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THE IMPACT OF PROCESSING ON SOME NUTRITIONAL PROPERTIES OF READY-TO-EAT MEALS MADE FROM COMPOSITE FLOUR

MAYADA A. MUSTAFA[1], ALMUJTABA H. M. ABDALLH[1], SUHA O. AHMED[2], MOHAMED M. ELTYEB[1], ELFADIL E. BABIKER[1]*

[1]*Faculty of Agriculture, University of Khartoum, Department of Food Science and Technology, Khartoum North 13314, Shambat, Sudan*

[2]*Department of Food Chemistry, Food Research Center, Ministry of Science and Communication, Khartoum North, P.O. Box 213, Shambat, Sudan*

*Correspondent author: elfadilbabiker@yahoo.com (Elfadil E. Babiker)

Abstract: Four meals were produced with sorghum cultivar (WadAhmed), defatted pumpkin seed pulp (PSP), and wheat flour to test the effect of component ratios, fermentation, and/or cooking on some nutritional qualities. Chemical compositions, total calories, minerals, anti-nutrients, and *in vitro* protein digestibility (IVPD) were assessed for ingredients before formulation and meals before and after processing. The sorghum cultivar was greater in carbohydrate (75.71%), phosphorus (296.55 mg/100 g), and calories (368.82 kcal) than wheat and PSP, and had significantly more antinutrients (tannin and phytate). Wheat had a greater IVPD than other components, but PSP was rich in ash (8.67%), protein (63.15%), fiber (7.09%), calcium (48.08 mg/100 g), and iron (26.37 mg/100 g). The addition of wheat and PSP flour decreased carbohydrate and antinutrient levels while improving ash, protein, fiber, fat, total and extractable minerals, and IVPD. Fermentation significantly reduced antinutrient levels ($p \leq 0.05$) and created high-quality meals with excellent IVPD. Cooking the fermented dough revealed even more considerable improvement. Cooking had a significant ($p \leq 0.05$) effect on chemical composition, resulting in a decrease in tannin and phytate and an increase in IVPD and mineral extractability. Organoleptically, the produced meals were highly accepted than the control meal ($p \leq 0.05$). The formulation with 10% wheat and 20% PSP flour (M3) was shown to be the most nutrient-dense and accessible meal.

Keywords: Sorghum; wheat; pumpkin; minerals; anti-nutrients

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1. Introduction

A common issue in developing nations, malnutrition affects a greater number of young people, expectant mothers, and the elderly who belong to the weaker segments of society. Consumer interest in healthful eating and food with valuable qualities has grown. As the most significant source of protein and energy, cereals and pulses comprise 60–70% of a person's entire diet (Baranwa, 2017). Along with milk, fruit, and vegetables, cereals are sometimes referred to as a "dietary staple" due to their substantial contributions to dietary energy and nutritional requirements (Thielecke *et al*., 2021). Malnutrition in poor nations should be addressed by increasing the bioavailability of nutrients in foods through correct processing and handling procedures. It can also be mitigated through enrichment with high-quality supplements. Because of its drought endurance, sorghum seeds or flour are the main supply of energy and protein for the vast majority of the population in the semi-arid tropics of Africa and Asia. They are also an important feed resource for poultry and cattle (de Morais Cardoso *et al*., 2017).

Sorghum is a major staple food for the impoverished in many nations. However, despite its promise as a dietary cereal, sorghum has lower nutritional value and organoleptic qualities than other popular cereals like maize and rice (Mugalavai *et al*., 2020). Sorghum's deficiency stems in part from its primary storage protein, kafirin, which is deficient in the important amino acid lysine and has low digestion in cooked diets (Taylor & Taylor, 2018). As a result, food-to-food fortification of sorghum flours has been implemented to enhance protein quality and micronutrient accessibility (Mugalavai *et al*., 2020). In comparison, wheat (*Triticum aestivum* L.) is the world's most significant and commonly grown crop, accounting for 20% of total calorie consumption. It is also a significant source of protein and minerals, with low levels of antinutrients (Hossain *et al*., 2021). Pumpkin pulp is rich in protein, carbohydrates, and lipids, with minimal antinutrients, and it contains a lot of minerals that are good for both people and animals (Shajan *et al*., 2024). On this basis, it has been suggested that pumpkin seeds can be used as a food ingredient to maximize the plant's potential (Shajan *et al*., 2024).

Antinutrients in plant-based diets have been shown to impair the uptake of nutrients (Nath *et al*., 2022). Food processing has several benefits, such as extending the shelf life of food, eliminating harmful bacteria and toxins from food, increasing the availability of nutrients, improving the quality of food by increasing its functional and sensory qualities, increasing convenience, and minimizing losses and waste (Augustin *et al*., 2016). Home cooking methods, including roasting, germination, fermentation, and malting, are excellent alternatives to increase the nutritional value of grains (Yousaf *et al*., 2021). In societies where cereals and legumes comprise a significant portion of the diet, many traditional food preparation techniques, like cooking and malting, improve the nutritive quality of whole grains by lowering certain antinutrients like tannin, phytic acid, and oxalic acid (Ertop and Bektaş, 2018).

