AJBAR Vol. 5(5), October 2026: 189-1201, ISSN: 2811-2881

Arid Zone Journal of Basic and Applied Research
Faculty of Science, Kashim Ibrahim University
Maiduguri, Nigeria

Journal homepage: https://www.azjournalbar.com

Research Article

Amino Acid Composition and *In-Vitro* Digestibility of *Sinasir* Formulated from Rice, Hungry Rice *(Digitaria exilis)* Fortified with Soybean (*Glycine max* L.)

Ilham Mohammad Shamsuddeen[1], Bintu Bukar Petrol[2], Ali Abdullahi Damasak[1], Sa'eedu Auwal Magaji[1], Amina Kachallah Modu[1]

[1]Department of Nutrition and Dietetics, University of Maiduguri, Bama Road, Maiduguri, Nigeria
[2]Department of Biochemistry, University of Maiduguri, Bama Road, Maiduguri, Nigeria
***Corresponding author's Email**: aliadamasak@unimaid.edu.ng, doi.org/10.55639/607.020100158

ARTICLE INFO:

Keywords:
Sinasir,
Hungry rice,
Digitaria exilis,
Soybean,
Amino-acids.

ABSTRACT

Sinasir is a traditional fermented cereal-based food commonly consumed in northern Nigeria. Incorporating hungry rice (*Digitaria exilis*) and soybean (*Glycine max* L.) into its formulation may influence its amino-acid composition and digestibility. However, information on these characteristics in hungry-rice-based *sinasir* remains limited. This study evaluated the amino-acid composition and *In-Vitro* carbohydrate and protein digestibility of *sinasir* produced from white rice, white hungry rice, brown hungry rice and their combinations with soybean. Nine *sinasir* formulations were prepared using different proportions of white rice, white hungry rice, brown hungry rice and soybean. Amino-acid composition was determined following acid hydrolysis using a Technicon Sequential Multi-Sample Amino Acid Analyzer. *In-Vitro* carbohydrate digestibility was assessed using an α-amylase-based starch hydrolysis procedure, while protein digestibility was determined using trypsin digestion at 1 and 6 h. Analyses were performed in triplicate and data were subjected to one-way analysis of variance. Significant differences ($p < 0.05$) were observed in the amino-acid profiles among the formulations. Among the essential amino acids, tryptophan ranged from 1.15 to 5.15 g/100 g protein, histidine from 0.83 to 3.23 g/100 g protein, leucine from 1.56 to 4.66 g/100 g protein, isoleucine from 1.65 to 4.03 g/100 g protein, phenylalanine from 5.88 to 9.64 g/100 g protein, valine from 2.72 to 4.98 g/100 g protein, lysine from 2.74 to 5.18 g/100 g protein, methionine from 2.65 to 7.37 g/100 g protein and threonine from 1.51 to 4.44 g/100 g protein across the formulations. The non-essential amino acids also varied, with tyrosine ranging from 3.15 to 11.12 g/100 g protein and glycine from 3.65 to 8.35 g/100 g protein. *In-Vitro* starch digestibility ranged from 70.42 ± 0.01% to 80.24 ± 0.01%, with brown hungry rice *sinasir* (78.84 ± 0.01%) showing the highest digestibility among the formulated products. Protein digestibility at 1 h ranged from 63.58 ± 0.09% to 78.91 ± 0.01%, while values at 6 h increased to 66.94 ± 0.01–82.62 ± 0.01%. The formulation containing 60 g white rice, 20 g brown hungry rice and 20 g soybean recorded the highest protein digestibility at both 1 h (78.91 ± 0.01%) and 6 h (82.62 ± 0.01%). The incorporation of hungry rice and soybean substantially influenced the amino-acid composition and *In-Vitro* digestibility of *sinasir*. Several formulations demonstrated improved amino-acid characteristics and protein digestibility compared with the white-rice control. These findings support the potential use of hungry rice and soybean in developing nutritionally improved *sinasir*, although the *In-Vitro* values should not be interpreted as direct estimates of human nutrient utilization.

Corresponding author: Ali Abdullahi Damasak, **Email**: aliadamasak@unimaid.edu.ng
Department of Nutrition and Dietetics, University of Maiduguri, Bama Road, Maiduguri, Nigeria

INTRODUCTION

Sinasir is a fermented, rice-based food consumed in northern Nigeria. The development of alternative formulations using indigenous cereals and legumes may support food diversification while retaining a familiar traditional product. Fonio (*Digitaria exilis*), also known as hungry rice, is an indigenous West African cereal whose nutritional and physicochemical properties have been investigated in relation to processing and food use (Jideani and Jideani, 2011; Bassey *et al.,* 2023). Research on stakeholder perspectives has also considered the potential role of fonio in strengthening local diets and food and nutrition security in Nigeria (Ibrahim Bio Yerima *et al.,* 2024).

Soybean (*Glycine max*) is used in many plant-based food formulations because it contributes protein and has an amino-acid profile that differs from that of cereal grains. Combining cereal and legume ingredients can therefore change the amino-acid composition of a food. Nevertheless, amino-acid concentration alone is not sufficient to establish protein quality: digestibility and the availability of indispensable amino acids also need to be considered (Gorissen *et al.,* 2018; Dong *et al.,* 2023).

Food processing can change the structure and properties of proteins and carbohydrates and may affect their accessibility to digestive enzymes. In cereal-based foods, starch digestion is influenced by the starch itself, the surrounding food matrix and interactions with other components (Singh *et al.,* 2010). Heating and other processing conditions can also influence in-vitro protein digestibility, although the direction and magnitude of change depend on the ingredient and processing conditions (Adeleye *et al.,* 2020; Li *et al.,* 2023). The effects of soaking, fermentation and cooking should consequently be evaluated in the finished product rather than assumed from the raw ingredients alone.

Although research has examined fonio's nutritional and physicochemical characteristics and has assessed cereal–legume food formulations, the literature identified for this study provides limited information on the amino-acid profile and in-vitro carbohydrate and protein digestibility of *sinasir* made from white rice, white fonio, brown fonio and selected combinations containing soybean. This study therefore evaluated nine formulations to compare their measured amino-acid composition and in-vitro digestibility. The findings may help guide further product development, while recognizing that in-vitro results do not directly predict human nutrient absorption or establish protein quality in clinical or dietary settings.

