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Assessment of amino acid profile, anti-nutritional compounds, and sensory properties of optimized locust beans with *Lactiplantibacillus plantarum* SAL-IMIE02 and *Limosilactobacillus fermentum* SAL-IMIE01 as starter cultures

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Abstract

The study aimed to evaluate the amino acid profile, anti-nutritional compounds, and sensory properties of optimized locust beans fermented using *Lactiplantibacillus plantarum* SAL-IMIE02 and *Limosilactobacillus fermentum* SAL-IMIE01 as starter cultures. Amino acid quantification was conducted using an Amino Acid Analyzer (Technicon Instruments Corporation, New York), while the determination of anti-nutritional compounds followed the method described by Ajayi (2011). The condiments were served to a panel of 60 untrained members familiar with the products, who evaluated attributes such as appearance, aroma, texture, and overall acceptability based on a 9-point Hedonic scale (Oke et al., 2023).Overall, there was a significant increase in the concentration of all amino acids—lysine, leucine, isoleucine, methionine, valine, glutamic acid, aspartic acid, and threonine—in locust beans produced through co-fermentation with *Lactiplantibacillus plantarum* SAL-IMIE02 and *Limosilactobacillus fermentum* SAL-IMIE01compared to the commercial sample. Additionally, the analysis of anti-nutritional compounds in the optimized samples and the control group revealed a significant decrease ($p < 0.05$) in Phytate content (from 45.4 ± 0.7 mg/100g to 15.9 ± 0.3 mg/100g), oxalate (from 160.5 ± 1.7 mg/100g to 49.4 ± 0.8 mg/100g), cyanide (from 12.8 ± 0.2 mg/kg to 4.3 ± 0.2 mg/kg), saponin (from 3.89 ± 0.03% to 1.78 ± 0.03%), phenolic compounds (from 4.08 ± 0.03 mg GAE/g to 2.38 ± 0.03 mg GAE/g), and flavonoids (from 3.06 ± 0.04 mg QE/g to 1.87 ± 0.02 mg QE/g). Furthermore, nitrate and tannin contents decreased significantly (from 15.6 ± 0.1 mg/kg to 6.7 ± 0.1 mg/kg; and from 8.18 ± 0.03 mg/g to 3.88 ± 0.03 mg/g, respectively) in the optimized locust beans.Locust beans fermented with Lactiplantibacillus plantarum SAL-IMIE02 and Limosilactobacillus fermentum SAL-IMIE01 received higher scores for color (8.83 ± 0.81), texture (8.33 ± 1.04), taste (7.53 ± 1.17), consistency (7.94 ± 1.30), and overall acceptability (9.31 ± 0.54) compared to the commercial locust beans. The results of this study support the fact that Lactobacillus spp. fermentation significantly enhances the composition and acceptability of fermented foods.

Keywords: Locust Beans; Amino Acid; Composition; Anti-Nutritional Compound; Nutritional Qualities

1. Introduction

Microbial contaminants and nutritional quality are major concerns of fermented locust beans large-scale production and commercialization. However, the use of probiotic lactobacillus as starter culture to play the role of preservation and Enrichment is a promising alternatives. Locust beans, a traditionally fermented African locust beans seeds are rich in

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protein but also contain anti-nutritional factors that greatly affects their nutritional benefits (Koledoye and Akanbi, 2015). It is a popular nutritive condiment that serve a major non-meat protein source, and is referred to as *Iru* in Yoruba, *dawadawa* in Hausa, *ogiri-igala* in Igbo, *kinda* in Sierra-Leone and *Kpalugu* in Ghana, Okpasa in Ivie-Imiegba (Odunfa, 1981), and it is used to prepare stews/soups. Traditionally processing techniques such as boiling up to 24 h, dehulling, steam blanching and fermentations (Christiana and Marcel, 2008) are used to reduce the anti-nutrients in them and improve the nutritional quality as well as organoleptic acceptability. However, the uncontrolled nature of these methods and the reliance on naturally occurring microorganisms lead to variation in the nutritive profile of each batch (Agunwah et al., 2024) necessitating the development of starter cultures and optimized conditions for controlled production.

