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Preparation and Evaluation of High-Nutritional Value Biscuits from Whole Wheat flour, Carrot and Whey Protein Powder

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ABSTRACT

Biscuits are among the most popular ready-to-eat foods globally, valued for their low cost and long shelf life. They are not only a convenient snack but also a versatile product that can be enriched to improve nutritional content, meet dietary needs, and address certain health concerns. This study aimed to develop nutrient-enriched biscuits by fortifying them with whole wheat flour (WWF), whey protein concentrate (WPC), and carrot powder (CP) in varying proportions: blend 1 (control, 100% WF), blend 2 (95% WWF + 5% CP), blend 3 (85% WWF + 5% CP + 10% WPC), blend 4 (80% WWF + 5% CP + 15% WPC), and blend 5 (75% WWF + 5% CP + 20% WPC). The results showed that WPC was high in crude protein and calcium and provided essential amino acids such as lysine and leucine. WWF, on the other hand, excelled in non-essential amino acids like glutamic acid and proline, while CP contributed high fiber and mineral content, including a notable amount of iron. Texture analysis indicated increased hardness and fracturability with WPC fortification. Sensory evaluation revealed that blends 3 and 4, which incorporated WPC, improved taste and overall acceptability. However, excessive WPC in blend 5 led to undesirable changes in texture and color. These findings suggest that WWF, WPC, and CP offer complementary nutritional profiles, which can enhance the nutritional value and sensory qualities of biscuits, resulting in a functional food product with potential health benefits.

1. Introduction

Wheat (*Triticum aestivum* L.), a member of the diverse Poaceae family, is the primary cereal crop for much of the global population. Cultivated worldwide, wheat supplies roughly half of the world's dietary calories and is rich in proteins (especially gluten), essential minerals (such as copper, magnesium, zinc, phosphorus, and iron), B-group and E vitamins, riboflavin, niacin, thiamine, and dietary fiber (El-Hadidy et al.,2023 and Khalid et al., 2023). Whole wheat, in particular, is a major source of plant-based protein, containing about 13% protein, which is high compared to other cereal grains. It is a substantial source of energy from its carbohydrates and gluten protein, while its bran layers contain phosphates and other essential minerals. Whole wheat also offers dietary fiber, supporting digestive health, and its protein, along with B and E vitamins, aids in muscle development and repair. The wheat germ, typically removed during refinement, is rich in vitamin E, a key nutrient (Iqbal et al., 2022; El-Hadidy et al., 2023 and Mospah et al., 2024). Studies show that whole wheat consumption is inversely related to the risk of chronic diseases such as obesity, type 2 diabetes, cardiovascular disease, and cancer. These health benefits are primarily due to its bioactive components, including phytochemicals and dietary fiber (El-Dreny et al., 2018 and Saini et al., 2021). *Daucus carota L.,* commonly known as the carrot, is a root vegetable celebrated for its nutritional value and health benefits.

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Carrots can be economically beneficial as an ingredient in products like biscuits, bread, cakes, and various functional foods. Studies show that carotenoids, polyphenols, and vitamins in carrots have anti-carcinogenic, antioxidant, and immune-boosting properties. Numerous in-vivo and in-vitro studies highlight carrots' broad health benefits, including cholesterol reduction, blood sugar management, blood pressure regulation, kidney and liver protection, and aiding liver fat and bile excretion. Additionally, extracts from carrot seeds exhibit antibacterial, antifungal, anti-inflammatory, cardioprotective, and hepatoprotective effects (El-Hadidy, et al., 2020 and Ikram et al., 2024). In its dehydrated form, carrot powder serves as a concentrated nutrient source. This nutrient-rich powder contains carotenoids, dietary fiber, and significant phenolic compounds, along with high levels of vitamin A (Jacobo, 2023). Carrot roots can be processed into various nutrient-dense products, such as juice, concentrate, dehydrated powder, canned goods, preserves, and pickles (Varshney and Mishra, 2022). Whey Protein Concentrate (WPC) is a nutrient-dense product with a high biological value, containing 34–80% protein. While WPC is widely used in protein-enriched foods like biscuits, excessive amounts may lead to an undesirably hard texture, which can reduce product palatability (Andoyo et al., 2023). Often utilized for muscle development and weight management, WPC is valued for its diverse amino acid profile (El-Aidie and Khalifa, 2024). It contains 162 amino acid residues, including all nine essential amino acids, with major components such as β-lactoglobulin (BLG), α-lactalbumin (α-LA), and bovine serum albumin (BSA) immunoglobulins. WPC is also rich in phospholipids. As protein concentration increases, lactose levels in WPC decrease, and its solubility is inversely related to temperature. Sodium caseinate in WPC helps it absorb water efficiently, and protein chain unfolding contributes to gel formation. Incorporating WPC into foods typically boosts protein, carbohydrate, and mineral content, while reducing fat and moisture levels (Chahatdeep et al., 2021). Commercially available biscuits typically contain high levels of fat, sugar, and various additives, but are low in fiber, vitamins, minerals, and protein (around 7–8%). This composition makes them an unhealthy snack option in the long term, potentially contributing to various health issues (Salihu et al., 2023). Protein content in biscuits can be increased by incorporating protein-rich ingredients, with whey protein being a prime option (Andoyo et al., 2024). This research aimed to develop nutritionally enhanced biscuits by incorporating high-value ingredients, such as whole wheat flour, carrot powder, and whey protein concentrate.Carrots can be economically beneficial as an ingredient in products like biscuits, bread, cakes, and various functional foods. Studies show that carotenoids, polyphenols, and vitamins in carrots have anti-carcinogenic, antioxidant, and immune-boosting properties. Numerous in-vivo and in-vitro studies highlight carrots' broad health benefits, including cholesterol reduction, blood sugar management, blood pressure regulation, kidney and liver protection, and aiding liver fat and bile excretion. Additionally, extracts from carrot seeds exhibit antibacterial, antifungal, anti-inflammatory, cardioprotective, and hepatoprotective effects (El-Hadidy, et al., 2020 and Ikram et al., 2024). In its dehydrated form, carrot powder serves as a concentrated nutrient source. This nutrient-rich powder contains carotenoids, dietary fiber, and significant phenolic compounds, along with high levels of vitamin A (Jacobo, 2023). Carrot roots can be processed into various nutrient-dense products, such as juice, concentrate, dehydrated powder, canned goods, preserves, and pickles (Varshney and Mishra, 2022). Whey Protein Concentrate (WPC) is a nutrient-dense product with a high biological value, containing 34–80% protein. While WPC is widely used in protein-enriched foods like biscuits, excessive amounts may lead to an undesirably hard texture, which can reduce product palatability (Andoyo et al., 2023). Often utilized for muscle development and weight management, WPC is valued for its diverse amino acid profile (El-Aidie and Khalifa, 2024). It contains 162 amino acid residues, including all nine essential amino acids, with major

