DEVELOPMENT AND QUALITY EVALUATION OF NUTRIENT ENRICHED BREAD FROM WHEAT, SOYBEAN, AND CARROT FLOUR BLENDS

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ABSTRACT

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| **Aim:** The increasing demand for nutritious, affordable, and sustainable food products calls for innovative strategies that enhance the resilience of food systems while improving livelihoods. This study investigates the development of nutrient-dense bread using composite flours made from wheat, soybeans, and carrots—three ingredients that are locally available and utilised in many regions.  **Materials and Methods:** Wheat flour, fresh carrots, and soybeans were the primary ingredients used for bread production. All ingredients were sourced from local markets. Four composite flour ratios were formulated and tested: 80:10:10, 70:25:5, 70:20:10, and 50:30:10 (wheat: soybean: carrot), using 100% wheat bread as the control. Proximate, mineral, vitamin, and antinutrient analysis were carried out on all the samples as well as functional and sensory evaluation.  **Result:** The proximate analysis revealed significant improvements in nutrient profiles with increased incorporation of carrot and soybean. Moisture content ranged from 5.42% to 7.15%, and ash content from 2.60% to 14.15%, indicating enhanced mineral density. Vitamin analysis revealed that the 80:10:10 formulation contained the highest levels of vitamin B1 (0.34 mg) and vitamin B3 (0.35 mg). Antinutrient levels such as tannins (0.4964–0.8802 mg) and oxalates (0.1795–0.2428 mg) remained within safe consumption limits. Functional properties such as water absorption (53.33%–91.33%) and swelling capacity (3.81–5.52%) improved with higher levels of carrot and soybean. Microbial analysis further indicated that the 50:30:10 sample had the lowest total plate count (1.50 × 10³ CFU/g), reflecting improved shelf stability and food safety. Sensory evaluation revealed that the 70:25:5 blend achieved the highest overall acceptability (6.40 ± 1.58), while the 70:20:10 sample scored best for texture, aroma, and color.  **Conclusion:** This study demonstrates that incorporating carrot and soybean into wheat-based bread enhances its nutritional and functional properties and provides a practical, low-cost solution for diversifying diets and improving food system resilience. |

*Keywords: (Composite flour, local crops, bread, sustainable food, nutrient-dense)*

1. INTRODUCTION

Bread is a widely consumed staple food across the globe, often serving as a significant source of energy and nutrients in many diets. Conventionally produced from wheat flour, bread offers appreciable amounts of refined carbohydrates and some essential nutrients. However, wheat-based bread is limited in certain nutrients, particularly dietary fiber, essential amino acids like lysine, and bioactive compounds such as β-carotene and antioxidants (Akinwande et al., 2022). In earlier civilisations, bread was seen as a vital and reliable source of nourishment and sustenance. However, nowadays, there are growing health concerns about bread consumption, particularly its possible links to a higher risk of overweight, type 2 diabetes, and cardiovascular diseases. As consumers become more health-conscious and demand more functional foods, there has been a growing interest in enriching bakery products with nutrient-dense plant-based ingredients (Adeyanju et al., 2021; Singh & Verma, 2023).

Soybeans (Glycine max) are legumes known for their high-quality protein content. It is one of the most affordable and nutrient-rich sources of plant protein available to help millions of people meet their nutritional needs (Ojo et al., 2022). Soybean is the only plant source that contains all of the essential amino acids and is particularly rich in lysine, making it an ideal complement to wheat’s amino acid profile. Soybean has a high fat content, predominantly composed of unsaturated fatty acids. Specifically, about 63% are polyunsaturated (mainly linoleic acid), 23% are monounsaturated (primarily oleic acid), and 14% are saturated (mainly palmitic acid) (Ojo et al., 2022). Soybeans also contribute B vitamins, minerals, and isoflavones that promote cardiovascular and metabolic health (Choi et al., 2023).

Carrots (Daucus carota) are root vegetables rich in dietary fiber, vitamins, especially provitamin A (β-carotene), and antioxidants, which can significantly improve the nutritional value and health benefits of baked products (Kumar et al., 2022). Carrots' dietary fiber is primarily made up of cellulose, along with smaller amounts of hemicelluloses, lignin, and starch. It also consists of natural sugars like sucrose, glucose, and fructose (Boadi et al., 2021). These dietary components contribute to overall health by promoting proper digestion, assisting in blood sugar regulation, and potentially lowering cholesterol levels and reducing the risk of cardiovascular diseases. In particular, fiber such as cellulose is essential for maintaining regular bowel movements and preventing conditions like constipation and diverticulosis. Additionally, Carrots are abundant in antioxidants like alpha- and beta-carotene, which contribute to it’s characteristic orange color (Boadi et al., 2021). The body transforms this carotene into vitamin A, which is vital for maintaining good vision, supporting immune function, and promoting healthy bones.

The integration of carrots and soybeans into wheat-based bread formulations has the potential not only to enhance the nutritional profile of the final product but also to influence sensory attributes such as flavor, texture, and color. Previous studies have shown that appropriate substitution levels can yield bread that is both nutritionally superior and organoleptically acceptable (Okafor & Uchegbu, 2021; Bello et al., 2020). However, the optimal blend ratios that maximize both nutritional enhancement and consumer acceptance remain under active investigation.

Thus, this study aims to evaluate the nutritional composition and sensory properties of bread produced from composite flours containing wheat, soybean, and carrot. The findings are expected to contribute to the development of affordable, functional bakery products that address nutritional deficiencies, particularly in low- and middle-income populations.

2. material and methods

**2.1 Materials**

Wheat flour, fresh carrots, and soybeans were the primary ingredients used for bread production. All ingredients were sourced from local markets in Ogbomoso, and other baking materials such as yeast, sugar, salt, and vegetable oil were of food-grade quality. The milling machine used to produce the composite flour is within the food processing laboratory of the Ladoke Akintola University of Technology, Ogbomoso, Oyo State, Nigeria.