Lactobacillus probiotics have gained unprecedented attention over the past decade for their ability to produce nutrition-enriched foods alongside their remarkable antimicrobial activity. However, data gaps exist regarding their effect on the nutritive quality of fermented African locust beans, as well as their potential to eliminate or reduce anti-nutritional factors. Lactobacillus are Gram-positive, rod shaped, catalase-negative and non-endospore forming lactic acid bacteria that are widely recognized as probiotic (Ma et al., 2025). They perform fermentative roles in several African fermented foods, characteristically affecting the sensory properties, safety, and shelf life of many fermented foods and beverages (Oguntoyinbo and Narbad, 2015; Adesulu-Dahunsi et al., 2018). This study therefore explore the potential of *L. plantarum strain* SAL-IMIE02 and *L. fermentum strain* SAL-IMIE01 for the controlled production of locust beans. The impact of these strains on the amino acid composition, anti-nutritional compound, and sensory properties of the optimized products were examined.

2. Materials and methods

2.1. Collection of Materials

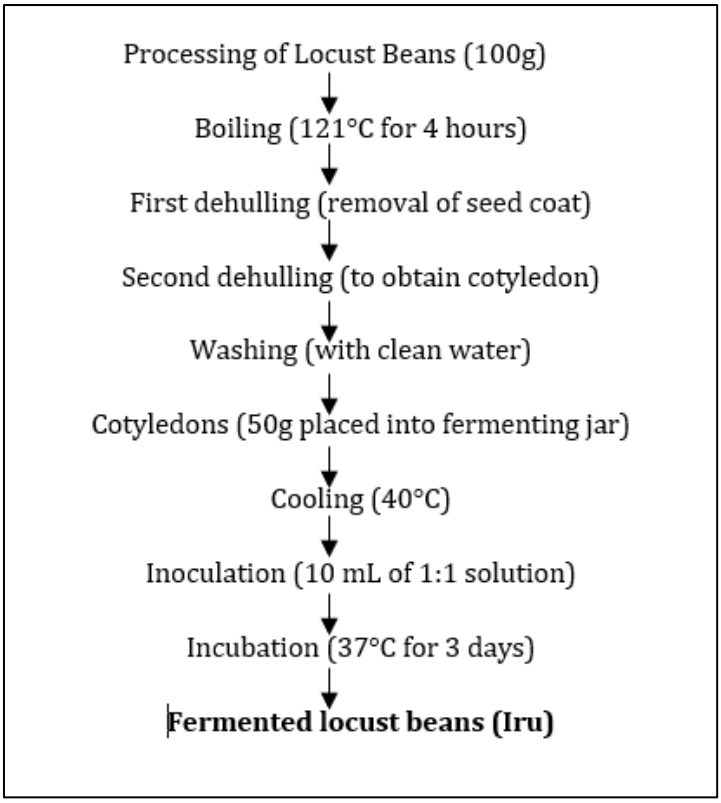
The locust bean seeds and traditionally fermented locust beans were purchased from the Imiegba Main Market, Edo State, Nigeria. The fermented locust beans were used as the **control**, while the locust bean seeds were identified by a botanist in the Department of Science and Biotechnology, Nasarawa State University, Keffi, Nasarawa State**.** *Lactiplantibacillus plantarum* SAL-IMIE02 and *Limosilactobacillus fermentum* SAL-IMIE01, which were previously isolated, characterized, identified, and preserved by Salmat Salisu Musah during her master's research, were used as starter cultures (Musah et al., 2022).

2.2. Preparation of inoculum starter culture

A loopful of an overnight colony each of *L. plantarum strain* SAL-IMIE02 and *L. fermentum strain* SAL-IMIE01 on MRS agar was transferred to 10 mL sterile MRS broth and incubated at 37°C. After, 24 h incubation, 100 µl was transferred into 10 mL MRS broth each and incubated at 37°C for 24 h (overnight). Aliquot of 5 ml of each of the starter cultures was used in the fermentation of the optimized fermented locust beans (Iru) (Akabanda et al., 2014).

2.3. Optimization Production of *Iru*

Hundred grams (100 g) of African locust beans was boiled in a pressure pot at 121°C. After 4 h boiling to soften seed coasts, excess water was decanted before dehulling was carried out. The seed coat was removed by gently smashing the cotyledons with a sterile metal pestle and mortar thereafter, washed with clean water. The cotyledons were then cooked for 1 hour at 121 °C. After. The excess water was drained, and 50 g of the cotyledons was transferred to sterile glass jars and allowed to cool to 40 °C. Thereafter, inoculated with 5 mL of a 1:1 mixture of *L. plantarum strain* SAL-IMIE02 and *L. fermentum strain* SAL-IMIE01 starter cultures (Sample M), and incubated (37 °C; 24hrs). This method was a slight modification of the procedure described by Salmat et al. (2025).