components such as β-lactoglobulin (BLG), α-lactalbumin (α-LA), and bovine serum albumin (BSA) immunoglobulins. WPC is also rich in phospholipids. As protein concentration increases, lactose levels in WPC decrease, and its solubility is inversely related to temperature. Sodium caseinate in WPC helps it absorb water efficiently, and protein chain unfolding contributes to gel formation. Incorporating WPC into foods typically boosts protein, carbohydrate, and mineral content, while reducing fat and moisture levels (Chahatdeep et al., 2021). Commercially available biscuits typically contain high levels of fat, sugar, and various additives, but are low in fiber, vitamins, minerals, and protein (around 7–8%). This composition makes them an unhealthy snack option in the long term, potentially contributing to various health issues (Salihu et al., 2023). Protein content in biscuits can be increased by incorporating protein-rich ingredients, with whey protein being a prime option (Andoyo et al., 2024).

This research aimed to develop nutritionally enhanced biscuits by incorporating high-value ingredients, such as whole wheat flour, carrot powder, and whey protein concentrate.

2. Materials and Methods

This study was conducted in 2024 at the Food Technology Research Institute, Bread and Pastries Technology Research Laboratory, Agricultural Research Center, Giza, Egypt

Materials

Wheat flour (*Triticum Aestivum*) (100% extraction) was purchased from the North Cairo Flour Mills Company, Egypt. Commercial whey protein concentrate (WPC-8000) was obtained from Hilmar Ingredients (Hilmar 8000, Hilmar, CA, USA) with approximately 80.4% protein**.** The necessary components for biscuit production, including butter, eggs, sugar (sucrose), baking powder, sodium bicarbonate, and vanilla, were sourced from a local market in Egypt. The chemicals and reagents used in this study were of high quality and obtained from Sigma-Aldrich, USA.

Methods

Preparation of whole wheat flour

Whole wheat grains were processed into flour following the method described by Ndife et al., (2014). Initially, the grains were cleaned to remove impurities like twigs, foliage, and grit. After rinsing with clean water, the grains were dehydrated at 55° C in a standard air oven. Subsequently, the dehydrated grains were ground using an attrition mill and sieved through a 425 µm mesh screen. The resulting flour was stored in an airtight container, labeled appropriately, and refrigerated at 4°C for future use.

Preparation carrot powder

The method adapted from Marvin (2009) was used to prepare carrot powder. Carrot roots were washed in tap water, peeled, and sliced. The slices were soaked in heated water containing sodium metabisulphite for 3 minutes to prevent browning and discoloration. After air-cooling, the slices were dried in a solar oven at 50 ± 5°C until the moisture content reached less than 8%. The dried material was milled using a Moulinex mill (Al Araby for Electronic Manufacture Company, Egypt) and stored in polyethylene bags at -18 ± 2°C until further analysis and use.

Preparation of sweet biscuits

To prepare the biscuits, sugar and butter were creamed together for 1 minute using a mixing machine. Whipped eggs and vanilla extract were then added to the creamed mixture and mixed at low speed for 5 minutes. Dry ingredients, such as wheat flour or its blends and baking powder, were combined and gradually incorporated into the wet ingredients, mixing continuously until a smooth dough formed. The dough was allowed to rest for 15 minutes. The rested dough was rolled out on a cookie sheet using a guide roll, cut into 5 cm diameter circles, and rolled to a thickness of 0.3 cm. The cut biscuits were transferred to a greased baking sheet and baked in an electric oven at 170°C for 12-15 minutes. After baking, the biscuits were cooled at room temperature for 1 hour before sensory evaluation (AACC, 2010).

The recipe for semi-hard sweet biscuits followed the standard procedure used by Bisco Misr Company in Cairo, Egypt, as outlined in Table 1 and described by El-Hadidy et al., (2022).