**2.2 Method**

**2.2.1 Production of Composite Flours**

Wheat flour was obtained by sorting the wheat, weighing, soaking, drying, and milling. Carrots were washed, peeled, and grated, while soybeans were cleaned, soaked, boiled, dehulled, dried, and milled into flour. Composite flours were prepared by blending wheat flour with varying proportions of carrot and soybean flours as follows: A= 85:10:5, B=70:20:10, C=60:25:15. D= 50:30:20 and E=100:0:0.

### **Table 1: Composite Flour Formulations for Bread Production**

| **Sample Code** | **Wheat Flour (%)** | **Soybean Flour (%)** | **Carrot Flour (%)** |
| --- | --- | --- | --- |
| A | 85 | 10 | 5 |
| B | 70 | 20 | 10 |
| C | 60 | 25 | 15 |
| D | 50 | 30 | 20 |
| E(Control) | 100 | 0 | 0 |

**2.2.2. Bread Preparation**

The bread was produced using the straight dough method. The ingredients were weighed and mixed to form a uniform dough, which was kneaded, proofed, molded, and baked at 180°C for 30 minutes. The loaves were cooled at room temperature before analysis.

**2.3. Analysis**

**2.3.1 Proximate Analysis**

Proximate composition (moisture, ash, crude protein, fat, and carbohydrate) was determined using standard AOAC (2019) methods. β-carotene content was analysed using spectrophotometric techniques.

**2.3.2 Mineral Analysis**

Mineral composition is a key indicator of the nutritional value of food products and provides essential information for evaluating dietary quality. In this study, the levels of iron, calcium, and potassium in the bread samples were determined using standard instrumental methods validated for food analysis (Adegboye et al., 2022; Onwuliri & Dike, 2021).

***2.3.2.1 Iron***

The iron content was determined using atomic absorption spectrophotometry (AAS), as described by Abubakar et al. (2020). Approximately 1 g of each composite flour sample was digested using a mixture of concentrated nitric acid (HNO₃) and hydrogen peroxide (H₂O₂) under controlled heating. The resulting solution was filtered and diluted appropriately. Iron concentration was measured at a wavelength of 248.3 nm using an atomic absorption spectrophotometer. Quantification was achieved by referencing a standard calibration curve prepared from iron standard solutions (Ogunyemi et al., 2023).

***2.3.2.2 Calcium***

Calcium determination was similarly carried out using AAS, following the method outlined by Fasakin and Olayinka (2022). The digested sample solutions were analyzed at a wavelength of 422.7 nm. Calibration was done using known concentrations of calcium standards, and results were expressed in mg/100 g of the sample. This method provides high specificity and sensitivity for calcium quantification in composite flour products (Okorie et al., 2021).

***2.3.2.3 Potassium***

Potassium content was assessed using flame photometry, a method suited for the detection of alkali metals in food matrices (Chinedu et al., 2023). The digested samples were aspirated into a flame photometer, and emission intensity was recorded at 766.5 nm. A calibration curve generated using potassium standard solutions was used to determine potassium concentration in the samples. Flame photometry remains a widely accepted method for potassium estimation due to its simplicity and rapid detection capacity (Nwankwo et al., 2020).

**2.3.3 Vitamin Analysis**

***2.3.3.1 Vitamin B1 (Thiamine)***

The vitamin B1 content was determined using a spectrophotometric method. The samples were digested in acid, and the solution was filtered. The absorbance of the filtrate was measured at 366 nm using a UV spectrophotometer. The concentration of vitamin B1 was calculated using a standard calibration curve.

***2.3.3.2 Vitamin B3 (Niacin)***

Vitamin B3 was analysed using the same spectrophotometric technique as vitamin B1. The samples were digested, and the absorbance was measured at a wavelength of 430 nm. The concentration of vitamin B3 was determined based on a standard curve.

***2.3.3.3 β-Carotene Analysis***

The β-carotene content of the bread samples was determined using the acetone–hexane extraction method as described by Rodriguez-Amaya and Kimura (2021), with slight modifications to suit the composite matrix. Approximately 1 g of the sample was homogenized with a mixture of cold acetone and hexane (2:3 v/v) to extract the carotenoids. The mixture was centrifuged at 4000 rpm for 10 minutes, and the upper layer containing the carotenoids was collected. The absorbance was measured at 453 nm using a UV-Vis spectrophotometer, and β-carotene content was calculated using a standard curve derived from pure β-carotene (Oluwatosin et al., 2023). This method is widely recognized for its accuracy in assessing provitamin A levels in plant-based and composite foods (Chinedu & Balogun, 2022).

**2.3.4 Antinutrient Analysis**

Antinutrients are bioactive compounds that may interfere with the bioavailability of essential nutrients. In this study, the levels of tannins and oxalates in the bread samples were determined using standard biochemical methods, as described by recent food science research (Onwuka, 2021; Ezeonu et al., 2023).

***2.3.4.1 Tannin Determination***

Tannin content was quantified using the Folin–Denis spectrophotometric method, which is reliable for plant-based foods (Okoye & Njoku, 2022). About 0.5 g of each sample was extracted with 50 mL of 70% methanol by shaking for 2 hours at room temperature. The extract was filtered, and 1 mL of the filtrate was mixed with 0.5 mL of Folin–Denis reagent and 1 mL of sodium carbonate solution. The mixture was incubated at room temperature for 30 minutes, and absorbance was measured at 760 nm using a UV-Vis spectrophotometer. Tannin content was calculated from a standard curve prepared with tannic acid (Ibrahim & Afolayan, 2020).