Source: Salmat *et* al. (2025)

Figure 1 Production of fermented locust beans (*Iru*)

2.4. Determination of Amino acid Profile

2.4.1. Sample preparation

The locus beans (sample M and commercial samples) was oven-dried at 60°C for 1 h. The dried samples were pulverized into using a mortar and pestle. The powder was sieve and stored in an airtight containers prior to acid hydrolysis. Briefly, 50 mL of 6 M hydrochloric acid (HCl) was added to 10 g each of the samples in 100 mL hydrolysis tube (Davidson, 2003). The tubes were flushed with nitrogen gas for 1 minute thereafter tightly sealed, and placed in an oven at 110°C. After 24 h incubation, the filtrate was collected using Whatman No. 1 filter paper, and dried under vacuum at 40°C. The dried residue was reconstituted with 0.02 M sodium citrate buffer (pH 2.2) and stored at 4°C for amino acid quantification.

Protein hydrolysates was injected into ion-exchange chromatography (0.50 cm^3/min flow rate; 60°C) of an Amino Acid Analyzer (Technicon Instruments Corporation, New York). The system was calibrated using standard amino acid mixtures with known concentrations. Ninhydrin derivatization was used to measure constituent amino acids at 570 nm (for most amino acids) and 440 nm (for proline). Regression equations obtained from the calibration curves was used to extrapolate concentrations and results expressed as mg amino acid per 100 g dry weight of sample (Oluwaniyi and Bazambo, 2016)).

2.5. Determination of Antinutritional Compounds

2.5.1. Phytate Determination

The phytate content of locust beans was determined using method described by Asunni et al., (2024). Briefly, 4 g sample was soaked in 100 mL 2% HCl, and filtered after 24 h. Thereafter, 25 mL of the mixture was reacted with 5 mL 0.3% ammonium thiocyanate solution. Distilled water (53 mL) was added before titration 0.01 normal (N) ferric chloride ($FeCl_3$) solution. The phytin phosphorus (mg per 100 grams of sample) was calculated using the formula: $Phytin\ phosphorus = Titrate\ value \times 1.19$.

The phytate content (mg per 100 grams of sample) was obtained using the formula:

$$Phytate\ content = Phytin\ phosphorus \times 3.55.$$

2.5.2. Oxalate Determination

The oxalate content was analyzed using the method of Jrand and Underwood (1986). A 1-gram portion of locust beans samples was added to 75 mL of 15% sulfuric acid (H_2SO_4) and stirred intermittently using a magnetic stirrer for 1 h. A 25 mL portion of the filtrate titrated against 0.1 normal (N) potassium permanganate ($KMnO_4$) solution until a faint pink color persisted for 30 seconds. The oxalate content (g per 100 grams of sample) was calculated using the formula:

$$\%\ Oxalate\ =\ Titer\ value\ \times\ 0.0045.$$

2.5.3. Cyanide Determination

The cyanide content of a powdered sample was determined using Bohm and Kocipai (1994) method. A 0.5-gram portion of locust beans samples was boiled 50 mL of distilled water for 30 minutes. The filtrate was heated at 90°C for 5 minutes and its absorbance was measured at 490 nm. The cyanide content (mg per 100 grams of sample) was calculated using a formula:

$$\text{Cyanide content}\ =\ \frac{\text{Absorbance of sample}}{\text{Absorbance of standard}}\ \times \text{Concentration of standard}$$

2.5.4. Saponin Determination

The saponin content was analyzed using the method described by Obadoni and Ochuko (2001). Five grams of locust bean samples were extracted in 100 mL of 20% ethanol and heated in a water bath at 55°C for 4 hours under continuous agitation. The resulting mixture was filtered, and the residue re-extracted with 200 mL of 20% ethanol. The combined filtrates were concentrated to 40 mL using a water bath at 90°C. The concentrate was transferred to a separating funnel, and 20 mL of diethyl ether was added. The ether layer was discarded, and the aqueous layer was further extracted with 60 mL of n-butanol. The butanol extract was washed with 10 mL of 5% sodium chloride (NaCl) solution and then evaporated to dryness. The weight of the residual saponin was recorded, and the percentage saponin content was calculated as:

$$\%\ Saponin\ =\ (Final\ weight\ of\ extract\ /\ Initial\ weight\ of\ sample)\ \times\ 100.$$

