

Effect of fermented chickpea flour substitution on the chemo-physical, rheological, and sensory characteristics of pan bread

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Abstract

Chickpea (*Cicer arietinum* L.) is an exceptional legume that has immense global utilization and serves as an outstanding reservoir of protein and amino acids. The present research studied the consequential outcomes that developed from the substitution of wheat flour with 0%, 5%, 10%, 15%, and 20% fermented chickpea flour (FCF) to prepare pan bread and its physicochemical and antioxidant activity as well as rheological and sensory properties. In terms of the visual traits of the bread, it was observed that the bread crust color turned progressively darker with increase in the level of FCF. Interestingly, despite this change in color, no significant impact was observed on the L^{*} value of bread crust. The incorporation of FCF at all concentrations resulted in a remarkable enhancement of total phenolic compounds and the antioxidant activity of the bread, compared to the control sample. This finding indicated that the inclusion of FCF in bread formulations not only improved its nutritional content but also enhanced its health benefits. It was found that the integration of up to 20% FCF in bread formulations increased its nutritional composition and preference in sensory evaluation. In conclusion, the incorporation of FCF to produce pan bread presents a good future for the food industry, paving the way for the creation of bread options that are both healthier and more nutritious for consumers.

Keywords: bread; fermented chickpea flour; physicochemical properties; sensory properties

Introduction

Globally, bread is a main element in the diet of a significant number of individuals, regardless of their social and economic standing. Numerous countries confront a disparity between the production and consumption of bread. As a result, many researchers (McWatters *et al.*, 2004) suggested the potential of wheat flour enrichment with alternative crops, such as barley, maize, and sorghum, as well as legume flours that are abundant in lysine. The authors observed an increase in dietary fiber and mineral content in the enhanced bread, making it a

healthier alternative. They also noted a slight decrease in loaf size and tear strength, indicating potential textural changes that need to be addressed. In a study conducted by Eissa *et al.* (2007), mixture of wheat flour and chickpea flour was used to produce bread. Studies on the bread made with wheat flour and chickpea flour revealed that the incorporation of chickpea flour into wheat flour improved the properties of bread dough and the resulting bread. In particular, the combination resulted in increased bread size and improved texture and aroma. It also resulted in decreased baking time and bread's increased dietary fiber content.

Globally, chickpea (*Cicer arietinum* L.) ranks as the third most important pulse seed, as pointed out by Boye *et al.* (2010), and is the second most cultivated legume. Chickpea seeds, which contain a high proportion of crude protein (17–22%), represent an affordable source of carbohydrates, minerals, and vitamins, thus making them a vital legume in certain tropical regions. Rachwa-Rosiak *et al.* (2015) illustrated that chickpea is grown mainly in the areas with temperate and semiarid climate. The protein found in chickpea is abundant in arginine but relatively low in sulfur-containing amino acids, such as methionine and cysteine. The starch content in chickpea typically ranges from 40% to 48%. Additionally, chickpea seeds are a rich source of vitamins, such as vitamin A, niacin, thiamine, and folate (Harsha, 2014).

Furthermore, a study conducted on the nutritional values of chickpea seeds by Zafar *et al.* (2015) showed that the seeds contain a substantial amount of protein, dietary fiber, vitamins (niacin, thiamine, and ascorbic acid), minerals, unsaturated fatty acids, and essential amino acids that are deficient in wheat flour. These properties contribute to reducing cholesterol, cardiovascular diseases, and cancer.

Although chickpeas have nutritional value and health benefits, it also contains anti-nutritional factors, such as chymotrypsin inhibitors, trypsin, phytates, flavonoids, phytic acid, lectins, α -amylase inhibitors, tannins, saponins, phenolics, and oxalic acid (Jukanti *et al.*, 2012). These factors reduce protein availability and digestibility by bonding with minerals. Therefore, it is imperative to reduce the levels of these anti-nutritional factors to enhance protein digestibility and availability.

Fermentation provides a diverse range of microbial and enzymatic transformation to food, leading to desirable properties, such as extended shelf life, safety, flavor, improved nutrition content, elimination of anti-nutrients, and enhanced health benefits. Olika *et al.* (2019) described the microorganisms and fermentation conditions that significantly impact the removal of phytates during fermentation. Solid-state fermentation (SSF) is a cost-effective and controlled method to enhance the nutritional value and functional properties of legumes and cereals. During fermentation, legumes undergo various biochemical and enzymatic changes. For instance, protease enzymes hydrolyze proteins, resulting in the production of short-chain compounds with low molecular weights.

Rhyu and Kim (2011) discussed how the digestibility, physicochemical properties, nutritional quality, and bioactivity of the resulting products are improved. In addition, Xiao *et al.* (2018) explained that treating chickpea seeds in various ways preserves or improves the value of reaction products.

The objective of this study was to investigate the impact of SSF on chickpea flour at different durations to enhance functional and nutritional properties. In addition, the study aimed to evaluate the effects of FCF on the physicochemical properties, antioxidant activity, and sensory acceptability of pan bread.

Materials and Methods

Materials

Chickpea seeds (*Cicer arietinum* L.) were procured from the local market situated in the Qassim Region of the Kingdom of Saudi Arabia (KSA). Kabuli-type chickpea, characterized by large owl/ram-head-shaped seeds with beige-color seed coat, was used. Wheat flour, with 80% extraction rate and 27% wet gluten content, was acquired from the Saudi Grains Organization (SAGO), Qassim Region, KSA. All remaining constituents, including white crystal sugar, vegetable palm shortening, instant active dry yeast, and salt, were bought from local market within the Qassim Region. The requisite chemicals were procured from Arkan Development Co. Ltd., located in the Qassim Region of KSA.

Methods

Solid-state fermentation of chickpea flour

Solid-state fermentation of chickpea flour was conducted in accordance with the procedure outlined by Xiao *et al.* (2015), albeit with certain modifications of the culture. The chickpea seeds were milled using a hammer mill to achieve a mesh size of 1 mm and subsequently sterilized at 121°C for 25 min, followed by cooling to the ambient temperature (23±2°C). Thereafter, the chickpea flour was inoculated with 1.4% of active dry yeast extract (*Saccharomyces cerevisiae*) along with 4% sucrose. In order to maintain moisture content of approximately 20%, the flour was subjected to a spraying process involving distilled water. The chickpea flour, spread in a thin layer measuring 0.5 cm, was then incubated at 30±2°C and a relative humidity of 85% from day 1 to day 4. Following the incubation process, the residual yeast was deactivated, and the fermented chickpea flour (FCF) was subsequently subjected to drying at 52°C in a hot air oven for 12 h. The dried FCF was then milled in a laboratory mill, sieved through a mesh size of 180 μ m, and stored in sealed containers prior to further analysis.