There have been varied effects of fermentation on cereals and food items based on legumes, and these effects are not reliant on the form of these foods. Food fermentation has been demonstrated to significantly improve the nutritional content of meals while lowering the concentrations of harmful and antinutritional ingredients. It may be a preferable option for reducing the negative impacts of these substances in diets (Adebiyi *et al*., 2019). While Nivetha *et al*. (2018) showed a reduction in the cyanogenic glycoside concentration (66%) of a linseed fermented beverage using *Lactobacillus acidophilus*, Ojha *et al*. (2018) also observed a reduction in hydrogen cyanide (52.3%) due to the fermentation of sorghum flour. The decrease in cyanogenic glycosides resulted from the hydrolysis and degradation of antinutritional components into smaller units by the activity of enzymes activated during the fermentation stage (Nivetha *et al*., 2018).

Sorghum's natural phytase activity destroyed phytates during fermentation, while microbial activity and phytate acyl hydrolases contributed to the decrease in tannin concentration (Ojha *et al*., 2018). Similarly, the amount of tannin in fermented sorghum decreased significantly, which was attributed to tannin structural rearrangement and depolymerization (Adebo *et al*., 2018). During fermentation, low-molecular-weight organic acids such as citric, malic, and lactic were found to have the potential to improve iron and zinc absorption by forming soluble ligands while also lowering the pH, which increases the activity of naturally occurring phytase in cereal and legume flours (Gan *et al*., 2023). Cooking, on the other hand, causes the crude protein to disintegrate into amino acids, and thus heat treatment causes modifications to the structure of the proteins, which can inactivate the antinutrients, increasing the digestibility and biological value of the bean protein (Marie *et al*., 2021). To take advantage of wheat's baking quality, pumpkin seed pulp's nutritional value, and fermentation process, the present study investigated the effect of wheat and pumpkin seed pulp formulation with sorghum in varied ratios on the nutritional quality of ready-to-eat meals.

2. Materials and Methods

Materials

Sorghum cultivar (WadAhmed) seeds were received from the Agricultural Research Corporation in Wad-Madani, Sudan. Wheat (Debaria) seeds were obtained from Seen Flour Mills in Khartoum, Sudan. Pumpkin seeds were bought from the Omdurman local market in Sudan. Extraneous components were removed from sorghum, wheat, and pumpkin seeds by hand. Pumpkin seeds were decorticated; pulps were defatted, and then ground in an electric miller (Kenwood Manufacturing Co. Ltd., UK) to pass through a sieve (0.4 mm) before being oven-dried at 60°C for 14 h in a laboratory oven (Fistreem International Co. Ltd, Leicestershire, UK). All chemicals utilized in this study were reagent-grade.

Preparation and processing of meals

Sorghum, wheat, and pumpkin seed pulp flour were used to produce different meals (M0, 100:0:0; M1, 70:20:10; M2, 70:15:15, and M3, 70:10:20). Each meal's ingredients were combined with distilled water (1:2 w/v) and allowed to naturally ferment for 24 h at room temperature. Following this incubation period, the fermented flour was placed in aluminum dishes and dried for another 24 h at 70°C in a hot air oven. The dried samples were then ground to pass a 0.4 mm screen and kept at 4ºC. Next, a portion of the dough that had fermented was cooked on a hot steel plate. The cooked meals were kept at 4°C for further analysis after being dried at 70°C in a hot oven and then ground to pass a 0.4 mm screen.

Proximate composition

The crude protein, fat, and ash content were evaluated using the AOAC (2006) standard techniques. The calculation of carbohydrate (nitrogen-free extract) was done using the difference. The energy was calculated using the Atwater factors as described by Novotny *et al.* (2012) there are 4 kcal in one gram of carbs, 4 kcal in one gram of protein, and 9 kcal in one gram of fat.

Determination of mineral contents

The meal samples' mineral contents were measured as described by Harris *et al*. (2017). Exactly 2 g of the material was heated to 550°C for 3 h and then cooled. The ash was treated with 10 mL of

50% HCl and 5 mL of 33% HNO_3, and then placed in a water bath at 100°C for 1 h. The sample was then placed in the water bath for another 15 min after adding 10 mL of HCl. After that, the mixture was put into a 100 mL volumetric flask that contained 100 mL of distilled water, and it was well mixed. The Fe content was determined by atomic absorption spectroscopy (Perkin-Elmer 2380). P content was calculated using the ammonium molybdate method, while Ca was determined using titration (Abdelseed *et al*., 2011).

Minerals HCI-extractability

The extractability of minerals in ingredients and meals was assessed using Kumar and Chauhan's (1993) approach. Approximately 1 g of the material was extracted in 10 mL of 0.03 N HCl at 37°C for 3 h. The clear extract was filtered through Whatman filter paper, oven-dried at 100°C, and then heated in a muffle furnace at 550°C for 3 h. The heated samples were cooled before adding around 3 mL of 3N HCI and diluting with distilled water to make the volume 50 mL. The extract's Fe, P, and Ca concentrations were determined as stated above. The minerals' extractability was calculated as a percentage of HCl extract to total mineral content.

Determination of tannin

The quantitative estimate of tannin was performed using the method provided by Dykes (2019). In a 100 mL conical flask containing 0.2 g of each sample, 10 mL of 1% HCl in methanol *(v/v)* was added, well mixed, and centrifuged at 2500 rpm for 5 min. Then, approximately 5 mL of vanillin-HCl reagent was added to 1 mL of supernatant. After 20 min of incubation at 300°C, the absorbance was measured at 500 nm with a spectrophotometer (PD-UV, Apel, Saitama, Japan). Tannin concentration was calculated using a standard curve, with the results expressed as catechin equivalent (CE mg/100 g).

