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Research Article

Effect of Sunflower Meal Protein Isolate (SMPI) Addition on Wheat Bread Quality

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Abstract

The effect of substituting wheat flour with 0%, 1%, 3%, 6% and 9% SMPI on wheat bread quality and nutritional value was studied. The partial substitution of wheat flour with SMPI influenced the structure of the bread ($P<0.05$), causing a small decrease in volume and higher hardness compared to 100% wheat bread. Addition of 9% SMPI in wheat increased the nutritional value of wheat bread in terms protein and amino acid content with only a small reduction in bread quality. Overall acceptability score revealed that bread supplemented with 1% SMPI was acceptable to the panelists and there was no significant difference in terms of taste and texture compared to the control. These findings are helpful and show how low-cost protein may improve the nutritional quality of wheat bread; it also increased potential applications of sunflower meal residue.

Keywords: Sunflower meal protein, wheat flour, nutritional value, bread, sensory evaluation.

Introduction

Bread has always been a critical contributor to human nutrition in many countries as it constitutes a significant source of nutrients like proteins, fibers, minerals and vitamins (Ahmed *et al.,* 2012; Thompkinson *et al.,* 2014). In the recent two decades, consumer eating trends towards healthy and functional food intake has been observed worldwide (Annapure *et al.,* 1998). The use of plant constituents of choice in various food matrices aimed at increasing their consumption in the human diet as well as explore and develop new functional foods has been on the rise. Bread, as a food matrix has been commonly used as a food vehicle to deliver many essential nutrients (Khoshgozaran-Abras *et al.,* 2014). Bread made from refined flour is nutritionally limited and does not sufficiently meet the necessities of many macro or micro-nutrients. It has been reported that bread made from refined wheat flour has low micronutrient composition (Al-Kanhal *et al.,* 1990; Isserliyska *et al.,* 2001). Additionally, wheat protein is deficient in essential amino acid content such as lysine, threonine and valine. Thus, there have been numerous ongoing researches to enhance the nutritional value of bread to enable compliance with the ever increasing recent dietary habits demands, such as the need for balanced protein content, mineral, vitamin and fiber content of the product.

Cereal products supplemented by different ingredients have been gaining a wide customer acceptance globally. Sunflower meal is a rich source of proteins that is considered a vegetable raw material, containing secondary materials such as oilseed meals; a residual material of the edible oil industries (Rodrigues *et al.,* 2012). It is a protein-rich constituent and can be applied to produce highly valuable proteins and other compounds for the development of functional bread and baked products that meet the conditions of the present functional food market. Based on this manner, bread will be an important source of functional proteins, carbohydrates, vitamins B-group and minerals (Chavan *et al.,* 1987). Food and Agriculture Organization of the United Nations (FAO, 2012), reported that soybean, rapeseed and sunflower seeds are among the world's most significant oilseeds in the world production of approximately 472 million tons in 2011/2012 (FAO, 2012). From a nutritive perspective, sunflower seeds contain about 20% crude proteins, however total protein content of the sunflower ranges from 30-50% when seeds are mechanically pressed (Dorrell and Vick, 1997). This high protein amount makes sunflower meal (SFM) an excellent source of proteins. The oil extraction technique determines the suitability of SFM proteins for food uses.

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Protein denaturation may happen during seed conditioning, expelling (temperature up to 140°C is reached) and roasting (Parrado *et al.,* 1993). Sunflower proteins have been assessed widely as food ingredients; however, their applications in food are still limited. These applications are mainly based on the supplementation of foods by sunflower meal, particularly meat and milk products, infant formulae, bakery products and pasta products (Bilani *et al.,* 1989; Bruckner and Mieth, 1984). Therefore, the main objective of this study was to produce low-cost protein from sunflower meal, obtained by mechanical press technique. Also, to examine the effect of partial substitution of wheat flour with the sunflower meal protein isolate (SMPI) on the bread making, bread quality, nutritional value and sensorial properties.

Materials and methods

Raw materials: Sunflower seeds (5.69% water content, 37.93 oil content and 24.20% protein content) were purchased from Shandong Lu Hua Group Ltd. (Shandong, China). The crude oil of sunflower seeds was removed by a mechanical press technique and sunflower meal was kept in a plastic bag for further analysis. Commercially obtainable refined wheat flour (12.80% protein, 0.51%, ash content, and 11.5% moisture content) was purchased from the local market.