MATERIALS AND METHODS

Materials and sample collection

White rice (*Oryza sativa*; Zabarmari), white hungry rice and brown hungry rice (*Digitaria exilis*), soybean (*Glycine max* L.), salt, sugar, potash and groundnut oil were used for the preparation of the *sinasir*. The raw materials were purchased from Maiduguri Monday Market during the dry season of 2023 and authenticated by a breeder/plant taxonomist at the Lake Chad Research Institute, Maiduguri.

Preparation of raw materials

The rice and hungry rice samples were manually sorted to remove stones, damaged grains and other extraneous materials and subsequently washed thoroughly with clean water. Soybean seeds were similarly sorted and washed before processing. These preliminary operations were performed to minimize physical impurities and prepare the ingredients for subsequent *sinasir* production.

Formulation of *sinasir*

Nine formulations were prepared from white rice, white hungry rice, brown hungry rice and soybean. Formulation A consisted entirely of white rice, B of white hungry rice and C of brown hungry rice. Formulations D–G contained combinations of rice, hungry rice and soybean, whereas H and I consisted of rice combined with either white or brown hungry rice without soybean. The formulation ratios are presented in Table 1.

Table 1: Formulation of Ratios for Preparation of *Sinasir*

Sinasir	White Rice	White Hungry Rice	Brown Hungry Rice	Soybeans	Total
A	100	-	-	-	100
B	-	100	-	-	100
C	-	-	100	-	100
D	20	60	-	20	100
E	20	-	60	20	100
F	60	20	-	20	100
G	60	-	20	20	100
H	50	50	-	-	100
I	50	-	50	-	100

Key: A: 100g of white rice
B: 100g of white hungry rice
C: 100g of brown hungry rice
D: 20g of white rice: 60g of white hungry rice: 20g of soybeans
E: 20g of white rice: 60g of brown hungry rice: 20g of soybeans
F: 60g of white rice: 20g of white hungry rice: 20g of soybeans
G: 60g of white rice: 20g of brown hungry rice: 20g of soybeans
H: 50g of white rice: 50g of white hungry rice
I: 50 g of white rice: 50 g of brown hungry rice

Preparation of *sinasir*

The preparation of *sinasir* was based on the method described by Chibuzo *et al.* (1996), with modifications to the ingredient proportions used in the present study, as presented in Table 1. For the rice-based control, 100 g of cleaned white rice was soaked in distilled water for 24 h at room temperature. A portion of the soaked rice was cooked until soft and subsequently combined with the remaining uncooked rice before wet milling to obtain a homogeneous batter. The batter was covered and allowed to ferment for 6 h in a warm environment. Following fermentation, the batter was thoroughly remixed and supplemented with salt, sugar and potash. The batter was subsequently cooked in a non-stick pan lightly coated with groundnut oil over low heat for approximately 3 min. The same general procedure was used for the remaining formulations, with the cereal and soybean proportions adjusted according to the formulation design.

The use of fermentation and heat treatment is relevant to the nutritional characteristics of the resulting products because processing can alter protein structure, amino-acid availability and carbohydrate accessibility. Recent studies have demonstrated that processing conditions can influence both amino-acid composition and *In-Vitro* protein digestibility of plant-based foods (Adeleye *et al.,* 2020).

Determination of Amino-Acid Composition

The amino acid profile in the known *sinasir* was measured. The known *sinasir* was dried to constant weight, defatted, hydrolyzed and evaporated in a rotary evaporator and load into the Technicon sequential Multi-*Sinasir* Amino Analyzer (TSM).

Defatting *Sinasir*

The *sinasir* was defatted using chloroform/methanol mixture of ratio 2:1. The *sinasir* was placed in extraction thimble and extracted for 15 hours in soxhlet extraction apparatus (AOAC, 2017).

Hydrolysis of the *Sinasir*

Acid hydrolysis of the defatted *sinasir* samples was performed using a hydrochloric acid hydrolysis procedure comparable to that described by Lucas and Sotelo (1982), with modifications. A known weight of the defatted *sinasir* was weighed into glass ampoule. 7 ml of 6 NHCl was added and oxygen was expelled by passing nitrogen into the ampoule (this is to avoid possible oxidation of some amino acids during hydrolysis e.g., methionine and cystine). The glass ampoule was scaled with Bunsen burner flame and put in an oven preset at 105^0C ($\pm$ 5^0 C)

for 22 hours. The ampoule was allowed to cool before broken open at the tip and the content was filtered to remove the humins. The filtrate was evaporated to dryness in hot air oven. The residue was dissolved with 5ml of acetate buffer (pH 2.0) and stored in plastic specimen bottles, which was kept in the freezer.

Loading of the Hydrolysate into TSM Analyzer

The amount loaded was between 5 to10 microliter. It was dispended into the cartridge of the analyzer. The TSM analyzer is designed to separate and analyzed free acidic, neutral and basic amino acids of the hydrolysate. The period of analysis lasted for 76 minutes.

Method of Calculating Amino Acid Values from the Chromatogram Peaks

The Amino Acid Values from the Chromatogram Peaks was calculated as described by Spackman *et al.* (1958). An integrator attached to the analyzer calculates the peak proportional to the concentration of each of the amino acids. Alternatively, the net height of each peak produced by the chart recorder of TSM (each representing and amino) was measured. Approximately area of each peak was obtained by multiplying the height with the width at half-height.

The norleucine equivalent (NE) for each amino acid in the standard mixture was calculated using the formula;

$$\mathrm{NE} = \frac{Area\ of\ Norleucine\ Peak}{Area\ of\ each\ Acid}$$

A constant S was calculated for esach amino acid in the standard mixture: where $S_{std} = NE_{stdx}$ Molecular weight xμ MAA_{std}. Finally, the amount of each amino acid present in the *sinasir* was calculated in g/16gNofg/100g protein using the following formula:

$$Concentration\ (g/100\ g\ protein) = \frac{NH\ x\ W\ @\ NH/2xSstdxC\ Dilution \times 16 \div NH \times W\ (Nleu)}{Sinasir\ Wt\ (g) \times N\% \times 10 \times Vol.Loaded}$$

Where: C = concentration, NH = net height, W = weight at half height, Nleu = norleucine

***In-Vitro* carbohydrate digestibility**

The method adopted by Laminu *et al.,* (2014) was used for in vitro carbohydrate digestibility. Into four sets of test tubes labelled as I, II, III and IV (0,30, 60 min and 6 h),1 ml of αamylase, 4 ml of phosphate buffer and 1 ml of sodium chloride was added and allowed to equilibrate at room temperature. After equilibration, 5 ml of 1% *sinasir* was added and mixed thoroughly. At intervals of 10, 30 and 60 min, 3 drops of Lugol‟s iodine was introduced into the reaction mixture and absorbance of starch-iodine complex was measured calorimetrically at 620 nm with Corning colorimeter 253 long sides the blank (Lugol‟s iodine). Unit of amylase activity was calculated as:

$$\textit{In-Vitro carbohydrate digestibility (\%)} = \frac{\mathrm{Ao}-\mathrm{At}}{\mathrm{Ao}} \times 100$$

Where: Ao = Absorbance of iodine-starch complex at zero time, At = Absorbance of iodine-starch at specified time intervals. Specific activity of amylase is expressed in unit /mg protein/minute.