Determination of phytate

The phytate content was determined using a spectrophotometer (PD-UV, Apel, Saitama, Japan), as described by Mandizvo & Odindo (2020) with slight modification. Phytate phosphorus was determined using a standard curve of varied $Fe(NO_3)_2$ concentrations plotted to compute ferric ion concentration, with a 4:6 iron: phosphorus molar ratio.

In vitro protein digestibility determination (IVPD)

The method provided by Monjula and John (1991) was applied to estimate the *in vitro* protein digestibility. Approximately 2 g of each ingredient, as well as the meals before and after processing, were digested at 37°C for 3 h with 1.5 mg pepsin (EC 3.4.23.1, 662 units/mg; enzyme/substrate, 1:20 (w/w); pH 2.0) in 15 mL of 0.1 M HCl. The mixture was neutralized with NaOH (0.5M) and treated with 4.0 mg pancreatin (8× USP; enzyme/substrate, 1:20 (w/w); pH 8.0) in 7.5 mL of 0.2 M phosphate buffer, pH 8.0, with 0.05% sodium azide; the mixture was then gently shaken and incubated at 37°C for 24 h. Following incubation, the sample was treated with 10% trichloroacetic acid (10 mL) and centrifuged at 5000 g for 20 min at ambient temperature. The nitrogen in the supernatant was measured using the Kjeldahl method (AOAC, 2006). The IVPD was estimated using the formula:

$$\text{Protein digestibility (\%)} = \frac{\text{N in supernatant} - \text{N in the blank}}{\text{N in sample}}$$

Sensory Evaluation

A panel of thirty semi-trained participants, selected from the faculty and staff of the University of Khartoum's Faculty of Agriculture, conducted the sensory evaluation. A variety of attributes were assessed, including general acceptability, texture, flavor, aroma, and appearance. The study employed a 9-point hedonic scale, where 1 denotes extreme dislike, 5 neither like nor dislike, and 9 represents high like. Before the procedure started, the panelists received a briefing and were told to rinse their mouths with water before assessing the next set of samples. Every member of the panel regularly consumed products made from wheat and sorghum.

Statistical analysis

The analysis of variance was performed on three data replicates using IBM SPSS Statistics 23.0 software (SPSS Inc., USA). The means and standard deviations of the data are shown. The multiple range tests of Duncan were performed to ascertain the significance between means. At $p \leq 0.05$, statistical significance was deemed acceptable.

3. Results and discussion

Proximate composition and total energy of ingredients and meals

As shown in Table 1, sorghum flour had a significantly ($p \leq 0.05$) higher content of carbohydrate (75.71%), phosphorus (296.55 mg/100 g), calories (368.82 kcal/100 g) than other ingredients. It also had a higher content of tannin (970.01 mg/100 g) and phytate (2590.42 mg/100 g), which coincided with a lower protein digestibility (34.49%). As reported by Tasie & Gebreyes (2020), Abah *et al*. (2020) and Al Juhaimi *et al*. (2019), sorghum is a rich source of minerals and carbohydrates, but it also contains a high concentration of antinutrients, which reduces the bioavailability of minerals and proteins. Wheat flour, on the other hand, had better protein digestibility (49.88%) and considerably ($p \leq 0.05$) higher carbohydrate content (71.71%) following sorghum. According to Biel *et al*. (2020), wheat is a valuable source of carbohydrates in most industrialized countries, as well as a protein and dietary fiber for human foods. It also contains minerals, lipids, and vitamins, all of which are sources of micronutrients. Furthermore, wheat showed minimal amounts of antinutrients, indicating a high nutritional value. Wheat flour had significantly ($p \leq 0.05$) higher *in vitro* protein digestibility than other ingredients, as reported by Ma and Baik (2021). Pumpkin seed pulp flour was characterized by low antinutrients that coincided with comparable *in vitro* protein digestibility and was higher in ash (8.67%), protein (63.15%), fiber (7.09%), calcium (48.08 mg/100 g), and iron (26.37 mg/100 g) contents than other ingredients. The results reported for pumpkin are similar to those of Adelerin *et al*. (2024). It also had lower levels of phytate (184.96 mg/100 g) and tannin (22.13 mg/100 g), which coincided with comparable protein digestibility. It has been reported that pumpkin seed pulp has a high mineral content that is beneficial to both humans and animals, and it is particularly rich in protein, carbohydrates, and fats with low antinutrients (Shajan *et al*., 2024). Moreover, El-Salhy *et al*. (2017) reported that pumpkin flour is a rich source of beta-carotene, iron, ascorbic acid, and protein.