Determination of phytic acid and free amino acids of chickpea flour

The concentration of phytic acid (mg/g) in the chickpea flour prior to and after fermentation was determined according to the method of the Association of

Official Analytical Chemists (AOAC, 2005) while the quantification of free amino acids (mg/g) was accomplished through the Cd-ninhydrin method outlined by Folkertsma and Fox (1992).

Rheological properties of bread dough

Following the 3 days of fermentation, the rheological properties, including texture, and behavior of wheat flour dough containing different proportions of FCF (0, 5, 10, 15, and 20%) were estimated during mixing of the dough according to the method described by the American Association of Cereal Chemists (Cereals & Grains Association) (AACC International, 2010) by using a Farinograph test apparatus (type 810107, Brabender OHG, Duisburg, Germany). The viscosity of wheat flour suspension with different proportions of FCF (0, 5, 10, 15, and 20%) was analyzed using an Amylograph apparatus (Brabender, Duisburg, Germany) according to the standard methods described by the International Association for Cereal Chemistry (ICC, 2001).

Preparing pan bread samples

Pan bread samples were prepared by the established straight-dough method 10-10.03, which has been approved by the AACC International (2010). The control bread dough comprised 100% wheat flour, 1.5% instant active dry yeast, 3% white crystal sugar, 3% vegetable shortening, and 1.5% table salt. Wheat flour was first mixed with dry ingredients in a mixer bowl for 1 min. Water was added followed by the addition of water up to 500 BU consistency of Farinograph (which expresses the best consistency of bread dough prepared from wheat flour) and the dough kneading was continued for 5 min. The dough was divided and placed in baking pans and subjected to proofing in a cabinet at 33°C and 80–85% relative humidity (RH). After 45 min of fermentation, the dough was baked at 230°C for 15 min in a conventional oven. Following the baking process, the pan bread was quickly cooled to a temperature of 30°C, achieving this within 60 min. To elucidate the impact of FCF on the overall quality bread, varying proportions of wheat flour (5, 10, 15, and 20%) were substituted with FCF (which has been previously fermented for 3 days under controlled conditions). Subsequently, the bread was placed in plastic bags for further analysis.

Evaluation of physicochemical properties of bread samples

The incorporation of FCF was based on the findings of preliminary investigations. The proportions of moisture, ash, protein, lipids, and dietary fibers in both wheat flour and chickpea flour, as well as in FCF, at different fermentation periods, and in bread samples, were determined according to the AACC International (2010) method No. 10-05.01. The nitrogen conversion factor to calculate the protein content was 5.70 for wheat flour and 6.25 for chickpea flour and bread samples. Moreover,

the carbohydrate content at dry basis was calculated by utilizing the following difference-based approach:

$$\text{Carbohydrate (\%)} = 100 - (\text{protein [\%]} + \text{lipids [\%]} + \text{ash [\%]} + \text{dietary fiber [\%]}).$$

$$\text{The energy value (kcal/100 g)} = 4 \times (\text{protein [g]} + \text{carbohydrate [g]}) + 9 \times \text{fat [g]}$$

Physical properties of pan bread samples, such as weight (g), volume (cm³), specific volume (cm³/g) (calculated by dividing volume by weight), and baking loss (%), were evaluated. These evaluations were conducted in accordance with AACC International (2010) method No. 10-05.01.

Determination of color properties of pan bread with FCF

The effect of adding FCF on the color attributes of crust and crumb of pan bread samples was evaluated. The lightness L* (ranging from 0 black to 100 white), redness a* (where a positive value [+60] indicates red and a negative value [-60] indicates green), and yellowness b* (where a positive value [+60] indicates yellow and a negative value [-60] indicates blue) values of the control pan bread, as well as the bread samples containing 5%, 10%, 15%, and 20% of FCF, were determined using a Hunter Lab Color QUEST II Minolta CR-400 (Minolta Camera Co. Ltd., Osaka, Japan). This analysis was conducted according to the methodology reported by Francis (1983).

Determination of phenolic content and antioxidant activity (DPPH)

Phenolic compounds in the bread samples were evaluated according to the method described by Bloor (2001). The total phenolic content was determined using the Folin–Ciocalteu assay, in which a spectrometer operating at a wavelength of 765 nm, as explained by Singleton *et al.* (1999), was employed. Gallic acid served as a standard for quantification, and the results were expressed in terms of milligram (mg) of Gallic acid equivalent per gram (g). Bread samples' ability to scavenge free radicals was evaluated following the procedure outlined by Blois (1958). The scavenging activity was calculated based on reduction in absorbance at 517 nm, compared to a 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical solution in methanol, using the following equation:

$$\text{Scavenging activity (\%)} = \frac{\text{Abs. control} - \text{Abs. sample}}{\text{Abs. control}} \times 100.$$

Evaluating the hardness of bread crumbs

The hardness of bread crumbs during a storage period of 1, 3, and 5 days at room temperature (23±2°C) was determined in accordance with the AACC International (2010)-approved method 74–09.01, using the Texture

analyzer TA-XT-Plus (Stable Microsystems, UK), and was expressed as crumb firmness (N). It is conducting a “measure of force in compression” test with an AACC 36 mm cylinder probe with radius (P/36R). To achieve this, bread slices, measuring 1.25 cm in thickness, were taken from the center of the pan bread. These slices were then subjected to compression, reducing their initial thickness by 50% while maintaining a test speed of 1 mm s⁻¹. The firmness or hardness of pan bread slices was quantified by recording the maximum force observed during the initial cycle of compression, referred to as F2.