Extraction of Sunflower Meal Protein Isolate (SMPI): The production process of SMPI was done according to the method described by (Ge *et al.,* 2000) with some modifications. Briefly, SMPI was mixed with deionized water (1:10, w/v) for two hours at room temperature. The suspension was adjusted to pH 10 by adding drops of 2M NaOH, followed by centrifugation at 8000 rpm for 30min at 4°C using a refrigerator centrifuge (Himac CR21 GLL, Japan). The supernatant was adjusted to pH 4.5 by adding drops of 2M HCl in order to precipitate the proteins, followed by centrifugation at 8000 rpm for 20 min at 4°C. The precipitates were washed three times with deionized water and then dispersed in deionized water and the pH adjusted to 7. The protein obtained was freeze-dried, sealed in an air-tight polyethylene bag and kept in a freezer (–20°C) until further analysis.

Preparation of bread: The basic recipe for bread making used follows ICC Standard No.131. Briefly, 300 g flour (wheat flour as control or wheat flour replaced with sunflower meal protein isolate (SMPI) powder; 1%, 3%, 6% and 9%) was mixed in a mixer bowl for 1 min. Afterwards, 9 g sugar, 3 g sodium chloride, 4.5 g fresh compressed yeast dissolved in water were added and water was added to achieve 500 BU consistency. Dough kneading process was done for 5 min, then 5% butter was added and kneaded for additional 3 min. The kneaded dough was placed in baking pans and proofed at 30°C for 60 min with 85% relative humidity. After proofing, the dough was punched down to eliminate gases and proofed for an additional 60 min, baked for 30 min at 205°C. After cooling, 5 pieces of bread were randomly selected from each sample and subsequently freeze-dried, crushed, sieved through an 80-mesh screen and stored at –20°C prior to analysis.

Bread quality analysis: Four hours after baking, analysis of the bread quality was determined. Loaves were mechanically cut transversely into 12 mm thick slices by an electric bread slicer machine (Sinmag Bakery Equipment, Wuxi Co., Ltd., Wuxi City, Jiangsu, China). Texture profile analysis (TPA) of bread parameters measured included hardness, resilience, cohesiveness, springiness and chewiness. A Texture Pro CT V 1.4 Build 17 (Brookfield Engineering Laboratory, Middleboro, MA, USA) fitted with an aluminum 25 mm diameter cylindrical probe was used. The slices from the center of each loaf were used to assess crumb texture. A stack of two slices (25 mm total) was prepared and compressed to 50% of its original thickness. The test conditions were pre-test speed, 2 mm/s; test speed, 0.5 mm/s; return speed, 0.5 mm/s; and trigger load, 7 g. Each sample was measured thrice and the final result was an average. Bread quality factors included weight, volume (carried out by rapeseed displacement) and specific volume was calculated.

Hunter color values of bread crumb and crust: Hunter color values of both crumb and crust were measured with a Minolta colorimeter (Lab Scan XE, Hunter Association Laboratory, Reston, VA, USA). Before measuring, the colorimeter was calibrated using a standard white plate. The color was measured from three different positions namely bread surface (crust) and inside (crumb). Color intensity was measured and expressed based on the values of L*, a* and b*. Where "L*" represents whiteness (value 100) or blackness (value 0); "a*" represents red (+a*) or green (–a*), and "b*" represents yellow (+b*) or blue (–b*).

Chemical analysis: Moisture content, ash content and total protein content of the bread was determined by following the ICC standard methods (Williams *et al.,* 2008). All the measurements of the analyzed samples were done in triplicate.

Amino acid composition of bread: Amino acid composition of bread samples was determined by a high-performance liquid chromatography (HPLC) using an Agilent 1100 (Agilent Technologies, Palo Alto, CA, USA) assembly system with UV detector operated at 338 nm. Amino acids were analyzed according to the method described by Huang *et al.* (2011) with some modifications.

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Table 1. Effect of SMPI substitution on the bread texture.

SMPI: WF	0:100	1:99	3:97	6:94	9:91
Hardness	1207.30±9.85[c]	1217.20±50.4[c]	1698.4±12.7[a]	1556.2±30.69[b]	1500.30±44.58[b]
Resilience	0.35±0.03[a]	0.33±0.02[ab]	0.31±0.03[ab]	0.32±0.01[ab]	0.31±0[b]
Cohesiveness	0.77±0.03[b]	0.79±0[ab]	0.77±0.03[ab]	0.82±0.02[a]	0.79±0.01[ab]
Springiness (mm)	0.95±0.05[c]	0.97±0.01[a]	0.95±0[c]	0.96±0.01[ab]	0.95±0[bc]
Chewiness (mj)	894.91±50.07[b]	970.61±41.24[b]	1298.1±6.59[a]	1268.3±3.82[a]	1220.2±50.84[a]

*Each observation is a mean ± SD of 3 replicate experiments.