***In-Vitro* protein digestibility**

In-vitro protein digestibility was determined using a trypsin-based method adapted from Naeem *et al.* (2022), with modifications where applicable. Briefly, 1 mL of 11% trypsin was mixed with 4 mL of phosphate buffer (pH 7.5) and 1 mL of 0.1 N hydrochloric acid. After equilibration, 1 mL of the 1% *sinasir* suspension was added. Digestion was assessed after 1 and 6 h. The reaction was terminated by adding neutralized formalin and the mixture was filtered. The nitrogen content of the undigested residue was determined using the Kjeldahl method. In-vitro protein digestibility was calculated as:

$$\%\ \textit{In vitro protein digestibility} = \frac{CP1-CP2}{CP1} \times 100$$

Where CP_1 represents the total protein content of the undigested *sinasir* sample and CP_2 represents the protein remaining after trypsin digestion.

Statistical analysis

All analytical determinations were performed in triplicate and results were expressed as mean ± standard error of mean (SEM). Differences among the *sinasir* formulations were evaluated using one-way analysis of variance (ANOVA). Data were analyzed by one-way analysis of variance (ANOVA). Where the ANOVA was significant, means were compared using Duncan multiple range test at $p < 0.05$.

RESULTS

Amino Acid Profile of the *Sinasir* Produced

Table 2 presents the essential and non-essential amino acid profiles of the nine *sinasir* formulations prepared from white rice, white hungry rice, brown hungry rice and soybean. The results revealed variations in amino acid concentrations among the formulations, with the magnitude of these differences depending on the amino acid and ingredient composition. Formulation C, prepared entirely from brown hungry rice, recorded the highest values for several essential amino acids, while formulations E and I also demonstrated comparatively high concentrations of selected amino acids.

Essential amino acids

Tryptophan concentrations ranged from 1.15 ± 0.01 g /100 g protein in formulation A (white rice control) to 5.15 ± 0.02 g /100 g protein in formulation C (100% brown hungry rice). Formulation I, comprising equal proportions of white rice and brown hungry rice, recorded 4.22 ± 0.01 g /100 g protein, followed by formulation H, containing equal proportions of white rice and white hungry rice, at 4.16 ± 0.01 g /100 g protein. Formulations D, E, F and G recorded 3.76 ± 0.02, 2.98 ± 0.01, 2.05 ± 0.01 and 2.63 ± 0.02 g /100 g protein, respectively, while formulation B contained 1.53 ± 0.01 g. Histidine concentrations ranged from 0.83 ± 0.01 g in formulation A to 3.23 ± 0.01 g in formulation C. Formulation E also recorded a relatively high concentration of 3.18 ± 0.01 g /100 g protein, whereas formulations D and I contained 1.66 ± 0.01 and 1.47 ± 0.01 g /100 g protein, respectively. Lower values were observed in formulations F (1.33 ± 0.01 g /100 g protein), H (1.31 ± 0.01 g /100 g protein) and G (1.12 ± 0.01 g /100 g protein). Formulation B recorded 0.85 ± 0.01 g /100 g protein, which was comparable to the control according to the superscript groupings.

Leucine concentrations varied from 1.56 ± 0.01 g /100 g protein in formulation A to 4.66 ± 0.01 g /100 g protein in formulation E. Formulation I recorded 4.65 ± 0.01 g /100 g protein, closely followed by formulation C at 4.42 ± 0.01 g. Formulations H and F contained 3.65 ± 0.01 and 2.84 ± 0.01 g /100 g protein, respectively, while formulations G, D and B recorded 2.25 ± 0.00, 1.95 ± 0.00 and 1.74 ± 0.01 g /100 g protein, respectively. For isoleucine, the highest concentration was recorded in formulation H (4.03 ± 0.01 g /100 g protein), followed by formulation I (4.02 ± 0.01 g /100 g protein) and formulation D (4.00 ± 0.00 g /100 g protein /100 g protein). Formulation C contained 3.96 ± 0.01 g, while formulation E recorded 2.55 ± 0.01 g /100 g protein. Formulations B and F each contained 2.01 ± 0.01 g, whereas the lowest concentration was observed in formulation A (1.65 ± 0.01 g /100 g protein). The superscript groupings indicate that some formulations with numerically different values were not significantly different from one another.

Phenylalanine was comparatively abundant across all nine formulations, with values ranging from 5.88 ± 0.01 g /100 g protein in formulation A to 9.64 ± 0.01 g /100 g protein in formulation E. Formulations I and H recorded 8.93 ± 0.01 and 8.92 ± 0.01 g /100 g protein, respectively, while formulation C contained 8.13 ± 0.01 g /100 g protein. Formulations D and G recorded 7.77 ± 0.01 and 7.33 ± 0.01 g /100 g protein, respectively. Lower concentrations were observed in formulations F (6.74 ± 0.04 g /100 g protein) and B (6.24 ± 0.01 g /100 g protein). Valine concentrations ranged from 2.72 ± 0.01 g in formulation A to 4.98 ± 0.01 g in formulation E. Formulation C recorded 4.33 ± 0.01 g /100 g protein, followed by formulation I at 4.16 ± 0.01 g /100 g protein. Formulations D, F, H and G contained 3.94 ± 0.01, 3.86 ± 0.01, 3.86 ± 0.01 and 3.84 ± 0.01 g /100 g protein, respectively. Formulation B recorded 2.75 ± 0.00 g /100 g protein /100 g protein.