***2.3.4.2 Oxalate Determination***

Oxalate content was determined using the permanganate titration method, a validated technique for evaluating oxalate levels in food (Adekunle et al., 2021). Approximately 1 g of the sample was digested with 75 mL of 3 M sulfuric acid for 1 hour and filtered. The filtrate was heated to 90°C, and oxalate was precipitated with calcium chloride. The precipitate was dissolved in hot distilled water and titrated against 0.05 M potassium permanganate until a pink color persisted for 30 seconds. Oxalate concentration was calculated and expressed in mg/100 g of sample (Chukwuma & Eze, 2023).

**2.3.5 Functional Properties**

Functional properties such as water absorption capacity, swelling capacity, and solubility index are critical in determining the behavior of flour blends during processing and their suitability for baked products like bread. These properties influence dough handling, texture, and moisture retention of the final product (Adeyemi & Olagunju, 2021; Ezeocha et al., 2023).

***2.3.5.1 Water Absorption Capacity (WAC)***

The water absorption capacity was determined following the modified method described by Okafor et al. (2020). One gram of each composite flour sample was mixed with 10 mL of distilled water in a centrifuge tube and allowed to stand at room temperature for 30 minutes. The mixture was then centrifuged at 3000 rpm for 30 minutes. The supernatant was carefully decanted, and the sediment-containing tube was weighed. The WAC was expressed as the weight of water retained per gram of dry sample. This parameter reflects the flour's ability to retain moisture and its potential influence on dough yield and bread texture (Oluwole et al., 2022).

***2.3.5.2 Swelling Capacity (SC*)**

Swelling capacity was assessed using the method outlined by Chikwendu and Nwankwo (2021). One gram of the flour sample was placed in a 10 mL graduated cylinder containing distilled water and allowed to stand for 24 hours at room temperature. The final volume of the swollen material was recorded, and the swelling capacity was calculated as the increase in volume (mL) per gram of flour. This property provides insight into the hydration behavior of the flour, which affects loaf expansion and crumb structure (Bello et al., 2022).

***2.3.5.3 Solubility Index (SI)***

The solubility index was measured using a modified procedure from Nwosu and Dike (2023). One gram of the flour sample was dispersed in 10 mL of distilled water and stirred continuously for 30 minutes. The mixture was left to settle before being filtered through Whatman No. 1 filter paper. The volume of the clear filtrate was measured, and the solubility index was expressed as the percentage of the sample that dissolved in water. The SI reflects the degree of solubilized components, such as proteins and soluble carbohydrates, which can influence flavor release and mouthfeel in baked products (Fasakin et al., 2021).

**2.3.6 Microbial Analysis**

Microbial analysis is essential for evaluating the safety and shelf stability of bread and other bakery products. The microbiological quality of the bread samples was assessed by determining the total coliform count, lactic acid bacteria (LAB), and fungal load using standard culture techniques, as described by recent guidelines (FAO, 2021; Olaniran et al., 2022; Ezeonu & Ogbonna, 2023).

***2.3.6.1 Coliform Count***

Coliform enumeration was performed using Eosin Methylene Blue (EMB) agar, following the method of Akinbinu and Bello (2020). Bread samples were serially diluted in sterile peptone water and plated in duplicates onto EMB agar. The plates were incubated at 37°C for 24 hours, and no visible colonies were seen after incubation. This method is widely used to assess sanitary quality and potential fecal contamination in food samples (Oluwatosin et al., 2023).

***2.3.6.2 Lactic Acid Bacteria Count***

Lactic acid bacteria were enumerated using de Man, Rogosa, and Sharpe (MRS) agar, a selective medium suitable for LAB growth (Olowe et al., 2022). Serially diluted samples were inoculated and incubated anaerobically at 37°C for 24 hours using an anaerobic jar system. Distinct colonies were counted, and the LAB count was expressed in CFU/mL. LAB are known for their role in food preservation, safety, and gut health (Chukwu et al., 2021).

***2.3.6.3 Fungal Count***

Fungal contamination was evaluated using Potato Dextrose Agar (PDA). The diluted samples were plated and incubated at room temperature (25–28°C) for 5–7 days as outlined by Onwuka et al. (2020). Fungal colonies, including molds and yeasts, were enumerated and recorded as CFU/g of the sample. Monitoring fungal load is critical due to the potential risk of mycotoxin production in bakery products (Ibrahim et al., 2022).

**2.3.7 Sensory Evaluation**

Sensory evaluation was conducted to assess consumer acceptability of the bread samples, focusing on key sensory attributes that influence purchase decisions and consumption. The evaluation was carried out using a structured 9-point hedonic scale, as recommended by sensory analysis protocols (ISO 13299:2016; Anyanwu & Nwosu, 2021).

A panel of 20 semi-trained individuals was selected from staff and students familiar with bread products. Each panelist evaluated the bread samples in terms of visual appeal (color and shape), aroma profile, taste, mouthfeel (texture), and overall acceptability. Samples were coded and presented in random order under standardized lighting and hygienic conditions to avoid bias (Okonkwo et al., 2023). Panelists were instructed to rinse their mouths with water between samples to minimize carryover effects.

3. results and discussion

**3.1 Proximate Analysis**

The result of the proximate composition is shown in Table.1. The proximate analysis evaluates the moisture, ash, protein, and fat content in the bread samples and is essential for understanding their nutritional makeup and overall quality. These components affect the shelf life, texture, and stability of food products. As reported by Fellows (2017), moisture content is a key factor in microbial growth and spoilage potential in food. Lower moisture levels generally extend shelf life by reducing bacterial growth. In this study, moisture content varied significantly among the samples, with Sample A having the highest moisture at 7.15%, while Sample C had the lowest at 5.42%. The higher moisture in Sample A suggests it may be more prone to spoilage, whereas the lower moisture in Sample C indicates better storage stability.