Table 1. Proximate composition (%), mineral contents (mg/100 g), total energy (kcal/100 g), anti-nutrients, and *in vitro* protein digestibility (IVPD) of meal ingredients

Flour type	Dry matter	Ash	Crude protein	Crude fiber	Fat	CHO	Ca	Fe	P	Total energy	Tannin (mg/100 g)	Phytate (mg/100 g)	IVPD (%)
WadAhmed	93.68[b] ± 0.07	1.51[a] ±0.01	10.41[c] ±0.15	2.46[b] ±0.07	3.78[a] ±0.24	75.71[a] ±0.19	20.89[b] ±0.29	5.76[c] ±0.27	296.55[a] ± 1.95	368.82[a] ± 2.38	970.01[a] ± 1.13	259.42[a] ±1.12	34.49[c] ±1.64

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Wheat flour	87.67[c]	0.89[c]	12.63[b]	0.44[c]	2.01[b]	71.71[b]	8.54[c]	10.86[b]	75.76[c]	347.12[b]	40.67[b]	43.42[c]	49.88[a]
	±0.03	±0.02	±0.14	±0.01	±0.21	±0.26	±0.69	±0.67	±0.55	±4.34	±0.64	±1.99	±0.82
Pumpkin	94.65[a]	8.67[b]	63.15[a]	7.09[a]	3.81[a]	11.92[c]	48.08[a]	26.37[a]	224.1[b]	334.59[c]	22.13[c]	184.96[b]	47.9[b]
seed	±0.09	±0.01	±0.13	±0.11	±0.12	±0.17	±0.55	±1.92	±0.51	±2.28	±0.86	±0.15	±0.81

Values are mean ± SD. Values with the same superscript letter in the column are not significantly different ($p \leq 0.05$). CHO = Carbohydrates

To compensate for nutritional deficiencies and to mitigate the variables that contribute to decreased nutrient bioavailability of sorghum, wheat, and PSP in varying ratios were mixed with sorghum flour, fermented, and cooked to provide nutrient-dense, ready-to-eat meals. Flour meals including M0 (100:0:0), M1 (70:20:10), M2 (70:15:15), and M3 (70:10:20) were created using varying percentages of sorghum, wheat, and PSP flour. The percentage of each ingredient was chosen with the sorghum as the major ingredient, accounting for more than 50%. The remaining percentage split between the other two ingredients, keeping in mind that pumpkin pulp should not exceed 20% to avoid bitterness. Kiharason *et al*. (2017) discovered that adding pumpkin flour at or above 40% resulted in a bitter taste and pungent odor, causing a general dislike of bakery products.

Table 2 depicts the changes in proximate composition and energy of sorghum flour during formulation and processing. The dry matter percent of sorghum flour meal (M0) did not change significantly after adding various ratios of wheat and PSP flour. Fermentation of both formulated and unformulated meals significantly decreased dry matter percent ($p \leq 0.05$) because microbial activities consume energy and nutrients during fermentation, resulting in a decrease in carbohydrate content (Simwaka *et al*., 2017). However, after cooking, the matter percentage increased significantly due to moisture loss. The addition of wheat and PSP flour considerably

Table 2. Proximate composition (%) and total energy (kcal) of a ready-to-use fermented and cooked meal

Chemical Composition (%)	Meal			
	M0	M1	M2	M3
	Sorghum: Wheat: Pumpkin seed pulp			
	100:0:0	70:20:10	70:15:15	70:10:20
Dry matter				
Raw	93.68^{aA} ± 0.65	92.59^{aA} ± 0.55	93.05^{aA} ± 0.34	93.41^{aA} ± 0.36

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Fermented	$91.92^{aC} \pm 0.35$	$91.59^{aB} \pm 0.62$	$91.67^{aB} \pm 0.84$	$91.85^{aC} \pm 0.67$
Cooked	$92.68^{aB} \pm 0.27$	$91.79^{aB} \pm 0.87$	$91.81^{aB} \pm 0.15$	$92.17^{aB} \pm 0.93$
Ash				
Raw	$1.51^{bB} \pm 0.13$	$2.49^{aA} \pm 0.31$	$2.91^{aA} \pm 0.21$	$2.11^{aB} \pm 0.09$
Fermented	$2.13^{bA} \pm 0.15$	$2.52^{bA} \pm 0.88$	$3.11^{aA} \pm 0.89$	$3.32^{aA} \pm 0.12$
Cooked	$2.53^{bA} \pm 0.27$	$2.74^{bA} \pm 0.05$	$3.07^{aA} \pm 0.86$	$3.52^{aA} \pm 0.69$
Crude protein				
Raw	$10.41^{dC} \pm 0.15$	$18.65^{bA} \pm 0.21$	$21.18^{aB} \pm 0.13$	$16.16^{c} \pm 0.07$
Fermented	$12.84^{dA} \pm 0.36$	$19.82^{cB} \pm 0.17$	$23.35^{aA} \pm 0.12$	$22.86^{bA} \pm 0.11$
Cooked	$11.34^{dB} \pm 0.18$	$16.62^{cC} \pm 0.13$	$19.25^{bC} \pm 0.24$	$22.65^{aA} \pm 0.94$
Crude fiber				
Raw	$2.46^{bA} \pm 0.66$	$2.85^{bA} \pm 0.59$	$3.26^{aA} \pm 0.31$	$2.82^{bC} \pm 0.54$
Fermented	$1.96^{dB} \pm 0.13$	$2.33^{cbA} \pm 0.16$	$2.51^{bB} \pm 0.44$	$2.03^{aB} \pm 0.28$
Cooked	$2.19^{bB} \pm 0.33$	$2.44^{bA} \pm 0.31$	$2.62^{bB} \pm 0.28$	$2.64^{aA} \pm 0.06$
Fat				
Raw	$3.78^{bB} \pm 0.24$	$4.27^{aA} \pm 0.08$	$4.61^{aA} \pm 0.04$	$3.93^{bB} \pm 0.13$
Fermented	$4.02^{aA} \pm 0.19$	$4.09^{aA} \pm 0.13$	$4.38^{aA} \pm 0.11$	$4.74^{aA} \pm 0.03$
Cooked	$3.96^{bA} \pm 0.47$	$4.05^{bA} \pm 0.11$	$4.36^{aA} \pm 0.11$	$4.43^{aA} \pm 0.21$
Carbohydrates				
Raw	$75.72^{aA} \pm 0.19$	$64.78^{cB} \pm 0.13$	$61.76^{cA} \pm 0.31$	$67.99^{bA} \pm 0.31$
Fermented	$70.99^{aC} \pm 0.26$	$64.91^{bB} \pm 0.98$	$61.43^{cA} \pm 0.46$	$58.34^{dB} \pm 0.98$
Cooked	$72.7^{aB} \pm 0.38$	$65.95^{bA} \pm 0.29$	$61.85^{cA} \pm 0.74$	$58.32^{dB} \pm 0.96$
Total energy (kcal/100 g)				
Raw	$378.82^{aA} \pm 2.35$	$372.54^{cB} \pm 1.18$	$373.82^{bB} \pm 1.38$	$371.01^{dA} \pm 1.08$
Fermented	$371.86^{cC} \pm 1.47$	$375.63^{bA} \pm 1.27$	$378.51^{aA} \pm 1.72$	$367.49^{cB} \pm 1.43$
Cooked	$371.42^{aC} \pm 1.68$	$366.91^{bC} \pm 1.48$	$363.76^{cC} \pm 1.57$	$363.84^{cC} \pm 1.76$