Phenolic Compounds Determination

Total phenolic content was determined using the Folin-Ciocalteu method. Locust bean samples (0.5 g) were extracted with 100 mL of 80% methanol, and the filtrate was reacted with Folin-Ciocalteu reagent and 2.5 mL of 7.5% of sodium carbonate. After incubation at 40°C for 30 minutes, absorbance was measured at 765 nm. Phenolic content (mg/100 g sample) was calculated using a gallic acid standard curve.

$$Phenolic\ content\ =\ \frac{Absorbance\ of\ sample}{Absorbance\ of\ standard}\times\ Concentration\ of\ standard.$$

2.5.5. Flavonoid Determination

Flavonoid content was determined using the Boham and Kocipai (1994) method. Locust bean samples (10 g) were extracted with 100 mL 80% methanol for 24 h, and the filtrate was evaporated at 40°C. The weight extract was recorded and the percentage flavonoid content was calculated as:

$$\%\ Flavonoid\ =\ \frac{Final\ weight\ of\ extract}{Initial\ weight\ of\ sample}\times\ 100$$

2.5.6. Nitrate Determination

Nitrate content was determined using the Joslyn (1970) method. Locust bean samples (0.1 g) was added to 10 mL of distilled water boiled for 30 minutes. The filtrate was measured using a UV-Vis spectrophotometer, and the nitrate content (mg per 100 grams of sample) was calculated using the formula:

$$Nitrate\ content\ =\ \frac{Absorbance\ of\ sample}{Absorbance\ of\ standard}\times\ Concentration\ of\ standard.$$

2.5.7. Tannin Determination

Tannin content was determined using the Joslyn (1970) method. Locust bean samples (100 mg) was added to 50 mL of distilled water and boiled for 30 minutes. The mixture final volume was adjusted to 50 mL with distilled water followed by incubation at room temperature for 20 minutes. The absorbance was measured at 760 nm using a UV-Vis spectrophotometer. The tannin content (mg per 100 grams of sample) was determined using the formula:

$$Tannin\ content\ = \frac{Absorbance\ of\ sample}{Absorbance\ of\ standard} \times\ Concentration\ of\ standard.$$

2.5.8. Sensory Evaluation

The condiments were served to a 60-member untrained panel that is familiar with the products to evaluate such attributes as appearance, aroma, texture and overall acceptability based on a 9-point Hedonic scale (Oke *et al.,* 2023).

2.6. Data Analysis

Experiments were done in triplicates. Means and standard deviations were calculated and data subjected to two sample T-test in order to separate their means, using Genstat (17th edition) (Musah *et al.,* 2023)

3. Results

3.1. Amino Acid Composition of Locust Beans (mg/100 g)

Table 1 presents the amino acid composition (mg/100g) of the commercial locust beans (Sample C) and the optimized fermented locust beans (produced by co-fermentation using *L. plantarum strain* SAL-IMIE02 and *L. fermentum strain* SAL-IMIE01. Over all, there is a significant increase in the concentration of all amino acids in locust beans produced by co-fermentation using *L. plantarum strain* SAL-IMIE02 and *L. fermentum strain* SAL-IMIE01 compared to commercial and non-fermented locust beans.

Lysine (5.80 ± 0.10 mg/100g), leucine (10.70 ± 0.15 mg/100g), isoleucine (3.20 ± 0.05^{b}), methionine (3.20 ± 0.05^{b}), valine (8.50 ± 0.10^{b}), and threonine (5.30 ± 0.10^{b}) were significantly higher in the optimized sample. Similarly, non-essential amino acids such as glutamic acid and aspartic acid also increased significantly, from 13.60 ± 0.10 to 16.40 ± 0.20 mg/100g and from 9.50 ± 0.15 to 11.80 ± 0.15 mg/100g, respectively.