Ash content represents the mineral content in the bread samples, which is important for assessing nutritional value. In agreement with work done by Sharma et al. (2016), higher ash content suggests a richer mineral composition. In this analysis, Sample B stood out with the highest ash content at 14.15%, suggesting a better supply of essential minerals like calcium and potassium compared to other samples. Sample C, with an ash content of 2.60%, exhibited the lowest mineral content, indicating that it may have fewer nutritional benefits in terms of mineral content.

Protein and fat content also play an important role in determining the nutritional quality of bread. As described by Slavin (2015), protein is essential for body repair and maintenance, while fat provides energy and helps in the absorption of fat-soluble vitamins. In this study, the protein content ranged from 12.91% in Sample E to 18.65% in Sample D, highlighting the variation in nutritional density between the samples. Fat content, while relatively consistent, ranged from 8.00% to 8.15%, contributing to the overall caloric content of the bread.

Overall, the proximate analysis shows that Sample B has the highest mineral density, while Sample C may appeal to those looking for a bread with lower moisture content and longer shelf life. However, Sample D demonstrates the highest protein content, making it a nutritionally dense option for consumers prioritizing protein intake. These findings align with the established importance of proximate composition in determining food quality and consumer preference as discussed by Sharma et al. (2016)

Table 2: Proximate Analysis

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **SAMPLE** | **CP%** | **CF%** | **EE%** | **MOISTURE%** | **ASH%** |
| A | 16.91±0.15ab | 5.20±0.14 a | 8.00±0.28 a | 7.15±0.07 d | 9.00±0.00 c |
| B | 16.49±0.12 b | 5.00±0.14 a | 8.15±0.07 a | 7.00±0.00 c | 14.15±0.07 d |
| C | 17.64±1.61 ab | 5.10±0.14 a | 8.15±0.07 a | 5.43±0.04 a | 2.60±0.28 a |
| D | 18.65±0.40 c | 4.75±0.35 a | 8.10±0.00 a | 6.15±0.07 b | 4.15±0.07 b |
| E | 12.91±0.01 a | 3.95±0.35 b | 8.05±0.07 a | 6.05±0.07 b | 2.50±0.14 a |

Mean values are of duplicates ± standard deviation. The value within the rows with different superscripts is significantly (P<0.05) different.

Key:

Sample A = 85% Wheat flour, 10% Soybean flour, and 5% Carrot flour

Sample B = 70% Wheat flour, 20% Soybean flour, and 10% Carrot flour

Sample C = 60% Wheat flour, 25% Soybean flour, and 15% Carrot flour

Sample D = 50% Wheat flour, 30% Soybean flour, and 20% Carrot flour

Sample E = 100% Wheat flour (Control)

**3.2 Mineral Analysis**

The result of the mineral analysis is shown in Table 3. Minerals are essential micronutrients required for various physiological functions, including bone development, muscle contraction, nerve function, and blood oxygenation. The mineral content of bread plays a significant role in determining its nutritional value. In this study, the key minerals analyzed were potassium, calcium, and iron, which are critical for maintaining good health. In agreement with research by Weaver et al. (2016), adequate levels of these minerals help prevent conditions such as osteoporosis, anemia, and hypertension.

In this analysis, Sample D exhibited the highest mineral content, particularly for potassium, calcium, and iron. Sample D had 256.92 mg of potassium, which is essential for maintaining proper heart function and regulating fluid balance within the body. As reported by Weaver et al. (2016), potassium is also effective in counteracting the negative effects of sodium on blood pressure, making Sample D a suitable choice for individuals aiming to maintain cardiovascular health. Sample D also had the highest calcium content at 235.74 mg, making it beneficial for bone health and preventing osteoporosis, a condition that affects bone density and strength, particularly in older adults. The iron content in Sample D was 1.14 mg, which is crucial for hemoglobin production, aiding in the transport of oxygen throughout the body and helping prevent iron-deficiency anemia.

Sample E had the lowest mineral content, with 84.53 mg of potassium, 96.29 mg of calcium, and 0.38 mg of iron. This suggests that Sample E may offer fewer health benefits, particularly for individuals who need higher intakes of these essential minerals. Low levels of calcium and potassium, in particular, could limit its effectiveness in promoting bone and heart health, respectively. In research by Abbaspour et al. (2018), lower iron content in foods is linked to a higher risk of developing anemia, especially in women and children, indicating that Sample E might not be suitable for those with higher mineral needs.

The other samples, A, B, and C, showed moderate mineral content. For example, Sample A had 198.25 mg of potassium and 156.45 mg of calcium, offering a balanced supply of these nutrients, though not as rich as Sample D. Sample B, with 206.71 mg of potassium and 194.56 mg of calcium, similarly provides a good nutritional profile, particularly for consumers looking for a balance between taste and nutritional benefits.

In conclusion, Sample D stands out as the most mineral-rich option, particularly for potassium, calcium, and iron, making it the best choice for consumers seeking a nutritionally dense bread option. Sample E, with its lower mineral content, may be less suitable for those with specific mineral requirements, while Samples A and B offer moderate amounts of essential minerals. These findings align with the importance of mineral content for overall health, as highlighted by Weaver et al. (20

Table 3: Minerals Analysis Result

|  |  |  |  |
| --- | --- | --- | --- |
| **SAMPLE** | **IRON**  **Mg/100g** | **CALCIUM**  **Mg/100g** | **POTASIUM**  **Mg/100g** |
| A | 0.90±0.01 c | 200.88±0.00 b | 203.34±4.04 c |
| B | 0.83±0.00 b | 235.74±0.00 c | 187.03±0.00 b |
| C | 1.12±0.04 d | 200.88±0.00 b | 252.26±8.07 d |
| D | 1.14±0.00 d | 235.74±0.00 c | 256.92±0.00 d |
| E | 0.38±0.04 a | 96.29±0.00 a | 84.53±8.07 a |

Mean values are of duplicates ± standard deviation. The value within the rows with different superscripts is significantly (P<0.05) different.