Sensory evaluation of various pan bread samples

Pan bread samples containing varying levels of FCF were subjected to sensory evaluation by a group of 20 semi-trained panelists (aged between 30 and 50 years) from the staff of the Department of Food Science and Human Nutrition, Faculty of Agriculture and Food, Qassim University. They were informed about the sensory attributes of bread, verbal definitions of attributes, and intensity of scales. These panelists were instructed to assess each loaf in terms of its visual appearance, crumb color, crumb texture, odor, taste, and the overall acceptability. In order to rate these characteristics, a 10-point scale was employed, where a rating of 10 indicated excellence and a rating of 1 indicated extreme dissatisfaction, following the guidelines outlined by AACC International (2010). Sensory evaluation was carried out within 8 h of baking in a separate room with white-painted walls, at a temperature of 24°C with incandescent lighting. Each panelist was provided with one complete slice (150-g loaf) presented in a tray covered with a plastic cling-film wrapper. A randomized 3-digit code was used to represent each sample served according to a complete block design. Lukewarm water served as palate cleanser between samples (Elia, 2011).

Statistical analysis

The statistical analysis was conducted using a randomized complete block design with three replications, except for the sensory evaluation, which had 10 replications. The analysis of variance was performed according to the methodology proposed by Gomez and Gomez (1984), utilizing the SAS (2004) program. The least significant difference at a significance level of ($p \leq 0.05$) (LSD) was used to compare the mean values of different treatments.

Results and Discussion

Proximate composition of wheat, chickpea flour, and FCF

The study analyzed the approximate chemical compositions of wheat, chickpea flour, and FCF at 30°C for 1, 2, 3 and 4 days and the obtained results are presented in

Table 1. The results demonstrated that wheat flour had much greater moisture content (11.46%) than other samples. After 1 day of fermentation, there was no significant ($p \leq 0.05$) change in the moisture content of fermented (5.60) and raw chickpea flour (6.11%) samples.

This trend persisted for 2- and 3-day fermentation with FCF, while 4th day fermentation resulted in a significant ($p \leq 0.05$) decrease in moisture content (2.76%), compared to all previous samples. Chickpea flour and FCF were characterized by a higher percentage ($p \leq 0.05$) of ash (3.43%), compared to wheat flour (0.64%).

The fermentation period did not significantly ($p \geq 0.05$) affect the ash content, which ranged between 3.03% and 3.57%. Chickpea flour had a greater protein percentage (17.61%) than wheat flour (10.40%). Chickpea flour's protein content increased gradually ($p \leq 0.05$) after fermentation for up to 3 days. However, after 4 days of fermentation, the protein concentration reduced (16.62%), which could be due to the strong hydrolytic activity of enzymes.

Similar results were also observed for fat content in FCF, compared to wheat flour. Chickpea flour and FCF had significantly ($p \leq 0.05$) higher crude fiber percentage ($p > 0.05$) at various fermentation periods, ranging from 6.68% to 9.09%, compared to 0.65% in wheat flour. These findings were comparable with those observed by Ali *et al.* (2021), who found that the proximate chemical composition of chickpea flour is 11.08% moisture, 19.00% crude protein, 6.65% crude fat, 3.67% ash, and 6.06% crude fiber.

Impact of fermentation in reducing phytic acid and boosting free amino acid

Figures 1A and 1B illustrate that wheat flour naturally contained the least phytic acid (2.95 mg/g) and free amino acids content (13.75 mg/g), while unfermented chickpea flour had considerably higher levels of phytic acid (7.47 mg/g) and free amino acids content (21.30 mg/g). Interestingly, the fermentation process with chickpea flour led to contrasting effects on these components. Phytic acid content steadily decreased with increasing fermentation period, dropping from 5.51 mg/g after 1 day fermentation to 2.52 mg/g after 4 days of fermentation (Kumar *et al.*, 2013).

Conversely, free amino acids increased progressively throughout the fermentation period, reaching 55.04 mg/g in 4 days, compared to 31.23 mg/g after 1 day. These findings suggested that fermentation could effectively modify the nutritional profile of chickpea flour, potentially enhancing its bioavailability and contributing

Table 1. Chemical components of wheat flour and chickpea flour prior to and after fermentation at 30°C (% of dry weight basis).

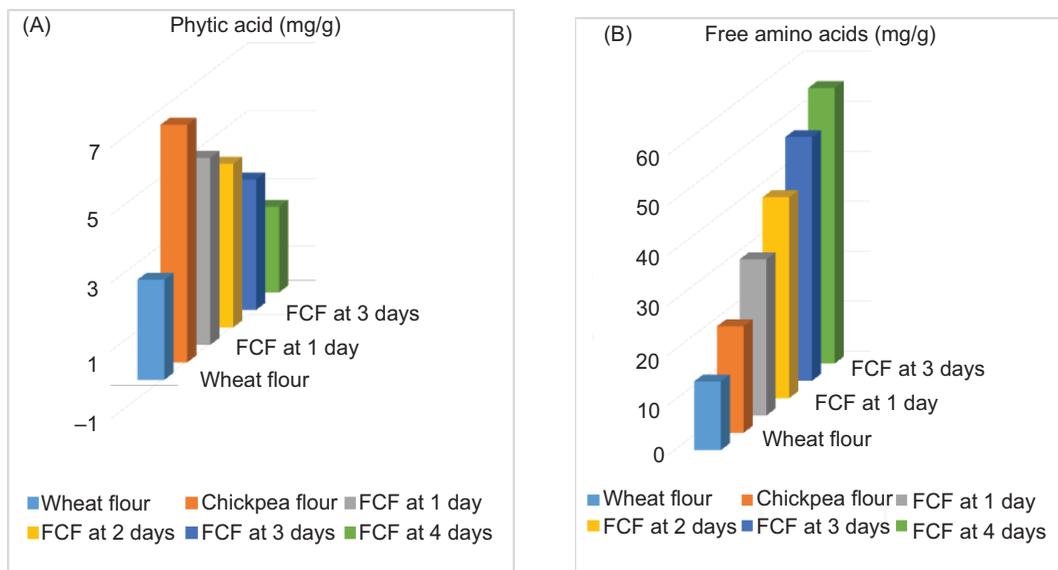
Treatments	Chemical composition (%)					
	Moisture	Ash	Crude protein	Lipids	Crude fiber	Carbohydrates*
Wheat flour	11.46 ± 0.09 ^a	0.64 ± 0.07 ^b	10.40 ± 0.09 ^e	1.86 ± 0.05 ^d	0.65 ± 0.06 ^c	86.45 ± 0.12 ^a
Chickpea flour	6.11 ± 0.26 ^b	3.43 ± 0.05 ^a	17.61 ± 0.66 ^{c,d}	4.96 ± 0.21 ^c	8.67 ± 0.33 ^a	65.32 ± 1.02 ^c
FCF at 1 day	5.60 ± 0.65 ^b	3.18 ± 0.38 ^a	18.40 ± 0.37 ^{b,c}	5.65 ± 0.14 ^{b,c}	8.41 ± 0.27 ^a	64.36 ± 0.22 ^c
FCF at 2 days	3.47 ± 0.58 ^c	3.03 ± 0.18 ^a	19.12 ± 0.32 ^b	5.89 ± 0.26 ^b	8.36 ± 0.56 ^a	63.61 ± 0.72 ^c
FCF at 3 days	3.20 ± 0.15 ^c	3.53 ± 0.21 ^a	20.72 ± 0.37 ^a	7.99 ± 0.38 ^a	9.09 ± 0.19 ^a	58.67 ± 0.33 ^d
FCF at 4 days	2.76 ± 0.25 ^d	3.57 ± 0.13 ^a	16.62 ± 0.44 ^d	5.79 ± 0.22 ^b	6.68 ± 0.37 ^b	67.35 ± 0.26 ^b

