the quantity of antioxidant properties of the same or different BGN cultivars as  
154 well as other legumes vary depending on various factors such as legume breed, soil  
155 characteristics, growing parameters and solvents used for extracting samples (Yusnawan *et al.*,  
156 2019; Okafor *et al.*, 2021). Total phenolic and flavonoid content of BGN grains are more reliant  
157 on their coat colour. Therefore, the darker the seed coat, the higher its antioxidant properties and

158 the lighter the seed coat, the lower its antioxidant properties (Parikh and Patel, 2018; Adedayo *et*  
159 *al.*, 2021). For instance, Adedayo *et al.* (2021) found higher flavonoid, phenolic and FRAP content  
160 of Black BGN variety compared to red and brown variety. Similar trends were also reported by  
161 Oyeyinka *et al.* (2021) for maroon, black, brown, and maroon BGN grains. Furthermore, Malik  
162 and Kapoor (2015) found that 70% acetone was the most effective solvent for extracting  
163 antioxidant properties of varieties of Indian lentils compared to pure methanol and 80% acetone.  
164 Similarly, Yusnawan and Kristiono (2019) found that 70% acetone generated higher flavonoid and  
165 phenolic content, as well as antioxidant activity of Mungbean varieties, compared to 70%  
166 methanol and 70% ethanol. Tsamo *et al.* (2018) found a range of total phenolic content (0.75–  
167 17.71 mg GAE/g) and total flavonoids content (0.01–2.51 mg GAE/g) in 21 BGN landraces. Since  
168 BGN grains contain considerable amount of phytochemicals, it is essential to further explore their  
169 utilisation and how new processing techniques affect their phytochemical composition (Oyedeji *et*  
170 *al.*, 2021).

## 171 **2.6. Limitation of Bambara groundnut production**

172 In underdeveloped areas, BGNs are essential part of people's diet by supplementing the  
173 shortage of protein in grains, stems, and tubers. Nevertheless, owing to the emergence of difficult  
174 to cook phenomena while storing at extreme conditions (above 26 °C) and relative humidity  
175 (above 75 °C) as is prevalent in African countries, limits the use of BGN (Azam-Ali *et al.*, 2001;  
176 Mubaiwa *et al.*, 2017; Mubaiwa *et al.*, 2018;). Two mechanisms occur at those temperatures, (1)  
177 lignification (the binding among branched-chain amino acids and liberated polyphenols in the cells  
178 results in lignin production and promotes cell wall strengthening) (Mubaiwa *et al.*, 2017;  
179 Chigwedere *et al.*, 2019), (2) pectin-phytase: phytase results in hydrolysis of positive charged  
180 phytate when stored, freeing bivalent charged ions. At the same time, pectin methyl esterase  
181 destroys the methyl esters of pectin, releasing liberated carboxyl group, that prefer to bind with  
182 bivalent positively charged ions. Therefore, the cell wall is solidified by the formation on non-  
183 soluble magnesium and calcium pectate compounds prevents cell detachment and weakens upon  
184 cooking (Chigwedere *et al.*, 2019; Tan *et al.*, 2020;). Grain outer layer, starch particles,  
185 polyphenols, phytic acid, proteins, lignins, tannins and cell wall are linked with difficult to cook  
186 phenomena (Aguilera and Rivera, 1992; Garcia *et al.*, 1998; Gwala *et al.*, 2019).

187 Individuals in affluent areas that have energy supplies and cutting-edge technology to use  
188 grains do not consider difficult-to-cook phenomena to be problematic because they can afford  
189 various products rich in protein (Chazovachii *et al.*, 2013; Mubaiwa *et al.*, 2018). Challenging to  
190 cook BGN grains need more time (3 to 4 h) to be boiled and thus elevated heat so that they can

191 be consumable. However, since most villagers are dependent on firewood for cooking, the longer  
192 boiling durations needed for BGN grains processing are not sustainable based on expenses and  
193 power usage. The need for fuel wood in several African areas keeps rising, rising in land usage,  
194 and cutting down of trees have limited their availability, forcing people to fetch firewood far away  
195 from their homes (Mubaiwa *et al.*, 2017; Temudo *et al.*, 2020).

196 Furthermore, difficult to cook BGN also display difficult to mill qualities and difficult to cook  
197 issues owing to the solid bonding among husks and cotyledons due to gums and mucilage at the  
198 interaction (Mubaiwa *et al.*, 2017; Muhammad *et al.*, 2020). Another impediment to the utilization  
199 of this significant legume is the low production of BGN in African countries (Khan *et al.*, 2016;  
200 FAOSTAT, 2020). Lack of modern equipment for post-harvest processes such as the removal of  
201 hulls, have also led to its under-use (Berchie *et al.*, 2010; Majola *et al.*, 2021). Altogether, these  
202 undesirable physical features contribute to reduction in BGN utilisation and reduces its application  
203 in various food products and increases its economic costs (Mubaiwa *et al.*, 2018; Temudo *et al.*,  
204 2020).

## 205 **2.7. Solutions to limitations of Bambara groundnut and future scope**

206 Difficult to cook occurrence in BGN grains can be managed by storing them at lower  
207 temperatures in refrigerators. However, individuals living in poor areas cannot access such ideal  
208 conditions.[26] Turning difficult to cook BGN grains to consumable and nutrient-rich products;  
209 efficient and effective preparation methods are needed (Annan *et al.*, 2003; Jideani and  
210 Diedericks, 2014). For instance, chemical procedures (cooking aids), biological procedures  
211 (germinating & fermenting), and physical procedures (milling, roasting, & canning) are all utilised  
212 when processing legumes (Mubaiwa *et al.*, 2017). Although these procedures minimise cooking  
213 time, Annan *et al.* (2003) found that most participants during sensory evaluation indicated that  
214 processing methods change the taste and texture of products. Due to BGN hard to cook  
215 phenomena which is a constraint for its utilisation, understanding advanced science using various  
216 fourth industrial revolution technologies, including green technology is essential (Jideani and  
217 Jideani, 2021).

218 Thus, to promote the use of BGN in areas with a shortage of resources, appropriate  
219 techniques to shorten cooking duration, conserving power, and grinding qualities should be  
220 designed and deployed in local circumstances (Mubaiwa *et al.*, 2018). Developing and promoting  
221 of local preparation procedures that can reduce existing post-harvest costs may be essential to  
222 the wide utilisation of BGN (Matsa and Mukoni, 2013; Mubaiwa *et al.*, 2018). Those procedures  
223 should conform to the standards for competence, digestion of nutrients, and customer satisfaction

224 requirements (Mubaiwa *et al.*, 2018). As a result, it is critical to learn from and improve on  
225 procedures which have proven successful in preparing difficult to cook grains (Gbaguidi *et al.*,  
226 2018; Mubaiwa *et al.*, 2018).

227         Between 1999 and 2018 there was a decline in the production of BGN which also resulted  
228 in less yielding cultivars and low standards grains (Annan *et al.*, 2003; Khan *et al.*, 2016).  
229 Therefore, initiating studies on breeding and the advancement of cultivars with improved  
230 agricultural methods targeting to improve yields is required. In future, trials to breed cultivars  
231 would be vital to examine local techniques, appropriateness, and personal participation of farming  
232 community to permit them to suggest viable lines to satisfy their urgent needs (Mubaiwa *et al.*,  
233 2018). Bambara groundnut is a climate smart plant because of thriving in different agricultural  
234 soils and varied climate conditions (Gbaguidi *et al.*, 2018; Tan *et al.*, 2020). Therefore, investors,  
235 sponsors, or departments of agriculture should assist farmers with funding to plant a lot of BGN  
236 crops by increasing labour and farming equipment. To improve the utilisation of BGN, food  
237 manufacturers with advanced fourth industrial revolution equipment machinery should collaborate  
238 with farmers to improve the availability of the product in the market.

## 239 **2.8 Effect of legumes on sensory attributes of meat products**

240         Meat tenderness, juiciness, flavour, colour, palatability, neatness, and cooking loss are  
241 some of the sensory attributes that consumers commonly use to assess meat quality (Calvo *et*  
242 *al.*, 2016). Tenderness and juiciness are two factors that influence total meat consuming quality,  
243 whereas meat pH and colour have an impact on meat storage quality and appearance (Geletu *et*  
244 *al.*, 2021). Meat patties are popular products eaten globally, often made with 70 % meat and about  
245 26 to 32 % fat as major additives (De Oliveira Fagundes *et al.*, 2017; Moghtadaei *et al.*, 2018).  
246 Meat patties are typically made from beef (Polizer-Rocha *et al.*, 2020), nevertheless novel  
247 formulas encompass of mutton and deer meat (Vargas-Ramella *et al.*, 2020). Reduced fat content  
248 in patties is linked to issues with hardness, flavour, and moistness (Heck *et al.*, 2019).

249         Nevertheless, it is important to keep in mind that not all customers are able to explore new  
250 products. Other customers are hesitant to explore that contain unique components or are made  
251 utilising new technology (Siegrist and Hartmann, 2020). Products neophobia consumers'  
252 segmentation enables for development of particular approaches depending on the likings of these  
253 individuals who are prepared to test novel or modified foods (Giacalone and Jaeger., 2023; Jin *et*  
254 *al.*, 2023). It is broadly understood that new formulations of foods should be indeed centered on  
255 the consumer and in addition to the consumer's interest in healthy food. Sensory analysis of novel

256 products is critical to their success and researchers have examined individual's perceptions on  
257 new products, with the majority of them focusing solely on product features (Câmara *et al.*, 2020).

258 Improving the nutritional composition of meat patties whilst preserving consumer approval  
259 has been demonstrated to be achievable. In light of this, Argel *et al.* (2020) found that employing  
260 legumes powder as partial pork meat substitute's improved overall acceptability of patties.  
261 Currently, Vargas-Ramella *et al.* (2020) also stated that customers did not notice any major  
262 changes among patties made with animal fat and those made by substituting fat with chia, linseed,  
263 and tiger nut oils. The sensory attributes of pork burgers treated with green pea, chickpea, bean,  
264 and lentil powder emulsions were determined satisfactory to consumers by Argel *et al.* (2020)  
265 with no significant variations between the pulse kinds. According to Alakali *et al.* (2010) there was  
266 no significant difference between any sensory properties of non-formulated and BGN flour  
267 formulated beef burgers over 21 day's storage period.

## 268 **2.9. Nutritional properties and health benefits of mutton meat**

269 Meat consumption is very significant in the diet of human with respect to completely  
270 developing and sustaining optimal physiological and mental performances. Mutton meat (Figure  
271 3) is composed of essential amino acids, water, lipids, vitamins and minerals, and small portion  
272 of carbohydrates (Mashau *et al.*, 2021). It is also comprised of fats, as well as wide range of  
273 essential omega-3- polyunsaturated fats (Ponnampalam *et al.*, 2021).



274

275 **Figure 3:** Boneless mutton meat composed of meat and fat with the ratio of 80/20, respectively  
276 (<https://www.murgaca.com/en/products/mutton-trunks-8020.html>).

### 277 **2.9.1. Minerals**

278 Red meat is very rich in micro-minerals, which are needed by the body in small quantity  
279 such as iron, copper, zinc, and selenium (Table 7). It also contains macro-minerals such as  
280 sodium, phosphorus, and sodium, which are needed by the body in large quantity. All these  
281 minerals are crucial since they serve significant part in antioxidative system and in the metabolism  
282 pathways (Zerabruk *et al.*, 2019).

283 **2.9.1.1 Iron**

284 Meat is very rich in iron, as well as heme iron that contain myoglobin and haemoglobin  
 285 proteins, which are crucial for respiratory systems and oxygenation of tissues (Fowler *et al.*, 2019;  
 286 Adeyeye *et al.*, 2020). Nevertheless, iron deficiency, which is responsible for anaemia, is  
 287 widespread globally, more especially in woman and is associated with lethargy, headache, stress,  
 288 and extreme fatigue when training (Tardy *et al.*, 2020; Abdelnabi, 2022). Furthermore, anaemia  
 289 also results in low productivity and lowers the health of an individual (Ahmad *et al.*, 2018).  
 290 However, consumption of mutton meat can help to prevent such disorders due to present of iron.

291 **Table 7.** Nutritional composition of mutton patties

Nutrients	Meat cut (Mutton, chop or meat, raw)	References
Protein (g)	18.42- 22.85	(Khomola <i>et al.</i> , 2021; Mashau <i>et al.</i> , 2021)
Saturated fat (g)	2.4- 2.47	(Sainsbury <i>et al.</i> , 2011; Ahmad <i>et al.</i> , 2018)
Fat (g)	4.80- 11.62	(Ahmad <i>et al.</i> , 2018; Mashau <i>et al.</i> , 2021).
Ash (g)	1.12- 2.10	(Khomola <i>et al.</i> , 2021; Mashau <i>et al.</i> , 2021)
Moisture (%)	65.50- 67.25	(Khomola <i>et al.</i> , 2021; Mashau <i>et al.</i> , 2021)
Energy (Kcal)	122- 524	(Sainsbury <i>et al.</i> , 2011; Ahmad <i>et al.</i> , 2018)
Vitamin B <sub>12</sub> (mcg)	2.0- 2.37	(Sainsbury <i>et al.</i> , 2011; Ahmad <i>et al.</i> , 2018)
Vitamin B <sub>1</sub> (mg)	0.04- 0.14	(Sainsbury <i>et al.</i> , 2011; Ahmad <i>et al.</i> , 2018)
Vitamin B <sub>2</sub> (mg)	0.04- 0.24	(Sainsbury <i>et al.</i> , 2011; Ahmad <i>et al.</i> , 2018)
Niacin (mg)	4.96- 4.99	(Sainsbury <i>et al.</i> , 2011; Ahmad <i>et al.</i> , 2018)
Pantothenic acid (mg)	0.6	(Ahmad <i>et al.</i> , 2018)
Vitamin B <sub>6</sub> (mg)	0.2- 0.3	(Sainsbury <i>et al.</i> , 2011; Ahmad <i>et al.</i> , 2018)
sodium (mg)	63 -83	(Sainsbury <i>et al.</i> , 2011; Ahmad <i>et al.</i> , 2018)
Zinc (mg)	3.56- 3.6	(Sainsbury <i>et al.</i> , 2011; Ahmad <i>et al.</i> , 2018)
Iron (mg)	1.9- 2.97	(Sainsbury <i>et al.</i> , 2011; Ahmad <i>et al.</i> , 2018)
Phosphorus (mg)	174- 221	(Sainsbury <i>et al.</i> , 2011; Ahmad <i>et al.</i> , 2018)
Copper (mg)	0.15	(Ahmad <i>et al.</i> , 2018)
Potassium (mg)	244- 275	(Sainsbury <i>et al.</i> , 2011; Ahmad <i>et al.</i> , 2018)
Magnesium (mg)	18.8- 22.7	(Sainsbury <i>et al.</i> , 2011; Ahmad <i>et al.</i> , 2018)
Calcium (mg)	12.5	(Ahmad <i>et al.</i> , 2018)

292

293 **2.9.1.2 Zinc and Selenium**

294 Zinc is also a part of superoxide dismutase, which is an antioxidant enzyme required for  
 295 the body immune system, having role in cell division, growth, and wound healing (Adeyeye *et al.*,  
 296 2020; Chafik *et al.*, 2021). An inadequate consumption of zinc results in exhaustion, deprived  
 297 development, rickets and weakened mental function of an individual (Ahmad *et al.*, 2018).  
 298 Consumption of red meat can prevent such disorders due to the availability of zinc. Selenium is  
 299 a crucial mineral required in a significant amount by individuals. Selenium is most utilised

300 antioxidant in food supplements to prevent oxidation of fat in meat. It is a key part of an enzyme  
301 glutathione peroxidase, which along with vitamin E protects the cells from free radicals (Habibian  
302 *et al.*, 2016; Xiao *et al.*, 2021). Via its integration into selenoproteins, it plays an important part in  
303 sustaining well-being of an individual (Mojadadi *et al.*, 2021)

### 304 **2.9.1.3 Vitamins**

305 Red meat such as mutton comprises of large quantities of vitamins that the body requires  
306 for better health (Libera *et al.*, 2021). It comprises of huge amount of B vitamins (B<sub>6</sub> and B<sub>12</sub>).  
307 Vitamin B<sub>6</sub> is required for proper operational of about 99 enzymes which catalyses crucial  
308 chemical reactions in individual's bodies. Also, it is crucial in creation of heme iron, which is  
309 constituent of haemoglobin, and it aids in the production of neurotransmitters (Juárez *et al.*, 2021).  
310 Vitamin B<sub>12</sub> is crucial for synthesis of Deoxyribonucleic acid that is essential for individual body's  
311 healthy growth and development. Vitamins found in mutton meat are required by all age groups  
312 (Ahmad *et al.*, 2018). Red meat also contains vitamin A, C, D and E (Juárez *et al.*, 2021). Vitamin  
313 A helps to strengthen and develop bones, as well as improving eyesight (Balami *et al.*, 2019).  
314 Vitamin D is crucial for formation and maintaining bones, as well as separation and variation of  
315 cells. Vitamin E protects cells against effects from free radicals as it is an antioxidant that acts as  
316 free radical terminator. Vitamin C is necessary for integrity of bone and collagen, which assist in  
317 maintaining skin (Ahmad *et al.*, 2018; Ahmad *et al.*, 2022).