Table 1. The formula used for preparing semi hard sweet biscuits

Ingredients (g)	Control 1	Blend 2	Blend 3	Blend 4	Blend 5
WWF	100	95	85	80	75
WPC	-	-	10	15	20
CP	-	5	5	5	5
Butter	15	15	15	15	15
Sugar	30	30	30	30	30
Fresh egg	24	24	24	24	24
Ammonium bicarbonate	0.66	0.66	0.66	0.66	0.66
Sodium bicarbonate	0.33	0.33	0.33	0.33	0.33
Skimmed milk	1	1	1	1	1
Vanilla	0.3	0.3	0.3	0.3	0.3

WWF= Whole wheat flour WPC=Whey protein concentrate CP=Carrot powder

Sensory characteristics of biscuits

Sensory evaluation was conducted to assess the appearance, texture, odor, taste, color, and overall acceptability of the product. A 20-point numerical hedonic scale (1 = very bad, 20 = excellent) was used, following the method described by Smith (1972). Twenty experienced judges from the Agricultural Research Center in Giza, Egypt, participated in the evaluation.

Determination of chemical analysis

Chemical composition analysis

Moisture, protein, ash, crude fat, crude fiber content, peroxide value, and acid value were determined according to the AOAC (2012) method.

- Available carbohydrates content was calculated by difference:

%Available carbohydrates = 100 - (%Ash+%Fiber+ %Fat + %Protein)

- Approximate energy content was calculated according to FAO/WHO (1974):

Total energy (Kcal/100g) = 4.1(%carbohydrate + % protein) + 9.1(%fat)

- Mineral content was determined by Atomic Absorption Spectrophotometer (3300 Perkin-Elmer) as described in AOAC (2012).

Amino acids analysis

Amino acids were determined according to AOAC (2012) using a high-performance Amino Acids Analyzer (Biochroom 30). Tryptophan content was determined colorimetrically in the alkaline hydrolyzate using P-dimethyl-amino-benzaldehyde (DMAB) following the method of Miller (1967).

Protein quality evaluation

Computed protein efficiency ratio (C-PER) was estimated using the equation reported by Alsmeyer et al. (1974):

C-PER = -0.4687 + 0.454(Leucine) - 0.105(Tyrosine)

Biological value (BV) was calculated as described by Farag et al. (1996):

$$BV = 49.9 + 10.53 * \text{C-PER}$$

Water activity measurement

Water activity (aw) was measured with a Rotronic (model Hy-grolab3-Switzerland). The ground biscuit sample was placed in a plastic cup, and the Hygroplam probe was inserted. After a few minutes, the display showed the water activity and temperature (Pigaet et al., 2005).

Color measurements of sweet biscuits

The crust color of the different biscuit types (WWF, WPC, CP, and blends) was determined using a method adapted from Tong et al. (2010). A Konica Minolta CR-410 Chroma meter was used to measure the L^*, a^*, and b^* values of the biscuit crusts. These values represent L^* (lightness, with higher values indicating lighter colors), a^* (red-green axis, with positive values indicating red and negative values indicating green), and b^* (yellow-blue axis, with positive values indicating yellow and negative values indicating blue).

Physical properties of sweet biscuits

According to El-Hadidy et al. (2022), the diameter (D) and thickness (T) of five biscuits were measured in millimeters by placing them edge-to-edge and stacking them, respectively. To obtain average measurements, the biscuits were rearranged and restacked. The spread ratio was calculated by dividing the diameter of the biscuit (mm) by its thickness (mm). After cooling, the weight of five biscuits was determined. The volume was calculated using the formula for the area of a circle:

$$A=\Pi r^2$$

where: r^2= square of the radius of biscuit circle

Specific volume was calculated by dividing volume (cm^3) by biscuit weight (g).

Texture profile analysis of sweet biscuits

Texture profile analysis was performed using a TA-XT2 Plus texture analyzer from Stable Microsystems. The test involved two compression cycles using a 75 mm diameter flat probe (P/75), with a 5-second interval between cycles. The test parameters were: 30 kg force load cell, pre-test, test, and post-test speeds of 1.0mm/s, a distance of 4 mm, and a trigger force of 0.1N. The textural properties of hardness and fracturability were calculated according to Correa et al., (2017).

Statistical analysis

Data from the different experiments were recorded as the means ± standard deviation (SD) of triplicate measurements and were analyzed by SPSS 26.0 software (SPSS, Inc., Chicago, IL, USA). The results were submitted to analysis of variance (ANOVA) and Duncan's multiple range test for means comparison at ($p \leq 0.05$) significance was applied

3. Results and Discussion

Chemical composition of WWF, WPC, and CP (g /100 g .D.W)

The data in Table 2 present the compositional analysis of WWF, WPC, and CP.

Results indicate that WWF has a moisture content of 13.50%, which is significantly higher than CP (7.60%) but is significantly higher than WPC (3.4%). Rodrigues et al., (2023) reported different moisture content for WWF (7.64%), though it was higher than that reported by Zaki and Hussien (2018). Meanwhile, WPC shows a markedly high crude protein content at 75.03%, significantly exceeding that of WWF (14.32%), a finding also observed by Beshir et al. (2021). The crude ether extract content in WPC (6.3%) is significantly higher than that in WWF (2.45%) and CP (1.94%). Conversely, CP exhibits the highest crude fiber content at 10.35%, significantly greater than that of WWF (3.12%) and WPC, which contains no fiber. This finding aligns with El-Hadidy, et al., 2020; Varshney and Mishra (2022); and Singh et al. (2021), who reported that carrots, as an important root crop, are rich in biochemical components like carotenoids and soluble fiber.