FCF: fermented chickpea flour.

Data are the mean values ± SE, n = 3.

Values followed by the same superscripted letters in the same column are not significantly different ($p \leq 0.05$).

*Carbohydrates calculated by the difference, Carbohydrate (%) = 100 - (protein [%] + lipids [%] + ash [%] + dietary fiber [%]).

**Figure 1.** Impact of chickpea flour fermentation on (A) phytic acid and (B) free amino acids.

to the development of unique flavor. Greiner *et al.* (2001) reported that FCF exhibited significant changes in both phytic acid and free amino acid content, compared to wheat flour and unfermented (raw) chickpea flour.

Effect of fermented chickpea flour on the farinograph of bread dough

Table 2 presents the farinograph parameters of wheat flour blended with different levels of FCF, ranging from 0% to 20%. The farinograph is an instrument used to measure the rheological properties of dough, such as its development time, stability, mixing tolerance, and water absorption. These properties are crucial for baking and influence the final quality of bread and other

baked goods. Resistance index (BU) measures the dough strength. The resistance index generally increases (33.00 BU) with increase in FCF level by up to 20%, indicating stronger dough. It could be due to the presence of protein and dietary fiber in chickpea flour, which contributed to gluten network development. Benali *et al.* (2019) observed similar trends with addition of milk powder. However, the highest value was observed at 10% FCF, followed by a decrease at higher FCF levels. This could be due to excessive fiber interfering with gluten formation. Stability time (min) reflects dough tolerance to mixing, and its higher value indicates better tolerance. The stability time increases with increasing FCF levels, with the highest value being at 20% FCF (7.99 min). This suggested that FCF could improve dough handling properties, as pointed out by Hassanien *et al.* (2012).

Table 2. Farinograph parameters of wheat flour containing different levels of fermented chickpea flour.

Treatment	Water absorption ratio (%)	Dough development time (min)	Stability time (min)	Resistance index (BU)	Farinograph No. quality
Wheat flour	63.57 ± 0.33 ^b	10.28 ± 0.06 ^a	10.83 ± 0.14 ^c	11.00 ± 0.58 ^e	126.00 ± 1.15 ^d
FCF 5%	60.88 ± 0.82 ^d	9.59 ± 0.01 ^c	13.06 ± 0.32 ^b	17.33 ± 0.88 ^d	136.66 ± 2.73 ^c
FCF 10 %	62.50 ± 0.06 ^c	10.03 ± 0.03 ^b	14.66 ± 0.35 ^a	23.00 ± 0.58 ^c	157.67 ± 3.48 ^a
FCF 15 %	63.80 ± 0.06 ^b	7.43 ± 0.03 ^e	9.46 ± 0.05 ^d	26.33 ± 0.88 ^b	148.00 ± 2.57 ^b
FCF 20 %	64.70 ± 0.15 ^a	8.33 ± 0.04 ^d	7.99 ± 0.44 ^e	33.00 ± 1.15 ^a	142.33 ± 0.88 ^{bc}

FCF: fermented chickpea flour.
Data are the mean values ± SE, n = 3.
Values followed by the same superscripted letters in the same column are not significantly different ($p \leq 0.05$).

Benali *et al.* (2019) studied the impact of adding chickpea flour to wheat flour on the rheological properties of toast bread. Dough development time (min) is the time required for the dough to achieve maximum consistency. It increases with increasing FCF levels, indicating that dough with a higher FCF content takes longer time to develop. This could be due to a dilution effect on gluten or interactions between chickpea flour components and gluten. Finally, absorption water ratio (%) indicates the amount of water required for optimal dough consistency. The water absorption ratio shows slight variations across treatments, with no clear trend with increasing FCF levels. This suggests a minimal impact of FCF on water absorption within the studied range.

Effect of fermented chickpea flour on the amylograph of bread dough

The impact of FCF on the amylograph properties of wheat flour is crucial for optimizing bread-making process and developing functional food products and bread quality parameters. Table 3 shows maximum viscosity (AU) values, which indicate the maximum resistance of flour–water mixture to flow during heating. A higher value of viscosity indicates stronger dough. In Table 3, the maximum

viscosity of wheat flour decreases significantly ($p \leq 0.05$) with increasing FCF content from 1812 AU for wheat flour to 1420.33 AU for wheat flour containing 20% FCF, suggesting the weakening of dough structure. Gelatinization temperature (°C) marks the onset of starch gelatinization, where starch granules begin to swell and lose their crystalline structure. The gelatinization temperature shows slight variations across treatments, with no clear trend found that the gelatinization temperature of wheat flour was 90.13°C. Regarding the begin of gelatinization temperature (°C), which indicates the start of the gelatinization process. As observed in Table 3, the begin of gelatinization temperature of wheat flour generally significantly ($p \leq 0.05$) increases up to 60.43°C with increasing FCF content, indicating a delay in starch gelatinization. The properties of baked goods are affected by adding chickpea flour to durum wheat (Hassanien *et al.*, 2012; Pasqualone *et al.*, 2016).

Chemical composition of different pan bread samples

Table 4 shows the significant influence of adding FCF at varying proportions to wheat flour on the nutritional composition and specific characteristics of the resulting pan bread. Nutrient contents, such as ash, protein, lipids, and crude fibers, exhibited a positive correlation

Table 3. Amylograph properties of wheat flour containing different levels of FCF.