## 318 **2.10. Mutton patties**

319 Mutton patties is basically meat that has been finely chopped or grounded by a chopping  
320 knife or meat mincer and flattened, usually round using patty maker, manually or petri dish (Alakali  
321 *et al.*, 2010; Bakieva *et al.*, 2019). The process of grinding makes the meat cuts which are tougher  
322 to become tender, and grinding fatty cuts in with lean cuts assist in reducing the dryness. Minced  
323 meat is highly perishable as compared to the whole cuts of mutton meat and must be carried or  
324 handled with particular care (Cauchi, 2020). Throughout process of grinding most of ground meat  
325 are prone to oxidation of fat. This is because grinding opens up channels of meat being  
326 susceptible to oxygen, as well as disrupting and exposing phospholipids of meat to pro-oxidants  
327 such as copper and iron (Wang *et al.*, 2021).

### 328 **2.10.1 Shelf-life of mutton meat**

329 Shelf-life refers to the time from when the products are packaged until their end utilisation  
330 date, whereby their attributes are still suitable for consumer's consumption. Shelf-life attributes  
331 may encompass the colour, flavour, texture, nutritional value (Meijer *et al.*, 2020; Shivakumar *et*

332 *al.*, 2022). However, meat products made from minced meat such as mutton patties is likely to  
333 have short shelf life as the large, exposed channels or surface area of meat may result in spoilage.  
334 The speed of degradation relies on nutritional content of meat, proper sanitary practices when  
335 slaughtering, mincing, and preparing and during storage temperatures or environment.  
336 Microbiological contamination is the leading cause of meat deterioration, and it has detrimental  
337 impact on safety and colour of meat (Fang *et al.*, 2017). Oxidation of fat and myoglobin results in  
338 colour deterioration, rancidity, and contributes to secondary defects of undesirable flavours and  
339 odours (Han *et al.*, 2018; Domínguez *et al.*, 2019). Minced meat products including meat patties  
340 are typically sold refrigerated at temperature ranging from 2- 5 °C. Main solutions of extending  
341 shelf-life of fresh meat and ensuring consumers safety is to prevent or reduce product  
342 contamination and retard or hinder growth of pathogenic and spoilage microorganisms (Kumar *et*  
343 *al.*, 2017; Santiesteban-López *et al.*, 2022).

## 344 **2.11 Physical properties of mutton meat**

### 345 **2.11.1 Colour of meat**

346 Consumers view colour as indication of freshness and healthiness of the product;  
347 therefore, colour has greater impact on meat buying choices than all other quality criterion  
348 (Neethling *et al.*, 2017; Ardeshiri and Rose, 2018). A link among colour liking and willingness to  
349 buy was observed, with consumers selecting against non-red beef and mutton (meat that is purple  
350 or brown) (Hughes *et al.*, 2019). Colour is initial quality attribute that attract consumers, and it can  
351 be quantified both subjectively and objectively. The red colour in meat is widely recommended by  
352 consumers, whereas brown, discoloured meat is not (Silvestri *et al.*, 2020; Xiong *et al.*, 2020).  
353 Consumers' viewpoint is to blame for these disparities in liking. When compared to discoloured  
354 meat, individuals view red meat to be healthier, fresher, and of higher consuming quality (Hastie  
355 *et al.*, 2020).

356 A prevalent misunderstanding between consumers is that meat colour signifies freshness,  
357 healthiness, and consuming quality (Liu *et al.*, 2022). Even though discolouration in meat can  
358 reflect its good value, it is not always the issue. Discoloured meat can be of higher-grade meat  
359 as compared to red meat. Meat colour is impacted by a variety of inherent and external influences;  
360 therefore, it is not always a reliable indicator of meat quality (Suman *et al.*, 2014; Poveda-Arteaga  
361 *et al.*, 2023). Nevertheless, consumers will continue to utilise meat colour to determine quality of  
362 the meat because is merely quality criterion they can examine at the moment of buying (Testa *et*  
363 *al.*, 2021). As a result, on-going investigation is required to determine the elements that affect

364 meat colour and stability of colour to guarantee that fresh meat have the best possible colour and  
365 shelf-life.

## 366 **2.12 Lipid oxidation in meat**

367 Lipid oxidation linked with formation of free radicals is an impulsive interaction of ambient  
368 oxygen with double bond of polyunsaturated fatty acid of meat. This results to oxidative damage  
369 and degradation of the quality of meat and meat products by decreasing the durability (Chaijan  
370 and Panpipat, 2017; Aminzare *et al.*, 2019), and development of unwanted sensory attributes,  
371 including as colour, nutritional value, texture, aroma, and taste resulting to rancidity. It is also  
372 accountable for off-flavours and bad taste, which are the most common causes for consumers to  
373 reject the products (Torriceo *et al.*, 2018; Dominguez *et al.*, 2019). Lipid oxidation occurs as a  
374 three-stage radical chain reaction, with initiation, propagation, and termination respectively  
375 (Cardoso-Ugarte and Sosa-Morales, 2022)

## 376 **2.13 Mechanisms of lipid oxidation in meat**

### 377 **2.13.1 Initiation stage**

378 A hydrogen atom is extracted from the carbon nearest to the dual bond of an unsaturated  
379 fatty acid resulting to formation of alkyl radical ( $R\cdot$ ) and free hydrogen atom ( $H\cdot$ ) at the initial phase  
380 of lipid oxidation, as indicated in equation one (E1) (Bumbadiya *et al.*, 2023). In the initial phase,  
381 a lag stage is usually detected when development fat oxidation in foods is gradual. This is owing  
382 to the sluggish generation of free radicals before build-up of hydroperoxides, as well as the fact  
383 that free radicals favourably attack or oxidise naturally antioxidants that shields fatty acids during  
384 early phases of oxidation (Cheng, 2016). Catalysts that speed up the reaction includes, metal  
385 ions, temperature (heating), and ultra-violet light (Amaral *et al.*, 2018; Hazrat *et al.*, 2021).

386  $RH \xrightarrow{\hspace{1.5cm}} R\cdot + H\cdot \dots\dots\dots$  (Equation 1).

### 387 **2.13.2 Propagation stage**

388 Propagation stage is the second step of lipid oxidation that takes place between the  
389 reaction of free alkyl radical ( $R\cdot$ ) formed throughout the initiation stage and molecular oxygen  
390 leading to the very reactive peroxy radical (Radical interacting with oxygen-  $ROO\cdot$ ) (Equation 2),  
391 which is able to react with unsaturated fatty acids and extract hydrogen atom, leading to formation  
392 of hydroperoxides ( $ROOH\cdot$ ) and alkyl radical ( $R\cdot$ ) (Equation 3) (Chaijan and Panpipat, 2017;

393 Hazrat *et al.*, 2021). Later as hydroperoxides decompose, volatile aromatic compounds are  
394 formed, which give meat its rancid odour and off-flavour (Flores *et al.*, 2021; Hazrat *et al.*, 2021).



### 397 2.13.3 Termination stage

398 Termination is the final stage of lipid oxidation which occurs between reaction of peroxy  
399 and alkyl radicals leading to development of non-radicals relatively stable products like alkanes,  
400 conjugated dienes, and ketones, which are produced when level of free radicals is adequately  
401 high (Equation 4, 5, and 6) (Karademir, 2019). Aldehydes production is assessed as being directly  
402 proportionate to degradation of meat colour and flavour, as well as functionality and stability of  
403 protein (Amaral *et al.*, 2018; Hussain *et al.*, 2021). Aldehydes can also cause malignancy,  
404 atherosclerosis, and purative mutagens (Ganesan and Xu, 2020).



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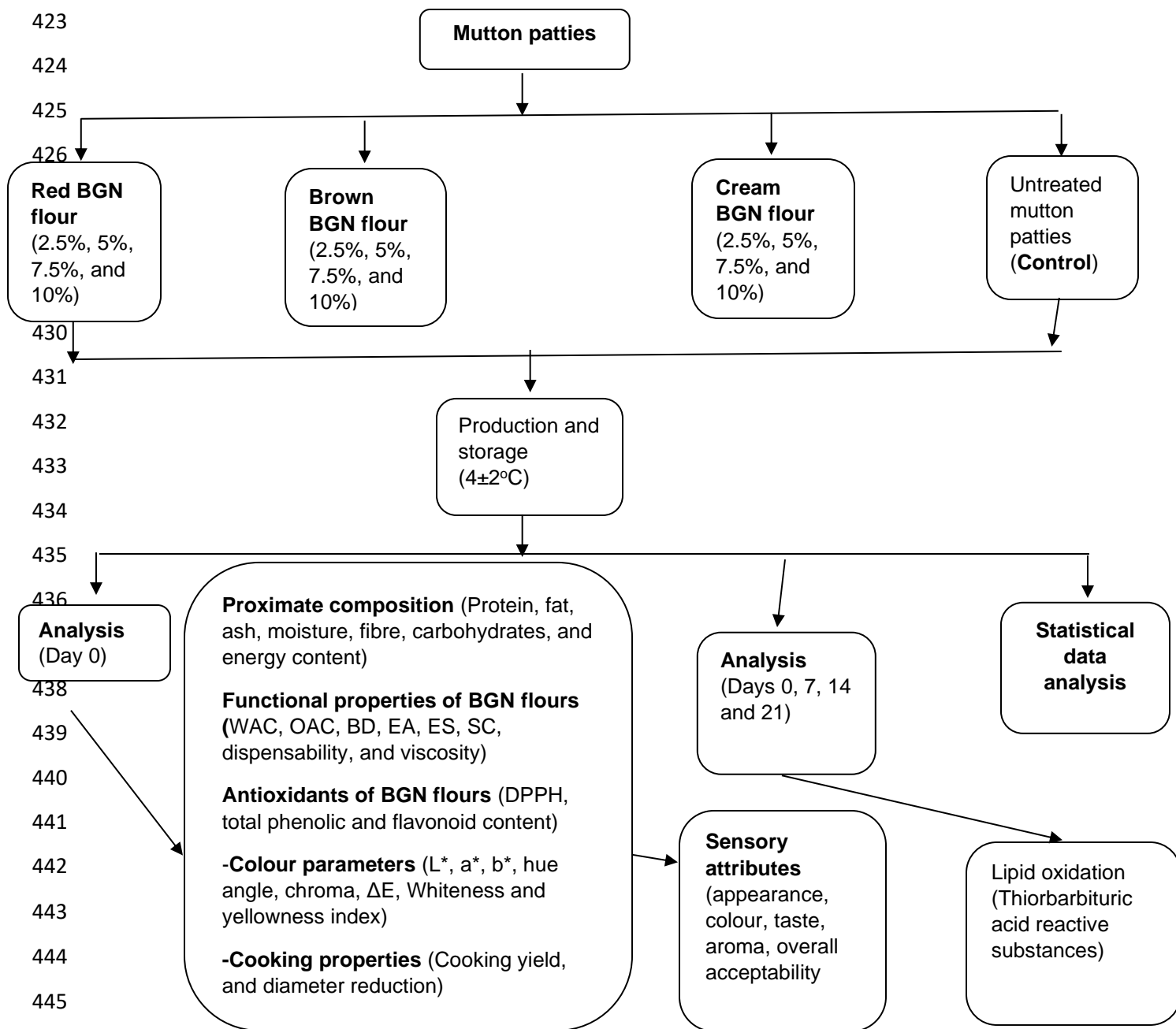
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419 **CHAPTER 3: MATERIAL AND METHODS**

420 **3.1. Study design**

421 The following figure summarises the processing of the samples and analysis conducted  
422 throughout the experiment.



447 **Figure 4:** Schematic diagram of the research project utilising factorial design. L\*= lightness; a= redness; b\*=  
448 yellowness; WAC= water absorption capacity; OAC= oil absorption capacity; EA= emulsion activity; ES=  
449 emulsion stability; BD= bulk density; SC= swelling capacity; DPPH= 2,2 Diphenyl-1-picryl-hydrazyl

## 450 **3.2 Raw materials, chemicals, and reagents**

451 Ten (10) kilogram of mixed BGN (brown, cream, and red-coated) grains was purchased  
452 from a supermarket in Thohoyandou, Limpopo Province, South Africa. The reagents acetone,  
453 acetic acid, methanol, ethanol, Folin-Ciocalteu, Hydrochloric and Thiorbarbituric acid were  
454 purchased from Merck (Pty, Ltd., Midrand, Gauteng province, South Africa). All the chemicals  
455 utilised were of analytical grade.

## 456 **3.3 Preparation of Bambara groundnut flour**

457 The BGN grains were grouped according to their colour (brown, cream, and red)  
458 separately and washed properly with tap water to remove dirt, small branches, and premature  
459 grains were physically discarded. The grains were then oven-dried at 37°C for 48 hours. The dried  
460 grains were ground utilising Retsch miller (ultra-centrifugal mill ZM 200, Germany) and sieved  
461 using a 212 µm sieve size to produce the final finer powder and packaged inside vacuum plastic  
462 bags and stored in a dry, cool area until utilisation (Alakali *et al.*, 2010).

## 463 **3.4 Preparation of mutton patties**

464 Ten (10) kilogram of deboned mutton meat was bought at Newco meat butchery in  
465 Shayandima, Limpopo Province, South Africa. Every ounce of subcutaneous and intramuscular  
466 fat as well as connective tissues were manually trimmed off and then the meat was chopped into  
467 small pieces and ground utilising meat mincer (P-22, Tallers Ramon, Barcelona, Spain) through  
468 5 mm plates. Throughout the mincing process, cold water with ice was added. Four (4) blends  
469 (for each BGN variety) were formulated by combining minced mutton meat with 2.5%, 5.0%, 7.5%,  
470 and 10% of BGN flours and 100% mutton patties was used as a control. To get a uniform mixture,  
471 each part was quietly mixed manually in a container for approximately 5 minutes. A petri-dish was  
472 utilised to shape the mix to form the patties (75 mm x 15 mm), two batches were produced and  
473 packaged in low-density polyethylene bags. For analysis of lipid oxidation, the patties were stored  
474 at refrigerator at  $4 \pm 2^\circ\text{C}$  and analysed at day 0, 7, 14, and 21 (Argel *et al.*, 2020).