Ash content is highest in CP at 6.43%, significantly differing from both WWF (1.6%) and WPC (2.87%).

Whole wheat flour has the highest available carbohydrate content at 74.38%, significantly greater than that of WPC (15.83%), although CP, with available carbohydrate content of 78.55%, significantly differ from WWF in this respect.

WPC has the highest energy content at (429.73Kcal/100g), significantly greater than that of Whole wheat flour (402.90Kcal/100g), although CP, with available carbohydrate content of (350.90Kcal/100g), significantly differ from WWF in this respect. Overall, these results are consistent with Zaki and Hussien (2018), who reported that WWF contained 12.92% moisture, 14.66% protein, 2.88% fat, 3.59% fiber, 1.58% ash, and 77.29% carbohydrates. However, the values reported here are generally higher than those observed by Rodrigues et al. (2023), except for total dietary fiber, which was 15.10% higher in Rodrigues et al.,'s findings.

Table 2.Chemical composition of WWF, WPC, and CP (g /100 g .D.W)

Materials	Moisture %	Crude Protein %	Crude ether extract %	Crude Fiber %	Ash %	ATC %	Energy (Kcal/100g)
WWF	13.50[a]±0.01	14.32[b]±0.03	2.45[b]±0.04	3.12[b]±0.06	1.60[c]±0.01	78.51[a]±0.04	402.90[b]±0.04
WPC	3.40[b]±0.09	75.03[a]±0.50	6.30[a]±0.05	----	2.87[b]±0.04	15.80[c]±0.04	429.73[a]±0.04
CP	7.60[c]±0.07	6.90[c]±0.05	1.94[c]±0.02	10.35[a]±0.10	6.43[a]±0.09	74.38[b]±0.04	350.90[c]±0.04

- Values followed by the same letter in columns are not significantly different at LSD at ($p \leq 0.05$).
- Each value was an average of three determinations ± standard deviation.
- WWF= Whole wheat flour WPC=Whey protein concentrate CP=Carrot powder ATC=Available total carbohydrates

Minerals content (mg /100g) of WWF, WPC, and CP

Minerals are vital components of our diet, serving a variety of functions, such as providing structural support for bones, facilitating nerve function, and regulating the body's water balance. They are also integral components of hormones, enzymes, and other biologically active compounds. Additionally, some minerals play key roles in the immune system; accordingly, mineral intake can influence susceptibility to infections and impact the development of chronic diseases (Weyh et al., 2022). The mineral content of WWF, WPC, and CP is presented in Table 3. Whole wheat flour is notable for its high potassium (K) content at 560 mg/100g, phosphorus (P) at 420 mg/100g, and magnesium (Mg) at 185 mg/100g. However, it has relatively lower concentrations of calcium (Ca) at 65 mg/100g and iron (Fe) at 7.5 mg/100g. Moreover, the mineral content of WWF was found to be higher than the values reported by Ertl and Goessler (2018) and Ahmed and Ashraf (2019). In contrast, WPC exhibits a higher calcium content of 650 mg/100g, which may enhance its value in calcium-rich dietary formulations. WPC also contains 590 mg of phosphorus and 240 mg of magnesium, though its iron content is markedly lower at 1.4 mg/100g. These findings align with those of Ahmed and Ashraf (2019), who reported that WPC had a low iron content. For CP, data show high calcium (690 mg/100g) and iron content (18 mg/100g), making it a promising candidate for dietary fortification, particularly for addressing iron deficiency. Notably, both WPC and CP have higher zinc (Zn) levels (8.9 mg and 7 mg, respectively) compared to WWF (4.9 mg). These values are higher than those reported by Purewal et al. (2023) and Ikram et al. (2024), yet consistent with findings by Kamel et al. (2023). These results highlight the importance of incorporating a variety of these sources to achieve optimal mineral intake for human health (El-Hadidy[b] et al.,2020).

Table 3. Minerals content (m g /100g) of WWF, WPC, and CP

Minerals (mg /100g)	WWF	WPC	CP
K	560[c]±1.90	850[a]±2.50	600[b]±3.70
Ca	65[c]±0.90	650[b]±1.60	690[a]±2.5
P	420[b]±1.50	590[a]±2.30	180[c]±0.9
Mg	185[b]±1.00	240[a]±2.40	165[c]±0.80
Fe	7.5[b]±0.4	1.40[c]±0.04	18[a]±0.05
Mn	6.40[a]±0.04	1.30[c]±0.03	3.50[b]±0.01
Zn	4.90[c]±0.05	8.90[a]±0.04	7.00[b]±0.01

- Values followed by the same letter in columns are not significantly different at LSD at ($p \leq 0.05$).
- Each value was an average of three determinations ± standard deviation.
- WWF= Whole wheat flour WPC=Whey protein concentrate CP=Carrot powder

Amino acids composition (g/100g Protein) of WWF and WPC

The amino acid composition of WWF and WPC is presented in Table 4. The data show that WWF contains 2.85g of lysine per 100g of protein, significantly below the FAO/WHO/UNU standard of 5.80g. This aligns with findings by Laze et al. (2019), who reported that WWF is notably low in lysine, tryptophan, and methionine.