Values are mean ± SD. Means not sharing a common superscript(s) a, b, or c in a row or A, B, or C in a column are significantly different at $p \leq 0.05$ as assessed by Duncan's Multiple Range Test. S: W: P = percent of sorghum (WadAhmed), wheat, and pumpkin seed flour in the meal

increased the ash content of the meals compared to the control meal ($p \leq 0.05$), with the greatest values observed for meals made with 20% PSP (M2). Fermented and cooked-formulated meals exhibited higher ash content than unformulated meals, with M3 having a substantially higher value ($p \leq 0.05$). The increase in ash concentration with the addition of wheat and PSP flour could be attributed to the high ash level found in PSP flour, as reported by Hamed *et al.* (2008). Furthermore, the current finding is consistent with Ike *et al.* (2020), who investigated the nutritional and microbiological properties of pumpkin seed composite flours and concluded that the increase in ash content among the blends could be attributed to the pumpkin seed flour's higher nutritive value than wheat flour.

Incorporating PSP flour at all ratios significantly increased protein and fiber contents ($p < 0.05$), with M2 providing more protein (21.18%) and fiber (3.26%) compared to other formulations. This is mostly due to PSP flour's high protein and fiber content, as demonstrated by Hamed *et al*. (2008). According to Ike *et al*. (2020), the protein and fiber content of the blends increases with the amount of pumpkin seed flour substituted. This anticipated increase served as the foundation for designing the blends, resulting in a finished product with not only more protein quantity but also higher protein quality. Fermentation of formed and unformed meals resulted in a considerable ($p \leq 0.05$) increase in protein content, with M2 containing more protein (23.35%) than the other meals. The increase in protein content before processing may be due to a high level of ingredients, as shown in Table 1. In addition, fermentation significantly ($p \leq 0.05$) improved protein content by synthesis and solubilization by microorganisms from metabolic intermediates, as reported by Kårlund *et al*. (2020) and Adebo *et al*. (2022). Moreover, Adelerin *et al*. (2024) reported that the higher protein level in the fermented flour sample could be attributed to microorganisms' metabolic activity during fermentation, which resulted in increased protein content. Also, Awolu *et al*. (2017) discovered an increase in the protein content of fermented kidney beans. Cooking, on the other hand, reduced the protein content of all meals, even the control, due to changes in protein structure during cooking (Barpete *et al*., 2021).

Crude fiber was reduced significantly ($p \leq 0.05$) during fermentation, but increased somewhat after heating all meals. The decrease in fiber content and the protein increase after fermentation agree with Makawi et al. (2019), who investigated the quality of cooked sorghum flour fermented with baobab fruit pulp flour as a starter. A study showed that a considerable ($p \leq 0.05$) loss in fiber content during fermentation may be attributed to breakdown by fermenting bacteria (Babalola and Giwa 2012), which is consistent with the current findings. Despite defatting pumpkin seed flour, adding wheat and PSP flour considerably increased ($p \leq 0.05$) the fat content of the unformulated meal, while marginally decreasing it after processing. The increase in fat content in raw meals may be due to high residual fat in PSP flour, but the decrease after processing may be related to a high rate of lipolysis to fatty acids and glycerol during fermentation. Although nutrient changes during fermentation depend on the inherent and accessible nutrients in the starting raw material, Adebo *et al.* (2022) report consistent trends. In cereals and legumes, fermentation generally increased these nutritive qualities such as fats and fatty acids. However, in certain instances, a decrease in these constituents was noted. Carbohydrates levels declined with the addition of wheat and PSP flour, owing to the low carbohydrate levels in both ingredients. An additional reduction was observed during processing, which could be attributed to fermenting micro-flora utilizing part of these nutrients for energy production (Olagunju *et al.*, 2018). The inclusion of wheat and PSP flour reduced the total energy of the unformulated meal, both before and after processing, which might be attributed to a drop in carbohydrates and a slower rate of fat increase.

