PROXIMATE COMPOSITION AND IN VITRO STARCH DIGESTIBILITY OF WHEAT AND GERMINATED-FERMENTED BROWN FINGER MILLET FLOUR BLENDS

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Abstract
The aim of this study was to investigate the proximate composition and in vitro starch digestibility of wheat flour and germinated-fermented brown finger millet flour blends. Wheat flour (WF) was substituted with germinated-fermented brown finger millet flour (GFBFMF) at different proportions (5%, 10%, 15%, 20%, 25% and 30%) and uniformly blended to obtain WF-GFBFMF samples. The proximate composition and starch characteristics of 100% wheat flour (control) and WF-GFBFMF blends were profiled using standard methods. Wheat flour containing 30% GFBFMF had the highest protein, ash, total dietary fiber, resistant starch (RS, 26.79%), slowly digestible starch (SDS, 29.25%) and lowest total starch (60.55 g/100g) and rapidly digestible starch (RDS, 43.96%) compared to 100% wheat flour. The results obtained demonstrated that substitution of wheat flour with germinated-fermented brown finger millet flour improved the protein, ash, total dietary content, and starch digestibility slowly of the blends compared to the control.

Keywords: Brown finger millet, Germination, Solid-state fermentation, proximate characteristics, Starch digestibility

4.0 Introduction

There is increasing interest in the consumption of foods with potential health properties due to the growing prevalence of various chronic and non-communicable diseases caused by significant increase in the consumption of high carbohydrate foods because of dietary patterns and changing lifestyles of people (Liu *et al.*, 2019; Cao *et al.*, 2022). Accordingly, research developments for food security targeting developing nations have been geared towards integrating functional ingredients from neglected and underutilized crops and modifying the food matrix to enhance nutrient bioavailability (Mensi and Udenigwe, 2021).

Brown finger millet is an underutilized grain cultivated in the tropical developing regions of Africa and Asia. It is rich in nutrients, micronutrients, phytochemicals, and antioxidant activities (Mitharwal *et al.*, 2021). Brown finger millet grains contain 52.71 g/100g starch, 7.54 g/100 g resistant starch and 45.17g/100g digestible starch (Azeez *et al.*, 2022) compared to refined wheat flour that contained higher total starch content (68.52 g/100 g) (Chinma *et al.*, 2023). Brown finger millet has low glycemic index and has been found to be useful in the management of diabetes and other chronic and non-communicable diseases (Singh *et al.*, 2012).

It has been reported that combined germinated and solid-state fermented brown finger millet flour (GFBFMF) contained low starch digestibility, residual antinutrients, improved physiochemical, phytochemical, antioxidant, functional and processing properties compared to the raw brown finger millet flour and could be utilized in the formulation of functional bakery products (Azeez *et al.*, 2022). These findings corroborate previous studies which showed that combined effects of germination and fermentation positively influence the properties of cereal and/or legume for diverse food applications (Rasane *et al.*, 2015). Therefore, substituting wheat flour with germinated-fermented brown finger millet flour may be promising to improve the proximate composition and starch digestibility of the resultant composite flours for improved nutrition, and promotion of health and wellness. However, there is paucity of information on the proximate composition and starch characteristics of wheat flour substituted with germinated-fermented brown finger millet flour. The objective of the study was to evaluate the proximate composition and starch digestibility of wheat and germinated-fermented brown finger millet flour blends.

2.0 Materials and Methods

2.1 Source of materials

Brown finger millet grains and wheat flour (Golden Penny Plc., Lagos, Nigeria) were procured from Central market Minna, Nigeria.