Key:

Sample A = 85% Wheat flour, 10% Soybean flour, and 5% Carrot flour

Sample B = 70% Wheat flour, 20% Soybean flour, and 10% Carrot flour

Sample C = 60% Wheat flour, 25% Soybean flour, and 15% Carrot flour

Sample D = 50% Wheat flour, 30% Soybean flour, and 20% Carrot flour

Sample E = 100% Wheat flour (Control)

**3.3 Vitamin Analysis**

The result of the vitamin analysis is shown in Table 4. Vitamins are essential nutrients that play a significant role in maintaining overall health, and their presence in bread can enhance its nutritional value. In particular, B vitamins like thiamine (B1) and niacin (B3) are important for energy metabolism, nerve function, and overall vitality. In research by Gropper et al. (2018), thiamine is identified as crucial for converting carbohydrates into energy, while niacin is essential for maintaining skin health, proper digestion, and improving cardiovascular function.

In this study, Sample A had the highest amount of vitamin B1 (0.34 mg) and vitamin B3 (0.35 mg), making it nutritionally superior in terms of vitamin content. As reported by Gropper et al. (2018), thiamine helps maintain a healthy nervous system, and its higher concentration in Sample A could provide better support for energy metabolism and neurological health compared to other samples. Vitamin B3, also present in higher amounts in Sample A, is beneficial for reducing cholesterol levels and improving skin and cardiovascular health, making it a valuable option for health-conscious consumers.

On the other hand, Sample E had the lowest content of both vitamins, with 0.32 mg of vitamin B1 and 0.12 mg of vitamin B3. In agreement with the work done by Miller et al. (2020), lower vitamin content could reduce the overall nutritional benefits of the bread, particularly for individuals looking to improve their vitamin intake for energy or skin health. Sample E might be less suitable for individuals with higher vitamin requirements, especially those needing a boost in thiamine and niacin.

Samples B, C, and D showed moderate levels of both vitamins. For example, Sample D had 0.33 mg of vitamin B1 and 0.31 mg of vitamin B3, making it a balanced option for individuals who want a bread that provides a decent amount of essential nutrients without being as vitamin-rich as Sample A. In research by Tang et al. (2019), a balanced vitamin content is crucial for maintaining health without excess, and these moderate levels could be appealing to a wider range of consumers.

In conclusion, Sample A stands out as the most vitamin-rich, offering the highest levels of both vitamin B1 and B3, making it an excellent option for consumers seeking to enhance their intake of essential vitamins. Sample E, while lower in vitamin content, may still be suitable for individuals with less demanding nutritional requirements. Samples B, C, and D provide moderate levels of vitamins, ensuring that they remain nutritionally relevant for a broad consumer base. These findings align with previous research, which highlights the importance of vitamin content in maintaining health and energy metabolism (Gropper et al., 2018).

Table 4: Vitamin Analysis

|  |  |  |
| --- | --- | --- |
| **SAMPLES** | **VITAMIN B1** | **VITAMIN B2** |
| A | 0.34±0.00 d | 0.35±0.00 e |
| B | 0.33±0.00 c | 0.25±0.00 c |
| C | 0.32±0.00 a | 0.22±0.00 b |
| D | 0.33±0.00 b | 0.31±0.00 d |
| E | 0.32±0.00 a | 0.12±0.01 a |

Mean values are of duplicates ± standard deviation. The value within the rows with different superscripts is significantly (P<0.05) different.

Key:

Sample A = 85% Wheat flour, 10% Soybean flour, and 5% Carrot flour

Sample B = 70% Wheat flour, 20% Soybean flour, and 10% Carrot flour

Sample C = 60% Wheat flour, 25% Soybean flour, and 15% Carrot flour

Sample D = 50% Wheat flour, 30% Soybean flour, and 20% Carrot flour

Sample E = 100% Wheat flour (Control)

**3.4 Antinutrient Analysis**

The result of the antinutrient analysis is shown in Table 5. Antinutrients, such as tannins and oxalates, are naturally occurring compounds in food that can interfere with the absorption of essential nutrients, particularly minerals like calcium and iron. These compounds can reduce the bioavailability of nutrients, which may affect the nutritional quality of food products (Sharma et al., 2019). In agreement with the work done by Kumar et al. (2020), the presence of antinutrients in food is particularly important in bread formulations where mineral fortification may be compromised by high levels of these compounds.

In this analysis, Sample E had the highest tannin content at 0.8802 mg, which may affect the absorption of iron and other essential minerals. As reported by Kumar et al. (2020), tannins can form insoluble complexes with iron, thereby inhibiting its absorption in the digestive tract. Consumers relying on iron-rich diets might find Sample E less suitable, as the high tannin content could impair iron absorption, potentially leading to issues such as iron deficiency anemia. Sample D, with the lowest tannin content at 0.4964 mg, presents a more favorable option for individuals concerned about mineral absorption.

Oxalate content, another antinutrient that binds to calcium, was found to be highest in Sample A at 0.2428 mg. In research by Noonan and Savage (2019), oxalates are known to form complexes with calcium, reducing its absorption and possibly contributing to the formation of kidney stones in susceptible individuals. Therefore, Sample A, despite its higher nutritional value in other categories, may not be ideal for individuals needing to maximize their calcium intake or those with a predisposition to kidney issues. Sample E, with the lowest oxalate content at 0.1795 mg, is a better option for those concerned with calcium absorption and bone health.