Treatment	Begin of gelatinization temp. (°C)	Gelatinization temp. (°C)	Maximum viscosity (AU)
Wheat flour	58.70 ± 0.30 ^d	90.13 ± 0.42 ^a	1812.67 ± 4.91 ^a
FCF 5%	59.30 ± 0.12 ^c	89.30 ± 0.06 ^{bc}	1752.33 ± 4.67 ^b
FCF 10%	59.87 ± 0.03 ^b	89.83 ± 0.58 ^{ab}	1678.00 ± 2.56 ^c
FCF 15%	60.27 ± 0.14 ^{ab}	89.11 ± 0.07 ^c	1595.00 ± 3.73 ^d
FCF 20%	60.43 ± 0.25 ^a	89.93 ± 0.13 ^{ab}	1420.33 ± 2.48 ^e

FCF: fermented chickpea flour.
Data are the mean values ± SE, n = 3.
Values followed by the same superscripted letters in the same column are not significantly different ($p \leq 0.05$).

Table 4. Chemical composition and energy content of pan bread fortified with varying levels of FCF (% on dry weight basis).

Treatments	Chemical composition (%)						Energy (kcal/100 g)
	Moisture	Ash	Crude protein	Lipids	Crude fiber	Carbohydrates*	
Control bread	38.54 ± 0.80 ^a	1.38 ± 0.03 ^d	12.78 ± 0.28 ^e	2.51 ± 0.13 ^e	1.72 ± 0.14 ^a	81.61 ± 2.39 ^a	400.19 ± 5.38 ^a
Bread with 5% FCF	36.66 ± 0.68 ^a	1.57 ± 0.05 ^d	14.40 ± 0.32 ^d	3.36 ± 0.16 ^d	3.73 ± 0.15 ^d	76.94 ± 1.57 ^b	395.57 ± 3.46 ^b
Bread with 10% FCF	33.32 ± 0.52 ^b	1.88 ± 0.03 ^c	16.38 ± 0.22 ^c	4.98 ± 0.16 ^c	4.85 ± 0.17 ^c	71.91 ± 0.28 ^c	397.95 ± 6.25 ^{a,b}
Bread with 15% FCF	27.64 ± 0.66 ^c	2.24 ± 0.13 ^b	18.18 ± 0.26 ^b	5.76 ± 0.38 ^b	5.78 ± 0.24 ^b	68.04 ± 0.56 ^d	396.76 ± 4.79 ^b
Bread with 20% FCF	24.98 ± 0.34 ^d	2.87 ± 0.09 ^a	19.29 ± 0.16 ^a	6.84 ± 0.32 ^a	6.91 ± 0.33 ^a	64.09 ± 0.83 ^e	395.11 ± 3.19 ^b

FCF: fermented chickpea flour.

Data are the mean values ± SE, n = 3.

Values followed by the same superscripted letters in the same column are not significantly different ($p \leq 0.05$). *Carbohydrates: calculated by difference. Carbohydrate (%) = 100 – (protein [%] + lipids [%] + ash [%] + dietary fiber [%]).

with increasing FCF content. The addition of FCF naturally enriches the bread with these elements. The bread containing 20% FCF showed maximum values (2.87% ash, 19.29% protein, 6.84% lipids, and 6.91% crude fibers), compared to control wheat flour bread. This aligned with the studies conducted by Kumar *et al.* (2013) and Yilmaz *et al.* (2016), demonstrating that incorporating chickpea flour into wheat bread positively impacted its nutritional profile. Carbohydrates and energy decreased steadily with increase in FCF inclusion. Control wheat flour bread had the highest carbohydrate content (81.61%) and energy value (400.19 kcal/100 g); with 20% FCF, these values dropped to 64.59% and 395.11 kcal/100 g, respectively, signifying respective reduction of 21% and 1.3%. This was due to the lower carbohydrate and energy content of FCF, compared to the contents of wheat flour. The control wheat flour bread had the highest moisture content (38.54%), which was statistically equivalent to the 5% FCF bread (38.66%). However, a gradual decline in moisture was observed with increasing FCF levels, reaching 24.98% at 20% FCF. This trend could be attributed to the moisture-absorbing properties of chickpea flour.

Effect of different levels of FCF on physical attributes of pan bread

Table 5 shows the effect of incorporating different levels of FCF on the physical properties of pan bread. The control bread (100% wheat flour) had the lowest loaf weight (172.34 g), while breads with 10%, 15%, and 20% FCF had the highest loaf weights with significantly increased which recorded 179.40, 180.47 and 181.76 g, respectively. As FCF content increased in bread making, loaf volume decreased gradually, which recorded 580.56, 511.27, 487.58, and 461.64 cm³ for 5%, 10%, 15%, and 20% of FCF, respectively. On the other hand, the control bread had the highest volume of 630.67 cm³. This decrease

was attributed to the disruption of gluten network by chickpea flour, hindering gas retention and expansion during baking (Brenan and Cleary, 2007; Simona *et al.*, 2015a). Specific volume (volume per unit weight) of bread followed a similar trend as volume. As FCF content increased in bread as 10% 15%, and 20%, specific volume decreased significantly, with 2.85, 2.70, and 2.54 cm³/g, respectively, compared to the control bread, which had the highest specific volume (3.66 cm³/g), followed by the bread with 5% FCF (3.32 cm³/g). This reflected the negative impact of FCF on gluten development and gas retention (Simona *et al.*, 2015b). Data also revealed that control bread had the highest baking loss (13.83%), indicating greater moisture evaporation during baking. Baking loss was 12.63%, 10.30%, 9.77%, and 9.12% with the increasing FCF content of 5%, 10%, 15%, and 20%, respectively. This could be due to the higher water absorption capacity of chickpea flour, leading to less moisture available for evaporation (Mohammed *et al.*, 2012a). The findings showed that incorporating FCF into pan bread negatively affected its volume, specific volume, and baking loss. This could be due to the disruption of gluten network by chickpea flour, hindering gas retention and expansion during baking. However, the addition of 5% FCF showed minimal impact on these properties, compared to the control bread. It's important to consider the sensory properties of bread, in addition to these physical characteristics. While FCF may impact volume and texture, it can also contribute to increased protein content, dietary fiber, and unique flavor profiles (Brenan and Cleary, 2007; Mohammed *et al.*, 2012a).