2.2. Preparation of germinated and fermented brown finger millet flour (GFBFMF)

The germinated-fermented brown finger millet flour was prepared as reported by Azeez *et al.* (2022). Cleaned brown finger millet grains were sterilized with 0.07 g/L food grade sodium hypochlorite solution for 30 min, drained and soaked in distilled water (for 12 h at 28 ± 2 ∘C) and germinated under darkness (at 25 ∘C for 48 h). The germinated grains were dried in an air-draft oven (Model TI2h, Genlab, Cheshire, U.K) at 40 ∘C for 24 h. The germinated brown finger millet grains were milled and sieved (100 µm mesh) to obtain germinated finger millet flour. Thereafter, germinated finger flour was used in the preparation of germinated-fermented finger millet flour following the method reported by Azeez *et al.* (2022). Briefly, dry baker's yeast (1 g) was mixed with 65 mL distilled water and the suspension was poured into 100 g of fermented finger millet brown and uniformly mixed in a glass beaker for 2 min. Thereafter, the slurry was covered with aluminium foil (Magic Wrap, Yuyao, Zhejiang, China) and fermented in a fermentation cabinet (National MEG Company, Lincoln, USA; at 27 ∘C for 16 h). Thereafter, the batter was dried in an air-draft oven at 40 ∘C for 24 h and the dried flour was blended and sieved through 100 µm mesh sieve to obtain germinated-fermented brown finger millet flour. The constituents of the germinated-fermented brown finger millet flour were profiled using standard methods. The GFBFMF contained 5.72 pH, 0.17% titratable acidity, 7.92 g/100 g moisture content, 13.15 g/100g protein, 2.91 g/100g ash, 1.13 g/100 g fat, 14.86 g/100 g total dietary fiber, 55.08 g/100g total starch, 9.33 g/100 g resistant starch, 54.95 g/100g rapidly digestible starch and 36.72 g/100 g slowly digestible starch.

2.3 *Formulation of blends*

The flour blends were formulated by substituting wheat flour with different levels of 0, 5, 10, 15, 20, 25 and 30% germinated-fermented brown finger millet flour. The flour blends were thoroughly mixed with a blender (BLX750RD, Kenwood, Sheffield, UK) and separately packaged in a plastic container with lids before analysis.

2.4 *Profiling of proximate composition and starch digestibility of flour blends*

The proximate composition (moisture, protein, ash, fat, total dietary fiber and carbohydrate content) of the samples was analyzed according to the method of AOAC (2005). Total starch kit (Megazyme, K-RSTA, Wicklow, Ireland) was used in profiling total starch content (Chinma *et al.*, 2023). Starch fractions (resistant, slowly and rapidly digestible starch) were assayed following a standard procedure (Englyst *et al.*, 1992) as detailed in a previous work by Chinma *et al.* (2023). Briefly, 200 mg of sample mixed with sodium acetate buffer (15 mL, pH 5.2) and allowed to equilibrate for 5 min at 30∘C. Resistant starch (RS), rapidly digested starch (RDS) and slowly digested starch (SDS) were determined following the hydrolysis of samples using glucose oxidase-peroxidase from Megazyme kit, Bray, Ireland. The percentage of resistant starch (RS), rapidly digestible starch (RDS) and slowly digestible starch (SDS) were calculated as described in equation 1.

$$RS (\%) = [(TS-RDS-SDS)/TS] \times 100 \quad (1)$$

2.5 *Statistical analysis*

Data obtained were subjected to analysis of variance (ANOVA) using statistical software (SPSS 20, IBM, Armonk, NY, USA) and Tukey's test was employed at 5% probability to determine significant differences among means.

3.0 Results and Discussion

3.1 Proximate composition of WF-GFBFMF blends

The proximate compositions of the 100% wheat flour and wheat-GFBFM flour blends are presented in Table 1. The moisture content of the blends was relatively low and suggests shelf stability of the flour blends during storage. The 100% wheat flour lower protein, ash and total dietary fiber and higher fat and total starch compared to the composite blends probably due to the inclusion effect caused by higher content of protein, ash and total dietary fiber in GFBFMF than 100%. Compared to the control (100% wheat flour), addition of GFBFMF (5-30%) caused a significant increase in the protein, ash and total dietary fiber content of the blends whereas fat and carbohydrate content decreased ($p \leq 0.05$) (Table 1). Higher protein and ash content of the composite flour samples could be beneficial, because good quality protein intake is essential for growth and physiological activities (Chinma *et al.*, 2023), whereas higher ash value suggests higher content of mineral elements required for the promotion of good health. Higher dietary fiber content of the composite flours than 100% wheat may partly be ascribed to the inclusion of a fiber rich GFBFMF, which could be beneficial, owning to the fact that foods rich in dietary fiber are important in improving human health and nutrition. The proximate results suggest that the blends would be useful in the formulation of nutritious wheat-based products.