Table 2. Effect of SMPI substitution on the bread characteristics.

SMPI: WF	0:100	1:99	3:97	6:94	9:91
Loaf weight (g)	84.8550±3.56[a]	87.7200±2.84[a]	89.8±1.98[a]	88.4950±1.77[a]	88.7550±1.78[a]
Loaf volume (mL)	334±5.66[a]	327±2.82[a]	302.5±4.62[b]	326±5.28[a]	303.5±5.80[b]
Loaf specific volume (mL/g)	4.04±0.057[a]	3.78±0.13[ab]	3.42±1.98[b]	3.68±0.022[ab]	3.65±0.32[ab]

One gram of sample was dissolved in 20mL of 6N HCl and poured into a hydrolysis tube with screw cap and then hydrolyzed for 22 h under a nitrogen atmosphere. The extracted sample was taken and dissolved in 4.8 mL 10M NaOH to neutralize for acid hydrolysis. After dilution to a known volume and filtration, the hydrolysate (1.0 µL) was injected into HPLC column.

Sensory evaluation: Sensory analysis was carried out on the bread 6 h after baking process. Bread samples were cut into equal sizes (6 cm × 6 cm) and presented to the panelist in color-coded plates. 20 students (12 females and 8 male) from the School of Food Science (Oil and protein laboratory, Jiangnan University), aged 24–35 years old were selected for sensory evaluation. Sensory evaluation was carried out in a laboratory under artificial daylight at ambient temperature. The panelists were requested to score the quality traits including; crumb color, crust color, taste, texture and overall acceptability of the bread using a nine-point hedonic scale, where 9 points to extremely like and 1 extremely dislike (Ihekoronye and Ngoddy, 1985).

Statistical analysis: The experimental data were subjected to analysis of variance using the SPSS statistical software, version 16.0 (SPSS, Chicago, Illinois, USA). All data was presented as the mean ± standard deviation (SD) of triplicate analyses, except for the sensory evaluation (n=20) and color measurements (n=6). Duncan's test was used to determine the significant differences amongst the samples means at the level of (P<0.05).

Results and discussion

Effect of SMPI substitution on the bread texture: Bread textural properties were assessed using a texture profile analysis. The results are presented in Table 1. Sensory texture attributes of bread such as appearance, taste and flavor are perceived by the consumer to indicate final product acceptability and palatability.

Product quality assessment can be determined through texture analysis, an essential tool for bread product improvement that creates a link between eating qualities, flavor and shelf life. In this study, wheat flour substitution with sunflower protein isolate resulted in significant (P<0.05) bread crumb hardening. At the highest substitution level (9% SMPI), the crumb was 24.26% harder than the wheat bread control. This may be attributed to the higher water absorption capacity of SMPI added, which has a dehydration effect on wheat dough thus leading to a harder texture. This is in accordance with Totosaus *et al.* (2013) who reported that supplemented proteins in wheat dough compete with gluten for available water which results in increased hardness. The product firmness when determined by the resilience value, can recover quickly from deformation, if it is small (Bhol and Bosco, 2014). Our findings revealed that the bread sample with 9% SMPI was firmer than the other bread samples, whereas cohesiveness and springiness were not significantly different among composite bread samples. Chewiness was higher as the amount of SMPI increased from 894.91 (control) to 1220.2 (9% SMPI).

Effect of SMPI substitution on the bread characteristics: Bread volume decreased significantly with increasing percentage of SMPI substitution (Table 2). The control wheat bread had higher volume compared with composite bread loaves as expected. This might be attributed to the high gluten content of wheat flour. There was a significant (P<0.05) decrease in specific volume among the composite bread loaves when 1%, 3%, 6% and 9% of SMPI were added (Table 2). The decrease in loaf volume of composite bread might be related to the dilution effect of SMPI on wheat dough gluten matrix during fermentation, resulting in less gas (CO_2) retention and subsequently a low loaf volume in bread after baking. In a previous study, a reduction in bread loaf volume due to the addition of fermented/germinated cowpea flour to the wheat dough was also reported (Hallen *et al.*, 2004).

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Table 3 Effect of SMPI substitution on color properties of bread.