Color attributes of pan bread crust and crumb fortified with FCF

The impact of incorporating FCF at various levels 5, 10, 15 and 20% on the color attributes (Lab values) of both

Table 5. Physical properties of pan bread samples prepared from wheat flour and different levels of fermented chickpea flour.

Treatments	Weight (g)	Volume (cm ³)	Baking loss (%)	Specific volume (cm ³ /g)
Control bread	172.34 ± 0.44 ^c	630.67 ± 1.20 ^a	13.83 ± 0.22 ^a	3.66 ± 0.02 ^a
Bread with 5% FCF	174.75 ± 1.52 ^{b,c}	580.56 ± 5.21 ^b	12.63 ± 0.76 ^{a,b}	3.32 ± 0.03 ^b
Bread with 10% FCF	179.40 ± 2.63 ^{a,b}	511.27 ± 7.26 ^c	10.30 ± 0.76 ^{b,c}	2.85 ± 0.09 ^c
Bread with 15% FCF	180.47 ± 0.92 ^a	487.58 ± 1.45 ^d	9.77 ± 0.45 ^c	2.70 ± 0.46 ^d
Bread with 20% FCF	181.76 ± 2.86 ^a	461.64 ± 1.66 ^e	9.12 ± 1.43 ^c	2.54 ± 0.05 ^e

FCF: fermented chickpea flour.

Data are the mean values ± SE, n = 3.

Values followed by the same superscripted letters in the same column are not significantly different ($p \leq 0.05$).

the crust and crumb of bread samples was investigated and the obtained results are shown in Table 6. For bread crust color, it could be noticed that lightness (L^*) was significantly ($p \leq 0.05$) decreased from 35.60 to 25.94 for all treatments with increasing FCF content from 5 to 20% FCF, respectively when compared to the control bread (37.36), samples containing FCF displayed. This can be attributed to an intensified Maillard reaction during baking due to the higher lysine content in FCF.

No significant differences were observed in bread crust redness (a^*) between the control (11.94) and FCF-containing breads which ranging from 11.31 to 12.37. In addition, significantly ($p \leq 0.05$) decreased in bread crust yellowness (b^*) as FCF concentration increased in bread samples. The control bread exhibited the highest yellowness value of 16.55, while bread with 20% FCF had the lowest value of 9.63 ($p \leq 0.05$).

Regarding bread crumb color, results indicated that no significant ($p \leq 0.05$) influence of FCF substitution on bread crumb lightness (L^*) was observed. Values of bread crumb lightness ranged from 64.90 for control sample to 63.61 for 20% of FCF. Both of redness (a^*)

and yellowness (b^*) values significantly ($p \leq 0.05$) gradually increased with increasing FCF levels. The control bread had a redness of -0.54 and a yellowness of 18.11, while the 20% FCF bread had values of 0.48 and 20.38, respectively. This indicates a shift towards a redder and yellower crumb color with FCF incorporation. These findings aligned with previous studies conducted by Fenn *et al.* (2010) and Mohammed *et al.* (2012b), who reported similar trends in redness, yellowness, and lightness values in FCF fortified bread.

Quantifying polyphenols and DPPH antioxidant activity in pan bread

Figures 2A and 2B demonstrate the potent effect of incorporating FCF into bread on its total phenolic content and antioxidant activity. The graph presents a striking upward trend in both total phenolic compounds and DPPH scavenging activity with increasing FCF levels (5, 10, 15, and 20%). Compared to the control bread devoid of FCF, FCF-enriched breads displayed a noteworthy elevation in total phenolic content. This increase ranged from 0.59 mg GAE/g for 5% FCF to a remarkable 1.01 mg

Table 6. Color properties of bread with varying levels of fermented chickpea flour.

Treatments	Bread crust color			Bread crumb color		
	L^*	a^*	b^*	L^*	a^*	b^*
Control bread	37.36 ± 0.04 ^a	11.94 ± 0.40 ^a	16.55 ± 0.13 ^a	64.90 ± 0.42 ^a	-0.54 ± 0.05 ^e	18.11 ± 0.04 ^d
Bread with 5% FCF	35.60 ± 0.32 ^{a,b}	11.33 ± 0.14 ^a	14.60 ± 0.72 ^{b,c}	64.84 ± 0.21 ^a	-0.32 ± 0.04 ^d	18.69 ± 0.02 ^{c,d}
Bread with 10% FCF	34.47 ± 0.86 ^b	12.37 ± 0.03 ^a	15.29 ± 0.52 ^{a,b}	65.48 ± 0.32 ^a	0.08 ± 0.03 ^c	19.24 ± 0.33 ^{b,c}
Bread with 15% FCF	29.64 ± 0.59 ^c	11.83 ± 0.46 ^a	13.34 ± 0.78 ^c	64.52 ± 0.43 ^a	0.32 ± 0.04 ^b	19.75 ± 0.27 ^{a,b}
Bread with 20% FCF	25.94 ± 0.85 ^d	11.31 ± 0.70 ^a	9.63 ± 0.52 ^d	63.61 ± 1.09 ^a	0.48 ± 0.02 ^a	20.38 ± 0.53 ^a

FCF: fermented chickpea flour.

Data are the mean values ± SE, n = 3.

Values followed by the same superscripted letters in the same column are not significantly different ($p \leq 0.05$).