3.2. Determination of Antinutritional Compounds in Sample C and M

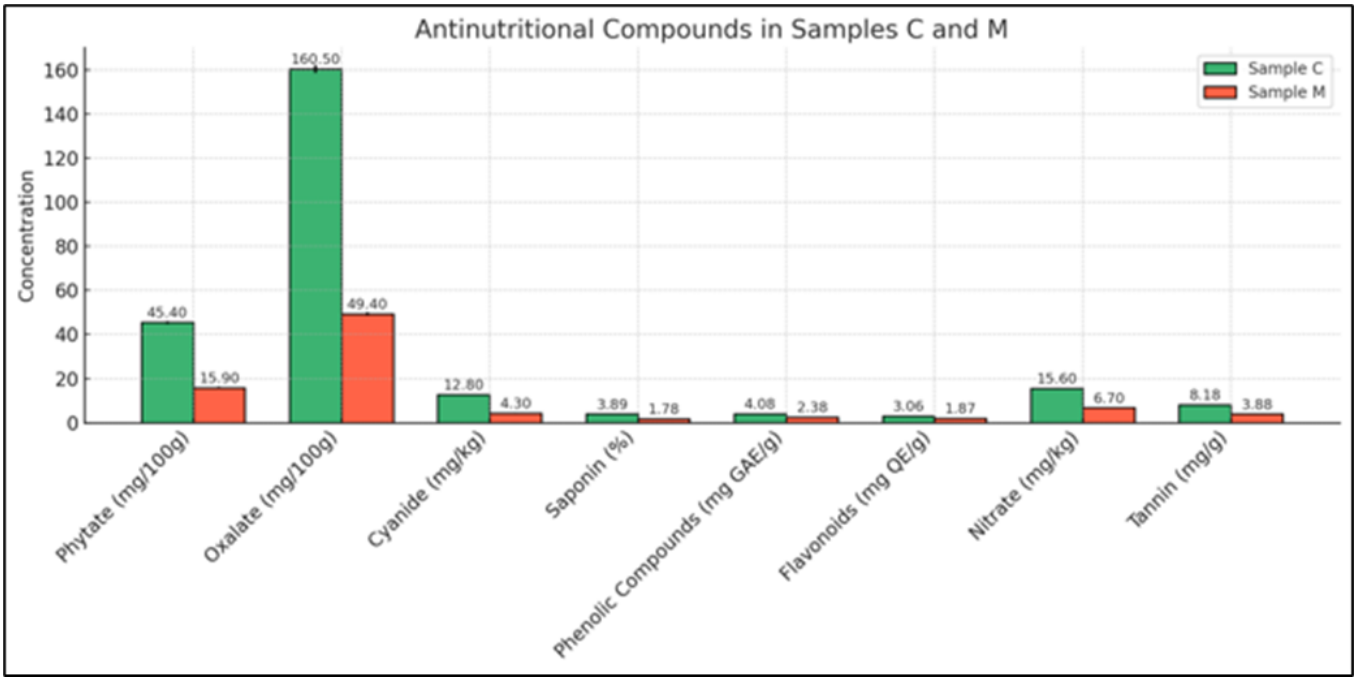
Figure 2 presents the levels of various antinutritional compounds in the commercial locust beans and the optimized fermented locust beans. Phytate content decreased specifically from 45.4 ± 0.7 mg/100g in commercial locust beans to 15.9 ± 0.3 mg/100g in locust beans produced through co-fermentation with *L. plantarum strain* SAL-IMIE02 and *L. fermentum strain* SAL-IMIE01. Similarly, oxalate levels decreased from 160.5 ± 1.7 mg/100g to 49.4 ± 0.8 mg/100g, and cyanide from 12.8 ± 0.2 mg/kg to 4.3 ± 0.2 mg/kg.

Significant reductions were also observed in saponin (3.89 ± 0.03% to 1.78 ± 0.03%), phenolic compounds (4.08 ± 0.03 mg GAE/g to 2.38 ± 0.03 mg GAE/g), and flavonoids (3.06 ± 0.04 mg QE/g to 1.87 ± 0.02 mg QE/g). Moreover, nitrate and tannin contents were lowered remarkably (15.6 ± 0.1 mg/kg to 6.7 ± 0.1 mg/kg; 8.18 ± 0.03 mg/g to 3.88 ± 0.03 mg/g) respectively.

Table 1 Amino Acid Composition of Sample C (Control) and Sample M (mg/100 g)