## 475 **3.5 Determination of functional properties of Bambara groundnut flour**

### 476 **3.5.1 Water absorption capacity and oil absorption capacity**

477 Water absorption capacity (WAC) and oil absorption capacity (OAC) were obtained in  
478 triplicate where one gram of sample was measured into 25 mL centrifuge tubes. A small amount  
479 of distilled and refined sunflower oil was added and stirred till the samples became separated with  
480 water or oil. The flours were centrifuged for 20 minutes at 3000 rpm and allowed to rest for 30  
481 minutes at ambient condition. The water or oil that was released as result of centrifugation was

482 emptied (Ferreira *et al.*, 2018). The OAC and WAC were calculated as follows in equation 1 and  
483 2:

484 Water absorption capacity (%) =  $\frac{\text{weight of water absorbed}}{\text{weight of the sample}} \times 100$  (Equation 1)

485 Oil absorption capacity (%) =  $\frac{\text{weight of oil absorbed}}{\text{weight of the sample}} \times 100$  (Equation 2)

### 486 3.5.2 Emulsifying activity

487 Emulsifying activity was obtained following the procedure outlined by Arise *et al.* (2015).  
488 One (1) gram of flour was put in 15 mL of water in centrifuge tubes and centrifuged at 20000 rpm  
489 for 30 s for homogenisation; then sunflower oil (15 mL) was poured, emulsified for 1.5 minutes,  
490 and centrifuged at 750 rpm for 5 min. Emulsifying activity was computed using equation 3:

491 Emulsifying activity (%) =  $\frac{\text{Height of the remaining emulsified layer}}{\text{Height of the whole tube content}} \times 100$  (Equation 3)

### 492 3.5.3 Emulsion stability

493 Emulsion stability was obtained similarly as the emulsifying activity and heated at 80°C for  
494 30 minutes and then cooled in tap water for 15 min and centrifuged at 750 rpm for 5 min. Emulsion  
495 stability was computed using the equation 4:

496 Emulsion stability (%) =  $\frac{\text{Height of the remaining emulsifying layer}}{\text{Height of the whole tube content}} \times 100$  (Equation 4)

### 497 3.5.4 Bulk density

498 Bulk density was measured using the procedure outlined by Jenfa and Akinrinde (2019). Twenty  
499 (20) grams of the powder was put in a 100 mL measuring cylinder and hit at the base with a hand.  
500 The ultimate volume was measured after the cylinder was put on a flat surface. The bulk density  
501 was determined using equation 5:

502 Bulk density =  $\frac{\text{Weight of sample g/ml}}{\text{Volume of the sample}}$  (Equation 5)

503

### 504 3.5.5 Swelling capacity

505 Swelling capacity was determined using the procedure explained by Arise *et al.* (2019).  
506 Ten grams of BGN flour was put in a 300 mL measuring cylinder, and the volume covered was  
507 measured. The powder was then poured with 150 mL of distilled water and set aside for 4 hours.  
508 Following swelling, the ultimate volume was measured. The swelling percentage was determined  
509 using equation 6:

510 Swelling capacity (%) =  $\frac{Final\ volume - Initial\ volume}{Initial\ volume} \times 100$  (Equation 6)

### 511 3.5.6 Dispensability

512 Dispensability was obtained using a procedure outlined by Jenfa and Akinrinde (2019). In  
513 a 100 mL measuring cylinder, 10 g of BGN flour was added, and 100 mL of distilled water was  
514 then poured. The mixture was aggressively agitated and let to rest for 3 h, then the volume of  
515 settled particles was recorded. The dispensability was determined using equation 7:

516 Dispensability (%) = 100 – The volume of settled particles (Equation 7)

### 517 3.5.7 Viscosity

518 The viscosity of the sample was obtained utilising the Brookfield viscometer (RUVDE230,  
519 USA). Three point five grams (3.5 g) of the sample (adjusted to 14 % moisture content) and  
520 distilled water was poured to get a mass of the sample of about 25 g. The sample was heated at  
521 50°C for one (1) minutes while stirring to enable complete dispersion. The resistance of the flour  
522 mixture to the mixing paddles of the viscometer was utilised to measure the viscosity (cP) (Mudau  
523 *et al.*, 2022).

## 524 3.6 Determination of antioxidants properties of Bambara groundnut flour

### 525 3.6.1 DPPH Radical Scavenging Activity of mutton patties

526 DPPH assay was determined using a procedure outlined by Oforu *et al.* (2020) with slight  
527 modifications. Two (2) grams of each flour was measured and placed inside beakers and 20 mL  
528 of methanol (30%) was poured and sornicated for 10 minutes in an ultrasonic bath. Afterwards,  
529 the samples were then centrifuged (Rotina 380R-Labotec Ecotherm, Midrand, South Africa) at  
530 3000 rpm for 10 min and filtered into beakers utilising Whatman filter paper. Two (2) millimeters  
531 of the sample extract was added to 2 mL of 0.1 Mm DPPH in 10 mL test tubes. The mixture was  
532 shaken vigorously, kept at ambient condition for 30 minutes, and absorbance was analysed at  
533 517 nm utilising UV-spectrophotometer (Shimadzu UV-1800, Japan). The DPPH content was  
534 computed using the following equation 8:

535 DPPH (%) =  $\frac{Absorbance\ of\ the\ control\ (methanol\ with\ DPPH\ solution) - Absorbance\ of\ the\ flour\ sample}{Absorbance\ of\ the\ control\ (methanol\ with\ DPPH\ solution)} \times 100$

536 (Equation 8)

### 537 3.6.2 Total phenolic content

538 Total phenolic content was analysed using a procedure outlined by Mahmoud *et al.* (2017)  
539 with slight modification. Two (2) grams of each flour was measured and deposited into the beakers

540 and 20 mL of methanol (30%) was then poured and sonicated for 10 minutes in an ultrasonic  
541 bath. The mixture was centrifuged at 3000 rpm for 10 min and filtered into beakers. Thereafter,  
542 0.5 mL of the sample was poured into test tubes and 1.5 mL of Folin-Ciocalteu reagent was also  
543 poured and let to stand for 5 minutes at ambient condition ( $25 \pm 2^\circ\text{C}$ ). After 5 minutes, 2 mL of  
544 sodium carbonate (7%) was poured and placed in an incubator for 50 minutes in a dim place with  
545 intermittent shake. The absorbance of the samples was determined utilising UV-  
546 spectrophotometer (Shimadzu UV- 1800, Japan) at 725 nm. The total phenolic content was  
547 computed using the standard curve and reported as gallic acid (mg GAE/g).

### 548 **3.6.3 Flavonoids content**

549 Total flavonoids content was analysed using a colorimetric procedure. Two grams of each  
550 flour was measured and placed into beakers and 20 mL of methanol (30%) was then poured and  
551 sonicated for 10 minutes in an ultrasonic bath. Afterwards, the mixture was centrifuged at 5000  
552 rpm for 10 min and filtered using Whatman paper. Then, 0.3 mL of sample extracts was added  
553 into a test tube and 3.4 mL of 30% methanol was added, and 0.15 mL of sodium nitrite ( $\text{NaNO}_2$ )  
554 was also added. Thereafter, 0.15 mL of Aluminum chloride hexahydrate ( $\text{AlCl}_3 \cdot 6\text{H}_2\text{O}$ ) was  
555 introduced to the mixture and let to rest for 5 minutes and 1 mL of NaOH was introduced. After  
556 that the absorbance was measured at 506 nm utilising UV-spectrophotometer (Mashau *et al.*,  
557 2021a). The total flavonoids content was computed using the standard curve and reported as  
558 catechin (mg CE/g).

## 559 **3.7 Determination of proximate composition of mutton meat**

### 560 **3.7.1 Moisture content**

561 Moisture content was obtained succeeding the procedure described in AOAC (2007)  
562 method 945.32 where 10 g of the sample was weighed and transferred into a blank crucible which  
563 was placed in an oven dryer at  $105 \pm 5^\circ\text{C}$  for 16-18 hours. Then the crucibles were placed in the  
564 desiccator to cool for an hour. After cooling, the weight loss was measured, and the moisture  
565 content was calculated in percentage using equation 9:

$$566 \text{ Moisture (\%)} = \frac{\text{fresh weight} - \text{dry weight}}{\text{fresh weight}} \times 100 \quad (\text{Equation 9})$$

### 567 **3.7.2 Ash content**

568 Ash content was analysed according to AOAC (2007) method no. 923.03. Ten grams (10 g) of  
569 sample was put into the empty crucibles which were put into a muffle furnace for 3 hours at  $550^\circ\text{C}$ .  
570 Therefore, the crucibles were placed in a desiccator to cool. After cooling, the weight loss was  
571 measured to determine ash content expressed in percentages using equation 10:

572 Ash content =  $\frac{\text{Weight of crucible+ash}(g)-\text{Weight of empty crucible}(g)}{\text{Weight of crucible+sample}(g)-\text{Weight of empty crucible}(g)} \times 100$  (Equation 10)

573 **3.7.3 Protein content**

574 Protein content was obtained using the Dumas procedure described in AOAC (2007)  
575 method no. 992.15. One gram (1.0 g) of the meat sample was put into a 528-203 ceramic  
576 combustion utilising a 604-494 plunger with a 502-210 tube. The boats containing samples were  
577 then transferred into the appropriate position in the autoloader. The analysis sequence was  
578 initiated to analyse the protein content.

579 **3.7.4 Fat content**

580 The fat content was determined utilising the Ankom technology described in AOCS (1989)  
581 official method no. 2. Two grams (2 g) of the sample was transferred into the filter bag and  
582 weighed. After that, the bag was sealed, followed by drying the sample in an oven drier at 102°C  
583 for 3 hours. Then the samples were placed in a desiccator for cooling and measured. The  
584 automated extraction was then performed using petroleum ether for 90 minutes at 60°C.  
585 Afterwards, the samples were dried at 105°C for 30 minutes in an oven drier, followed by cooling  
586 in a desiccator and measured. The fat content was calculated using equation 11:

587 Fat (%) =  $\frac{\text{Weight of pre-dried smple} - \text{Weight of dried sample after extraction}}{\text{Weight of the sample}} \times 100$  (Equation 11)

588 **3.7.5 Crude fibre**

589 Crude fibre of the samples was determined following AOAC (2007) method no. 985.33.  
590 One gram 1 g of each sample was measured in triplicates into 400 ml beakers. Therefore, the  
591 samples were heated for 30 minutes with 100 mL solution of Sulphuric acid (H<sub>2</sub>SO<sub>4</sub>) (1.25 %)  
592 using reflux. The mixture was filtered through filter paper and then rinsed with hot water to clear  
593 the base remains completely. Therefore, the filtrate was transferred into the clean conical flask  
594 by washing it with 100 mL of NaOH (1.25 %), and then boiled again for 30 minutes. After boiling  
595 the mixture was filtered again and then washed again with hot water to completely remove the  
596 acidic residues. The residue was left on filter paper and transferred into crucibles and placed on  
597 the hot plate to evaporate excess water and measured. The remains were burnt in the muffle  
598 furnace for 3 hours at 550 °C, and then cooled in a desiccator for an hour and then measured.  
599 The fibre content was then calculated using equation 12:

600 Crude fibre =  $\frac{\text{Weight of crucible with fibre} - \text{Weight of crucible with ash}}{\text{Weight of sample}}$  (Equation 12)

601 **3.7.6 Carbohydrates content**

602 Carbohydrates amount was obtained by calculating the difference of percentage of moisture, fat,  
603 protein, ash, and crude fibre, using equation 13 (Sultana, 2020):

604 Carbohydrates (%) = 100- (moisture + fat + protein + Crude fibre). (Equation 13)

605 **3.7.7 Energy content**

606 The total energy was calculated using equation 14: (Kinyuru, 2021).

607 Energy value (Kcal) = g/ 100 g carbohydrate x 4 + g/ 100 g fat x 9 + g/ 100 g protein x 4 (Equation  
608 14)

609 **3..8 Determination of physical properties of mutton patties**

610 **3.8.1 Cooking yield**

611 Raw mutton patties were weighed, then cooked and surface-dried with a filter and  
612 measured once more utilising an analytical scale balance (Ali *et al.*, 2022). The cooking yield was  
613 calculated using equation 15:

614 
$$\text{Cooking yield} = \frac{\text{Weight of cooked mutton patties}}{\text{Weight of raw mutton patties}} \times 100$$
 (Equation 15)

615 **3.8.2 Diameter reduction of patties during cooking**

616 Diameter was determined following a method described by Kilincceker (2018) using equation 16:

617 
$$\text{Diameter Reduction (\%)} = \frac{(\text{Diameter of cooked raw patties} - \text{Diameter of cooked patties})}{\text{Diameter of raw patties}} \times 100$$

618 (Equation 16)

619 **3.9 Colour attributes**

620 Colour attributes were determined at three randomly chosen spots of the samples through  
621 a method outlined by Neethling *et al.* (2016) using Hunter Lab colourflex (Reston VA, U.S.A, D65).  
622 The instrument was initially calibrated using white and black tiles to determine the colour  
623 parameters such as L\* (lightness), a\* (redness), b\* (yellowness). The whiteness index (WI), and  
624 yellowness index (YI) were determined using the equations (Mehdizadeh *et al.*, 2020):

625

626 
$$\text{Hue} = \tan^{-1} (b^*/a^*)$$
 (Equation 17)

627 
$$\text{Chroma} = (a^{*2} + b^{*2})^{1/2}$$
 (Equation 18)

628 
$$\Delta E = \sqrt{(\Delta L)^2 + (\Delta a)^2 + (\Delta b)^2}$$
 (Equation 19)

629  $WI = 100 - \sqrt{(100 - L^*)^2 + (a)^2 + (b)^2}$  (Equation 20)

630  $YI = \frac{142.86b}{L^*}$  (Equation 21)

### 631 **3.10 Texture profile analysis**

632 The texture characteristics of grilled patties were carried out using a method outlined by  
633 Andrés *et al.* (2011). The patties were pressed two times to 30% of their initial height amongst  
634 levelled plates. The texture analyser (Stable Micro Systems, London, UK) with a 75 mm diameter  
635 probe  $\text{mms}^{-1}$  (SMSP/75), linked up with a PC, and software provided by texture operating system  
636 corporation (New York, United States) was used. Hardness (maximum power of first compression  
637 phase, N), springiness (length of the discovered height of the sample on the succeeding (2<sup>nd</sup>)  
638 pressing split by the initial pressing length, mm/mm), cohesiveness (proportion of opposite of  
639 negative areas of the number two cycle to an area of the first cycle, J/J), chewiness (hardness  $\times$   
640 cohesiveness  $\times$  springiness, N), and resilience (area through the removal of the initial pressing  
641 split by the area of the initial pressing, J/J) were obtained.

### 642 **3.11 Determination of lipid oxidation**

643 Thiorbarbituric acid reactive substance (TBARS) values were obtained following the procedure  
644 described by Mashau *et al.* (2021b) for 21 days (0, 7, 14 & 21) where standard solution of TBA  
645 was prepared in glacial acetic acid by dissolving 57.66 mg of TBA in 100 mL of glacial acetic acid.  
646 TBA solution was prepared on each day of the analysis. One gram of each raw patties sample  
647 was put in 25 mL test tube and 50% glacial acetic acid (solvent). BHT (0.01 %) was utilised to  
648 prevent the sample from oxidising. The samples mixture was centrifuged (Rotina 380R-Labotec  
649 Ecotherm, Midrand, South Africa) at 3000 rpm for 15 minutes and filtered into test tubes using  
650 Whatman papers. The mixture was boiled in water bath at 95 °C for 60 min, and test tubes were  
651 allowed to cool at ambient temperature ( $25 \pm 2$  °C) and absorbance was read at 532 nm using  
652 UV-visible spectrophotometer (Shimadzu UV-1800, Japan).