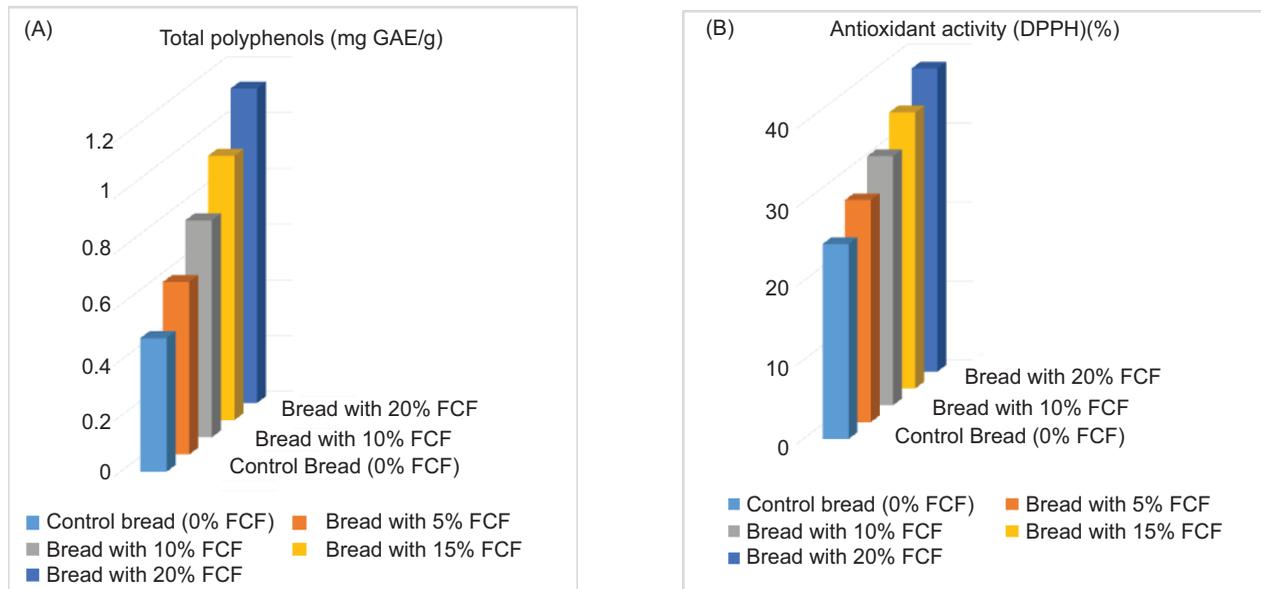


Figure 2. Unveiling the synergistic effect of fermented chickpea flour on bread's (A) total polyphenol content and (B) antioxidant activity (DPPH).

GAE/g for 20% FCF, representing a substantial enhancement. Correspondingly, the DPPH scavenging activity mirrored this upward trend, exhibiting a significant climb from 28.74% at 5% FCF to an impressive 39.29% at 20% FCF.

This phenomenon could be attributed to the inherent richness of FCF in phenolic compounds, a direct consequence of the fermentation process. In addition, changes in protein hydrolysis products also had an important role in antioxidant activity expressed as DPPH scavenging ability. Moreover, free tyrosine and tryptophan and peptides containing these amino acids reacted with Folin–Ciocalteu reagent, so the increase in total polyphenol content was a result of protein hydrolysis. As highlighted in the studies conducted by Fosschia *et al.* (2016) and Kumar *et al.* (2013), fermented chickpeas feature a higher concentration of these health-promoting compounds, compared to their unfermented counterparts. Dordevic *et al.* (2010) further emphasized that fermentation actively promotes the development and diversification of bioactive compounds within FCF. Interestingly, the findings of this study aligned with those reported by Sayaslan and Şahin (2018), who demonstrated that incorporating fermented chickpeas not only enhanced the nutritional profile of wheat flour bread but also positively impacted its volume and texture characteristics. In essence, Figure 2 serves as compelling visual evidence supporting the utilization of FCF as a means to enrich bread with valuable phenolic compounds and bolster its antioxidant potential, thereby contributing to the development of more nutritious and health-conscious bakery products.

Impact of FCF flour on pan bread staling rate

The investigation focused on the effects of incorporating 5%, 10%, 15%, and 20% FCF on hardness of bread (Figure 3). The objective was to comprehend how this incorporation influenced the crumb's hardness throughout a 5-day storage period at room temperature ($23\pm 2^{\circ}\text{C}$). The outcomes, as illustrated in Figure 3, revealed an interesting disparity in hardness between the control bread and the breads containing 5% and 10% FCF on day 1, with hardness values of 829.13 N, 846.60 N, and 902.10 N, respectively. However, the inclusion of 15% and 20% FCF led to significantly higher hardness values of 1223.58 N and 1399.23 N, respectively, compared to the control sample. This trend persisted on days 3 and 5, with all FCF concentrations resulting in the increased hardness of breads. The bread samples containing 5% and 10% FCF remained relatively softer and more palatable even on day 5 of storage. These findings suggested that FCF could contribute to the retention of moisture within the gluten network, consequently impacting the hardness of the crumb. This discovery aligned with the research conducted by Shrivastava and Chakraborty (2018), who observed that higher levels of FCF led to increased hardness of crumbs. The hardness was attributed to the formation of cross-links between gluten proteins and starch, with moisture acting as a plasticizer (Mohammadi *et al.*, 2014). In situations where water concentrations are lower, the cross-linking between proteins and starch intensifies, resulting in a more solid bread structure. Eliasson (2006) explained that starch could undergo retrogradation, wherein amylose and amylopectin molecules

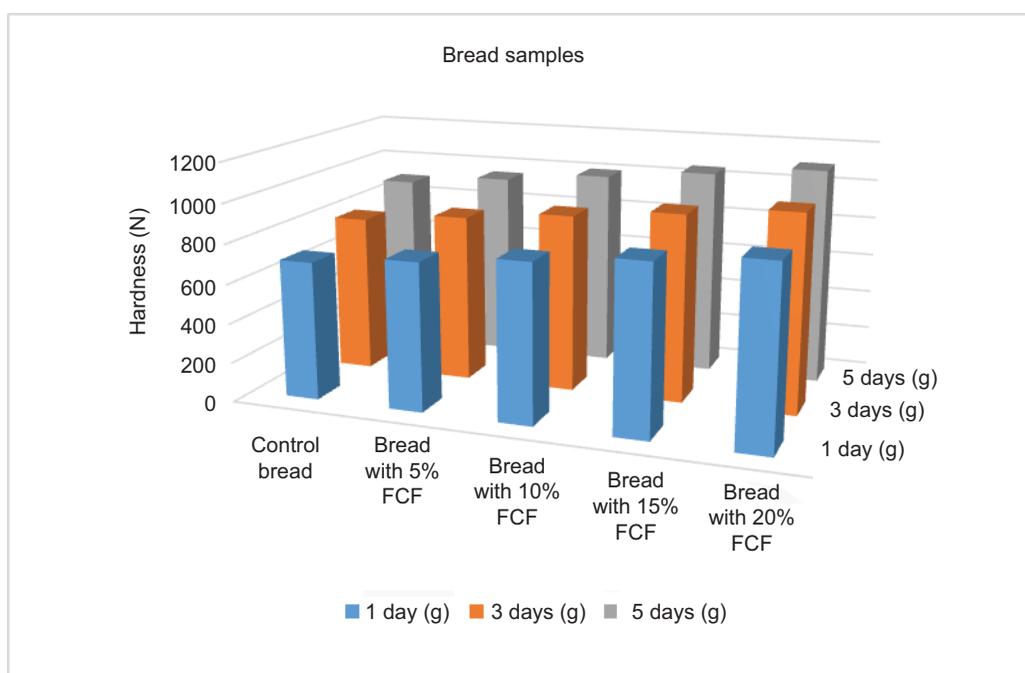


Figure 3. Effect of fermented chickpea flour on bread hardness (N) during storage period.

reassembled, forming crystalline structures and expelling water molecules, ultimately leading to a hardened bread structure.