Overall, antinutrient levels varied across the samples, with Sample E having the highest tannin content and Sample A the highest oxalate content. These results suggest that while certain samples may offer higher mineral or vitamin content, the presence of antinutrients like tannins and oxalates could limit their bioavailability. As reported by Sharma et al. (2019), balancing the benefits of nutritional content with the impact of antinutrients is critical in ensuring that food products meet dietary needs effectively. Sample D, with its lower tannin and moderate oxalate content, offers a balanced option that minimizes the negative effects of antinutrients while still providing essential nutrients.

Table 5: Antinutrient Analysis

|  |  |  |
| --- | --- | --- |
| SAMPLE | TANNIN  Mg/g | OXALATE  Mg/g |
| A | 0.6247±0.00 b | 0.2428±0.00 c |
| B | 0.7694±0.00 c | 0.2407±0.00 c |
| C | 0.5052±0.00 a | 0.2313±0.00 c |
| D | 0.4964±0.00 a | 0.2022±0.00 b |
| E | 0.8802±0.00 d | 0.1795±0.00 a |

Mean values are of duplicates ± standard deviation. The value within the rows with different superscripts is significantly (P<0.05) different.

Key:

Sample A = 85% Wheat flour, 10% Soybean flour, and 5% Carrot flour

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Sample D = 50% Wheat flour, 30% Soybean flour, and 20% Carrot flour

Sample E = 100% Wheat flour (Control)

**3.5 Functional Properties Analysis**

The result of the functional properties is shown in Table 6. Functional properties such as water absorption capacity, swelling capacity, and solubility index play a crucial role in determining the usability and performance of composite flours in bread production. These properties affect the dough’s handling characteristics, texture, and overall quality of the final product. In agreement with the work done by Kinsella et al. (2017), high water absorption capacity indicates the ability of flour to retain moisture during mixing and baking, which is crucial for producing soft, moist bread.

In this analysis, Sample D showed the highest water absorption capacity at 91.33%, indicating that this formulation can hold more water, resulting in bread that is likely to be softer and retain moisture for a longer time. Sample E had the lowest water absorption at 53.33%, suggesting that bread made from this formulation might be drier and more suitable for applications where a crispier texture is desired.

Swelling capacity is another important functional property that affects the volume and texture of bread. It measures how much a product increases in volume when water is absorbed. As reported by Adebowale et al. (2015), higher swelling capacity is desirable for products like bread, where a light and fluffy texture is preferred. Sample D once again had the highest swelling capacity at 5.52%, suggesting that it would produce bread with a good rise and a lighter crumb. Sample E, with the lowest swelling capacity at 3.81%, would likely produce a denser, more compact loaf, which might appeal to consumers looking for a firmer bread texture.

The solubility index reflects the ability of the flour to dissolve in water, which is particularly important in products that require easy hydration during processing. In research by Tang and Zhai (2019), high solubility is linked to better dispersibility, making it an important property for flours used in beverages or batters. In this study, Sample D also ranked highest in solubility index at 10.67%, making it ideal for bread formulations where quick hydration and smooth dough formation are necessary. Sample C, with the lowest solubility index at 4.33%, may not dissolve as easily, which could affect the smoothness of the dough during preparation.

In conclusion, Sample D emerged as the best-performing sample in terms of functional properties, with high water absorption, swelling capacity, and solubility index, making it a versatile option for producing soft, moist, and well-risen bread. Sample E, while lower in these properties, could be useful for producing denser, crispier bread. These findings are in agreement with the functional requirements outlined by Kinsella et al. (2017), which highlight the importance of these properties in determining bread quality.

Table 6: Functional Properties

|  |  |  |  |
| --- | --- | --- | --- |
| **SAMPLES** | **WATER ABSORPTION CAPACITY (%)** | **SWELLING CAPACITY (%)** | **SOLUBILITY INDEX**  **(%)** |
| A | 79.33±0.57 c | 5.42±0.04 c | 4.67±1.15 a |
| B | 70.66±1.15 b | 4.55±0.37 b | 5.33±0.58 a |
| C | 85.00±1.00 d | 5.38±0.14c | 4.33±0.58 a |
| D | 91.33±1.53 e | 5.52±0.13 c | 10.67±1.15 c |
| E | 53.33±1.53 a | 3.81±0.04 a | 6.67±1.15 b |

Mean values are of duplicates ± standard deviation. The value within the rows with different superscripts is significantly (P<0.05) different.

Key:

Sample A = 85% Wheat flour, 10% Soybean flour, and 5% Carrot flour

Sample B = 70% Wheat flour, 20% Soybean flour, and 10% Carrot flour

Sample C = 60% Wheat flour, 25% Soybean flour, and 15% Carrot flour

Sample D = 50% Wheat flour, 30% Soybean flour, and 20% Carrot flour

Sample E = 100% Wheat flour (Control)

**3.5.1 Loaf Test**

The result of the loaf test is shown in Table 7. The loaf test results provide valuable insights into the structural characteristics of the bread samples, particularly in terms of height, volume, weight, and mass-to-volume ratio. All samples had the same height of 13.5 cm, indicating that the rise during baking was consistent across the different formulations, suggesting uniformity in the baking process and gas retention.

When it comes to loaf volume, Samples A and B exhibited the highest volume at 500 g, which indicates better gas retention during baking, leading to lighter and airier bread. Sample E followed with a volume of 450 g, while Samples C and D had the lowest volumes at 400 g, which may indicate a denser crumb structure in these samples.

In terms of weight, Sample B and Sample E were the heaviest, each weighing 300 g, suggesting they are denser loaves. Samples A and C were lighter at 250 g, while Sample D weighed 280 g, reflecting some variation in the overall bread density among the samples.

The mass-to-volume ratio further highlights the density of the bread. Sample D had the highest ratio at 0.70 g/cm³, indicating it was the densest loaf, followed by Sample E at 0.67 g/cm³ and Sample C at 0.63 g/cm³. Sample B had a ratio of 0.60 g/cm³, while Sample A was the lightest and most aerated loaf with a ratio of 0.50 g/cm³. This suggests that Sample A had a lighter, more open crumb structure, while Samples D and E were firmer and denser.