Lysine concentrations varied from 2.74 ± 0.00 g /100 g protein /100 g protein in formulation F to 5.18 ± 0.76 g /100 g protein in formulation I. Formulation C recorded 5.16 ± 0.01 g /100 g protein, while formulations H and D contained

4.25 ± 0.01 and 4.22 ± 0.01 g, respectively. Formulation G recorded 4.01 ± 0.01 g /100 g protein, followed by formulation B (3.86 ± 0.01 g /100 g protein), formulation A (3.85 ± 0.01 g /100 g protein) and formulation E (3.71 ± 0.01 g /100 g protein). Although formulation I had the highest reported mean, its comparatively large standard error warrants verification against the original laboratory results. Methionine concentrations ranged from 2.65 ± 0.01 g /100 g protein in formulation A to 7.37 ± 0.26 g /100 g protein in formulation C. Formulation I recorded 5.85 ± 0.01 g, followed by formulation H at 5.23 ± 0.01 g /100 g protein. Formulations E and G each contained 3.77 ± 0.01 g /100 g protein, while formulations F and D recorded 3.71 ± 0.01 and 3.65 ± 0.01 g /100 g protein, respectively. Formulation B contained 2.71 ± 0.01 g /100 g protein. Threonine concentrations ranged from 1.51 ± 0.01 g /100 g protein in formulation A to 4.44 ± 0.01 g in formulation C. Formulation I recorded 4.16 ± 0.01 g, while formulation H contained 3.85 ± 0.01 g /100 g protein. Formulations E, F and D recorded 3.61 ± 0.01, 2.71 ± 0.01 and 2.56 ± 0.01 g, respectively. The lowest concentrations after the control were observed in formulations G (1.86 ± 0.01 g /100 g protein) and B (1.53 ± 0.01 g /100 g protein).

Non-essential amino acids

Asparagine concentrations ranged from 3.60 ± 0.00 g /100 g protein /100 g protein in formulation A to 6.52 ± 0.01 g /100 g protein in formulation I. Formulation H recorded 6.33 ± 0.01 g /100 g protein, followed by formulation E at 5.49 ± 0.03 g /100 g protein and formulation C at 4.96 ± 0.01 g /100 g protein. Formulations F, D and G contained 4.61 ± 0.01, 4.35 ± 0.00 and 4.23 ± 0.01 g /100 g protein, respectively, while formulation B recorded 3.61 ± 0.01 g. Arginine concentrations ranged from 2.73 ± 0.01 g /100 g protein in formulation A to 4.56 ± 0.01 g /100 g protein in formulation E. Formulations D and F recorded 4.00 ± 0.00 and 3.93 ± 0.01 g, respectively, while formulation I contained 3.81 ± 0.01 g. Formulation C recorded 3.77 ± 0.01 g /100 g protein, followed by formulation H at 3.44 ± 0.01 g and formulation G at 3.12 ± 0.01 g /100 g protein. Formulation B contained 2.80 ± 0.01 g /100 g protein.

Alanine concentrations varied from 0.55 ± 0.00 g /100 g protein /100 g protein in formulation A to 2.96 ± 0.01 g /100 g protein in formulation C. Formulation E also recorded a high concentration of 2.83 ± 0.01 g /100 g protein, while formulations F and D contained 1.53 ± 0.01 and 1.44 ± 0.01 g /100 g protein, respectively. Formulation I recorded 1.37 ± 0.01 g, followed by formulation H at 0.97 ± 0.01 g /100 g protein and formulation G at 0.70 ± 0.00 g /100 g protein /100 g protein. Formulation B contained 0.57 ± 0.01 g /100 g protein. Aspartate concentrations ranged from 3.21 ± 0.01 g in formulation A to 5.92 ± 0.01 g /100 g protein in formulation I. Formulation H recorded 5.82 ± 0.01 g, followed by formulation C at 5.65 ± 0.01 g /100 g protein. Formulations F and E contained 5.14 ± 0.01 and 5.12 ± 0.01 g /100 g protein /100 g protein, respectively, while formulation G recorded 4.61 ± 0.01 g /100 g protein /100 g protein. Formulation D contained 4.11 ± 0.01 g /100 g protein /100 g protein and formulation B recorded 3.22 ± 0.01 g /100 g protein /100 g protein.

Glutamate concentrations ranged from 1.44 ± 0.01 g /100 g protein in formulation A to 4.15 ± 0.00 g /100 g protein in both formulations H and I. Formulation C recorded 3.71 ± 0.01 g /100 g protein, followed by formulation E at 3.23 ± 0.01 g /100 g protein and formulation G at 2.72 ± 0.01 g /100 g protein. Formulation D contained 2.43 ± 0.01 g /100 g protein, while formulations B and F recorded 1.76 ± 0.01 and 1.71 ± 0.01 g /100 g protein, respectively. Glycine concentrations varied from 3.65 ± 0.00 g /100 g protein in formulation A to 8.35 ± 0.00 g /100 g protein in formulation I. Formulation H recorded 7.34 ± 0.01 g /100 g protein, followed by formulation C at 7.15 ± 0.01 g /100 g protein and formulation D at 6.85 ± 0.00 g /100 g protein. Formulation G contained 6.51 ± 0.01 g /100 g protein, while formulation F recorded 5.82 ± 0.01 g /100 g protein. Formulations B and E contained 4.57 ± 0.01 and 3.75 ± 0.01 g /100 g protein, respectively. Tyrosine concentrations showed substantial variation, ranging from 3.15 ± 0.01 g /100 g protein in formulation G to 11.12 ± 0.01 g /100 g protein in formulation I. Formulation C recorded 6.46 ± 0.01 g /100 g protein, followed by formulation D at 5.81 ± 0.01 g /100 g protein and formulation E at 5.66 ± 0.01 g /100 g protein. Formulation F contained 4.62 ± 0.01 g /100 g protein, while formulations B, H and A recorded

4.43 ± 0.01, 4.11 ± 0.01 and 4.10 ± 0.00 g /100 g protein, respectively.