### 653 **3.12 Sensory analysis**

654 Sensory evaluation was undertaken by 60 consumer panellists with an age range of 18 to  
655 50 years, encompassing university students and staff members who had previous knowledge with  
656 sensory analysis of meat. The formed patties were cooked by grilling in a Hobart CN85-19  
657 convection oven (Hobart Corp., Troy, Ohio, USA) for 20 min at 160°C to a core temperature of  
658 80°C as measured at the geometrical centre. To obtain homogeneous cooking, the patties were  
659 flipped over every 10 min. The prepared mutton patties were kept on warm trays in sealed plates

660 for less than 10 min. Warmed portions (10 g) of each of the patties were placed in the white  
661 polystyrene plates and given to every participant with 3-digit numbers in a random and consistent  
662 way for analysis. The panellists were given tap water to rinse their mouth after tasting every  
663 sample. Sensory testing was carried out in a well-lit and well-designed environment. To evaluate  
664 appearance, taste, texture, colour, and total acceptability, analysis was undertaken using a 9-  
665 point hedonic scale (9= extremely like; 5= indifferent; 1= extremely dislike). All participants were  
666 given written information about the study and the products to be tasted before giving their informed  
667 consent (Argel *et al.*, 2020).

### 668 **3.13 Statistical analysis**

669 The experiment and analyses were duplicated to validate the results. The data obtained  
670 was analysed by one way analysis of variance (ANOVA) utilising Statistical Package for the Social  
671 Sciences (SPSS) version 28 for Bambara groundnut flours. Two-way analysis of variance was  
672 used for mutton patties. The disparity between the mean of each analysis done in triplicates was  
673 determined utilising Duncan's multiple range test and Ryan-Einot-Gabriel-Welsch (REGWQ) test  
674 for BGN flours, and mutton patties, respectively. The acceptable significant difference was at  $p \leq$   
675 0.05 level. The spearman rank order correlation test was utilised to determine the correlation  
676 between cooking yield and diameter reduction. Primary sensory evaluation data were subjected  
677 to statistical calculations and on their bases, regression analysis was performed for each of the  
678 tested mutton patties samples.

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688 **CHAPTER 4: RESULTS AND DISCUSSION**

689 **4.1 Bambara groundnut flour characteristics**

690 The nutritional, functional and physical properties of BGN flours are indicated in Table 8.  
 691 The moisture content of red BGN flour (7.13%) was significantly lower ( $p < 0.05$ ) than that of  
 692 cream BGN flour (8.85%), whereas brown BGN flour was not significantly ( $p < 0.05$ ) different from  
 693 cream and red BGN flours. However, the moisture content of all BGN flours was typically less and  
 694 between the anticipated range (0-14%) in pulse flours (Shanono and Muhammed, 2015; Buckman  
 695 *et al.*, 2018; Skřivan *et al.*, 2021).

696 **Table 8.** Bambara groundnut flours properties.

Properties	Cream	Red	Brown
Moisture (%)	8.85 ± 0.92 <sup>b</sup>	7.13 ± 0.10 <sup>a</sup>	8.17 ± 0.58 <sup>ab</sup>
Ash (%)	2.89 ± 0.42 <sup>a</sup>	3.14 ± 0.35 <sup>a</sup>	3.39 ± 0.10 <sup>a</sup>
Protein (%)	19.67 ± 0.21 <sup>a</sup>	19.03 ± 0.70 <sup>a</sup>	21.07 ± 0.98 <sup>b</sup>
Fat (%)	6.67 ± 0.07 <sup>b</sup>	6.70 ± 0.27 <sup>b</sup>	6.42 ± 0.07 <sup>a</sup>
Fibre (%)	4.76 ± 0.03 <sup>a</sup>	7.73 ± 0.07 <sup>c</sup>	5.89 ± 0.05 <sup>b</sup>
Carbohydrates (%)	57.45 ± 0.11 <sup>c</sup>	56.27 ± 0.08 <sup>b</sup>	55.06 ± 0.47 <sup>a</sup>
Energy (kcal/100g)	368.48 ± 1.17 <sup>b</sup>	361.50 ± 4.43 <sup>a</sup>	362.29 ± 2.89 <sup>ab</sup>
L*	88.07 ± 0.08 <sup>c</sup>	81.82 ± 0.59 <sup>a</sup>	83.43 ± 0.12 <sup>b</sup>
a*	1.27 ± 0.02 <sup>a</sup>	2.95 ± 0.08 <sup>b</sup>	2.91 ± 0.05 <sup>b</sup>
b*	10.48 ± 0.06 <sup>c</sup>	7.18 ± 0.05 <sup>a</sup>	8.39 ± 0.12 <sup>b</sup>
Hue angle	82.78 ± 0.66 <sup>c</sup>	67.01 ± 0.81 <sup>a</sup>	70.79 ± 0.62 <sup>b</sup>
Chroma	10.56 ± 0.06 <sup>c</sup>	7.77 ± 0.07 <sup>a</sup>	8.70 ± 0.34 <sup>b</sup>
WI	84.07 ± 0.09 <sup>c</sup>	80.23 ± 0.57 <sup>a</sup>	81.20 ± 0.12 <sup>b</sup>
YI	17.00 ± 0.10 <sup>c</sup>	12.54 ± 0.18 <sup>a</sup>	14.37 ± 0.21 <sup>b</sup>
WAC (%)	182.49 ± 1.03 <sup>a</sup>	204.38 ± 2.24 <sup>b</sup>	201.91 ± 3.23 <sup>b</sup>
OAC (%)	166.87 ± 1.11 <sup>a</sup>	169.03 ± 3.72 <sup>ab</sup>	174.21 ± 3.50 <sup>b</sup>
EA (%)	49.64 ± 0.31 <sup>a</sup>	51.73 ± 1.35 <sup>b</sup>	52.83 ± 1.05 <sup>b</sup>
ES (%)	51.37 ± 1.05 <sup>a</sup>	49.32 ± 1.18 <sup>a</sup>	51.26 ± 0.79 <sup>a</sup>
BD (g/ml)	0.66 ± 0.01 <sup>a</sup>	0.68 ± 0.01 <sup>b</sup>	0.70 ± 0.00 <sup>c</sup>
SC (g/g)	0.41 ± 0.03 <sup>b</sup>	0.24 ± 0.05 <sup>a</sup>	0.18 ± 0.13 <sup>a</sup>
Dispensability (%)	57.33 ± 2.31 <sup>b</sup>	52.00 ± 2.00 <sup>a</sup>	48.00 ± 2.00 <sup>a</sup>
Viscosity (cP)	2666 ± 5.57 <sup>a</sup>	3402 ± 7.21 <sup>b</sup>	3464.67 ± 4.51 <sup>b</sup>
DPPH (%)	95.15 ± 3.67 <sup>b</sup>	87.19 ± 0.87 <sup>a</sup>	87.54 ± 0.74 <sup>a</sup>
TPC (mg GAE/g)	0.79 ± 0.01 <sup>a</sup>	0.98 ± 0.01 <sup>b</sup>	0.91 ± 0.06 <sup>b</sup>
TFC (mg CE/g)	0.06 ± 0.00 <sup>ab</sup>	0.05 ± 0.00 <sup>a</sup>	0.07 ± 0.01 <sup>b</sup>

697 Distinct small letters (superscripts) on the same row display that mean values are significantly different ( $P$   
 698  $< 0.05$ ). BGN= Bambara groundnut; L\*= lightness; a= redness; b\*= yellowness; WI= whiteness index; YI=  
 699 yellowness index; WAC= water absorption capacity; OAC= oil absorption capacity; EA= emulsion activity;  
 700 ES= emulsion stability; BD= bulk density; SC= swelling capacity; DPPH= 2,2 Diphenyl-1-picryl-hydrazyl;  
 701 TPC= Total phenolic content; TFC= Total flavonoids content.

702 The low moisture content suggested that BGN flours would have an extended shelf-life and be  
703 less susceptible to the proliferation of microorganisms during storage if they are packaged and  
704 stored appropriately (Sruthi and Rao, 2021; Mashau *et al.*, 2022). Similarly, Oyeyinka *et al.* (2021)  
705 found higher moisture in cream BGN flour than in red and brown BGN flours. The ash content of  
706 all BGN flours was not significantly ( $p < 0.05$ ) different from each other with values varying from  
707 2.89 to 3.39%.

708 The protein content of brown BGN flour (21.07%) was significantly ( $p < 0.05$ ) higher than  
709 that of cream (19.67%) and red BGN (19.03%) flours. Differences in the amount of protein of BGN  
710 flours might be due to variations in the type of the cultivar or unique genetic characteristics if  
711 cultivated in comparable conditions (Adeleke *et al.*, 2018; Ikegwu *et al.*, 2023). Oyeyinka *et al.*  
712 (2017) found significantly higher protein content in brown than red BGN flour. High protein content  
713 makes BGN flour suitable for inclusion in protein-rich diets or as a supplement for diets that lack  
714 adequate protein (Adeleke *et al.*, 2017).

715 The fat content of brown BGN flour (6.42%) was less than that of cream (6.67%) and red  
716 BGN (6.70%) flours. The low-fat content in BGN flours was anticipated given that BGNs are dry  
717 grains and typically have lower than 10% fat when compared to oily grains like groundnuts and  
718 soybeans (Nwadi *et al.*, 2020). Kaptso *et al.* (2015) stated that the low amount of fat of BGN flours  
719 might be because of their respective great amount of protein content, as supported by results in  
720 Table 8. A low amount of fat suggested that BGN flour could be preserved with minimal loss of  
721 quality until future use (Deepa and Umesh Hebbar, 2017).

722 The crude fibre of red BGN flour (7.73%) was significantly higher ( $p < 0.05$ ) than that of  
723 brown (5.89%), and cream BGN (4.76%) flours, respectively. Variations in the amount of crude  
724 fibre of BGN flours might be because of the type of the cultivar (Ikegwu *et al.*, 2023). Similarly,  
725 Shanono and Muhammad (2015) found higher crude fibre content in red BGN flour than in brown  
726 BGN flour. Significant crude fibre content in BGN flours is important in reducing hypertension and  
727 serum cholesterol levels and providing protection from cardiovascular conditions. It also promotes  
728 healthy bowel function and increases faecal bulk (Tan *et al.*, 2021; Aremu *et al.*, 2022).

729 The carbohydrate content of cream BGN flour (57.45%) was significantly higher ( $p < 0.05$ )  
730 than that of red (56.27%) and brown BGN (55.06%) flours. Regardless of the type of cultivar,  
731 carbohydrate was the most predominant component in all BGN flours. Adeleke *et al.* (2017) found  
732 higher carbohydrate in white (cream) BGN flour than in red BGN flour. Consuming BGNs can  
733 supply the body with enough energy due to their high carbohydrate content (Schulz and Slavin *et al.*,  
734 2021). The energy content of red BGN (361.50 kcal/100g) flour was less than that of cream  
735 BGN (368.48 kcal/100g) flour but not significantly different ( $p < 0.05$ ) from brown BGN flour

736 (362.29 kcal/100g). Generally, the energy content of all BGN flours was high. The high-energy  
737 content of cream BGN flour was due to its respective high carbohydrate (Bunmee *et al.*, 2022).

738 The BGN flour's colour properties are indicated in Table 8. The cream BGN (88.07) flour  
739 was significantly higher ( $p < 0.05$ ) in lightness ( $L^*$ ) value than brown (83.43) and red BGN (81.82)  
740 flours. Therefore, BGN flours with less  $L^*$  values, displayed significantly higher ( $p < 0.05$ )  $a^*$   
741 values, probably due to their relatively higher total phenolic content (Table 8) which is responsible  
742 for the red colour (Bamshaiye *et al.*, 2011; Parikh and Patel, 2018). The two BGN flour (red and  
743 brown) varieties were significantly higher ( $p < 0.05$ ) in  $a^*$  values than cream BGN flour. The higher  
744 the total phenolic content of the grain the higher the  $a^*$  value and less  $L^*$  value (Tsamo *et al.*,  
745 2018). Oyeyinka *et al.* (2017) reported similar trends in the colour of red, brown, and cream BGN  
746 flours with respect to their phenolic content. The yellowness ( $b^*$ ) value of cream BGN (10.48) flour  
747 was significantly higher ( $p < 0.05$ ) than that of brown and red BGN flours which had values of 8.39  
748 and 7.18, respectively. The higher  $b^*$  value of the flour might be due to greater amount of flavonoid  
749 content as shown in Table 8 which is responsible for the yellow colour in the grain (Khatun, 2021).

750 The chroma values of cream BGN flour was significantly higher ( $p < 0.05$ ) than that of  
751 brown and red BGN flours, varying from 7.77 to 10.56. The high chroma values of cream BGN  
752 flour was probably because of its relatively high  $b^*$  value since it depends on it (Falade and  
753 Akeem, 2020; Ramashia *et al.*, 2021). The higher chroma value of the flour displays the highly  
754 intense colour of the sample as perceived by consumers (Mubaiwa *et al.*, 2018; Falade and  
755 Akeem, 2020). A similar trend of chroma and  $b^*$  values was reported by Falade and Akeem (2020)  
756 in African mesquite bean powders. The hue angle of cream BGN flour was significantly higher ( $p$   
757  $< 0.05$ ) than that of red and brown BGN flours.

758 The WI of cream BGN flour was significantly higher ( $p < 0.05$ ) than that of brown and red  
759 BGN flours ranging from 80.23 to 84.07. The greater WI value of cream BGN flour was probably  
760 because of its relatively high  $L^*$  value (Suliman *et al.*, 2023). The higher WI value of cream BGN  
761 flour might also be due to its relatively lower phenolic content as well as genetic differences in the  
762 grain varieties (Mang *et al.*, 2015; Uchechukwu-Agua *et al.*, 2015). Falade and Akeem (2020)  
763 found a higher WI of African mesquite bean flour due to its relatively high  $L^*$  value. The YI of  
764 cream BGN flour was significantly higher ( $p < 0.05$ ) than that of brown and red BGN flours. The  
765 higher YI value of cream BGN flour was probably due its correspondingly high  $b^*$  value  
766 (Uchechukwu *et al.*, 2015). A comparable pattern of YI and  $b^*$  was also reported by Uchechukwu-  
767 Agua *et al.* (2015) in different cassava flour varieties.

768

769 The functional properties of BGN flours are indicated in Table 8. The WAC of brown and  
770 red BGN flours was significantly higher ( $p < 0.05$ ) than that of the cream BGN flour ranging from  
771 182.49 to 204.38%. The greater polar amino acid residues of protein which are water-loving might  
772 be the reason for the higher WAC of brown and cream BGN flours (Adeleke *et al.*, 2017; Arise *et*  
773 *al.*, 2017). Thus, BGN flour with higher protein levels (Table 8) might have drawn up more water  
774 than those with lower protein content (Marikkar *et al.*, 2021). Iwe *et al.* (2016) stated that the  
775 disparity in WAC of various flours might be because of variations in their protein content, structural  
776 properties, and ability to absorb water. Furthermore, Adeleke *et al.* (2017) found higher WAC in  
777 brown than in white BGN flour. The OAC of brown BGN flour was significantly higher ( $p < 0.05$ )  
778 than that of red and cream BGN flours, with values varying from 166.89 to 174.21%. The disparity  
779 in the OAC of BGN flours might be due to the difference in the availability of non-polar side chains  
780 that might have attached to the hydrocarbon side chain of the oil amongst the BGNs flours  
781 (Chandra *et al.*, 2015; Awuchi *et al.*, 2019).

782 The emulsion activity (EA) of cream BGN flour was significantly lower ( $p < 0.05$ ) than that  
783 of red and brown BGN flours. The lower EA of cream BGN flour might be because of its respective  
784 less protein content in comparison to other BGN flours (Argel *et al.*, 2020). In addition, proteins  
785 with higher solubility and less surface hydrophobicity levels result in weaker interconnection  
786 surface covering lipid droplets (low EA) (Argel *et al.*, 2020; Abiala *et al.*, 2022). This result might  
787 also be due to the variety of the sample (Iwe *et al.*, 2016). The EAs of BGN flours are higher than  
788 the EAs of cowpea, soya bean, and peanut (Abiala *et al.*, 2022).