In contrast, WPC has a lysine content of 9.20 g, which exceeds the recommended level, indicating its superior lysine concentration. Both WWF and WPC meet or exceed the isoleucine and leucine recommendations, with WWF containing 4.35 g and 6.63 g, respectively, and WPC containing 4.80 g and 11.80 g. The amino acid content in WWF and WPC is generally higher than values reported by Ahmed and Ashraf (2019) and is consistent with those found by Hulmi et al., (2010) and El-Azab (2019). Auestad and Layman (2021) highlighted that WPC has gained popularity as a protein supplement for muscle health due to its high concentration of leucine (12% w/w). Whey proteins, in highly purified forms, are also widely included in infant formulas to enhance protein quality. On the other hand, WWF exceeds the histidine recommendation (2.30g vs. 1.90g) but falls short in methionine (1.40 g vs. 2.20g), while WPC meets this requirement (2.60g). Additionally, WPC has higher levels of tryptophan and cysteine compared to WWF. Although whole wheat flour provides a total essential amino acid (EAA) content of 35.58 g, its lysine and methionine deficiencies underscore the importance of dietary variety to ensure adequate EAA intake, particularly for isoleucine and leucine. WWF contains 30.80 g of glutamic acid per 100 g of protein, which is considerably higher than the 16.50 g found in WPC. However, whey protein has higher levels of serine and aspartic acid (3.40 g and 10.00 g, respectively) compared to whole wheat flour (4.80g and 5.56 g, respectively), while proline is significantly higher in whole wheat flour (9.50g) than in WPC (4.90g). Additionally, glycine content is greater in whole wheat flour (5.19g) than in WPC (1.60g). Whey protein has a slightly higher alanine content (4.70g) compared to whole wheat flour (3.60g), while arginine is more abundant in WWF (4.80g) than in whey protein (2.40g). These findings suggest that while whole wheat flour is rich in several non-essential amino acids, whey protein provides a more balanced profile of essential amino acids. This supports the importance of combining both sources for a comprehensive amino acid intake, consistent with Park et al. (2020) and in alignment with the FAO/WHO/UNU (1985) recommendations. Zhao et al. (2022) reported that whey proteins have a high biological value, exceeding that of egg protein, and are a rich source of essential and branched-chain amino acids.

Table 4. Amino acids composition (g amino acid / 100 g Protein) of WWF and WPC

Amino acids	WWF	WPC	FAO/WHO/UNU (1985) pattern
Lysine	2.85	9.20	5.80
Isoleucine	4.35	4.80	2.80
Leucine	6.63	11.80	6.60
Phenylalanine	4.80	3.20	6.30
Tyrosine	2.70	3.50	
Histidine	2.30	1.80	1.90
Valine	4.40	4.90	3.5
Threonine	2.90	4.70	3.40
Methionine	1.40	2.60	2.20
Tryptophan	1.30	2.50	1.00
Cysteine	1.950	3.00	
Total (EAA)	35.58	52	
Aspartic acid	5. 56	10.00	
Glutamic acid	30.80	16.50	
Serine	4.80	3.40	
Proline	9.50	4.90	
Glycine	5.19	1.60	
Alanine	3.60	4.70	
Arginine	4.80	2.40	
Total (NEAA)	64.02	43	
C-PER	2.26	4.52	
BV	73.70	97.50	

-EAA: Essential amino acids. NEAA: Nonessential amino acids -C-PER = Computed protein efficiency ratio.
- BV = Biological value - WWF= Whole wheat flour WPC=Whey protein concentrate

Color parameters of WWF, WPC, and CP

The data in Table 5 present the color characteristics of WWF, WPC, and CP as measured by their *L**, *a**, and *b** values. The *L** value, which represents lightness, was highest for WPC at 63.45, indicating it was the lightest among the samples. In contrast, CP had the lowest *L** value at 18.22, making it the darkest material, while WWF had an intermediate lightness value of 25.91. Regarding the *a** value (redness or greenness), CP displayed the highest redness with an *a** value of 12.48, whereas WPC had a slightly greenish hue with a negative *a** value of -0.24. WWF showed moderate redness with an *a** value of 4.3. Finally, the *b** value (yellowness or blueness) was most pronounced in CP at 41.69, highlighting its strong yellow coloration. WPC also exhibited notable yellowness with a *b** value of 15.87, while WWF had the lowest *b** value at 12.38, indicating a relatively muted yellow hue.

Table 5. Color parameters of WWF, WPC, and CP

Materials	L* (Lightness)	a* (Redness／greenness)	L* (Yellowness／blueness)
WWF	25.91^{b}±0.03	4.30^{b}±0.08	12.38^{c}±0.21
WPC	63.45^{a}±0.02	-0.24^{c}±0.02	15.87^{b}±0.03
CP	18.22^{c}±0.07	12.48^{a}±0.17	41.69^{a}±0.03

- Values followed by the same letter in columns are not significantly different at LSD at ($p \leq 0.05$).
- Each value was an average of three determinations ± standard deviation.
- WWF= Whole wheat flour WPC=Whey protein concentrate CP=Carrot powder

Chemical composition of biscuits produced from of WWF, WPC, and CP (on dry weight basis)