3.2 In vitro starch digestibility of WF-GFBFMF blends

Table 2 shows that a significant difference was recorded in the starch fractions (resistant, slowly digestible and rapidly digestible starch) between 100% wheat flour and the composite samples containing GFBFMF. The 100% wheat flour contained higher total starch and rapidly digestible starch with lower resistant starch and slowly digestible starch than the WF-GFBFMF samples (Table 2). The RS and SDS increased steadily with the addition of GFBFMF while total starch (TS) and RDS significantly decreased. The decreased starch digestibility recorded in WF-GFBFMF samples may be partly ascribed to higher resistant starch and total dietary fiber content in GFBFMF. Higher dietary fiber and resistant starch have been previously reported to cause decreased starch digestibility in foods by reducing the diffusion of amylase enzyme, and the release of the products of starch hydrolysis because these constituents can stick on the starch granules acting as physical barriers for enzyme degradation (Chinma *et al.*, 2021). The improved starch digestibility of the WF-GFBFMF samples suggests their potential in promoting good health because the samples would not cause a spike in the blood sugar level. Therefore, there is the need to explore their potential in the development of functional wheat-based foods.

4.0 Conclusion

The results obtained in this study demonstrated that the addition of increasing level of germinated-fermented brown finger millet flour to wheat flour significantly improved the protein, ash, total dietary fiber and starch digestibility of the blends. This suggests that the WF-GFBFMF blends could serve as functional flours. However, future work on the rheological and techno-functional properties of the flour blends are needed to determine their specific food application during food product development.

Table 1: Proximate composition of wheat and germinated-fermented brown finger millet flour blends

Samples	100WF	95WF: 5 GFBFMF	90WF: 10 GFBFMF	85WF: 15 GFBFMF	80WF: 20 GFBFMF	75WF: 25 GFBFMF	70WF: 30 GFBFMF
Moisture content (g/100g)	8.10±0.01^{a}	8.05±0.03^{a}	7.80±0.05^{b}	8.01±0.03^{a}	7.92±0.04^{ab}	8.01±0.02^{a}	8.05±0.03^{a}
Protein (g/100 g)	11.04±0.04^{g}	11.36±0.01^{f}	11.47±0.01^{e}	11.56±0.01^{d}	11.63±0.02^{c}	11.82±0.03^{b}	11.97±0.02^{a}
Ash (g/100g)	0.75±0.03^{g}	0.88±0.01^{f}	1.03±0.01^{e}	1.14±0.01^{d}	1.36±0.00^{c}	1.45±0.01^{b}	1.73±0.03^{a}
Fat (g/100g)	1.22±0.02^{a}	1.19±0.00^{a}	1.14±0.01^{a}	1.09±0.01^{ab}	1.03±0.01^{b}	0.97±0.00^{b}	1.02±0.01^{b}
Total dietary fiber (g/100g)	1.54 ±0.01^{g}	2.28±0.02^{f}	3.02±0.01^{e}	4.46±0.01^{d}	5.11±0.02^{c}	5.92±0.01^{b}	6.68±0.05^{a}
Carbohydrate content (g/100g)	77.35 ±0.23^{a}	76.24±0.44^{b}	75.53±0.31^{c}	73.74±0.56^{d}	72.95±0.49^{e}	71.83±0.37^{f}	70.55±0.36^{g}

Mean ± standard=deviation of triplicates. Values with different superscripts in a row are significantly different ($p \leq 0.05$). 100WF = 100% wheat flour (control); GFBFMF= Germinated-fermented brown millet flour; 95WF:5GFBFMF = 95% wheat flour : 5% Germinated-fermented brown finger millet flour, 90WF:10GFBFMF = 90% wheat flour : 10% Germinated-fermented brown finger millet flour; 85WF:15GFBFMF = 85% wheat flour : 15% Germinated-fermented brown finger millet flour; 80WF:20GFBFMF = 80% wheat flour : 20% Germinated-fermented brown finger millet flour; 75WF:25GFBFMF = 75% wheat flour : 25% Germinated-fermented brown finger millet flour and 70WF:30GFBFMF = 70% wheat flour : 30% Germinated-fermented brown finger millet flour.