789 The bulk density (BD) of brown BGN flour was significantly higher ( $p < 0.05$ ) than that of  
790 red and cream BGN flours with values varying from 0.66 to 0.70 g/mL. The disparity in BD might  
791 be because of the presence of starch content (Iwe *et al.*, 2016), therefore a large amount of  
792 carbohydrate might account for a greater amount of BD in BGN flours (Adeleke *et al.*, 2017). The  
793 greater the BD, the smaller packing volume and the stiffer the packaging material is needed, and  
794 vice versa (Mi and Ejeh, 2018; Awuchi *et al.*, 2019). Adeleke *et al.* (2018) found higher BD in  
795 brown than white BGN flour. The BDs of BGN flours were comparable to the BD of soya bean  
796 and cowpea flours (Abiala *et al.*, 2022).

797 The swelling capacity (SC) of cream BGN flour was significantly higher ( $p < 0.05$ ) than  
798 that of red and brown BGN flours varying from 0.18 to 0.41 g/g. Higher SC of cream BGN flour  
799 might be caused by high starch content which is indicated by high carbohydrates (Table 8),  
800 particularly in starches that contain more branches of amylopectin (Iwe *et al.*, 2016). The  
801 dispensability of cream BGN flour was significantly higher ( $p < 0.05$ ) than red and brown BGN  
802 flours. The high dispensability of BGN flours might be due to their relatively low WAC (Table 8)

803 and higher gelling ability of flours (Nzuta *et al.*, 2022). The dispensability of BGN flour was less  
804 than that of the cowpea powder found by Jenfa *et al.* (2019).

805 The viscosity of brown and red BGN was significantly higher ( $p < 0.05$ ) than that of cream  
806 BGN flours ranging from 2666 to 3464.67 cP. The variation in viscosity may be caused by the  
807 intermolecular network's fragility which might have led to the disintegration of BGN flours granules  
808 when gelatinised in hot water, resulting in a paste with a relatively less viscosity (Nzuta *et al.*,  
809 2022). Similarly, Falade and Nwajei (2015) reported higher viscosity in the brown variety than in  
810 the cream variety.

811 The antioxidant properties of BGN flours are indicated in Table 8. The DPPH content of  
812 cream BGN flour was significantly ( $p < 0.05$ ) higher than brown and red BGN flours varying from  
813 87.19 to 95.15%. The high DPPH of cream BGN flour followed by the red BGN flour might be due  
814 to their relatively high flavonoid content than brown BGN flour (Table 8) (Ahmed *et al.*, 2015).  
815 Additionally, differences in BGN composition might have an impact on how hydrogen atoms were  
816 donated to DPPH, therefore affecting the DPPH content of BGN flour (Klompong and Benjakul,  
817 2015; Ramatsetse *et al.*, 2023). Oyeyinka *et al.* (2021) reported higher DPPH content in maroon  
818 BGN than in black and brown BGN flours with their respective high flavonoid content. Nyau *et al.*  
819 (2015) reported higher DPPH content in red BGN than in brown BGN flour.

820 The total phenolic content (TPC) of red and brown BGN flours was significantly higher ( $p$   
821  $< 0.05$ ) than that of cream BGN flour ranging from 0.79 to 0.98 mg GAE/g. The difference in BGN  
822 coat colours might account for the disparity in TPC (Oyeyinka *et al.*, 2021). Therefore, BGN grains  
823 that are darker in coat colours possess higher TPC than those with lighter coat colours (Parikh *et*  
824 *al.*, 2018; Adedayo *et al.*, 2021). Oyeyinka *et al.* (2017) found greater TPC in red BGN than in  
825 brown BGN flour. Tsamo *et al.* (2018) also reported higher TPC in red BGN than in brown and  
826 cream BGN flours, respectively. Therefore, consuming BGN grains may assist in fighting the  
827 negative effects caused by free radicals by functioning as free radical scavengers (Moyo *et al.*,  
828 2018; Adedayo *et al.*, 2021). The total flavonoids content of brown and cream BGN flour was  
829 significantly higher ( $p < 0.05$ ) than that of red BGN flour ranging from 0.05 to 0.07 mg/g CE. The  
830 differences in total flavonoids content of BGN flours might possibly be caused by differences in  
831 grain cultivars, assay procedure and conditions of extracting (Ramatsetse *et al.*, 2023).

#### 832 **4.2 Proximate composition of mutton patties**

833 The proximate composition of raw mutton patties is shown in Table 9. The increase in  
834 percentage of BGN flours decreased the moisture content of patties compared to the control  
835 sample ranging from 69.86 to 67.66%.

836

**Table 9.** Proximate composition of mutton patties incorporated with different levels of Bambara groundnut flour.

Samples	Moisture (%)	Ash (%)	Protein (%)	Fat (%)	Fiber (%)	CHO (%)	Energy (kcal/100g)
Control	69.86±0.10 <sup>iD</sup>	1.16±0.05 <sup>Aa</sup>	19.64±0.06 <sup>cC</sup>	7.20±0.02 <sup>eB</sup>	0±0.00 <sup>aA</sup>	2.14±0.11 <sup>aA</sup>	151.92±0.39 <sup>bcdefAB</sup>
Brown 2.5%	69.35±0.10 <sup>h</sup>	1.45±0.03 <sup>c</sup>	19.57±0.02 <sup>b</sup>	7.11±0.02 <sup>cd</sup>	0.15±0.01 <sup>bc</sup>	2.37±0.15 <sup>abc</sup>	151.74±0.42 <sup>bcde</sup>
Brown 5%	69.11±0.06 <sup>fg</sup>	1.78±0.00 <sup>de</sup>	19.53±0.01 <sup>ab</sup>	7.06±0.03 <sup>bc</sup>	0.31±0.01 <sup>def</sup>	2.21±0.08 <sup>ab</sup>	150.50±0.05 <sup>a</sup>
Brown 7.5%	68.72±0.23 <sup>d</sup>	1.85±0.01 <sup>f</sup>	19.53±0.01 <sup>ab</sup>	6.94±0.02 <sup>ab</sup>	0.42±0.01 <sup>fg</sup>	2.49±0.24 <sup>bcd</sup>	150.95±0.75 <sup>ab</sup>
Brown 10%	68.25±0.04 <sup>bc</sup>	1.99±0.00 <sup>g</sup>	19.51±0.02 <sup>a</sup>	6.89±0.12 <sup>a</sup>	0.52±0.01 <sup>gh</sup>	2.79±0.04 <sup>de</sup>	151.68±0.12 <sup>bcd</sup>
Mean brown	68.86±0.44 <sup>C</sup>	1.77±0.21 <sup>D</sup>	19.54±0.03 <sup>A</sup>	7.02±0.07 <sup>A</sup>	0.35±0.14 <sup>C</sup>	2.47±0.26 <sup>B</sup>	151.22±0.66 <sup>A</sup>
Red 2.5%	69.19±0.06 <sup>fgh</sup>	1.41±0.03 <sup>bc</sup>	19.59±0.02 <sup>bc</sup>	7.12±0.03 <sup>cd</sup>	0.19±0.00 <sup>bcd</sup>	2.50±0.08 <sup>bdc</sup>	152.41±0.42 <sup>cdefg</sup>
Red 5%	68.77±0.11 <sup>de</sup>	1.77±0.00 <sup>d</sup>	19.56±0.00 <sup>ab</sup>	7.08±0.04 <sup>cd</sup>	0.40±0.01 <sup>f</sup>	2.43±0.15 <sup>abc</sup>	151.65±0.33 <sup>bcd</sup>
Red 7.5%	68.14±0.03 <sup>b</sup>	1.81±0.01 <sup>ef</sup>	19.55±0.02 <sup>ab</sup>	6.99±0.03 <sup>ab</sup>	0.55±0.01 <sup>h</sup>	2.95±0.07 <sup>ef</sup>	152.91±0.12 <sup>fg</sup>
Red 10%	67.66±0.06 <sup>a</sup>	1.98±0.01 <sup>g</sup>	19.54±0.01 <sup>ab</sup>	6.94±0.02 <sup>a</sup>	0.79±0.18 <sup>i</sup>	3.10±0.18 <sup>f</sup>	153.03±0.80 <sup>g</sup>
Mean red	68.44±0.61 <sup>A</sup>	1.74±0.21 <sup>C</sup>	19.56±0.02 <sup>B</sup>	7.03±0.08 <sup>A</sup>	0.48±0.24 <sup>D</sup>	2.74±0.32 <sup>C</sup>	152.50±0.70 <sup>B</sup>
Cream 2.5%	69.21±0.10 <sup>gh</sup>	1.38±0.01 <sup>b</sup>	19.56±0.02 <sup>ab</sup>	7.14±0.03 <sup>de</sup>	0.12±0.00 <sup>b</sup>	2.59±0.06 <sup>cd</sup>	152.84±0.53 <sup>fg</sup>
Cream 5%	68.98±0.02 <sup>gh</sup>	1.75±0.00 <sup>d</sup>	19.55±0.02 <sup>ab</sup>	7.08±0.04 <sup>cd</sup>	0.24±0.01 <sup>cde</sup>	2.39±0.04 <sup>abc</sup>	151.43±0.20 <sup>abc</sup>
Cream 7.5%	68.40±0.02 <sup>c</sup>	1.78±0.01 <sup>de</sup>	19.54±0.01 <sup>ab</sup>	6.98±0.03 <sup>ab</sup>	0.35±0.01 <sup>ef</sup>	2.95±0.06 <sup>ef</sup>	152.77±0.09 <sup>efg</sup>
Cream 10%	68.12±0.02 <sup>b</sup>	1.95±0.01 <sup>g</sup>	19.53±0.04 <sup>ab</sup>	6.92±0.03 <sup>a</sup>	0.42±0.01 <sup>ef</sup>	3.06±0.03 <sup>ef</sup>	152.68±0.25 <sup>defg</sup>
Mean cream	68.67±0.46 <sup>B</sup>	1.72±0.21 <sup>B</sup>	19.55±0.02 <sup>AB</sup>	7.03±0.09 <sup>A</sup>	0.28±0.12 <sup>B</sup>	2.75±0.28 <sup>C</sup>	152.45±0.62 <sup>B</sup>

Values expressed as mean ± standard deviation. Means in the same column with distinct lowercases (superscripts) indicate a significant difference ( $p < 0.05$ ). Brown, red, and cream (2.5%, 5%, 7.5%, and 10%) indicate different concentrations of different cultivars of Bambara groundnut flours. Whereas means in the same column with distinct uppercases (superscripts) indicate a significant difference ( $p < 0.05$ ) of the type of varieties. CHO = carbohydrate

The brown BGN variety significantly decreased ( $p < 0.05$ ) the moisture content of patties than the cream and red BGN varieties, respectively. The reduction in the amount of moisture of formulated patties may be because of the low moisture content of BGN flours as shown in Table 8. Correspondingly, Kasaiyan *et al.* (2023) observed the reduction of moisture content in lamb sausage due to the inclusion of chickpea powders.

The ash content of formulated patties significantly increased ( $p < 0.05$ ) with the increase in percentage of BGN flours varying from 1.16 to 1.99%. The brown BGN flour significantly increased the ash content than the red and cream BGN flours, respectively. The increased in ash content of patties with BGN substitution might be due to the high amount of ash in BGN flours as shown in Table 8. Similarly, Novello *et al.* (2019) reported an increase in the ash content of beef patties due to the inclusion of golden flaxseed powder. Furthermore, Alakali *et al.* (2010) found comparable data in beef patties added with BGN flour.

The protein content of formulated patties significantly decreased ( $p < 0.05$ ) with the increase in the percentage of BGN flours ranging from 19.64 to 19.51%. The brown BGN flour decreased the protein content of the mutton patties than the red BGN variety, respectively. The decreased in protein content of formulated patties might be due to substitution of meat fat by BGN flours since meat fat holds a substantial amount of protein (Vargas-Ramella *et al.*, 2020). However, the low protein was still around the expected protein content (19%) of meat (Rama *et al.*, 2019). Saikia *et al.* (2019) found a decrease in the protein content of duck meat patties as the amount of black gram powder increased. Jamaly *et al.* (2017) observed low protein content of beef meatballs due to the addition of wheat powder.

The fat content of formulated patties significantly decreased ( $p < 0.05$ ) with an increase in the percentage of BGN flours with values ranging from 7.20 to 6.89%. Overall, all BGN varieties decreased the fat content of mutton patties equally. The control sample had a higher fat content of 7.20%. Gao *et al.* (2020) stated that the low-fat content of meat patties might be due to the low-fat of legumes incorporated, as supported by the low-fat content of BGN flours in Table 8. Morbos *et al.* (2019) found a decrease in the amount of fat of pork patties with an increase in the inclusion of Mung bean powder.

The fibre content of formulated patties significantly increased ( $p < 0.05$ ) with an increase in the percentage of BGN flours compared to the control samples varying from 0.00 to 3.10%. The red BGN variety increased the fibre content of the mutton patties than the brown and cream BGN varieties, respectively. The improvement in the amount of fibre of formulated patties might be due to the amount of fibre content in BGN flours as shown in Table 8. Similarly, Kahraman *et al.* (2023) found an increase in the amount of fibre of kirkclareli meatballs incorporated with cowpea powder.

The carbohydrate content (CHO) of control sample (2.14%) was significantly less ( $p < 0.05$ ) than that of formulated samples, except for the sample added with 5% brown BGN flour. The red and cream BGN varieties increased the CHO content of the mutton patties than the brown BGN variety. This was due to high CHO content in BGN flours as shown in Table 8. However, there was inconsistency trend of CHO in patties with the inclusion of BGN flours because CHO content is calculated by the difference (Sousa *et al.*, 2023). Therefore, samples with higher sums of proximate composition (protein, ash, moisture, and fibre) had lower CHO content, and vice versa. Embaby *et al.* (2016) found a similar trend in beef burgers added with lentil coat powder.

There was no consistent trend in energy content of patties with the inclusion of BGN flours with values ranging from 150.50 to 153.03 kcal/100g. This is because the energy relies mostly on fat (9 kcal/100g), carbohydrate (4kcal/100g), and protein (4kcal/100g) (Kwon *et al.*, 2020) and there was no effective difference in fat content, therefore the sample with higher sum of multiplication factor yielded higher energy content. The red and cream BGN varieties improved the energy content of the patties than the brown BGN variety. However, the control sample was not significantly ( $p < 0.05$ ) different from samples formulated with brown 2.5% and 10%, and red 5% BGN flours. Öztürk-Kerimoğlu *et al.* (2020) reported similar results in low-fat sausages incorporated with quinoa flour.

#### **4.3 Cooking yield and diameter reduction of mutton patties**

The cooking yield and diameter reduction of mutton patties are indicated in Table 10. The cooking yield of mutton patties increased ( $p < 0.05$ ) with an increase in the percentage of BGN flours with values ranging from 76.39% for control and 86.78% for patties added with 10% cream flour variety. Overall, all BGN varieties increased the cooking yield of the mutton patties equally. The improvement of cooking yield in formulated patties might be due to the availability of dietary fibre, WAC, and OAC in BGN flours that absorbed or retained the fat and water throughout cooking, thereby forming a firmer component of the mutton patties matrix (Xu, 2017., Essa and Elsebaie, 2018; Mashau *et al.*, 2021b). Similarly, Chandler and McSweeney (2022) reported an increase in cooking yield of chicken patties incorporated with yellow pea powder.

The increase in the percentage of BGN flours limited the diameter reduction of mutton patties with values ranging from 17.73 to 13.89%. All BGN varieties significantly decreased ( $p < 0.05$ ) the diameter reduction of the mutton patties equally. The decrease in diameter reduction of formulated patties might be because of greater water-holding and swelling ability of BGN flours as shown in Table 8 (Younis and Ahmad, 2018). Additionally, the decrease in diameter reduction

might be attributed to the BGN flour's ability to bind thereby keeping the meat components intact and preventing the patties dimensions from changing (Bunmee *et al.*, 2022).