Amino Acid	Sample C (mg/100 g)	Sample M (mg/100 g)	Sample U (mg/100 g)	R^2 Value	Peak Area	RT (min)	LOD (mg/mL)	LOQ (mg/mL)
Lysine	4.52^{b}±0.12	5.80^{c}± 0.10	4.10^{a}±0.10	0.998	13560	7.2	0.02	0.06
Leucine	8.95^{b}±0.15	10.70^{c}±0.15	8.00^{a}±0.10	0.997	16240	8.5	0.03	0.08
Isoleucine	6.38^{b}±0.10	8.00^{c}±0.15	5.90^{a}±0.13	0.996	14050	9.1	0.03	0.09
Methionine	2.45^{b}±0.05	3.20^{c}±0.05	2.20^{a}±0.12	0.995	11800	10.3	0.02	0.07
Phenylalanine	5.62^{b}±0.15	7.10^{c}±0.10	5.00^{a}±0.14	0.998	15320	7.8	0.02	0.06
Threonine	4.08^{b}±0.10	5.30^{c}±0.10	3.60^{a}±0.16	0.997	13610	6.9	0.03	0.08
Tryptophan	1.85^{b}±0.05	2.60^{c}±0.05	1.60^{a}±0.10	0.995	11080	12.5	0.02	0.07
Valine	6.90^{b}±0.15	8.50^{c}±0.10	6.10^{a}±0.10	0.998	14500	8.8	0.02	0.06
Histidine	3.10^{b}±0.05	4.10^{c}±0.05	2.90^{a}±0.10	0.996	11560	7.4	0.03	0.08
Alanine	5.80^{b}±0.10	7.30^{c}±0.10	4.10^{a}±0.10	0.997	13300	8.2	0.02	0.07
Aspartic	9.50^{b}±0.15	11.80^{c}±0.15	8.00^{a}±0.12	0.998	16840	6.4	0.02	0.06
Glutamic	13.6^{b}±0.10	16.40^{c}±0.20	5.90^{a}±0.12	0.999	20050	5.9	0.02	0.06
Glycine	4.70^{b}±0.10	6.00^{c}±0.10	2.20^{a}±0.12	0.996	12780	9.6	0.03	0.08
Proline	3.90^{b}±0.10	5.10^{c}±0.10	5.00^{a}±0.11	0.995	12180	10.9	0.03	0.08
Serine	4.40^{b}±0.10	5.70^{c}±0.10	3.60^{a}±0.10	0.997	13510	7.0	0.02	0.07
Tyrosine	3.20^{b}±0.05	4.50^{c}±0.05	1.60^{a}±0.10	0.996	11540	7.6	0.02	0.06

Note: Values with different superscript letters are significantly different at $p < 0.05$; Key: Sample C (commercial locust beans); Sample M (locust beans fermented with *L. plantarum strain* SAL-IMIE02 and *L. fermentum strain* SAL-IMIE01); and Samples 2 and 5 (locust beans fermented with *L. plantarum strain* SAL-IMIE02 and *L. fermentum strain* SAL-IMIE01); RT (Retention time); Sample U (Non-fermented locust beans)



Note: Values with different superscript letters are significantly different at $p < 0.05$. Key: Sample C (commercial locust beans); Sample M (locust beans fermented with *L. plantarum strain* SAL-IMIE02 and *L. fermentum strain* SAL-IMIE01.

Figure 2 Determination of Antinutritional Compounds in Sample C and M

3.3. Sensory properties of fermented locust beans samples

The sensory attributes of the optimized fermented locust beans and the commercial locust beans were presented in Table 2. Locust beans fermented with *L. plantarum strain* SAL-IMIE02 and *L. fermentum strain* SAL-IMIE01 recorded significantly higher scores for colour (8.83 ± 0.81), texture (8.33 ± 1.04), taste (7.53 ± 1.17), consistency (7.94 ± 1.30), and overall acceptability (9.31 ± 0.54) compared to the commercial locust beans. In contrast, Sample C had higher aroma score (8.67 ± 1.06) compared to Sample M (6.68 ± 1.61).

Table 2 Sensory properties of fermented locust beans samples

Samples	**M**	**C**
Colour	8.83 ± 0.81^{a}	6.68 ± 1.61^{b}
Texture	8.33 ± 1.04^{a}	7.47 ± 1.45^{b}
Taste	7.53 ± 1.17^{a}	5.57 ± 2.02^{b}
Aroma	6.68 ± 1.61^{b}	8.67 ± 1.06^{a}
Consistency	7.94 ± 1.30^{a}	6.93 ± 1.30^{b}
Overall acceptability	9.31 ± 0.54^{a}	7.50 ± 1.49^{b}

Value are means ± standard deviation, value with the same superscript in the same column are not significantly different from each other at $P \leq 0.05$.; Key: Sample C (commercial locust beans); Sample M (locust beans fermented with *L. plantarum strain* SAL-IMIE02 and *L. fermentum strain* SAL-IMIE01