Table 6 presents data on the nutritional composition of biscuits produced from WWF, WPC, and CP. Crude protein content varied significantly among all biscuit samples supplemented with different levels of WPC. Blend 5 had the highest protein content at 16.71%, followed by blend 4 at 14.94%, blend 3 at 13.1%, blend 2 at 9.77%, and the control at 9.98%. Blend 2 and the control had the lowest protein levels, with significant difference between them. On the other hand, ether extract values show significant variation across the biscuit samples: the control had 11.49%, blend 2 had 11.43%, blend 3 had 11.7%, blend 4 had 11.86%, and blend 5 had 11.91%. These results differ from those reported by Munaza et al. (2012), who found that the fat content of wheat/ whey protein biscuits slightly decreased with increased WPC levels. Apart from this, Crude fiber content also show significant differences among the biscuit samples, with values ranging from 7.97% in blend 3 to 7.66% in the control. This may be due to the lack of fiber in WPC. Similarly, ash content, representing total mineral content, remained consistent across all biscuit samples, with significant differences. Ash values ranged from 1.10% in the control to 1.36% in blend 5. These results align with those reported by Raghul (2023) for ash content but are lower in protein and ether extract content and higher in fiber content. Available carbohydrates content also show significant differences among the biscuit samples, with values ranging from 69.69% to 62.08% and 69.77%the control. Not only-but also, energy content also show significant differences among the biscuit blends, with values ranging from 429.80 to 431.42(Kcal/100g) and 431.53(Kcal/100g) the control. Zaki and Hussien (2018) reported that whole wheat flour biscuits contained 3.12% moisture, 1.56% ash, 17.22% protein, 13.54% fat, 3.15% fiber, and 64.53% carbohydrate.

Color parameters of biscuits produced from of WWF, WPC, and CP

Color is a crucial quality attribute in foods, influencing consumer perception and acceptance or rejection of products. In biscuits, color develops through the Maillard reaction during baking, which is responsible for the formation of color, taste, and especially aroma in thermally processed foods (Starowicz and Zieliński, 2019 and El-Hadidy et al., 2020). Table 7 presents the colorimetric values, including Lightness (*L**), Redness/Greenness (*a**), and Yellowness/Blueness (*b**), for biscuits made from WWF, WPC, and CP. The control biscuit sample exhibited the highest *L** value at 74.93, making it the lightest among all samples. Blend 2 showed a significant decrease in lightness to 62.9 compared to the control sample. Blends 3, 4, and 5 displayed progressively lower but significant lightness values at 58.47, 56.61, and 54.7, respectively. The reduction in *L** values may result from the formation of colored compounds through the Maillard reaction between lactose in WPC and free amino groups from lysine in the protein ingredients. The addition of WPC reduced lysine availability and increased color development, aligning with findings by Kheto et al., (2024). The *a** values, indicating redness or greenness, vary significantly across biscuit samples. Blend 5 had the highest *a** value at 8.14, while blends 2, 3, and 4 had *a** values of 7.38, 7.52, and 7.53, respectively. The increase in redness could be due to caramelization of sugars and the darkening effect of WPC during high-temperature baking, consistent with findings by Aryajaya (2020). On the other hand, the control sample had *b** value of 33.78, which was significantly lower than those of the other blends. The *b** values showed a significant increase across blends 2, 3, 4, and 5, with means ranging from 40.68 in blend 2 to 46.67 in blend 5. Overall, these results indicate that blend modifications had a more pronounced effect on Lightness and Yellowness, while Redness remained relatively stable.

Table 6. Chemical composition of biscuits produced from of WWF, WPC, and CP (on dry weight basis)

Materials		Crude Protein %	Ether Extract %	Crude Fiber %	Ash %	T.C %	ATC %	Energy (Kcal/100 g)
Biscuit's samples	Control 1	9.98^{d}±0.01	11.49^{d}±0.05	7.66^{d}±0.01	1.10^{e}±0.01	77.43^{b}±0.02	69.77^{a}±0.10	431.53^{a}±0.10
	Blend 2	9.77^{e}±0.01	11.43^{e}±0.03	7.90^{c}±0.02	1.21^{d}±0.01	77.59^{a}±0.05	69.69^{a}±0.15	429.80^{c}±0.05
	Blend 3	13.10^{c}±0.03	11.70^{c}±0.02	7.97^{b}±0.03	1.28^{c}±0.02	73.92^{c}±0.03	65.95^{b}±0.10	430.58^{b}±0.04
	Blend 4	14.94^{b}±0.03	11.86^{b}±0.01	7.91^{c}±0.02	1.32^{b}±0.01	71.88^{d}±0.02	63.97^{c}±0.02	431.46^{a}±0.20
	Blend 5	16.71^{a}±0.02	11.91^{a}±0.06	7.94^{a}±0.01	1.36^{a}±0.01	70.02^{e}±0.09	62.08^{d}±0.08	431.42^{a}±0.15

-Values followed by the same letter in columns are not significantly different at LSD at ($p \leq 0.05$).
-Each value was an average of three determinations ± standard deviation.
-ATC=Total carbohydrate = 100- (Crude Protein +Fiber+ Ether extract + Ash).
-ATC= Available Total carbohydrate

Table 7. Color parameters of biscuits produced from of WWF, WPC, and CP

Blends	*L** (Lightness)	*a** (Redness/ greenness)	*b** (Yellowness/blueness)
Control 1	74.93^{a}±0.10	6.33^{d}±0.03	33.78^{e}±0.05
Blend 2	62.9^{b}±0.20	7.38^{c}±0.01	40.68^{d}±0.03
Blend 3	58.47^{c}±0.30	7.52^{b}±0.05	43.55^{c}±0.04
Blend 4	56.61^{d}±0.20	7.53^{b}±0.02	45.79^{b}±0.02
Blend 5	54.70^{e}±0.25	8.14^{a}±0.05	46.67^{a}±0.05

-Values followed by the same letter in columns are not significantly different at LSD at ($p \leq 0.05$).
-Each value was an average of three determinations ± standard deviation.