**Table 10.** The cooking yield and diameter reduction of mutton patties incorporated with different levels of Bambara groundnut flours and their correlation.

Samples	Cooking yield (%)	Diameter reduction (%)	R
Control	76.39 ± 0.03 <sup>Aa</sup>	17.73 ± 0.03 <sup>eB</sup>	-0.985
Brown 2.5%	81.28 ± 0.06 <sup>b</sup>	16.49 ± 0.03 <sup>d</sup>	
Brown 5%	83.21 ± 0.03 <sup>d</sup>	15.54 ± 0.02 <sup>c</sup>	
Brown 7.5%	84.03 ± 0.04 <sup>e</sup>	15.04 ± 0.03 <sup>b</sup>	
Brown 10%	86.75 ± 0.03 <sup>g</sup>	13.99 ± 0.58 <sup>a</sup>	
Mean brown	83.82 ± 2.27 <sup>B</sup>	15.28 ± 0.94 <sup>A</sup>	
Red 2.5%	81.39 ± 0.03 <sup>c</sup>	16.25 ± 0.04 <sup>d</sup>	
Red 5%	83.23 ± 0.02 <sup>d</sup>	15.56 ± 0.04 <sup>c</sup>	
Red 7.5%	84.12 ± 0.04 <sup>ef</sup>	15.06 ± 0.02 <sup>b</sup>	
Red 10%	86.80 ± 0.03 <sup>g</sup>	13.94 ± 0.04 <sup>a</sup>	
Mean red	83.89 ± 2.25 <sup>B</sup>	15.20 ± 0.91 <sup>A</sup>	
Cream 2.5%	81.31 ± 0.02 <sup>bc</sup>	16.48 ± 0.02 <sup>d</sup>	
Cream 5%	83.31 ± 0.03 <sup>d</sup>	15.42 ± 0.03 <sup>bc</sup>	
Cream 7.5%	84.12 ± 0.03 <sup>f</sup>	15.10 ± 0.02 <sup>b</sup>	
Cream 10%	86.78 ± 0.16 <sup>g</sup>	13.89 ± 0.03 <sup>a</sup>	
Mean cream	83.88 ± 2.27 <sup>B</sup>	15.22 ± 0.97 <sup>A</sup>	

Values expressed at mean ± standard deviation. Mean in the same column with distinct lowercases (superscripts) indicates a significant difference ( $p < 0.05$ ). Brown, red, and cream (2.5%, 5%, 7.5%, and 10%) indicate different concentrations of different cultivars of Bambara groundnut flours. Whereas means in the same column with distinct uppercases (superscripts) indicate a significant difference ( $p < 0.05$ ) of the type of varieties. R, is correlation coefficient of cooking yield and diameter reduction.

High diameter reduction value of the control sample was probably due to the point that during cooking, the protein denatured, water evaporated, and fat drips leading to weight loss and shrinkage of the patties (Chandler and McSweeney, 2022). Park *et al.* (2017) reported the decrease in diameter reduction in pork patties formulated with black rice powder. Furthermore, there was negative correlation ( $r = -0.985$ ) between the diameter reduction and cooking yield of the patties. This was anticipated since BGN flours has the ability to absorb water thereby improving cooking yield and limiting the diameter reduction of mutton patties during cooking.

#### 4.4 Colour properties of mutton patties

The colour properties of raw mutton patties are depicted in Table 11 while Figure 5 shows images of mutton patties incorporated with different BGN flours varieties. The L\* values of mutton patties significantly increased ( $p < 0.05$ ) with the addition of BGN flours ranging from 54.04 to

58.59. The cream and brown BGN varieties significantly increased ( $p < 0.05$ ) the  $L^*$  value of the patties than red BGN variety. However, patties added with 2.5% brown and red BGN flours did not differ with the control sample. The patties sample with the most increased  $L^*$  value was the one added with 10% cream BGN flour. The  $a^*$  values of formulated patties significantly decreased ( $p < 0.05$ ) with an addition of BGN flours compared to control. The cream BGN variety decreased the  $a^*$  values of the patties than the red and brown BGN varieties, respectively. Therefore, the increase in  $L^*$  values and reduction in  $a^*$  values might be attributed to the dominant light colour of BGN flours that diluted the red colour of mutton patties causing reduction in meat myoglobin (Saikia *et al.*, 2019). Da Silva Felix *et al.* (2021) found an improvement in lightness and a reduction in redness of lamb patties with the inclusion of cassava flour. The  $b^*$  values of mutton patties decreased with the inclusion of BGN flours ranging from 18.38 to 15.49. All BGN varieties decreased the  $b^*$  values of patties equally. However, patties added with 2.5% BGN flours did not significantly differ with the control sample. The decrease in  $b^*$  values might be due to the low-fat content of formulated patties as shown in Table 9. Similarly, Salarkarimi *et al.* (2019) found a decrease in  $b^*$  values of beef burgers incorporated with chestnut powder.

Chroma of mutton patties significantly decreased ( $p < 0.05$ ) with an increase in the percentage of BGN flours with values ranging from 23.66 to 17.82. All BGN varieties decreased the chroma values of patties equally. The reduction in chroma values might be attributed to the reduction in  $a^*$  values of formulated mutton patties (Mashau *et al.*, 2021b). Thus, the inclusion of BGN flours did not boost the colour intensity of patties since chroma decreased. However, the values obtained are tolerable, since the acceptable chroma value by consumers is 3 and above (King *et al.*, 2023). Ball *et al.* (2021) found the reduction in chroma values of ground beef with an inclusion of pea protein. The hue angle of the control sample was lower than that of the formulated samples ranging from 50.98 to 60.39°. The brown, and cream BGN varieties increased the hue angle of patties than the red BGN variety. The increase in hue angle was due to decrease in  $a^*$  values of formulated patties, as higher hue angle represents lower redness of meat products (Ball *et al.*, 2021). Furthermore, lower  $a^*$  and chroma values with high hue angle denote the browning of meat because of their strong correlation with metmyoglobin concentration (Mashau *et al.*, 2021a; Khomola *et al.*, 2021).

The colour difference of formulated patties significantly increased ( $p < 0.05$ ) with the addition of BGN flours varying from 0.00 to 8.21. The cream BGN variety increased the colour difference of patties than the brown and red varieties. The noticed disparities in the colour of formulated mutton patties might be due to the presence of colour pigments of BGN flours (Adewumi *et al.*, 2022). Since the colour differences of formulated patties are above 2, it

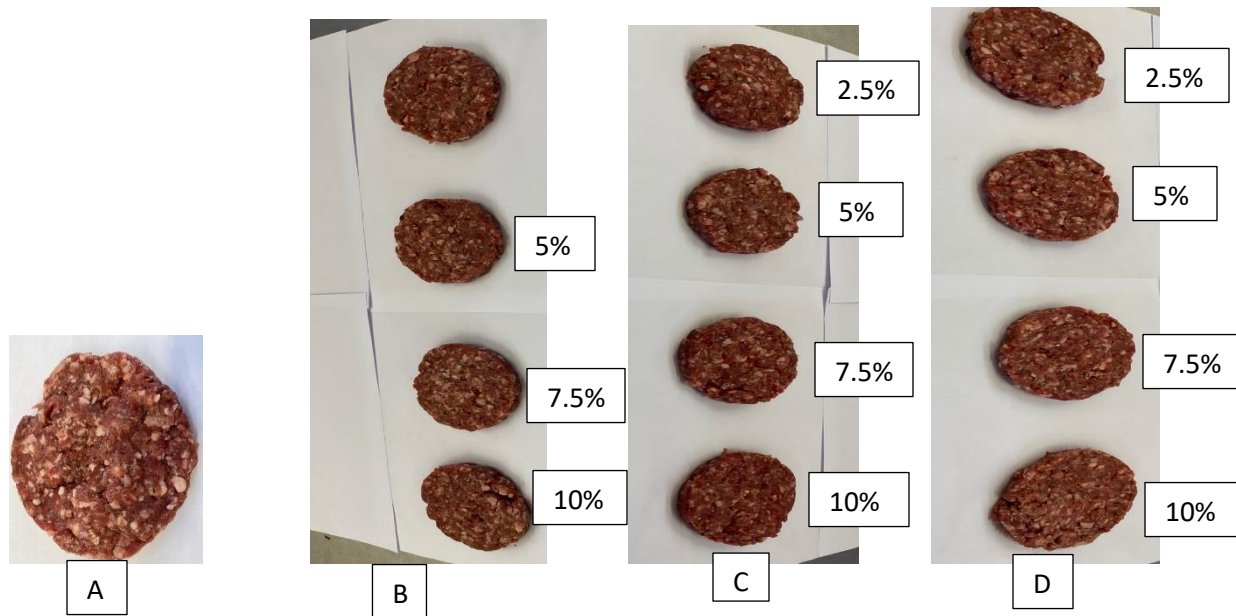
indicates that they are noticeable (Moarefian *et al.*, 2013; Mashau *et al.*, 2021a). Similarly, Argel *et al.* (2020) found an increase in colour difference of low-fat pork burgers with an increase in lentil flour concentration.

**Table 11.** Colour properties of mutton patties incorporated with different levels of Bambara groundnut flour.

Samples	L*	a*	b*	Chroma	Hue angle	$\Delta E$	WI	YI
Control	54.04±0.96 <sup>Aa</sup>	14.90±0.46 <sup>dC</sup>	18.38±0.43 <sup>dB</sup>	23.66±0.63 <sup>eB</sup>	50.98±0.24 <sup>aA</sup>	0.00±0.00 <sup>aA</sup>	48.31±1.14 <sup>aA</sup>	48.61±2.03 <sup>eB</sup>
Brown 2.5%	55.47±1.11 <sup>abc</sup>	11.17±1.35 <sup>bc</sup>	17.68±0.15 <sup>cd</sup>	20.93±0.86 <sup>cd</sup>	57.79±2.87 <sup>bc</sup>	4.26±1.51 <sup>bc</sup>	50.79±0.73 <sup>bc</sup>	45.54±0.59 <sup>de</sup>
Brown 5%	55.93±1.67 <sup>abcd</sup>	11.18±1.43 <sup>bc</sup>	16.72±0.89 <sup>abc</sup>	20.12±1.53 <sup>bcd</sup>	56.33±1.98 <sup>b</sup>	4.85±1.42 <sup>bc</sup>	51.52±0.95 <sup>bcd</sup>	42.70±1.10 <sup>bcd</sup>
Brown 7.5%	56.99±0.10 <sup>bcde</sup>	10.70±0.45 <sup>bc</sup>	16.30±0.99 <sup>abc</sup>	19.51±0.63 <sup>abcd</sup>	56.67±2.60 <sup>b</sup>	5.65±1.10 <sup>bcd</sup>	52.77±0.32 <sup>de</sup>	40.85±2.53 <sup>abc</sup>
Brown 10%	57.72±0.37 <sup>cde</sup>	9.97±0.42 <sup>ab</sup>	16.13±0.68 <sup>abc</sup>	18.96±0.79 <sup>ab</sup>	58.26±0.29 <sup>bc</sup>	6.57±1.34 <sup>cd</sup>	53.66±0.02 <sup>ef</sup>	39.91±1.44 <sup>abc</sup>
Brown mean	56.53±1.27 <sup>B</sup>	10.76±1.02 <sup>B</sup>	16.71±0.90 <sup>A</sup>	19.88±1.15 <sup>A</sup>	57.27±2.03 <sup>C</sup>	5.33±1.46 <sup>BC</sup>	52.18±1.27 <sup>C</sup>	42.25±2.62 <sup>A</sup>
Red 2.5%	54.96±0.71 <sup>ab</sup>	12.05±0.39 <sup>c</sup>	17.54±0.45 <sup>cd</sup>	21.29±0.31 <sup>d</sup>	55.50±1.36 <sup>b</sup>	3.17±0.90 <sup>b</sup>	50.38±0.48 <sup>b</sup>	45.61±1.54 <sup>de</sup>
Red 5%	55.07±0.67 <sup>ab</sup>	11.25±0.64 <sup>bc</sup>	16.42±0.21 <sup>abc</sup>	19.91±0.54 <sup>bcd</sup>	55.62±1.14 <sup>b</sup>	4.06±1.12 <sup>bc</sup>	51.22±0.58 <sup>bcd</sup>	42.61±0.05 <sup>bcd</sup>
Red 7.5%	55.41±0.41 <sup>abc</sup>	10.82±0.33 <sup>bc</sup>	15.72±0.10 <sup>ab</sup>	19.08±0.18 <sup>abc</sup>	55.46±0.90 <sup>b</sup>	5.10±0.90 <sup>bc</sup>	51.34±0.48 <sup>bcd</sup>	40.52±0.43 <sup>abc</sup>
Red 10%	56.21±0.66 <sup>abcde</sup>	10.22±0.29 <sup>abc</sup>	15.26±0.32 <sup>a</sup>	18.37±0.37 <sup>ab</sup>	56.17±0.28 <sup>b</sup>	6.03±0.37 <sup>cd</sup>	52.16±0.62 <sup>bcde</sup>	38.78±1.21 <sup>ab</sup>
Red mean	55.21±0.74 <sup>A</sup>	11.09±0.78 <sup>B</sup>	16.24±0.94 <sup>A</sup>	19.66±1.18 <sup>A</sup>	55.69±0.91 <sup>B</sup>	4.59±1.34 <sup>B</sup>	51.27±0.80 <sup>B</sup>	41.88±2.79 <sup>A</sup>
Cream 2.5%	56.11±0.42 <sup>abcd</sup>	10.79±0.77 <sup>bc</sup>	17.03±0.42 <sup>bcd</sup>	20.17±0.45 <sup>bcd</sup>	57.67±2.14 <sup>bc</sup>	4.89±0.30 <sup>bc</sup>	51.74±0.34 <sup>bcd</sup>	43.36±0.73 <sup>cd</sup>
Cream 5%	56.65±0.05 <sup>bcde</sup>	10.24±0.11 <sup>abc</sup>	16.66±0.35 <sup>abc</sup>	19.55±0.35 <sup>abcd</sup>	58.41±0.38 <sup>bc</sup>	5.64±0.85 <sup>bcd</sup>	52.32±0.21 <sup>cde</sup>	42.01±0.86 <sup>bcd</sup>
Cream 7.5%	57.97±1.03 <sup>de</sup>	10.13±0.17 <sup>ab</sup>	16.37±0.36 <sup>abc</sup>	19.25±0.34 <sup>abcd</sup>	58.25±0.64 <sup>bc</sup>	6.57±1.57 <sup>cd</sup>	53.79±1.00 <sup>ef</sup>	40.34±0.33 <sup>abc</sup>
Cream 10%	58.59±1.35 <sup>e</sup>	8.79±0.34 <sup>a</sup>	15.49±0.80 <sup>ab</sup>	17.82±0.71 <sup>a</sup>	60.39±1.63 <sup>c</sup>	8.21±1.76 <sup>d</sup>	54.84±1.83 <sup>f</sup>	37.82±2.79 <sup>a</sup>
Cream mean	57.33±1.28 <sup>B</sup>	9.99±0.85 <sup>A</sup>	16.39±0.74 <sup>A</sup>	19.20±0.99 <sup>A</sup>	58.68±1.60 <sup>C</sup>	6.33±1.69 <sup>C</sup>	51.27±0.80 <sup>D</sup>	40.88±1.21 <sup>A</sup>

Values expressed as the mean± standard deviation. Means in the same column with distinct lowercases (superscripts) indicate a significant difference ( $p < 0.05$ ). L\*= lightness; a\*= redness; b\*= yellowness;  $\Delta E$ = colour difference; WI= whiteness index; YI= yellowness index. Brown, red, and cream (2.5%, 5%, 7.5%, and 10%) indicate different concentrations of different cultivars of Bambara groundnut flours. Whereas means in the same column with distinct uppercases (superscripts) indicate a significant difference ( $p < 0.05$ ) of the type of varieties

The WI of formulated patties significantly increased ( $p < 0.05$ ) with the inclusion of BGN flours with values ranging from 48.31 to 54.84. Overall, the cream BGN variety decreased the WI of the patties than the brown, red BGN variety, and control, respectively. The increase in WI might be due to an increase in  $L^*$  values caused by the inclusion of BGN flours which diluted the myoglobin of meat thereby increasing WI (Saikia *et al.*, 2019). Saricoban and Yilmaz (2010) found an improvement in WI of beef meatballs with an increase in lightness. However, the YI of formulated patties significantly decreased ( $p < 0.05$ ) with the inclusion of BGN flours ranging from 48.61 to 37.82. All BGN varieties decreased YI of patties equally. The decrease in YI might be because of the reduction in  $b^*$  values and fat content of the patties, as well as dilution of myoglobin content (Kumar and Kumar., 2020). A similar trend was reported by Saricoban and Yilmaz (2010) in beef meatballs.