Changes in antinutrients and in vitro protein digestibility (IVPD) during the processing of meals

Table 3 shows the impact of including wheat and PSP flour on anti-nutritional components and IVPD in formulated and unformulated meals. When compared to other ingredients, raw sorghum flour contained the highest levels of tannin (970.01 mg/100 g) and phytate (259.42 mg/100 g). Also, sorghum flour had a lower IVPD than the other components. The addition of wheat and PSP flour significantly ($p \leq 0.05$) lowered tannin and phytate levels. Additionally, increasing ingredient levels resulted in a significant ($p \leq 0.05$) decrease in anti-nutrients and was accompanied by a sharp increase in IVPD. Further significant ($p \leq 0.05$) reduction in antinutrients and an increase in IVPD were observed when the meals were fermented.

Adelerin *et al.* (2024) found that the enzymes tannase and phytase generated by bacteria during fermentation are responsible for the breakdown of tannin and phytate complexes, increasing protein digestibility. However, fermentation had little effect on the saponin concentration of pumpkin flour. Olagunju *et al.* (2018) found that fermentation significantly reduced phytic acid concentration, tannin content, and trypsin inhibitor activity. This could be attributed to the action of phytase, which is released during fermentation. Furthermore, the reduction in tannin levels in prepared meals could be attributed to the action of the enzyme tannase produced by microorganisms during fermentation, as described by Makawi *et al.* (2019). According to Annor *et al.* (2017), processing reduces anti-nutrient levels and degrades proteins, while increasing protein digestibility.

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Table 3. Ant-nutrients and *in vitro* protein digestibility (IVPD) of a ready-to-use fermented and cooked meal

Anti-nutrients /IVPD	Meal			
	M0	M1	M2	M3
	S: W: P			
	100:0:0	70:20:10	70:15:15	70:10:20
Tannin (mg/100 g)				
Raw	$970.01^{dA} \pm 1.13$	$810.04^{cA} \pm 2.03$	$691.87^{bA} \pm 3.09$	$687.91^{aA} \pm 3.18$
Fermented	$370.04^{dB} \pm 2.01$	$490.11^{cB} \pm 1.02$	$310.21^{bB} \pm 2.13$	$320.03^{aB} \pm 1.08$
Cooked	$150.21^{dC} \pm 2.21$	$330.25^{cC} \pm 1.32$	$240.33^{bC} \pm 1.07$	$280.26^{aC} \pm 2.14$
Phytate (mg/100 g)				
Raw	$259.421^{aA} \pm 1.15$	$211.31^{dA} \pm 0.97$	$218.18^{cA} \pm 0.73$	$224.44^{bA} \pm 0.32$
Fermented	$98.67^{dB} \pm 0.41$	$98.32^{cB} \pm 0.99$	$105.06^{bB} \pm 0.55$	$111.65^{aB} \pm 0.53$
Cooked	$67.17^{dC} \pm 0.13$	$76.42^{cC} \pm 0.67$	$82.61^{bC} \pm 0.26$	$89.82^{aC} \pm 0.24$
IVPD (%)				
Raw	$34.49^{bC} \pm 1.62$	$38.91^{aC} \pm 0.95$	$38.81^{aC} \pm 0.98$	$38.72^{aC} \pm 0.92$
Fermented	$52.03^{aA} \pm 1.32$	$50.72^{bA} \pm 0.84$	$49.96^{bA} \pm 0.65$	$49.26^{bA} \pm 0.75$
Cooked	$49.07^{dB} \pm 1.98$	$42.65^{aB} \pm 0.89$	$41.11^{bB} \pm 0.31$	$40.05^{cB} \pm 0.45$

Values are mean ± SD. Means not sharing a common superscript(s) a, b, or c in a row or A, B, or C in a column are significantly different at $p \leq 0.05$ as assessed by Duncan's Multiple Range Test. S: W: P = percent of sorghum (WadAhmed), wheat, and pumpkin seed flour in the meal

Protein digestibility during fermentation was reported to be enhanced by the release of protein from plant tissues by the enzymatic breakdown of dietary fibers and the reduction or degradation of polyphenols, tannins, and phytic acid by the action of enzymes produced by microbes (Annor *et al*., 2017). Suarti and Budijanto (2021) proposed that a reduction in pH values during fermentation may improve peptidase enzyme activity and activate endogenous proteases, resulting in increased peptide and free amino acid concentrations and protein solubility. Several seeds with low protein digestibility, caused by tannins, phytic acid, and trypsin inhibitors, can be made more digestible through boiling or fermentation. Among these methods, fermentation is the most efficient technique for lowering tannin, phytic acid, and trypsin inhibitor activity (Joye, 2019). A comparable impact on the digestibility of protein in fermented sorghum flour was noted by Ogodo *et al*. (2019). The scientists postulated that the highly insoluble storage proteins had been changed into simpler and soluble molecules by hydrolytic enzymes. Additionally, the peptidase enzyme activity and protein solubility may be enhanced by the pH decrease that occurs during fermentation (Ogodo *et al*., 2019).