4. Discussion of Results

Microbial contaminants and nutritional quality remains a major concerns of fermented limiting the large-scale production and commercialization of African locust beans. To address this challenge, this study was design explore the probiotic lactobacillus capabilities to control locust beans production, safety and nutritional qualities. Results analysis revealed that the amino acid composition of the locust beans fermented with *L. plantarum strain* SAL-IMIE02 and *L. fermentum strain* SAL-IMIE01 was significantly higher ($p < 0.05$) than that of the control sample and non-fermented locust beans across all investigated amino acids, indicating improved nutritional quality (Table 5). The improvement in amino acid composition of optimized sample relative to the control sample underlines the role of fermentation on the

nutritional quality of locust beans (*Parkia biglobosa*). More so, the observed significant increases in essential amino acids (lysine, leucine, isoleucine, methionine, valine as well as the non-essential amino acids (glutamic and aspartic acids) is a pointer to enhanced protein quality and functionality of the optimized locust beans. Notably, R^2 values ranged between 0.995 and 0.999, indicating excellent linearity in the calibration curves. Furthermore, the low values of LOD (0.02–0.03 mg/mL) and LOQ (0.06–0.09 mg/mL) confirm the sensitivity and precision of the detection method. Notably, the increase in glutamic acid may contributes to the development of flavor, which will no doubt increased it palatability. Previous studies by Oboh *et al*. (2008); Ijarotimi and Keshinro (2012) and Zebedee *et al*. (2022) indicated that protein value, particularly the amino acid composition increased significantly following fermentation hence the finding of this study corroborates existing literature.

The determination of antinutritional compounds in optimized samples and the control revealed significant differences ($p < 0.05$) across all measured parameters (Figure 2). The decrease in antinutrient concentrations in optimized locust beans in the present work may be due to enzymatic hydrolysis during the course of fermentation (Adegbehingbe *et al.,* 2017). Phytate levels were significantly higher in control (45.4 mg/100g) than in optimized sample (15.9 mg/100g), indicating a considerable disparity in their mineral bioavailability potential, as phytates are known to chelate essential minerals like calcium and zinc and reduce their absorption. Similarly, oxalate contents were markedly elevated in control (160.5 mg/100g) relative to optimized sample (49.4 mg/100g), which has implications for calcium bioavailability and kidney stone risk due to oxalate's potential to form insoluble complexes with calcium.

The cyanide content of the locust bean samples revealed a significant reduction in the optimized sample (Sample M: 4.3 mg/kg) compared to the control (Sample C: 12.8 mg/kg) (Figure 2). This substantial difference highlights the potential cyanogenic toxicity risk associated with un-optimized processing, as high cyanide levels can interfere with cellular respiration and pose serious health threats if not adequately detoxified (Akinmutimi, 2007; Oboh and Elusiyan, 2007). The reduced cyanide levels in optimized sample may reflect the effectiveness of fermentation in breaking down cyanogenic glycosides, a phenomenon previously documented in leguminous seeds (Odetokun et al., 2010).

Similarly, the levels of saponins, phenolic compounds, and flavonoids—all known for their dual roles as bioactive and antinutritional compounds—were significantly higher in the control sample (Figure 2). While moderate concentrations of these phytochemicals may contribute to antioxidant and antimicrobial activity (Amarowicz et al., 2004; Edeoga et al., 2005), excessive levels can inhibit nutrient bioavailability and negatively affect sensory properties such as taste and texture, particularly by increasing bitterness (Gupta et al., 2015; Fasoyiro et al., 2005).

Furthermore, nitrate and tannin concentrations were markedly higher in the control sample (15.6 mg/kg and 8.18 mg/g, respectively) than in the optimized sample (6.7 mg/kg and 3.88 mg/g) (Figure 2). Elevated nitrate levels raise health concerns due to their potential conversion to nitrites, which can impair oxygen transport in the blood, particularly in infants (Greer & Shannon, 2005). High tannin content, on the other hand, is well-documented to reduce protein digestibility by forming indigestible protein-tannin complexes (Addisu, 2016; Adejoro, 2019).

The consistent pattern of elevated antinutrients in the control sample emphasizes the necessity of improved processing or detoxification methods, such as controlled fermentation, to enhance both the safety and nutritional value of locust beans. The lower concentrations observed in optimized sample affirm the potential of optimized fermentation—particularly with selected lactic acid bacteria strains—as a viable method to minimize antinutritional factors while preserving beneficial compounds. This is particularly important for target populations such as children and pregnant women, who are vulnerable to nutrient deficiencies and more susceptible to the adverse effects of dietary toxins (Ijarotimi and Keshinro, 2012).