Cysteine concentrations ranged from 2.12 ± 0.01 g /100 g protein in formulation F to 6.24 ± 0.01 g /100 g protein in formulation I. Formulation H recorded 6.05 ± 0.01 g /100 g protein, followed by formulation C at 4.41 ± 0.01 g /100 g protein and formulation D at 3.94 ± 0.01 g /100 g protein. Formulation G contained 3.41 ± 0.01 g /100 g protein, while formulations B and E each recorded 3.14 ± 0.01 g /100 g protein. Formulation A contained 3.00 ± 0.00 g /100 g protein. Proline concentrations ranged from 1.70 ± 0.00 g /100 g protein /100 g protein in formulation A to 3.80 ± 0.00 g /100 g protein in formulation E. Formulation I recorded 3.30 ± 0.00 g /100 g protein, followed by formulation H at 2.28 ± 0.01 g /100 g protein and formulation C at 2.26 ± 0.01 g /100 g protein. Formulation D contained 2.00 ± 0.00 g /100 g protein, while formulations G and F recorded 1.82 ± 0.01 and 1.74 ± 0.00 g /100 g protein, respectively. Formulation B contained 1.71 ± 0.01 g /100 g protein. Serine concentrations ranged from 2.05 ± 0.01 g /100 g protein in formulation F to 4.92 ± 0.01 g /100 g protein in formulation I. Formulation H recorded 4.83 ± 0.01 g /100 g protein, followed by formulation D at 3.76 ± 0.01 g /100 g protein and formulation C at 3.23 ± 0.01 g /100 g protein. Formulation G contained 3.12 ± 0.01 g /100 g protein, while formulation E recorded 2.98 ± 0.01 g /100 g protein. Formulations B and A contained 2.75 ± 0.01 and 2.60 ± 0.00 g /100 g protein, respectively.

Table 2. Amino Acid Profile of the *Sinasir* Produced (g/100 g protein)

Amino Acids	A	B	C	D	E	F	G	H	I
Tryptophan	1.15±0.01^{i}	1.53±0.01^{h}	5.15±0.02^{a}	3.76±0.02^{d}	2.98±0.01^{e}	2.05±0.01^{g}	2.63±0.02^{f}	4.16±0.01^{c}	4.22±0.01^{b}
Histidine	0.83±0.01^{g}	0.85±0.01^{g}	3.23±0.01^{a}	1.66±0.01^{c}	3.18±0.01^{b}	1.33±0.01^{e}	1.12±0.01^{f}	1.31±0.01^{e}	1.47±0.01^{d}
Leucine	1.56±0.01^{h}	1.74±0.01^{g}	4.42±0.01^{b}	1.95±0.00^{f}	4.66±0.01^{a}	2.84±0.01^{d}	2.25±0.00^{e}	3.65±0.01^{c}	4.65±0.01^{a}
Isoleucine	1.65±0.01^{g}	2.01±0.01^{f}	3.96±0.01^{c}	4.00±0.00^{b}	2.55±0.01^{e}	2.01±0.01^{f}	2.84±0.01^{d}	4.03±0.01^{a}	4.02±0.01^{ab}
Phenylalanine	5.88±0.01^{h}	6.24±0.01^{g}	8.13±0.01^{c}	7.77±0.01^{d}	9.64±0.01^{a}	6.74±0.04^{f}	7.33±0.01^{e}	8.92±0.01^{b}	8.93±0.01^{b}
Valine	2.72±0.01^{h}	2.75±0.00^{g}	4.33±0.01^{b}	3.94±0.01^{d}	4.98±0.01^{a}	3.86±0.01^{e}	3.84±0.01^{f}	3.86±0.01^{e}	4.16±0.01^{c}
Lysine	3.85±0.01^{b}	3.86±0.01^{b}	5.16±0.01^{a}	4.22±0.01^{b}	3.71±0.01^{b}	2.74±0.00^{c}	4.01±0.01^{b}	4.25±0.01^{b}	5.18±0.76^{a}
Methionine	2.65±0.01^{e}	2.71±0.01^{e}	7.37±0.26^{a}	3.65±0.01^{d}	3.77±0.01^{d}	3.71±0.01^{d}	3.77±0.01^{d}	5.23±0.01^{c}	5.85±0.01^{b}
Threonine	1.51±0.01^{h}	1.53±0.01^{h}	4.44±0.01^{a}	2.56±0.01^{f}	3.61±0.01^{d}	2.71±0.01 e	1.86±0.01^{g}	3.85±0.01^{c}	4.16±0.01^{b}
Asparagine	3.60±0.00^{h}	3.61±0.01^{h}	4.96±0.01^{d}	4.35±0.00^{f}	5.49±0.03^{c}	4.61±0.01^{e}	4.23±0.01^{g}	6.33±0.01^{b}	6.52±0.01^{a}
Arginine	2.73±0.01^{h}	2.80±0.01^{h}	3.77±0.01^{e}	4.00±0.00^{b}	4.56±0.01^{a}	3.93±0.01^{c}	3.12±0.01^{g}	3.44±0.01^{f}	3.81±0.01^{d}
Alanine	0.55±0.00^{h}	0.57±0.01^{h}	2.96±0.01^{a}	1.44±0.01^{d}	2.83±0.01^{b}	1.53±0.01^{c}	0.70±0.00^{g}	0.97±0.01^{f}	1.37±0.01^{e}
Aspartate	3.21±0.01^{g}	3.22±0.01^{g}	5.65±0.01^{c}	4.11±0.01^{f}	5.12±0.01^{d}	5.14±0.01^{d}	4.61±0.01^{e}	5.82±0.01^{b}	5.92±0.01^{a}
Glutamate	1.44±0.01^{h}	1.76±0.01^{f}	3.71±0.01^{b}	2.43±0.01^{e}	3.23±0.01^{c}	1.71±0.01^{g}	2.72±0.01^{d}	4.15±0.00^{e}	4.15±0.00^{e}
Glycine	3.65±0.00^{i}	4.57±0.01^{g}	7.15±0.01^{c}	6.85±0.00^{d}	3.75±0.01^{h}	5.82±0.01^{f}	6.51±0.01^{e}	7.34±0.01^{b}	8.35±0.00^{a}
Tyrosine	4.10±0.00^{g}	4.43±0.01^{f}	6.46±0.01^{b}	5.81±0.01^{c}	5.66±0.01^{d}	4.62±0.01^{e}	3.15±0.01^{h}	4.11±0.01^{g}	11.12±0.01^{a}
Cysteine	3.00±0.00^{g}	3.14±0.01^{f}	4.41±0.01^{c}	3.94±0.01^{d}	3.14±0.01^{f}	2.12±0.01^{h}	3.41±0.01^{e}	6.05±0.01^{b}	6.24±0.01^{a}
Proline	1.70±0.00^{h}	1.71±0.01^{h}	2.26±0.01^{d}	2.00±0.00^{e}	3.80±0.00^{a}	1.74±0.00^{g}	1.82±0.01^{f}	2.28±0.01^{c}	3.30±0.00^{b}
Serine	2.60±0.00^{h}	2.75±0.01^{g}	3.23±0.01^{d}	3.76±0.01^{c}	2.98±0.01^{f}	2.05±0.01^{i}	3.12±0.01^{e}	4.83±0.01^{b}	4.92±0.01^{a}

Values are presented as mean ± SEM, n=3. Values with different superscripts along the column are significantly different (P<0.05)

Key: A: 100g of white rice

B: 100g of white hungry rice

C: 100g of brown hungry rice

D: 20g of white rice: 60g of white hungry rice: 20g of soybeans

E: 20g of white rice: 60g of brown hungry rice: 20g of soybeans

F: 60g of white rice: 20g of white hungry rice: 20g of soybeans

G: 60g of white rice: 20g of brown hungry rice: 20g of soybeans

H: 50g of white rice: 50g of white hungry rice

I: 50 g of white rice: 50 g of brown hungry rice.