Changes in mineral contents and extractability during the processing of meals

Table 4 shows a considerable ($p \leq 0.05$) increase in mineral content, specifically Ca, Fe, and P, after incorporating wheat and PSP flour. Fermentation and heating of the meals resulted in an additional and considerable increase in mineral contents and extractability. Adebiyi *et al*. (2017) found that fermented pearl millet had a drop in total ash content after fermentation, but an increase in mineral elements such as Ca, Na, Cu, Fe, Zn, and K was observed. The leaching of soluble salts resulted in a reduction in ash, whereas an increase in mineral elements was caused by better mineral extractability and availability as a result of fermentation. Ilowefah *et al*. (2015) reported a rise in the mineral and ash contents of fermented rice, with the increase in ash content attributed to increased mineral solubility and bioavailability. The increased P content in fermented meals could be attributed to the action of phytase, which hydrolyzes phytate and produces more phosphorus (Humer & Schedle, 2016). The phytate analysis entails removing phosphate groups from the phytate's inositol ring, which lessens the strength of mineral binding to phytate and thus increases essential mineral bioavailability.

The mineral content of cooked meals increased as the ratio of ingredients increased, particularly PSP flour, which contains a high quantity of minerals (Hamed *et al*., 2008). Among the minerals studied, the extractability study revealed that P was the most extractable mineral in raw meals, followed by Ca and Fe. Fermentation considerably ($p \leq 0.05$) enhanced mineral extractability in meals. Sokrab *et al*. (2014) demonstrated that fermenting both high and low-phytate corn improved mineral extractability.

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Table 4. Total (mg/100 g) and extractable (%) minerals of ready-to-use fermented and cooked meals

Minerals (mg/100g)	Meal			
	M0	M1	M2	M3
	S: W: P			
	100:0:0	70:20:10	70:15:15	70:10:20
Ca				
Raw				
Total	$20.89^{dC} \pm 0.95$	$28.57^{cC} \pm 0.13$	$32.01^{bB} \pm 0.77$	$36.75^{aB} \pm 0.45$
Extractable	$34.43^{dq} \pm 0.85$	$40.34^{cr} \pm 0.57$	$44.88^{br} \pm 0.59$	$49.12^{ar} \pm 0.89$
Fermented				
Total	$29.78^{dA} \pm 0.95$	$32.03^{cA} \pm 0.48$	$34.21^{bA} \pm 0.17$	$38.37^{aA} \pm 0.74$
Extractable	$84.78^{dp} \pm 1.27$	$87.36^{cp} \pm 0.65$	$89.97^{bp} \pm 0.69$	$91.95^{ap} \pm 0.87$
Cooked				
Total	$25.16^{dB} \pm 0.56$	$28.03^{cC} \pm 0.21$	$32.01^{bB} \pm 0.56$	$36.03^{aB} \pm 0.42$
Extractable	$30.13^{dr} \pm 0.98$	$42.46^{cq} \pm 0.49$	$51.23^{bq} \pm 0.49$	$56.37^{aq} \pm 0.59$
Fe				
Raw				
Total	$5.76^{dC} \pm 0.28$	$7.88^{cC} \pm 0.24$	$10.43^{bC} \pm 0.45$	$12.6^{aC} \pm 0.02$
Extractable	$16.81^{dr} + 0.49$	$29.34^{cr} \pm 0.45$	$34.12^{br} \pm 0.58$	$41.13^{ar} \pm 0.29$
Fermented				
Total	$9.99^{dB} \pm 0.71$	$12.01^{cB} \pm 0.16$	$13.12^{bB} \pm 0.26$	$15.29^{aB} \pm 0.59$
Extractable	$27.78^{dq} \pm 0.89$	$38.79^{cq} \pm 0.59$	$41.68^{bq} \pm 0.57$	$51.45^{aq} \pm 0.35$
Cooked				
Total	$16.42^{dA} \pm 0.58$	$17.53^{cA} \pm 2.28$	$18.16^{bA} \pm 1.73$	$20.72^{aA} \pm 1.96$
Extractable	$41.18^{dp} \pm 0.94$	$44.68^{cp} \pm 0.57$	$47.79^{bp} \pm 0.48$	$54.67^{ap} \pm 0.89$
P				
Raw				
Total	$296.55^{aA} \pm 1.95$	$243.63^{dC} \pm 4.53$	$255.77^{cC} \pm 4.22$	$265.71^{bC} \pm 3.27$
Extractable	$44.24^{dq} \pm 0.94$	$46.56^{cq} \pm 0.52$	$47.99^{br} \pm 0.69$	$49.47^{ar} \pm 0.58$
Fermented				
Total	$385.9^{cB} \pm 5.98$	$373.45^{dA} \pm 2.75$	$394.3^{bA} \pm 2.02$	$429.13^{aA} \pm 2.96$
Extractable	$67.67^{dp} \pm 0.86$	$69.88^{cp} \pm 0.59$	$72.58^{bp} \pm 0.38$	$75.26^{ap} \pm 0.67$
Cooked				
Total	$284.8^{dC} \pm 5.65$	$303.8^{cB} \pm 5.08$	$386.6^{bB} \pm 2.19$	$412.21^{aB} \pm 0.98$
Extractable	$41.23^{dr} \pm 0.88$	$43.78^{cr} \pm 0.85$	$51.03^{bq} \pm 0.66$	$57.97^{aq} \pm 0.77$

Values are mean ± SD. The means not sharing a common superscript(s) a, b, or c in a row or A, B, or C for total and or p, q or r for extractable for each element in a column are significantly different at $p \leq 0.05$ as assessed by Duncan's Multiple Range Test. S: W: P = percent of sorghum (WadAhmed), wheat, and pumpkin seed flour in the meal

According to Makawi *et al.* (2019), the increased mineral extractability of meals could be attributed to the hydrolysis of phytate and tannin by the enzymes phytase and tannase during fermentation. Furthermore, the rise in mineral content could be attributed to the loss of dry matter during fermentation, as bacteria break down carbs and protein (Day & Morawicki 2018).