Table 2: In vitro starch of wheat and germinated-fermented brown finger millet flour blends

Samples	100WF	95WF: 5 GFBFMF	90WF: 10 GFBFMF	85WF: 15 GFBFMF	80WF: 20 GFBFMF	75WF: 25 GFBFMF	70WF: 30 GFBFMF
Total starch (g/100g)	75.84±0.14^{a}	65.82±0.10^{b}	64.60±0.14^{c}	63.88±0.10^{d}	62.73±0.11^{e}	61.39±0.13^{f}	60.55±0.12^{g}
Resistant starch (%)	4.38±0.16^{g}	23.15±0.13^{f}	24.07±0.11^{e}	24.92±0.13^{d}	25.64±0.16^{c}	26.05±0.10^{b}	26.79±0.13^{a}
Rapidly digestible starch (%)	63.90±0.12^{g}	52.29±0.17^{f}	50.05±0.15^{e}	48.44±0.12^{d}	46.17±0.10^{c}	45.25±0.15^{b}	43.96±0.14^{a}
Slowly digested starch (%)	23.72±0.17^{g}	24.56±0.19^{f}	25.88±0.13^{e}	26.63±0.19^{d}	28.19±0.12^{c}	28.70±0.17^{b}	29.25±0.11^{a}

Mean ± standard=deviation of triplicates. Values with different superscripts in a row are significantly different ($p \leq 0.05$). 100WF = 100% wheat flour (control); GFBFMF= Germinated-fermented brown millet flour; 95WF:5GFBFMF = 95% wheat flour : 5% Germinated-fermented brown finger millet flour, 90WF:10GFBFMF = 90% wheat flour : 10% Germinated-fermented brown finger millet flour; 85WF:15GFBFMF = 85% wheat flour : 15% Germinated-fermented brown finger millet flour; 80WF:20GFBFMF = 80% wheat flour : 20% Germinated-fermented brown finger millet flour; 75WF:25GFBFMF = 75% wheat flour : 25% Germinated-fermented brown finger millet flour and 70WF:30GFBFMF = 70% wheat flour : 30% Germinated-fermented brown finger millet flour.

References

AOAC. (2005) Official Methods of Analysis (18th ed.). The Association of Official Analytical Chemists.

Azeez, S.O., Chinma, C.E., Bassey, S.O., Eze, U.R., Makinde, A.F., Sakariyah, A.A., ... & Adebo, O.A. (2022). Impact of germination alone or in combination with solid-state fermentation on the physicochemical, antioxidant, in vitro digestibility, functional and thermal properties of brown finger millet flours. *LWT-Food Science and Technology*, 154, 112734.

Cao, H., Wang, C., Li, R., Guan, X., Huang, K., & Zhang, Y. (2022). Influence of sprouted oat flour substitution on the texture and in vitro starch digestibility of wheat bread. *Food Chemistry*: X, 15, 100428.

Chinma, C. E., Ezeocha, V. C., Adedeji, O. E., Ayo-Omogie, H. N., Oganah-Ikujenyo, B. C., Anumba, N. L., Enimola, G. E., Adegoke, D. O., Alhassan, R., & Adebo, O. A. (2023). Germinated Bambara groundnut (*Vigna subterranea*) flour as an ingredient in wheat bread: Physicochemical, nutritional, and sensory properties of bread. *Journal of Food Science*, 88(6), 2368-2384.

Englyst, H.N., Kingman, S.M., & Cummings, J.H. (1992). Classification and measurement of nutritionally important starch fractions. *European Journal of Clinical Nutrition*, 46, 33–50.

Liu, X., Lu, K., Yu, J., Copeland, L., Wang, S., & Wang, S. (2019). Effect of purple yam flour substitution for wheat flour on in vitro starch digestibility of wheat bread. *Food Chemistry*, 284, 118-124.

Mensi, A., & Udenigwe, C. C. (2021). Emerging and practical food innovations for achieving the Sustainable Development Goals (SDG) target 2.2. *Trends in Food Science and Technology*, 111, 783-789.

Mitharwal, S., Kumar, S., & Chauhan, K. (2021). Nutritional, polyphenolic composition and in vitro digestibility of finger millet (*Eleusine coracana* L.) with its potential food applications: a review. *Food Bioscience*, 44, 101382.

Singh, K. P., Mishra, A., & Mishra, H. N. (2012). Fuzzy analysis of sensory attributes of bread prepared from millet-based composite flours. *LWT- Food Science and Technology*, 48(2), 276–282.

Rasane P, Jha A, Kumar A, Sharma N (2015) Reduction in phytic acid content and enhancement of antioxidant properties of nutricereals by processing for developing a fermented baby food. *Journal of Food Science and Technology*, 52(6):3219–3234.