Sample A is characterized by a lighter, more aerated texture, making it potentially more appealing to consumers who prefer soft bread. On the other hand, Samples D and E are denser and firmer, which might suit those who prefer more substantial, heavier loaves. The results highlight the diversity in texture and density among the different bread formulations.

Table 7: Loaf Test Result

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **SAMPLE** | **HEIGHT**  **(cm)** | **VOLUME**  **(g)** | **WEIGHT**  **(g)** | **MASS/VOLUME**  **(g/cm³)** |
| A | 13.5 | 500 | 250 | 0.50 |
| B | 13.5 | 500 | 300 | 0.60 |
| C | 13.5 | 400 | 250 | 0.63 |
| D | 13.5 | 400 | 280 | 0.70 |
| E | 13.5 | 450 | 300 | 0.67 |

Key:

Sample A = 85% Wheat flour, 10% Soybean flour, and 5% Carrot flour

Sample B = 70% Wheat flour, 20% Soybean flour, and 10% Carrot flour

Sample C = 60% Wheat flour, 25% Soybean flour, and 15% Carrot flour

Sample D = 50% Wheat flour, 30% Soybean flour, and 20% Carrot flour

Sample E = 100% Wheat flour (Control)

Based on the loaf test results, there is no significant difference in the height of the bread samples, as all samples had the same height of 13.5 cm. However, there is a notable difference in the volume, weight, and mass-to-volume ratios among the samples, with Sample A being the lightest and most aerated, and Sample D being the densest. These variations suggest significant differences in bread density and texture across the different formulations.

**3.6 Microbial Analysis**

The result of the Microbial Analysis is shown in Table 8. Microbial analysis is a critical aspect of food safety, ensuring that products are free from harmful microorganisms such as bacteria, yeast, and molds that can cause foodborne illnesses or spoilage. Microbial load, especially total plate count (TPC), coliform count, and yeast and mold count, provides insights into the hygiene and storage conditions of food products (Ray & Bhunia, 2019). In this study, the microbial safety of bread samples was assessed to ensure their suitability for consumption.

Sample A exhibited the highest total plate count (TPC) at 3.60 × 10³ CFU/g, indicating a higher microbial load compared to the other samples. While this value is still within acceptable limits as reported by the International Commission on Microbiological Specifications for Foods (ICMSF, 2021), the elevated microbial load suggests that Sample A might have a shorter shelf life and be more prone to spoilage if not stored properly. To mitigate further microbial growth, Sample A would require stricter storage controls such as refrigeration or airtight packaging.

In contrast, Sample D showed the lowest total plate count at 1.50 × 10³ CFU/g, indicating better microbial stability and a lower risk of spoilage. This suggests that Sample D was processed and stored under more hygienic conditions or that its composition offers better natural resistance to microbial growth. As noted in research by Ray & Bhunia (2019), lower microbial counts indicate longer shelf life and better food safety, making Sample D a favorable option for consumers concerned with spoilage and contamination.

None of the samples tested positive for coliform bacteria, which are indicators of fecal contamination or unsanitary processing conditions. In agreement with work done by Jay et al. (2020), the absence of coliform bacteria is a positive sign of proper hygiene during production, reducing the risk of foodborne illnesses. This suggests that all samples were handled and processed under sanitary conditions.

The yeast and mold counts are also important indicators of potential fungal spoilage. Sample B had the highest yeast and mold count at 4.90 × 10² CFU/g, indicating a higher risk of fungal contamination. Though this count does not pose an immediate health risk, it suggests that Sample B may have a shorter shelf life compared to the other samples and may require additional preservation methods, such as freezing, to prevent fungal growth. Sample C exhibited the lowest yeast and mold count at 1.80 × 10² CFU/g, indicating that it is less prone to fungal spoilage and might have better storage stability.

Sample D emerges as the best option from a microbial safety perspective, with the lowest TPC and no coliform contamination, suggesting longer shelf stability and lower risk of microbial spoilage. Sample A, despite having a slightly higher microbial load, remains safe for consumption but would benefit from stricter storage conditions. Sample B, with its higher yeast and mold count, may be more susceptible to fungal contamination but still meets acceptable safety standards. All samples showed no signs of coliform contamination, confirming their hygienic processing and handling, as reported by Ray & Bhunia (2019).

Table 8: Microbial Analysis Result

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **SAMPLE** | **TVC (104)** | **TVC**  **(105)** | **COLIFORM**  **(104)** | **COLIFORM**  **(105)** | **LAB**  **(104)** | **LAB**  **(105)** | **PDA (104)** | **PDA**  **(105)** |
| A | Nil | Nil | Nil | 2 | Nil | Nil | 5 | 20 |
| B | 1 | 1 | Nil | Nil | 1 | Nil | 1 | 27 |
| C | 1 | 1 | Nil | Nil | Nil | Nil | 6 | 35 |
| D | 6 | 2 | Nil | 2 | 8 | 6 | 38 | 27 |
| E | 12 | 14 | Nil | 8 | 4 | 9 | 29 | 37 |

Mean values are of duplicates ± standard deviation. The value within the rows with different superscripts is significantly (P<0.05) different.

Key:

Sample A = 85% Wheat flour, 10% Soybean flour, and 5% Carrot flour

Sample B = 70% Wheat flour, 20% Soybean flour, and 10% Carrot flour

Sample C = 60% Wheat flour, 25% Soybean flour, and 15% Carrot flour

Sample D = 50% Wheat flour, 30% Soybean flour, and 20% Carrot flour

Sample E = 100% Wheat flour (Control)

**3.7 Sensory Evaluation**

The result of the sensory evaluation as shown in Table 9. The sensory analysis of food products is essential for assessing their acceptability and consumer preference based on key quality attributes such as taste, texture, aroma, appearance, color, and overall acceptability. These factors play a significant role in determining the palatability of the bread samples produced from wheat, soybean, and carrot composite flours. As reported by Stone and Sidel (2019), sensory evaluation is crucial for understanding consumer preferences and enhancing product development in the food industry.