Exploring the sensory characteristics of pan bread fortified with fermented chickpea flour

Table 7 shows that the control bread (without FCF) scored highest in all categories, with panelists finding it most appealing in appearance, texture, taste, color, odor, and the overall acceptability. Generally, sensory scores decreased with increased levels of FCF. While 5% FCF maintained good scores for most attributes, except for a slight decrease in appearance and odor, compared to the control.

Belobrajdic and Bird (2013) reported improved sensory properties of bakery products with fermented chickpeas, indicating potential benefits of FCF in certain contexts. With the addition of 10% FCF, there was a further decline in scores across most categories, particularly in texture and crumb color. However, with 15% and 20% FCF, there was a significant reduction in all sensory scores, indicating significant disapproval from panelists. Overall, in terms of appearance, control bread had the highest score, followed by 5% FCF. Scores progressively decreased with increased levels of FCF. While crumb texture had a similar trend as that of appearance, with control bread superior and addition of FCF leading to decreased scores. Increased FCF could alter texture of bread, making it denser or less crumbly, impacting texture scores.

Table 7. Sensory evaluation scores for pan bread samples containing varying levels of fermented chickpea flour.

Treatments	Appearance (10)	Crumb texture (10)	Taste (10)	Crumb color (10)	Odor (10)	Overall acceptability (10)
Control bread	9.4 ± 0.16 ^a	9.2 ± 0.25 ^a	9.4 ± 0.21 ^a	9.3 ± 0.21 ^a	9.6 ± 0.22 ^a	9.6 ± 0.16 ^a
Bread with 5% FCF	8.7 ± 0.21 ^b	8.7 ± 0.21 ^a	9.1 ± 0.26 ^a	9.0 ± 0.33 ^{a,b}	9.3 ± 0.42 ^a	8.6 ± 0.35 ^b
Bread with 10% FCF	8.1 ± 0.23 ^b	7.9 ± 0.24 ^b	8.1 ± 0.23 ^b	8.3 ± 0.31 ^{b,c}	8.3 ± 0.46 ^b	8.3 ± 0.21 ^b
Bread with 15% FCF	6.8 ± 0.25 ^c	6.2 ± 0.29 ^c	7.5 ± 0.31 ^b	7.5 ± 0.34 ^c	8.2 ± 0.25 ^b	7.4 ± 0.27 ^c
Bread with 20% FCF	5.9 ± 0.28 ^d	5.8 ± 0.20 ^c	6.5 ± 0.37 ^c	6.6 ± 0.31 ^d	7.9 ± 0.28 ^b	6.1 ± 0.24 ^d

FCF: fermented chickpea flour.

Data are the mean values ± SE, n = 3.

Values followed by the same superscripted letters in the same column are not significantly different ($p \leq 0.05$).

Regarding taste of bread, control bread again scored highest, followed by 5% and 10% FCF. Taste scores declined substantially at 15% and 20% FCF. The fermented chickpeas might introduce off-flavors at higher concentrations, reducing taste and odor scores. As for the crumbly color, control bread had the best color, followed by 5% and 10% FCF. Higher FCF levels resulted in significantly lower color scores. FCF at higher levels might impart an undesirable color to the bread, affecting appearance and crumb color scores. Control bread excelled in odor, followed by 5% FCF. Odor scores decreased with increasing FCF levels. Control bread had the highest score for the overall acceptability, followed by 5% FCF. Acceptability scores dropped significantly at higher FCF levels. While Sayaslan and Şahin (2018) observed improved cell structure and uniformity in whole-wheat bread with 15% and 30% levels of fermented chickpeas, suggesting potential textural benefits of FCF at specific concentrations. Da Costa *et al.* (2020) observed no sensory differences in bread with varying chickpea flour concentrations, suggesting potential of FCF use without compromising acceptability.

Conclusion

It could be concluded that the fermentation process of chickpea flour until the third day under controlled conditions showed a high efficiency in increasing the content of free amino acids and significantly reducing the percentage of phytic acid, which is considered an anti-nutritional compound. Thus, it is possible to obtain FCF suitable for use in the manufacture of various baked products. Also, addition of FCF in bread-making leads to an increase in nutritional and health values, as the proportions of protein, ash, fat, and dietary fiber were increased. The substitution of wheat flour with FCF at various ratios does not significantly alter the overall lightness (L^*) of bread crust. In addition, bread samples containing different ratios of FCF exhibited a higher concentration of health-promoting compounds, such as total phenolic content and antioxidant activity, compared to control bread. The results of sensory evaluation of bread samples containing FCF indicated that the panelists accepted all sensory characteristics under study, with no abnormal properties of breads, compared to the control bread. Finally, it was concluded that fermenting chickpeas and adding FCF to bread-making showed many nutritional benefits because of the higher levels of proteins and bioactive compounds, which contributed to solving the problem of malnutrition among masses. It also contributed economically to reducing the amount of imported wheat, especially in countries that suffer the shortage of locally grown wheat.

Data Availability

All relevant data used to support the current research findings are included in the article. The raw data are available at the Department of Food Science and Human Nutrition, College of Agriculture and Food, Qassim University, KSA.

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Author Contributions

Conceptualization: Mohamed G.E. Gadallah and Ali A. Aljebreen; data collection and preparation: Mohamed G.E. Gadallah and Ali A. Aljebreen; creation and writing the initial/original draft: Mohamed G.E. Gadallah and Ali A. Aljebreen; methodology: Mohamed G.E. Gadallah and Ali A. Aljebreen; and application of statistics and supervision and fund acquisition: Mohamed G.E. Gadallah. Both authors read and agreed to the published version of the manuscript.

Conflicts of Interest

The authors declared no conflict of interest.

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