***In-Vitro* Carbohydrate Digestibility**

Table 3 presents the in-vitro starch digestibility of the nine *sinasir* formulations. The results showed variation in starch digestibility across the formulations, with values ranging from 70.42 ± 0.01% to 80.24 ± 0.01%. Formulation A, prepared exclusively from white rice, recorded the highest digestibility (80.24 ± 0.01%), followed by formulation C, made entirely from brown hungry rice, at 78.84 ± 0.01%. Formulation I, containing equal proportions of white rice and brown hungry rice, also exhibited relatively high starch digestibility (76.99 ± 0.00%).

The white hungry rice formulation (B) had a digestibility value of 72.36 ± 0.01%. Among the composite formulations, formulation E, comprising white rice, brown hungry rice and soybean, recorded a value of 72.16 ± 0.01%, followed by formulation F, containing white rice, white hungry rice and soybean, at 72.04 ± 0.01%. Formulation D, which consisted of white rice, white hungry rice and soybean in proportions of 20:60:20, had a digestibility value of 71.15 ± 0.01%. Formulation H, containing equal proportions of white rice and white hungry rice, recorded 70.84 ± 0.01%, while formulation G, comprising white rice, brown hungry rice and soybean in proportions of 60:20:20, had the lowest value (70.42 ± 0.01%) (Table 3).

Table 3. *In-Vitro* Carbohydrate Digestibility of the *Sinasir* Produced

Sinasir	Starch digestibility
A	80.24±0.01[a]
B	72.36±0.01[d]
C	78.84±0.01[b]
D	71.15±0.01[g]
E	72.16±0.01[e]
F	72.04±0.01[f]
G	70.42±0.01[i]
H	70.84±0.01[h]
I	76.99±0.00[c]

Values are presented as mean ± SEM, n=3. Values with different superscripts along the column are significantly different (P<0.05).
Key: A:100g of white rice
B:100g of white hungry rice
C:100g of brown hungry rice
D: 20g of white rice: 60g of white hungry rice: 20g of soybeans
E:20g of white rice: 60g of brown hungry rice: 20g of soybeans
F:60g of white rice: 20g of white hungry rice: 20g of soybeans
G:60g of white rice: 20g of brown hungry rice: 20g of soybeans
H:50g of white rice: 50g of white hungry rice
I: 50 g of white rice: 50 g of brown hungry rice.

***In-Vitro* Protein Digestibility**

Table 4 presents the in-vitro protein digestibility of the nine *sinasir* formulations after 1 and 6 hours of enzymatic digestion. At 1 hour, protein digestibility ranged from 63.58 ± 0.09% to 78.91 ± 0.01%. Formulation G, containing 60 g white rice, 20 g brown hungry rice and 20 g soybean, recorded the highest digestibility (78.91 ± 0.01%), followed closely by formulation I (78.75 ± 0.01%), comprising 50 g white rice and 50 g brown hungry rice. Formulation H, containing equal proportions of white rice and white hungry

rice, recorded 77.92 ± 0.01%, while formulation E, consisting of 20 g white rice, 60 g brown hungry rice and 20 g soybean, had a value of 76.85 ± 0.01%. Formulation C, prepared entirely from brown hungry rice, recorded 75.42 ± 0.01%. Formulations B and D had similar digestibility values of 73.82 ± 0.67% and 73.85 ± 0.00%, respectively. Formulation F recorded 72.53 ± 0.01%, while the white-rice control (A) had the lowest digestibility at 63.58 ± 0.09%.

After 6 hours of digestion, protein digestibility increased across all nine formulations, with values ranging from 66.94 ± 0.01% to 82.62 ± 0.01%. Formulation G again recorded the highest value (82.62 ± 0.01%), followed by formulation H (81.77 ± 0.01%). Formulations E and I both recorded 80.16 ± 0.01%, while formulation C had a value of 79.12 ± 0.01%. Formulation D recorded 78.04 ± 0.01%, whereas formulations B and F had digestibility values of 76.44 ± 0.01% and 76.22 ± 0.01%, respectively. The white-rice control (A) remained the lowest, with a value of 66.94 ± 0.01%. Formulation G exhibited the highest in-vitro protein digestibility at both 1 and 6 hours, whereas the white-rice control recorded the lowest values at both digestion periods. The increase in digestibility values between 1 and 6 hours across all formulations indicates greater protein digestion with longer exposure to the enzyme under the experimental conditions. According to the superscript letters presented in Table 4, formulations G and I were not significantly different at 1 hour, while formulations E and I were not significantly different at 6 hours ($p > 0.05$).

Table 0. *In-Vitro* Protein Digestibility of the *Sinasir* Produced

Sinasir	Protein digestibility @ 1hr	Protein digestibility @ 6hrs
A	63.58±0.09^{g}	66.94±0.01^{h}
B	73.82±0.67^{e}	76.44±0.01^{f}
C	75.42±0.01^{d}	79.12±0.01^{d}
D	73.85±0.00^{e}	78.04±0.01^{e}
E	76.85±0.01^{c}	80.16±0.01^{c}
F	72.53±0.01^{f}	76.22±0.01^{g}
G	78.91±0.01^{a}	82.62±0.01^{a}
H	77.92±0.01^{b}	81.77±0.01^{b}
I	78.75±0.01^{a}	80.16±0.01^{c}

Values are presented as mean ± SEM, n=3. Values with different superscripts along the column are significantly different ($P<0.05$).