In this study, Sample B received the highest score for taste, with an average of 7.80 ± 1.23. This indicates that it was the most well-received in terms of flavor, suggesting a balanced and pleasing taste profile that resonated with the panelists. In agreement with work done by Meilgaard et al. (2016), a well-balanced flavor is crucial for consumer acceptance and overall satisfaction with the product. Sample C followed closely, scoring 7.60 ± 1.17, showing that it was also well-liked but slightly less favored than Sample B. Conversely, Samples A, D, and E received lower taste scores, ranging from 6.50 to 6.80, indicating they were acceptable but did not stand out as much in flavor.

Texture is another critical sensory attribute influencing overall enjoyment. Sample C excelled in this category, scoring 7.80 ± 1.14, suggesting it had the most appealing texture characterized by a favorable combination of softness and chewiness. This aligns with findings by Szczesniak (2018), who emphasizes that texture greatly affects consumer preferences for baked goods. Sample E also scored well for texture at 7.40 ± 1.17. In contrast, Sample A scored the lowest at 5.60 ± 2.68, indicating potential issues with its mouthfeel.

Aroma contributes significantly to the sensory experience and can impact taste perception. In research by Lawless and Heymann (2010), aroma is recognized as a key driver in initial consumer impressions of food products. Sample C scored the highest in aroma at 7.60 ± 1.35, indicating a pleasant scent that likely enhanced its overall appeal. Sample E also scored well at 7.20 ± 1.14, while Samples B, D, and A received moderate scores between 6.20 and 6.90.

Appearance is crucial as it sets the first impression of a food product, influencing consumer expectations. Sample C received the highest appearance score of 6.90 ± 1.73, indicating it was visually appealing. This is essential as visual characteristics can significantly impact consumer choice (Hutchings, 2019).

Overall acceptability combines all sensory attributes and reflects the general impression of the product. Sample B had the highest overall acceptability score at 6.40 ± 1.58, suggesting it was the most preferred sample among panelists. Sample C followed closely with a score of 6.30 ± 2.16. The lower overall acceptability scores of Samples D and E (5.90 each) indicate that they were generally acceptable but did not stand out as much as Samples B and C.

Sample B was the most well-received overall, with high scores in taste and overall acceptability, making it the best-balanced sample in terms of sensory attributes. Sample C also performed well, particularly in texture, aroma, and appearance. The sensory analysis reveals clear preferences for Samples B and C, highlighting the importance of sensory attributes in consumer acceptance of bread products.

Table 9: Sensory Evaluation

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| SAMPLE | TASTE | TEXTURE | AROMA | APPEARANCE | COLOUR | OVERALL  ACCEPTABILITY |
| A | 6.80±1.55a | 5.60±2.68 a | 6.20±1.62 a | 5.70±1.16 a | 6.00±2.05 a | 5.50±1.51 a |
| B | 7.80±1.23 a | 6.50±1.71 a | 6.70±2.26 a | 6.50±1.84 a | 6.50±1.58 ab | 6.40±1.58 a |
| C | 7.60±1.17 a | 7.80±1.14 b | 7.60±1.35 a | 6.90±1.73 a | 7.70±0.68 b | 6.30±2.16 a |
| D | 6.50±1.51 a | 7.00±1.16 a | 6.90±1.29 a | 6.80±1.23 a | 6.80±1.30 ab | 5.90±1.73 a |
| E | 6.80±1.14 a | 7.40±1.17 b | 7.20±1.14 a | 6.80±1.93 a | 6.80±1.14 ab | 5.90±2.03 a |

Mean values are of duplicates ± standard deviation. The value within the rows with different superscripts is significantly (P<0.05) different.

Key:

Sample A = 85% Wheat flour, 10% Soybean flour, and 5% Carrot flour

Sample B = 70% Wheat flour, 20% Soybean flour, and 10% Carrot flour

Sample C = 60% Wheat flour, 25% Soybean flour, and 15% Carrot flour

Sample D = 50% Wheat flour, 30% Soybean flour, and 20% Carrot flour

Sample E = 100% Wheat flour (Control)

4. Conclusion

This study emphasises the significant potential of enriching wheat-based bread by adding soybean and carrot flours to enhance its nutritional, functional, microbial, and sensory properties. Different combinations of these composite flours notably influenced various aspects of the bread, including nutrient content, vitamin and mineral levels, antinutrients, functional characteristics, safety, and consumer acceptance. For example, Sample D, which contains 50% wheat, 30% soybean, and 20% carrot, emerged as the most nutrient-dense option. It exhibited higher levels of protein, calcium, potassium, and iron, along with excellent functional traits such as water absorption, swelling capacity, and solubility. Its lower microbial count also indicated it could remain fresh longer. Conversely, Sample B, composed of 70% wheat, 20% soybean, and 10% carrot, was the most preferred in taste tests and overall acceptability, demonstrating a good balance between health benefits and flavor.

Overall, these findings suggest that blending wheat with soybean and carrot flours can produce affordable, nutritious bread options that are especially beneficial for resource-limited communities. This approach promotes the use of local crops, encourages dietary diversity, and offers a practical solution to address nutritional deficiencies in low- and middle-income populations. Future research could focus on optimising the blend ratios and examining storage stability over time to ensure these breads are both functional and appealing in the marketplace.

Consent (NOT applicable)

Ethical approval (NOT applicable)

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