Water activity (aw) of WWF biscuit supplemented with CP and different levels WPC

Water activity (a_w) is a crucial factor in controlling microbial hazards or spoilage, as it prevents or limits microbial growth. In many cases, aw is the primary parameter influencing food stability, modulating microbial response, and determining the types of microorganisms present in food (Tapia et al., 2020). Table 8 shows the water activity (a_w) values of the control biscuit sample and various WWF biscuit blends supplemented with CP and different levels of WPC.

Results indicate that the control sample exhibited the highest aw at 0.55, closely followed by blend 2 with an a_w value of 0.54, with no significant difference between them. Blend 3 had a slightly lower aw at 0.53, followed by blend 4 at 0.46. Blend 5 had a significantly lower aw than all other biscuit samples, in line with findings by Beshir et al., (2021). The a_w values for all biscuits are below the recommended levels for bacterial growth (a_w > 0.91) and mold growth (a_w > 0.81), which could impact the shelf life, texture, and susceptibility to microbial growth. Generally, lower aw values are associated with increased stability and longer shelf life (Tapia et al., 2020).

Table 8. Water activity (a_w) of whole wheat flour biscuit supplemented with carrot powder and different levels whey protein concentrate

Blends	Water activity	°C
Control1	0.55^a ±0.004	33.26 - 33.30
Blend 2	0.54^a ±0.004	33.16 – 33.24
Blend 3	0.53^b ±0.005	33.33 – 33.37
Blend 4	0.46^c ±0.004	33.25 – 33.31
Blend 5	0.36^d ±0.004	33.16 – 33.22

-Values followed by the same letter in columns are not significantly different at LSD at ($p \leq 0.05$).
-Each value was an average of three determinations ± standard deviation.

Texture Analysis of WWF biscuit supplemented with CP and different levels WPC

Data in Table 9 illustrate the texture analysis of WWF biscuits supplemented with CP and various levels of WPC, focusing on two key parameters: hardness and fracturability. The control sample exhibited the lowest values for both hardness and fracturability, at 38.66 and 24.66, respectively, indicating a softer texture. These results are consistent with those reported by Rodrigues et al., (2023). There was a significant increase in both hardness and fracturability with the addition of WPC to the biscuit blends. Hardness values ranged from 38.66 for the control to 81.17 for blend 5, while fracturability values ranged from 24.66 for the control to 98.33 for blend 5. The highest values were observed in blend 5, with a hardness of 81.17 and fracturability of 98.33, indicating the hardest and most fracturable texture among all samples. These progressive increases suggest that the blend modifications significantly impact the texture of the product. These findings align with those of Andoyo et al., (2023), who reported that excessive WPC use may result in a harder texture, potentially reducing product palatability.

Table 9. Texture Analysis of WWF biscuit supplemented CP and different levels WPC

Blends	Hardness (N)	Fracturability (N)
Control1	38.66^e ± 0.08	24.66^e ± 1.21
Blend 2	72.59^d ± 0.59	63.64^d ± 0.73
Blend 3	75.90^c ± 0.18	72.40^c ± 0.75
Blend 4	79.29^b ±0.45	84.44^b ±0.83
Blend 5	81.17^a ± 0.42	98.33^a ±1.11

-Values followed by the same letter in columns are not significantly different at LSD at ($p \leq 0.05$).
-Each value was an average of three determinations ± standard deviation.

Sensory evaluation of WWF biscuit supplemented with CP and different levels WPC

Quality is not a single, well-defined attribute but comprises many properties or characteristics. Appearance is one of the primary factors consumers use to assess food product quality. Food appearance, mainly determined by surface color, is the first sensation that consumers perceive and use as a basis for acceptance or rejection. The visual appearance of food, reflected in its color, strongly influences consumer perceptions of quality.

Color can be associated with other quality attributes, such as sensory appeal, nutritional value, and the presence of visual or non-visual defects, allowing for immediate quality control (Bredariol et al., 2019). Table 10 presents the sensory evaluation of WWF biscuits supplemented with CP and various levels of WPC. The data show notable differences among the blends. Blend 4 achieved the highest appearance (18.84), taste (18.82), and odor (18.96) values compared to the control biscuits, suggesting that adding WPC may enhance flavor attributes. Conversely, Blend 5 showed significantly lower scores for texture (17.38) and color (16.42), indicating that higher WPC levels may negatively affect the physical properties of the biscuits. This could be due to the Maillard reaction between amino acids and reducing sugars, which can cause color deterioration (El-Hadidy et al.,2020). These results align with findings from Parate et al. (2011), who reported a decrease in sensory scores for appearance and color with increasing WPC levels, and they are consistent with Raghul (2023).