Fermentation increases the bioavailability of calcium, phosphorus, and iron, most likely due to the destruction of tannin and phytates, which are complexed with minerals and reduce their bioavailability. The mineral extractability of cooked meals was increased gradually and significantly ($p \leq 0.05$) with the ingredient quantities. This rise may be due to the components' high mineral extractability.

Sensory attributes of meals formulate

Table 5 compares the sensory quality of fermented and cooked meals using varied amounts of defatted pumpkin seed pulp and wheat flour. When compared to sorghum meal, wheat, and defatted pumpkin seed pulp-formulated meals, all sensory properties were superior and statistically different from the control meal (M0). This could be due to the mixture of wheat and pumpkin, which considerably boosted such qualities. According to the present investigation, adding wheat and pumpkin to sorghum could improve meals while retaining their organoleptic features. Overall acceptability findings indicate that panelists favored flour blend formulations M1, M2, and M3, while the control sample (M0) was the least accepted.

Table 5. Sensory data of meals formulated from sorghum, wheat, and pumpkin seed flour mix

Meal	Appearance	Aroma	Taste	Flavor	Texture	Overall acceptability
M0 (100 % sorghum)	6.17 ± 1.21^{b}	6.05 ± 1.3^{a}	6.63 ± 1.53^{b}	6.45 ± 1.25^{b}	6.11 ± 1.16^{a}	6.18 ± 1.37^{b}
M1 (S: W: P, 70:20:10)	7.29 ± 1.15^{a}	7.37 ± 1.2^{a}	7.75 ± 1.16^{a}	7.56 ± 1.24^{a}	7.69 ± 1.04^{a}	7.53 ± 1.18^{a}
M2 (S: W: P, 70:15:15)	7.47 ± 1.13^{a}	7.56 ± 1.05^{a}	7.45 ± 1.13^{a}	7.43 ± 1.13^{a}	7.41 ± 1.21^{a}	7.46 ± 1.27^{a}
M3 (S: W: P, 70:10:20)	7.34 ± 1.51^{a}	8.06 ± 1.0^{a}	7.55 ± 1.32^{a}	7.63 ± 1.32^{a}	7.54 ± 1.31^{a}	7.62 ± 1.31^{a}

Values are mean ± SD. Means not sharing a common superscript(s) a, b, or c in a column are significantly different at $p \leq 0.05$ as assessed by Duncan's Multiple Range Test. S: W: P = percent of sorghum, wheat, and pumpkin seed flour in the meal

These findings imply that including a high proportion of pumpkin seed pulp flour into fermented sorghum flour increases the sensory rating of the resulting flour blend compositions. The findings coincide with Kiharason *et al*. (2017), who found that including pumpkin flour in wheat at levels of 40% or higher resulted in a bitter taste and pungent odor, leading to a general dislike of bakery products. In contrast, substituting pumpkin flour at 20% and 30% produced consumer-acceptable food products. In contrast, Adeyeye *et al*. (2019) examined the impact of fermentation on the nutritional composition, anti-nutritional factors, and acceptability of cookies containing fermented sorghum and soybean flour blends, discovering that fermentation influenced all sensory characteristics as well as overall acceptability. Moreover, Adobofor *et al*. (2018) studied the anti-nutritional factors, mineral composition of pumpkin pulp, and functional properties of composite pumpkin wheat flour for bread preparation. They concluded that sensory evaluation showed that consumers preferred the control sample and bread replaced with 5% pumpkin pulp flour.

4. Conclusion

The findings of this study demonstrated the nutritional impact of integrating PSP and wheat flour into sorghum flour after fermentation and cooking. When PSP and wheat flour were added to the flour mix formulations, their protein, iron, calcium, phosphorus, and total energy content rose. Following processing, the resulting flour blend formulations exhibited high nutritional quality due to the elimination of antinutrients. They were successful in meeting preschool children's daily protein, iron, and calcium nutrient needs. This shows that such a formulation can be used as a fortifying agent to improve the nutrient quality of low-nutrition cereal meals, addressing protein, calcium, and iron deficiencies in children. Sensory studies demonstrated that the composite flour had significantly improved sensory qualities; hence, such formulations, particularly those comprising 20% PSP and 10% wheat flour (M3), have the potential to increase the acceptance of sorghum-based products. More research is needed to assess the usefulness of these mixed formulations in improving the vitamin status of preschool children. In addition, *in vivo* assimilation

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experiments are needed to assess the actual amounts of nutrients absorbed in the body after the intake of meals containing a sorghum-pumpkin-wheat flour mixture.

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