Key: A:100g of white rice
B:100g of white hungry rice
C:100g of brown hungry rice
D: 20g of white rice: 60g of white hungry rice: 20g of soybeans
E:20g of white rice: 60g of brown hungry rice: 20g of soybeans
F:60g of white rice: 20g of white hungry rice: 20g of soybeans
G:60g of white rice: 20g of brown hungry rice: 20g of soybeans
H:50g of white rice: 50g of white hungry rice
I: 50 G OF WHITE RICE: 50 G OF BROWN HUNGRY RICE

DISCUSSION

Amino Acid Profile of the *Sinasir*

The amino acid composition of the *sinasir* formulations demonstrates that the choice and proportion of cereal ingredients, as well as their combination with soybean, can influence the amino acid characteristics of the final product. However, the effects were not uniform across all formulations, indicating that the nutritional contribution of an ingredient depends partly on the other components with which it is combined. This finding supports the importance of

evaluating the amino acid composition of formulated foods rather than assuming that the nutritional characteristics of individual ingredients will be retained unchanged in the finished product.

The differences observed between the white and brown hungry rice formulations suggest that the form of hungry rice used may be an important consideration in product development. Differences in grain composition, processing characteristics and the proportions of constituent nutrients could contribute to variation in the amino acid profiles of the resulting products. Research on fonio has documented variation in its nutritional and physicochemical characteristics, reinforcing the importance of considering grain type and processing history when developing fonio-based foods (Bassey *et al.,* 2023; Jideani and Jideani, 2011).

The inclusion of soybean provides a further explanation for the variation among the composite formulations. Cereal grains and legumes differ in their protein composition and amino acid distribution; consequently, combining them can modify the overall amino acid profile of a food. Soybean is particularly relevant in cereal-based formulations because its protein composition differs from that of rice and hungry rice. Nevertheless, the present findings indicate that the effect of soybean incorporation depends on the proportions of the ingredients used. The results should therefore not be interpreted as evidence that every soybean-containing formulation necessarily has a superior amino acid profile to every formulation prepared without soybean.

This interpretation is consistent with Malomo and Abiose (2020), who investigated the effects of substituting maize with acha (*Digitaria exilis*) and soybean in masa and reported changes in the chemical composition of the resulting products. Although masa and *sinasir* differ in their preparation and formulation, both studies illustrate the potential of incorporating indigenous cereals and legumes into traditional cereal-based foods. The comparison also highlights the importance of evaluating the specific food matrix rather than generalizing the nutritional effects of ingredient combinations across different products. The amino acid findings also require careful interpretation in relation to protein quality. The presence of indispensable amino acids is nutritionally important, but their measured concentrations alone do not establish whether a food provides an adequate balance of these amino acids relative to human requirements. Such an assessment requires a clearly defined reporting basis, comparison with an appropriate reference amino acid pattern and consideration of the digestibility of individual indispensable amino acids. Gorissen *et al.* (2018) demonstrated that the protein content and amino acid composition of plant-based protein ingredients vary considerably, underscoring the need for product-specific evaluation.

Furthermore, the measured amino acid profile should be interpreted in the context of the analytical procedure. Conventional acid hydrolysis can cause the loss of certain amino acids, particularly tryptophan, while asparagine and glutamine may be converted to their corresponding acidic forms. Accordingly, the determination of tryptophan generally requires an appropriate separate analytical procedure, while the reported aspartate and glutamate values may represent the combined contributions of their respective amino acids and amides. These methodological considerations are important when interpreting individual amino acid measurements and comparing the findings with those of other studies. The amino acid profile suggests that hungry rice, particularly when combined with other cereal ingredients and, where appropriate, soybean, offers opportunities for diversifying the composition of traditional *sinasir*. However, identifying the formulation with the best protein quality requires more than comparing individual amino acid concentrations. Further assessment should establish the indispensable amino acid pattern relative to an appropriate reference standard and determine the extent to which those amino acids remain available after digestion.

***In-Vitro* Carbohydrate Digestibility**

The variation in starch digestibility among the formulations indicates that replacing white rice with hungry rice, with or without soybean, altered the susceptibility of starch to enzymatic hydrolysis. The lower hydrolysis observed in several hungry-rice-containing formulations may reflect differences in starch characteristics and

interactions between starch and other components of the food matrix. Such effects are important because starch digestion is governed not only by total carbohydrate concentration but also by starch structure, accessibility of digestive enzymes and interactions with proteins and other food components.

The relatively high digestibility of the brown hungry-rice formulation suggests that the characteristics of the grain itself can influence starch accessibility. Fonio varieties have been shown to possess different nutritional and physicochemical characteristics and processing can further modify their functional properties (Bassey *et al.,* 2023). The present findings therefore support the need to consider grain variety when developing cereal-based traditional foods. The effect of soybean addition was not uniform across the formulations. This may be associated with interactions between soybean components and starch during processing, which can modify the accessibility of starch to hydrolytic enzymes. Consequently, the carbohydrate-digestibility response of the blends cannot be attributed solely to the presence or absence of soybean.

***In-Vitro* Protein Digestibility**

The greater protein digestibility observed in several formulated products compared with the rice control suggests that the cereal–legume combinations provided protein that was more accessible to enzymatic hydrolysis under the conditions used. The result is consistent with the recognised contribution of soybean as a protein-rich ingredient and with evidence that processing can substantially influence soy protein quality. A quantitative review by Gorissen *et al.* (2018) found considerable variation in the digestibility and protein-quality scores of soybean products and showed that processing and product form can influence protein quality.

The differences among formulations further indicate that the response was dependent on the specific combination of ingredients. Formulation G, which combined white rice, brown hungry rice and soybean, showed the most favourable digestibility response, whereas formulations containing different cereal proportions produced lower values. This pattern suggests that the nutritional effect of soybean cannot be considered independently of the cereal matrix. The structural properties of soy-based foods have been shown to influence the accessibility of protein to digestive enzymes, providing a possible explanation for formulation-specific differences (Li *et al.,* 2023). The increase in protein digestibility with longer enzymatic exposure is consistent with progressive hydrolysis of susceptible protein fractions. Processing before digestion may also have facilitated enzyme accessibility by modifying the physical structure of the fermented product. Recent evidence indicates that processing-induced changes in protein conformation, aggregation and food microstructure can substantially influence *In-Vitro* protein digestibility (Li *et al.,* 2023). Therefore, the combined effects of fermentation, heating and ingredient composition are likely to have contributed to the differences observed among the *sinasir* formulations.