Table 10. Sensory evaluation of WWF biscuit supplemented with CP and different levels WPC

Blends	Texture 20	Oder 20	Taste 20	Color 20	Appearance 20
Control 1	$18.90^{a}\pm 0.02$	$17.00^{d}\pm 0.02$	$16.94^{e}\pm 0.01$	$18.90^{a}\pm 0.03$	$16.86^{d}\pm 0.06$
Blend 2	$18.88^{a}\pm 0.03$	$16.84^{e}\pm 0.07$	$16.88^{d}\pm 0.02$	$16.84^{d}\pm 0.01$	$16.50^{e}\pm 0.09$
Blend 3	$18.74^{b}\pm 0.04$	$17.66^{c}\pm 0.04$	$17.62^{c}\pm 0.05$	$18.42^{b}\pm 0.03$	$17.96^{c}\pm 0.01$
Blend 4	$18.42^{c}\pm 0.03$	$17.96^{b}\pm 0.01$	$18.82^{b}\pm 0.04$	$17.82^{c}\pm 0.02$	$18.84^{b}\pm 0.04$
Blend 5	$17.38^{d}\pm 0.09$	$18.96^{a}\pm 0.03$	$19.02^{a}\pm 0.16$	$16.42^{e}\pm 0.03$	$19.06^{a}\pm 0.08$

-Values followed by the same letter in columns are not significantly different at LSD at ($p \leq 0.01$).
-Each value was an average of twenty determinations ± standard deviation.

Physical properties of WWF biscuit supplemented with CP and different levels WPC

The data in Table 11 show the physical properties of WWF biscuits supplemented with CP and varying levels of WPC. The control sample had the largest diameter (5.09 cm) and thickness (0.74 cm), which were significantly greater than the values observed in the other biscuit blends. This could be attributed to the gluten strength in wheat flour. On the other hand, blend 5 had the smallest diameter (4.90 cm) but showed a slight increase in thickness to 0.69 cm, possibly due to a compensating expansion in thickness from the higher whey protein addition. These results align with Parate et al., (2011), who found that the diameter of WPC-fortified biscuits was smaller than control biscuits, while thickness tended to increase. The findings also agree with those of Raghul (2023).

For the spreading ratio, blend 2 had the highest value (7.88), which differed significantly from the control ratio of 6.85, suggesting that increasing WPC may enhance spread ability. Blends 3 and 4 showed moderate, significant differences in spreading ratios (7.71 and 7.51, respectively), while blend 5 had the lowest spread ratio (7.41). The reduction in spread ratio could be due to the increased thickness of the biscuits resulting from WPC incorporation. These findings align with Parate et al. (2011) but contrast with those of Ahmed and Ashraf (2019).

The weight of all blends showed significant change, ranging from 6.70 g for blend 5 to 7.50 g for the control sample. The slight weight change in WPC biscuits may be due to the addition of extra water in the dough, as WPC dough has a higher water absorption capacity than the control dough.

Similarly, volume values showed significant differences, ranging from 18.88 cm^3 for blend 5 to 20.35 cm^3 for the control sample. Specific volume values ranged from 2.72 cm^3/g for the control sample to 2.83 cm^3/g for blend 5, also significant changes. These results are consistent with those reported by Beshir et al. (2021).

Table 11. Physical properties of WWF biscuit supplemented with CP, and different levels WPC

Blends	Diameter (cm)	Thickness (cm)	Spread ratio	Weight (g)	Volume (cm^3)	Specific volume (cm^3/ g)
Control 1	$5.09^{a}\pm 0.02$	$0.74^{a}\pm 0.02$	$6.85^{e}\pm0.06$	$7.50^{a}\pm0.02$	$20.35^{a}\pm 0.02$	$2.72^{c}\pm 0.01$
Blend 2	$5.07^{a}\pm 0.01$	$0.64^{b}\pm 0.02$	$7.88^{a}\pm0.02$	$7.39^{b}\pm0.07$	$20.19^{b}\pm 0.02$	$2.73^{c}\pm 0.02$
Blend 3	$4.98^{b}\pm 0.02$	$0.65^{b}\pm 0.03$	$7.71^{b}\pm 0.05$	$7.06^{c}\pm0.03$	$19.51^{c}\pm 0.01$	$2.76^{b}\pm 0.03$
Blend 4	$4.97^{b}\pm 0.02$	$0.66^{b}\pm 0.01$	$7.51^{c}\pm0.04$	$6.91^{d}\pm0.01$	$19.50^{c}\pm 0.02$	$2.82^{a}\pm 0.01$
Blend 5	$4.90^{c}\pm 0.01$	$0.69^{b}\pm 0.01$	$7.41^{d}\pm 0.01$	$6.70^{e}\pm0.04$	$18.88^{d}\pm 0.03$	$2.83^{a}\pm 0.02$

-Values followed by the same letter in columns are not significantly different at LSD at ($p \leq 0.05$).
-Each value was an average of three determinations ± standard deviation.
Weight: W (g); Diameter: D (mm); Thickness: T (mm); Expansion factor: (D/T); biscuit Volume's (cm^3);

4. Conclusion

Based on the overall results, it can be concluded that WWF biscuits supplemented with CP and varying levels of WPC demonstrated improvements in chemical composition, sensory attributes, physical properties, and color parameters. The inclusion of CP and different levels of WPC in the WWF biscuit formula enhanced its nutritional value, color characteristics, and overall quality. Organoleptic evaluations indicated that the biscuits supplemented with CP and varying levels of WPC were acceptable and showed significant differences compared to the control in terms of overall acceptability, color, thickness, texture, taste, and odor. Therefore, it is possible to produce high-quality bakery products using carrot powder (CP), varying levels of whey protein concentrate (WPC), and whole wheat flour that are suitable for consumers.

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