The sensory evaluation revealed significant differences between the optimized locust bean sample and the control across most parameters (Table 2), and this indicates enhanced acceptability of the optimized product. Colour was rated significantly higher in optimized product (8.83 ± 0.81) than in control (6.68 ± 1.61). Colour plays a crucial role in consumer perception and acceptance of food products. The observed improved appearance of optimized product signaled better fermentation control that result in more appealing browning and less discoloration which can occur during microbial spoilage or oxidation. This observation supports the findings of Adegbehingbe et al. (2017), who identified control fermentation of locust beans often produced a more appealing colour and uniformity compared to those traditionally processed.

Further, texture was also rated higher in optimized sample (8.33 ± 1.04) compared to control (7.47 ± 1.45) (Table 2). Controlled fermentation with starter cultures such as Lactiplantibacillus spp. could produce locust beans with improved softening of the seed coat with a smoother mouth feel. This is in line with the work of Ijarotimi and Keshinro (2012), where optimized fermentation enhanced the tenderness and palatability locust beans.

Additionally, taste showed a significant improvement in optimized sample (7.53 ± 1.17) compared to the control (5.57 ± 2.02). This is likely due to the metabolic potential of the selected probiotic strains including *L. plantarum strain* SAL-IMIE02 and *L. fermentum strain* SAL-IMIE01 used in fermentation. The activity of lactobacillus spp. have been attributed to reduce bitterness, enhance umami as well as savory flavors associated to proteins and carbohydrates degradation. This supports Kayitesi et al. (2023) who found that microbial fermentation significantly enhance the taste profile of fermented foods.

Though, aroma was rated higher in control (8.67 ± 1.06) compared to optimized sample (6.68 ± 1.61) (Table 2). Traditional fermentation encourages a wider range inherent microbes to participate and contribute to the characteristic pungent smell associated with locally processed *iru*. This finding reflects the trade-off between traditional flavor complexity and improved hygienic, nutritional, and sensory control often found in standardized fermentation (Omafuvbe et al., 2002).

Consistency described the spread ability and cohesiveness of a paste, was rated higher in the optimized sample (7.94 ± 1.30) compared to control (6.93 ± 1.30) (Table 2). Improved consistency in optimized can also be linked to enhanced protein and carbohydrates degradation that accompany controlled fermentation. Lastly, the overall acceptability was significantly higher for optimized sample (9.31 ± 0.54) compared to the control (7.50 ± 1.49), while implied that the optimized sample was generally liked and further affirm the fact that controlled fermentation have the prospect to significantly enhance the sensory appeal and consumer acceptability of locust beans.

5. Conclusion

Nutritional quality assessment revealed that the optimized *iru* showed significantly enhanced amino acid profiles—lysine (5.80 ± 0.10 mg/100g), leucine (10.70 ± 0.15 mg/100g), isoleucine (3.20 ± 0.05 mg/100g), methionine (3.20 ± 0.05 mg/100g), valine (8.50 ± 0.10 mg/100g), and threonine (5.30 ± 0.10 mg/100g).

Our findings also revealed that fermentation with *L. plantarum strain* SAL-IMIE02 and *L. fermentum strain* SAL-IMIE01 resulted in significant decreases in the in anti-nutritional factors, saponin (3.89 ± 0.03% to 1.78 ± 0.03%), phenolic compounds (4.08 ± 0.03 mg GAE/g to 2.38 ± 0.03 mg GAE/g), and flavonoids (3.06 ± 0.04 mg QE/g to 1.87 ± 0.02 mg QE/g). in addition, nitrate and tannin contents were lowered remarkably (15.6 ± 0.1 mg/kg to 6.7 ± 0.1 mg/kg; 8.18 ± 0.03 mg/g to 3.88 ± 0.03 mg/g) respectively in the optimized sample.

Further, *L. plantarum* 103150 and *L. fermentum* CIP 102980 optimized *iru* had a significant high scores for colour (8.83 ± 0.81), texture (8.33 ± 1.04), taste (7.53 ± 1.17), consistency (7.94 ± 1.30), as well as overall acceptability (9.31 ± 0.54) compared to the commercial locust beans.

Recommendations

- Large-scale production of *iru* using *L. plantarum strain* SAL-IMIE02 and *L. fermentum strain* SAL-IMIE01 is encouraged to boost the economic potentials of the product.
- Owing the potential of the two probiotic strains, commercial starter culture formulations should be develop and publicized to create awareness amongst local processors, with the ultimate of standardizing traditional methods of production.
- Local producer should be trained on hygiene and aseptic techniques that will ensure the microbiological safety and wide acceptability of Africa originated fermented food

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest to be disclosed.

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