The relationship between amino-acid composition and digestibility is particularly important when interpreting these findings. A food may contain appreciable concentrations of essential amino acids but still provide limited nutritional value if those amino acids are poorly digested or absorbed. Conversely, improved digestibility does not necessarily indicate a superior indispensable amino-acid pattern. Contemporary approaches to protein-quality assessment therefore consider both amino-acid composition and digestibility rather than relying on either measure alone (Gorissen *et al.,* 2022). The findings indicate that formulation with hungry rice and soybean can modify the protein and carbohydrate characteristics of *sinasir*. The results are consistent with previous evidence that cereal–legume combinations and processing can alter nutritional quality, while also demonstrating that the magnitude and direction of these effects depend on the specific formulation. Further research using standardized indispensable amino-acid digestibility and protein-quality measures, particularly DIAAS, would provide a stronger basis for determining the nutritional significance of the observed differences.

CONCLUSION

The study demonstrates that incorporation of white and brown hungry rice (*Digitaria exilis*) and soybean into *sinasir* can substantially influence its amino-acid composition and *In-Vitro* carbohydrate and protein digestibility. The

different formulations produced distinct nutritional characteristics, indicating that the choice and proportion of cereal and soybean ingredients are important determinants of the nutritional quality of the final product. The findings further highlight the potential of hungry rice and soybean combinations as alternatives to conventional rice-based *sinasir*. However, the observed differences in *In-Vitro* digestibility should be interpreted as preliminary indicators of nutrient accessibility rather than direct measures of human nutrient utilization. The study supports the use of hungry rice and soybean in the development of nutritionally improved *sinasir* and provides a basis for further investigation using standardized amino-acid digestibility and protein-quality assessments.

CONFLICT OF INTEREST

- None

ACKNOWLEDGMENT

- The authors are grateful to the Department of Biochemistry, Faculty of Life Science, University of Maiduguri, for providing access to laboratory facilities and a conducive research environment.

REFERENCES

Adeleye, O. F., Awodiran, S. T., Ajayi, A. O. and Ogunmoyela, T. F. B. (2020). Effect of high-temperature, short-time cooking conditions on in vitro protein digestibility, enzyme inhibitor activity and amino acid profile of selected legume grains. *Heliyon,* 6(11), e05419. https://doi.org/10.1016/j.heliyon.2020.e05419

AOAC, (2017). *Association of Official Analytical Chemists.* 19th ed. Official methods of analysis. Washington D.C:

Bassey, S. O., Chinma, C. E., Ezeocha, V. C., Adedeji, O. E., Jolayemi, O. S., Alozie-Uwa, U. C., Adie, I. E., Ofem, S. I., Adebo, J. A. and Adebo, O. A. (2023). Nutritional and physicochemical changes in two varieties of fonio (*Digitaria exilis* and Digitaria iburua) during germination. *Heliyon,* 9(6), e17452. https://doi.org/10.1016/j.heliyon.2023.e17452

Chibuzo, E. C., Godowoli, F. A. and Kure, R. J. (1996). Nutrient and nutritional evaluation of sinasin prepared in two ways. *Plant Foods for Human Nutrition,* 49(2), 139–145. (https://doi.org/10.1007/BF01091971?utm_source=chatgpt.com)

Gorissen, S. H. M., Crombag, J. J. R., Senden, J. M. G., Waterval, W. A. H., Bierau, J., Verdijk, L. B. and van Loon, L. J. C. (2018). Protein content and amino acid composition of commercially available plant-based protein isolates. *Amino Acids, 50*(12), 1685–1695. https://doi.org/10.1007/s00726-018-2640-5

Ibrahim Bio Yerima, A. R., Oselebe, H., Nnamani, C. V., Ifekwe, C., Adje, C. O. A., Kwon-Ndung, E. H., Afiukwa, C. A., Uyoh, E. A., Abdul, S. D., Opaluwa, H. I., Dachi, S. and Achigan-Dako, E. G. (2024). Stakeholders' perceptions of and preferences for utilizing fonio (*Digitaria exilis*) to enrich local diets for food and nutritional security in Nigeria. *Genetic Resources and Crop Evolution,* 71, 999–1011. https://doi.org/10.1007/s10722-023-01837-9

Jideani, I. A. and Jideani, V. A. (2011). Developments on the cereal grains *Digitaria exilis* (hungry rice) and Digitaria iburua (iburu). *Journal of Food Science and Technology*, 48(3), 251–259.

Laminu H., Sheriff, M. and Alhaji, M. A. (2014). Evaluation of the chemical composition, anti-nutrients and mineral element level of a composite meal from pearl millet, wheat, cowpea and groundnut. *Sky Journal of Food Science,* 3(6), 61-70.

Li, M., Wang, J., Zhang, J., Lv, Y., Guo, S. and Van der Meeren, P. (2023). In vitro protein digestibility of different soy-based products: Effects of microstructure, physico-chemical properties and protein aggregation. *Food and Function*, 14(24), 10964–10976. https://doi.org/10.1039/D3FO02410E

Lucas, B. and Sotelo, A. (1982). Amino acid determination in pure proteins, foods and feeds using two different acid hydrolysis methods. *Analytical Biochemistry,* 123(2), 349–356. [DOI: 10.1016/0003-

2697(82)90457-2](https://doi.org/10.1016/0003-2697(82)90457-2?utm_source=chatgpt.com)

Malomo, A. and Abiose, S. (2020). The effect of the substitution of acha (*Digitaria exilis*) and soybean on the chemical composition and sensory properties of maize masa. *Croatian Journal of Food Science and Technology,* 12(1), 101–109. https://doi.org/10.17508/CJFST.2020.12.1.13

Naeem, M., Un-Nisa, M., Ahmad, N., Imran, M., Anwar, H. and Manzoor, M. F. (2022). Preparation of weaning foods by replacing plant proteins with egg protein. *Food Science and Technology,* 42, e44920. (https://doi.org/10.1590/fst.44920?utm_source=chatgpt.com)

Singh, J., Dartois, A. and Kaur, L. (2010). Starch digestibility in food matrix: A review. *Trends in Food Science and Technology*, 21(4), 168–180. https://doi.org/10.1016/j.tifs.2009.12.001

Yates, H. S. A., Carter, J. F., Hungerford, N. L. and Fletcher, M. T. (2023). Ion chromatography and ion chromatography/mass spectrometry as a complementary analysis technique for amino acid analysis in food: A review. *Food Chemistry Advances*, 3, 100415. https://doi.org/10.1016/j.focha.2023.100415

Zielińska-Dawidziak, M., Białas, W., Piasecka-Kwiatkowska, D., Staniek, H. and Niedzielski, P. (2023). Digestibility of protein and iron availability from enriched legume sprouts. *Plant Foods for Human Nutrition*, 78, 270–278. https://doi.org/10.1007/s11130-023-01045-x