**Figure 5.** Mutton patties formulated with BGN flours (2.5%, 5%, 7.5%, and 10%). A = 100% mutton patties, B = mutton patties added with brown BGN flour, C = mutton patties added with cream BGN flour and D = mutton patties added with red BGN flour, BGN = Bambara groundnut

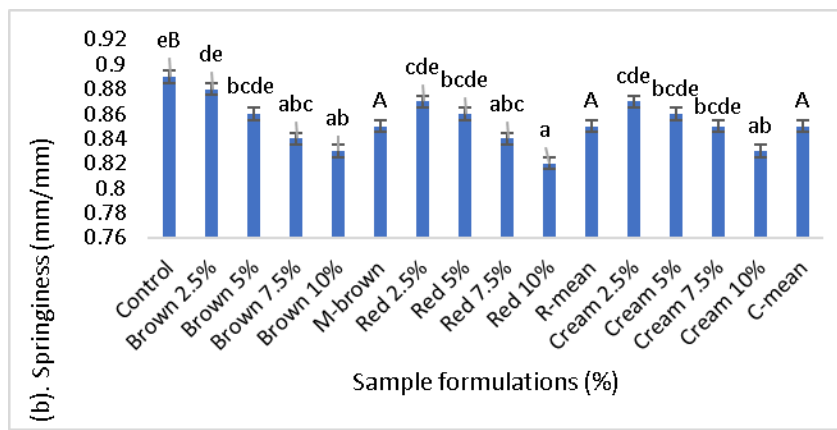
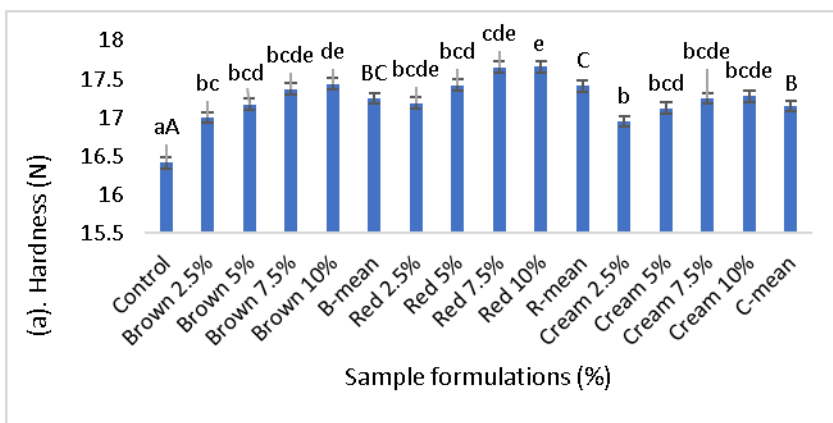
#### 4.5 Textural properties of mutton patties

The textural properties of raw mutton patties are indicated in Figure 6. The increase in the percentage of all BGN flours significantly increased ( $p < 0.05$ ) the hardness (a) and resilience (e) of patties compared to the control sample ranging from 16.41 to 17.66 N, and from 0.35 to 0.48 J/J, respectively. However, the resilience of the control was not different from sample added with 2.5% of brown and cream BGN flours. The red and brown BGN varieties increased the hardness of the patties than the cream BGN variety, respectively. On the other hand, the red and cream BGN varieties increased the resilience of the patties than brown BGN variety. The significant increase ( $p < 0.05$ ) in hardness and resilience might be due to high WAC and dry matter of BGN flours (Table 8) that absorbed water in meat patties, thereby decreasing the moisture content of patties (Table 9) making them harder (Siddiq *et al.*, 2009; Nuhriawangsa *et al.*, 2023).

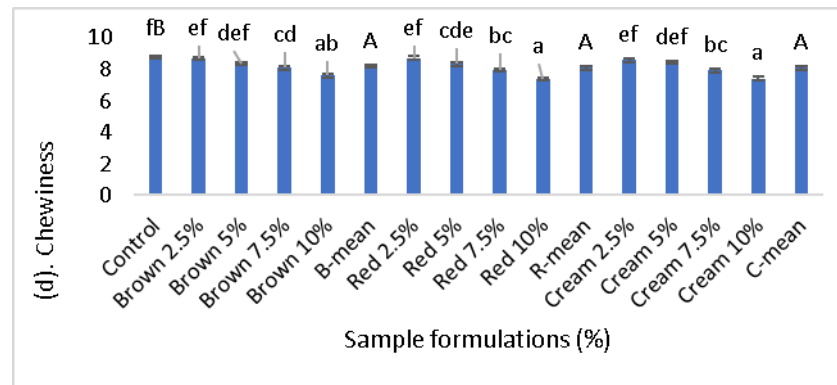
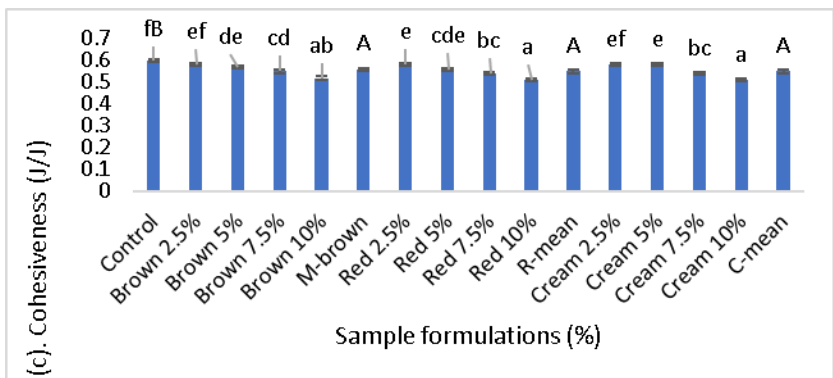
Moreover, the improvement of dietary fibre due to the addition of BGN flours might have improved water holding and binding abilities of patties, thereby improving hardness and resilience (Choi *et al.*, 2010; Lopez-Vargas *et al.*, 2014; Younis and Ahmad., 2018). Tahmasebi *et al.* (2016) found an increase in the hardness of sausages because of the inclusion of pigeon pea powder. Younis and Ahmad (2018) reported an increase in hardness and resilience of buffalo patties with an increase of apple pomace flour. The addition of BGN flours significantly decreased ( $p < 0.05$ ) springiness (b) of mutton patties with values ranging from 0.89 to 0.82 mm/mm. All BGN varieties decreased the springiness of patties equally. The decrease in springiness might be due to the inclusion of BGN flours which increased the fibre content of patties, thereby reducing the speed of deformed patties to spring back after initial compression (Hautrive *et al.*, 2019).

The increase in the percentage BGN flours significantly decreased ( $p < 0.05$ ) the cohesiveness (c) of patties compared to control sample ranging from 0.60 to 0.51 J/J. Overall, all BGN varieties decreased the cohesiveness of patties equally. The decrease in cohesiveness (c) of patties might be due to the inclusion of BGN flours which led to weakening of inner bond structure of patties by reducing the stronger bonds formed between meat particles, resulting in less dense and homogeneous structure of meat (Chandler and McSweeney, 2022; Argel *et al.*, 2022). Similarly, Ghribi *et al.* (2018) reported a reduction in cohesiveness of sausages incorporated with chickpea protein isolates. Varma *et al.* (2019) reported a reduction in cohesiveness of goat nuggets due to incorporation of amaranth seed powder.

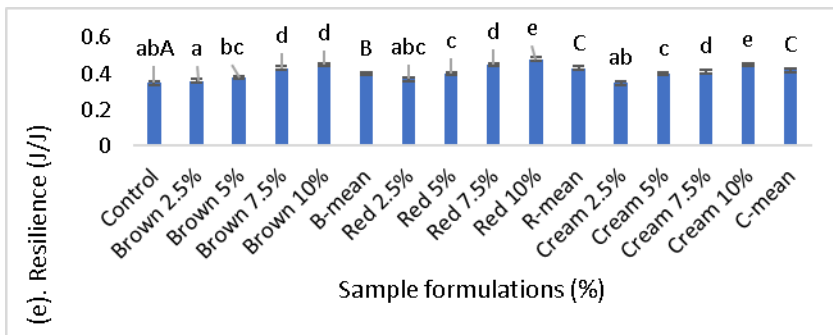
1



2



3



**Figure 6.** The texture properties of mutton patties incorporated with different levels of Bambara groundnut flour based on their colours. (a) Hardness, (b) Springiness, (c) cohesiveness, (d) Chewiness, and (e) Resilience, respectively. Means with distinct lowercases (Superscripts) indicates significant difference ( $p < 0.05$ ). Whereas means in the same column with distinct uppercases (superscripts) indicate a significant difference ( $p < 0.05$ ) of the type of the varieties

The increase in the addition of BGN flours led to a significantly decrease ( $p < 0.05$ ) in chewiness (d) of patties with values varying from 8.77 to 7.38. However, there was no significant variation among the control and patties added with 2.5% brown and red BGN flours. All BGN varieties decreased the chewiness of patties equally. The decrease in chewiness might be due to less difficult and more effective integration of BGN flours in meat matrix (Sayas-Barbera *et al.*, 2021). Correspondingly, Öztürk-Kerimoğlu *et al.* (2020) found a reduction in the chewiness of beef sausages with the incorporation of teff powder. Moreover, Sayas-Barbera *et al.* (2021) found a reduction in the chewiness of beef patties with the incorporation of black quinoa powder.

#### 4.6 Lipid oxidation of mutton patties

Thiobarbituric acid reactive substances (TBARS) values of raw mutton patties are indicated in Table 12. There was no significant difference ( $p < 0.05$ ) in all samples on day zero.

**Table 12.** Lipid oxidation of mutton patties incorporated with different levels of Bambara groundnut flour (mg MDA/kg).

Samples	Storage days			
	0	7	14	21
Control	0.06 ± 0.00 <sup>a</sup>	0.20 ± 0.00 <sup>g</sup>	0.63 ± 0.00 <sup>p</sup>	0.88 ± 0.00 <sup>w</sup>
Control mean				0.44 ± 0.34 <sup>C</sup>
Brown 2.5%	0.05 ± 0.00 <sup>a</sup>	0.14 ± 0.00 <sup>f</sup>	0.53 ± 0.01 <sup>mn</sup>	0.77 ± 0.01 <sup>tu</sup>
Brown 5%	0.06 ± 0.00 <sup>a</sup>	0.12 ± 0.00 <sup>cdef</sup>	0.49 ± 0.02 <sup>l</sup>	0.76 ± 0.01 <sup>t</sup>
Brown 7.5%	0.06 ± 0.00 <sup>a</sup>	0.12 ± 0.00 <sup>cdef</sup>	0.45 ± 0.02 <sup>j</sup>	0.70 ± 0.01 <sup>r</sup>
Brown 10%	0.05 ± 0.00 <sup>a</sup>	0.10 ± 0.00 <sup>abc</sup>	0.39 ± 0.02 <sup>i</sup>	0.60 ± 0.00 <sup>o</sup>
Brown mean				0.38 ± 0.27 <sup>B</sup>
Red 2.5%	0.05 ± 0.00 <sup>a</sup>	0.13 ± 0.00 <sup>ef</sup>	0.51 ± 0.01 <sup>lm</sup>	0.78 ± 0.02 <sup>u</sup>
Red 5%	0.05 ± 0.00 <sup>a</sup>	0.12 ± 0.00 <sup>cdef</sup>	0.50 ± 0.01 <sup>l</sup>	0.74 ± 0.01 <sup>s</sup>
Red 7.5%	0.05 ± 0.00 <sup>a</sup>	0.11 ± 0.00 <sup>cde</sup>	0.44 ± 0.02 <sup>j</sup>	0.65 ± 0.02 <sup>q</sup>
Red 10%	0.06 ± 0.00 <sup>a</sup>	0.08 ± 0.01 <sup>b</sup>	0.35 ± 0.02 <sup>h</sup>	0.54 ± 0.01 <sup>n</sup>
Red mean				0.36 ± 0.26 <sup>A</sup>
Cream 2.5%	0.05 ± 0.00 <sup>a</sup>	0.14 ± 0.00 <sup>f</sup>	0.54 ± 0.01 <sup>n</sup>	0.79 ± 0.01 <sup>v</sup>
Cream 5%	0.06 ± 0.00 <sup>a</sup>	0.13 ± 0.00 <sup>ef</sup>	0.50 ± 0.01 <sup>l</sup>	0.77 ± 0.00 <sup>tu</sup>
Cream 7.5%	0.05 ± 0.00 <sup>a</sup>	0.11 ± 0.00 <sup>cde</sup>	0.47 ± 0.01 <sup>k</sup>	0.70 ± 0.01 <sup>r</sup>
Cream 10%	0.06 ± 0.00 <sup>a</sup>	0.10 ± 0.00 <sup>abc</sup>	0.40 ± 0.02 <sup>i</sup>	0.63 ± 0.00 <sup>p</sup>
Cream mean				0.38 ± 0.27 <sup>B</sup>

Values expressed as mean ± standard deviation. Means in the same column with distinct lowercases (superscripts) indicate significant difference ( $p < 0.05$ ) within that day. Means in the same row with distinct lowercases (superscripts) indicate a significant difference ( $p < 0.05$ ). TBARS = Thiobarbituric acid reactive substances. Brown, red, and cream (2.5%, 5%, 7.5%, and 10%) indicate different

concentrations of different cultivars of Bambara groundnut flours. Whereas means in the same column with distinct uppercases (superscripts) indicate a significant difference ( $p < 0.05$ ) of the type of varieties

However, from day 7 to 21 the TBARS values of formulated mutton patties increased but was still less than that of the control sample within storage days which had values varying from 0.08 to 0.88 mgMDA/kg. The red BGN variety significantly ( $p < 0.05$ ) delayed the lipid oxidation of the patties effectively than the brown and cream BGN varieties. Mutton patties incorporated with 10% red BGN flour delayed lipid oxidation more effectively than the samples added with brown and cream BGN flours from day 7 to 21. The low TBARS values of formulated patties within storage days compared to control might be due to the decrease in the amount of fat in formulated patties samples as shown in Table 9 with the increase of BGN flours (Faid, 2019). Furthermore, low TBARS values might be because of the greater amount of polyphenolic compound in BGN flours (Table 8) which acted as free radical terminators and oxygen scavengers, thereby slowing down the process of lipid oxidation in mutton patties (Al-Juhaimi *et al.*, 2017; Bing *et al.*, 2022).

The significant increase ( $p < 0.05$ ) in TBARS values during the storage period might influence oxidation of double bonds of polyunsaturated fatty acid as well as the production of volatile compounds in meat (Dilnawaz *et al.*, 2017; Mashau *et al.*, 2021a). Although TBARS values increased in all samples during the storage days, the values were still within the permissible limits of less than 1 MDA/kg (Dilnawaz *et al.*, 2017; Cerón-Guevara *et al.*, 2020). Comparable data were found by Dilnawaz *et al.* (2017) in mutton blocks incorporated with green coffee bean powder. Furthermore, Alakali *et al.* (2010) found a delay in lipid oxidation of beef patties incorporated with BGN flour.

#### 4.7 Sensory properties of mutton patties

Sensory properties of mutton patties formulated with BGN flours are indicated in Table 13. There was no significant difference ( $p < 0.05$ ) in the appearance, aroma, texture, and overall acceptability scores of formulated patties compared to the control sample. Furthermore, there was no noticeable difference in the taste of formulated patties (except 10% brown and 7.5% cream BGN flours) compared to the control sample. The liked sensory attributes of formulated patties by panellists might be due to high OAC of BGN flours as shown in Table 8 which improves the sensation of chewing, the taste, and preserves the flavour in meat (Arise *et al.*, 2019; Adewumi *et al.*, 2022). The taste scores of mutton patties samples formulated with 10% brown BGN flour (6.45), and 7.5% cream BGN flour (6.27) were less than that of the control sample. The lower taste score might be due to the stronger flavour of BGNs which panellists disliked, this dominated the flavour of formulated mutton patties (Motamedi *et al.*, 2015; Novikasari *et al.*, 2022). Nevertheless, the sensory properties of all samples scored above the acceptable level of 5 in the 9-hedonic scale.

**Table 13.** Sensory properties of mutton patties incorporated with different levels of Bambara groundnut flour.

Samples	Appearance	Aroma	Taste	Texture	Overall acceptability
Control	7.05 ± 1.61 <sup>abc</sup>	6.66 ± 1.83 <sup>ab</sup>	7.53 ± 1.93 <sup>b</sup>	7.23 ± 1.31 <sup>a</sup>	7.31 ± 1.62 <sup>a</sup>
Brown 2.5%	7.60 ± 1.42 <sup>abc</sup>	7.02 ± 1.56 <sup>ab</sup>	7.55 ± 1.31 <sup>b</sup>	7.45 ± 1.48 <sup>a</sup>	7.47 ± 1.32 <sup>a</sup>
Brown 5%	7.06 ± 1.84 <sup>abc</sup>	6.85 ± 1.49 <sup>ab</sup>	6.87 ± 1.42 <sup>ab</sup>	6.98 ± 1.73 <sup>a</sup>	6.81 ± 1.77 <sup>a</sup>
Brown 7.5%	6.79 ± 1.99 <sup>ab</sup>	6.63 ± 1.67 <sup>ab</sup>	6.79 ± 1.85 <sup>ab</sup>	6.87 ± 1.65 <sup>a</sup>	6.74 ± 1.86 <sup>a</sup>
Brown 10%	6.92 ± 1.86 <sup>c</sup>	6.77 ± 1.82 <sup>ab</sup>	6.45 ± 2.23 <sup>a</sup>	6.81 ± 2.02 <sup>a</sup>	6.76 ± 1.84 <sup>a</sup>
Red 2.5%	6.87 ± 1.48 <sup>abc</sup>	6.76 ± 1.75 <sup>ab</sup>	6.79 ± 1.77 <sup>ab</sup>	6.97 ± 1.64 <sup>a</sup>	7.05 ± 1.47 <sup>a</sup>
Red 5%	7.46 ± 1.32 <sup>ab</sup>	7.23 ± 1.56 <sup>b</sup>	6.95 ± 1.83 <sup>ab</sup>	7.28 ± 1.40 <sup>a</sup>	7.25 ± 1.52 <sup>a</sup>
Red 7.5%	7.24 ± 1.74 <sup>abc</sup>	6.73 ± 1.71 <sup>ab</sup>	6.79 ± 2.11 <sup>ab</sup>	7.23 ± 1.48 <sup>a</sup>	7.00 ± 1.82 <sup>a</sup>
Red 10%	6.89 ± 1.96 <sup>abc</sup>	7.03 ± 1.85 <sup>ab</sup>	6.77 ± 2.20 <sup>ab</sup>	7.13 ± 1.52 <sup>a</sup>	6.76 ± 2.13 <sup>a</sup>
Cream 2.5%	7.18 ± 1.82 <sup>abc</sup>	6.94 ± 1.88 <sup>ab</sup>	7.03 ± 2.40 <sup>ab</sup>	7.19 ± 1.84 <sup>a</sup>	7.02 ± 1.92 <sup>a</sup>
Cream 5%	6.61 ± 2.26 <sup>a</sup>	6.81 ± 1.61 <sup>ab</sup>	6.77 ± 2.15 <sup>ab</sup>	6.95 ± 1.87 <sup>a</sup>	6.90 ± 1.91 <sup>a</sup>
Cream 7.5%	7.00 ± 1.70 <sup>abc</sup>	6.40 ± 2.04 <sup>a</sup>	6.27 ± 2.35 <sup>a</sup>	7.00 ± 1.98 <sup>a</sup>	6.73 ± 2.09 <sup>a</sup>
Cream 10%	7.29 ± 1.63 <sup>abc</sup>	6.90 ± 1.58 <sup>ab</sup>	6.79 ± 2.07 <sup>ab</sup>	7.18 ± 1.47 <sup>a</sup>	7.03 ± 1.75 <sup>a</sup>

Values expressed as mean ± standard deviation. Mean in the same column with distinct lowercases (superscripts) indicates a significant difference ( $p < 0.05$ ). Brown, red, and cream (2.5%, 5%, 7.5%, and 10%) indicate different concentrations of different cultivars of Bambara groundnut flours.

Therefore, the findings of the study imply that BGN flours may be utilised as a fat substitute in low-fat mutton patties without causing drastic changes in the sensory attributes of the meat. Similar results were reported by Argel *et al.* (2020) in low-fat pork patties incorporated with lentil powder. Furthermore, Alakali *et al.* (2010) found comparable results in beef patties incorporated with 5% BGN powder.

## CHAPTER 5. CONCLUSION AND RECOMMENDATIONS

The inclusion of BGN flour varieties in mutton patties can be utilised as natural preservative, fat substitute, ash, and fibre booster. The increase in percentages of BGN flours decreased the protein content of mutton patties, but it was still within expected protein content (19%) of meat. The cooking yield and diameter reduction of mutton patties were also improved due to the increase in percentage of BGN flours up to 10%. Furthermore, the inclusion of BGN flour from 2.5 to 10% was successful in delaying the lipid oxidation, indicating that it may be utilised to improve the shelf-life of mutton patties because of its antioxidant's properties.

The sensory properties (appearance, taste, texture, colour, and overall acceptability) of formulated patties were not significantly different from the control sample; however, they were above the acceptable score of 5. All BGN varieties had positive effects on the mutton patties, more especially red BGN variety followed by brown, and cream BGN varieties, respectively. Furthermore, the inclusion of 10% red BGN flour in mutton patties is highly recommended since it retains the colour, reduces the moisture content, delays lipid oxidation, increases the ash, fibre, carbohydrates, and energy contents, as well as hardness, and resilience than other BGN varieties. The results of the study show that Bambara groundnut can be used as an additive in mutton patties without detrimental effects on the quality parameters. However, difficult to cook and mill phenomena of BGN are major constraints for its utilization, therefore, it is recommended to utilise biological and physical procedures to minimize cooking time and advanced milling equipment should be implemented. Furthermore, extensive research on advanced processing technologies such as high-pressure processing should be explored.

Meat industries can utilise BGN flours as fat-substitute and natural preservative to meet the demands of consumers for healthy meat products. The current study will open new opportunities to use Bambara groundnut flours low-fat for processed meat in meat or food industry. Further research should be conducted to discover the effect of different BGN flour varieties on other meat products and their potential influence on pH, antioxidants, microbiological properties, vitamins, and minerals of such products. Modern technologies such three-dimensional (3D) food printing should also be used in the production of mutton patties incorporated with bambara flours. Since BGN is rich in nutritional composition and has high oil absorption capacity, further research is also warranted to manufacture plant-based meat analogue from BGN flours using 3D printing and analysed for nutritional composition, lipid oxidation, sensory attributes, and textural properties.

Gas chromatography-mass spectrometry (GC-MS) should also be used to extract the volatile organic compounds which contributes to the flavour and aroma such as aldehydes (2-Methylpropanal, Hexanal, and Nonanal), ketones (2,3-Butanedione and 2,3-pentanedione) that will also determine the quality and shelf-life of the mutton patties added with different



varieties of BGN flours. Nevertheless, future research is necessary to ascertain the influence of incorporating distinct types of legumes in low-fat meat products.

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## Appendix A

ETHICS APPROVAL CERTIFICATE

RESEARCH AND INNOVATION  
OFFICE OF THE DIRECTOR

NAME OF RESEARCHER/INVESTIGATOR:  
**Mr KE Ramatsetse**

STUDENT NO:  
**16014230**

PROJECT TITLE **Effect of partial mutton meat substitution by Bambara groundnut flour on physicochemical properties, lipid oxidation and sensory acceptability of low-fat patties.**

ETHICAL CLEARANCE NO: **FSEA/22/FST/07/1711**

SUPERVISORS/ CO-RESEARCHERS/ CO-INVESTIGATORS

NAME	INSTITUTION & DEPARTMENT	ROLE
Dr SE Ramashia	UNIVEN, Food Science and Technology	Supervisor
Mr M Mashau	UNIVEN, Food Science and Technology	Co - Supervisor
Mr. KE Ramatsetse	UNIVEN, Food Science and Technology	Investigator - Student

Type: **Masters Research**

Risk: **Minimal risk to humans, animals, or environment (Category 2)**

Approval Period: **November 2022 - November 2023**

The Human and Clinical Trials Research Ethics Committee (HCTREC) hereby approves your project as indicated above.

**General Conditions**

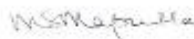
While this ethics approval is subject to all declarations, undertakings and agreements incorporated and signed in the application form, please note the following:

- The project leader (principal investigator) must report in the prescribed format to the REC:
  - Annually (or as otherwise requested) on the progress of the project, and upon completion of the project.
  - Within 48hrs in case of any adverse event (or any matter that violates sound ethical principles) during the course of the project.
  - Annually a number of projects may be randomly selected for an external audit.
- The approval applies strictly to the protocol as stipulated in the application form. Should any changes to the protocol be deemed necessary during the course of the project, the project leader must apply for approval of these changes at the REC. Would there be deviations from the project protocol without the necessary approval or such changes, the ethics approval is immediately and automatically terminated.
- The date of approval indicates the first date that the project may be started. Would the project have to continue after the expiry date, a new application must be made to the REC and new approval received before or on the expiry date.
- In the interest of ethical responsibility, the REC retains the right to:
  - Request access to any information or data at any time during the course or after completion of the project.
  - To ask further questions; Seek additional information; Require further modification or monitor the conduct of your research or the informed consent process.
  - withdraw or postpone approval if:
    - Any unethical principles or practices of the project are revealed or suspected.
    - It becomes apparent that any relevant information was withheld from the REC or that information has been false or misrepresented.
    - The required annual report and reporting of adverse events was not done timely and accurately.
  - New institutional rules, national legislation or international conventions deem it necessary.

ISSUED BY:  
UNIVERSITY OF VENDA, RESEARCH ETHICS COMMITTEE  
Date Considered: **November 2022**

Name of the HCTREC Chairperson of the Committee: **Prof MS Maputle**

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### A review on health benefits, antimicrobial and antioxidant properties of Bambara groundnut (*Vigna subterranean*)

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#### ABSTRACT

Bambara groundnut (*Vigna subterranean*) is an African legume that belongs to the family and subfamily of *Fabaceae* and *Faboidea*, respectively. It is becoming one of the most utilized legumes after being underutilized for many years in Africa. It is the third most significant legume after groundnut (*Arachis hypogaea*) and cowpea (*Vigna hypogaea*). Bambara groundnut (BGN) is a complete food because of its richness in nutritional properties such as crude fiber, iron, protein, carbohydrate, fat and minerals. Moreover, BGN possesses antioxidants properties as it contains phytochemicals, such as tannins, flavonoids and phytic acids which possess health benefits for humans such as preventing diabetes, stroke, atherosclerosis, heart disease, cancer, Alzheimer and cardiovascular diseases. In addition, these phytochemicals extend the shelf life of food products and have antimicrobial properties which can inhibit microbial growth. This paper highlights the current findings on BGN nutritional composition, health benefits, antimicrobial and antioxidant properties. Therefore, it is advisable for food industry to exploit BGN grain to its full potential to tackle challenges of food security and nutrition globally.

#### ARTICLE HISTORY

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#### KEYWORDS

Bambara groundnut;  
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#### Introduction

Bambara groundnut (*Vigna subterranean*) is a legume belonging to the family and subfamily of *Fabaceae* and *Faboidea*, respectively.<sup>[1]</sup> Bambara groundnut is a legume indigenous to Africa and it is believed that its origin is between west and central Africa.<sup>[2]</sup> It grows in the wild in northern Nigeria and eastwards to southern Sudan and is currently grown across tropical Africa and to a smaller range in tropical portions of America, Asia, and Australia.<sup>[3]</sup> Bambara groundnut (BGN) is the third most significant legume after groundnut (*Arachis hypogaea*) and cowpea (*Vigna hypogaea*).<sup>[4,5]</sup> It plays a crucial socio-economic impact in the semi-arid regions globally and contains sufficient protein. Therefore, if combined with some native protein sources, it could help improve the nutritional issues globally.<sup>[6,7]</sup>

Bambara groundnut grain is a proper healthy food due to its high iron (4.9–48 mg/100 g) and protein contents (18.0–24.0%) when compared to other legume products, with great amount of amino acids content, fiber (5.0–12%), fat (5.0–7.0%), carbohydrate (57.43–63.09%) calcium (95.8–99 mg/100 g), potassium (1144–1935 mg/100 g), and sodium (2.9–12.0 mg/100 g).<sup>[7,8]</sup> Bambara groundnuts diverse nutritional composition suggests that it can meet dietary needs of people globally.<sup>[9]</sup> Its high protein content maintains a greater focus on improving procedures for processing its grains and expanding food applications.<sup>[9]</sup> Bambara groundnut also contains several polyphenols such as flavonoids (medioresinol, catechin, catechin

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This article has been republished with minor changes. These changes do not impact the academic content of the article.

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## Appendix C

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Nutrition - Manuscript ID  
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ref: SE-8-a] Inbox



WOA Admin 2023/10/31

to mpho.mashau, me, s... 



31-Oct-2023

Dear Mr. Mashau:

Your revised manuscript entitled "Effect of partial mutton meat substitution with Bambara groundnut (*Vigna subterranea* (L.) Verdc.) flour on physicochemical properties, lipid oxidation and sensory acceptability of low-fat patties" by Ramatsetse, Kgaogelo; Ramashia, Shonisani; Mashau, Mpho, has been successfully submitted online and is presently being given full consideration for publication in Food Science & Nutrition.

Co-authors: Please contact the Editorial Office as soon as possible if you disagree with being listed as a co-author for this manuscript.

## Appendix D

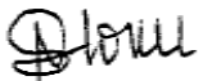
### Editing and Proofreading Report

09 July 2022

This letter serves to confirm that I, Dr I. Ndlovu of the Department of English, Media Studies and Linguistics at the University of Venda, have proofread and edited an article titled “Bambara groundnut (*Vigna subterranean*): Nutritional composition, health benefits, antioxidants, antimicrobial properties, and food applications” by Kgaogelo Edwin Ramatsetse, Eugenia Shonisani Ramashia and Mpho Edward Mashau.

I carefully read through the document, focusing on proofreading and editorial issues. The recommended suggestions are clearly highlighted and can either be accepted or rejected using the Microsoft Track Changes Function.

Yours Sincerely



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## Appendix E

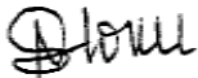
### Editing and Proofreading Report

14 August 2023

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