	Crust color			Crumb color		
SMPI:WF	L*	a*	b*	L*	a*	b*
0:100	83.91 ± 0.11[a]	3.3 ± 0.07[a]	16.73 ± 0.17[a]	91.67 ± 1.41[a]	0.28 ± 0[a]	11.47 ± 0.54[a]
1:99	82.98 ± 1.04[a]	4.26 ± 0.03[b]	12.75 ± 0.62[b]	82.62 ± 0.34[b]	1.65 ± 0.03[c]	13.84.75 ± 0.16[b]
3:97	75.76 ± 0.62[b]	6.67 ± 0.02[c]	10.93 ± 0.02[c]	75.46 ± 1.71[c]	2.17 ± 0.02[d]	16.54 ± 0.45[d]
6:94	71.34 ± 0.05[c]	6.55 ± 0.05[c]	10.19 ± 0.04[c]	72.6 ± 0.33[d]	2.11 ± 0.03[d]	18.59 ± 0.10[c]
9:91	70.08 ± 1.18[b]	8.42 ± 0.27[d]	5.95 ± 0.02[d]	73.38 ± 0.23[cd]	0.24 ± 0.02[b]	21.63 ± 0.50[a]

Table 4. Chemical composition (%) of composite bread.

SMPI: WF	0:100	1:99	3:97	6:94	9:91
Moisture	33.75±0.16[d]	34.87±0.06[c]	37.71±0.09[a]	36.79±0.24[b]	37.36±0.06[a]
Protein	18.84±0.03[d]	19.82±0.05[c]	22.95±0.05[b]	23.10±0.14[b]	24.12±0.17[a]
Ash	0.97±0.01[a]	0.97±0[a]	0.98±0[a]	0.98±0[a]	0.98±0[a]

However, the specific volume of bread is an important quality factor because it is connected to dough-inflating capacity and oven spring and must not be large or small as it affects the crumb structure (Bhol and Bosco, 2014).

Effect of SMPI substitution on color properties of bread: The results effect of SMPI substitution on the color properties of wheat bread is presented in Table 3. The color properties were determined by Hunter the L*, a*, and b* values related to lightness, redness, and yellowness, respectively. In the crumb, the L* values gradually decreased, while a* and b* values increased with the addition of SMPI in wheat bread. The crumb of the control bread was lighter (L*) with less red color (a*) than any of the other bread samples. In the crust, an increase in SMPI substitution resulted in a* reduction in the L* and b*. In the bread made at substitution ratios of 6% and 9%, the lightness (L*) and yellowness (b*) values of the crust were significantly different ($P<0.05$) from the other samples. Additionally, the higher values were observed among the composite bread samples. The lower L* and higher a* values in the crumb and crust of composite bread may be attributed to their higher protein and lysine content due to the addition of SMPI as well as the Maillard and caramelization reactions during baking (Aboshora *et al.*, 2016).

Amino acid composition of bread: In this study, SMPI was added to bread formula to enhance its nutritional value. From systematical analysis of the information, several studies on the biological or nutritional value of bread have shown that white bread is limited in some essential amino acids (Shchekoldina and Aider, 2014). The effect of SMPI substitution on the amino acid composition of wheat bread is shown in Fig. 1. The addition of SMPI in the wheat bread formula improved the content of both essential and non-essential amino acids.

Bread sample supplemented with wheat flour: SMPI, 91:9; had a higher essential and non-essential amino acid contents (lysine, threonine, arginine, leucine, and phenylalanine). This was followed by sample supplemented with 94:6, 97:3, 99:1 and lowest was the control sample. Similarly, valine (1.22 g/100) and methionine (0.465 g/100) content were increased in bread containing 9% SMPI supplemented bread. Histidine and Glycine increased by 44.77% and 45.45%, respectively, compared with the control bread. The overall amino acid content increased at the all supplementation levels. Hence, the addition of SMPI to wheat bread formula significantly ($P<0.05$) increased the amino acid profile of composite SMPI-wheat flour bread, especially the essential ones. This could be attributed to higher amino acid contents of SMPI than wheat flour which suggests that substitution of wheat flour with SMPI in bread improved the nutritional contents. Similarly, El-Adawy (1997) and Alu'datt *et al.* (2012) reported that addition of barley protein isolate and sesame protein concentrate/isolate respectively increased amino acid composition of wheat bread, Alu'dattet *et al.* (2012) found that total amino acid of 100% wheat bread and composite bread containing 10% barley protein isolate were 28.18 and 42.50 g/100 g, respectively.

Chemical composition of composite bread: The effect of substitution of SMPI on the proximate composition of the control and SMPI-WF bread is presented in Table 4. The moisture content increased significantly ($P<0.05$) with increasing percentage of SMPI, while ash content was not affected. The higher moisture content of composite SMPI-wheat flour bread compared with the control bread might be attributed to high water absorption capacity of SMPI (Chinma *et al.*, 2015). The higher protein content of composite SMPI-wheat flour bread samples was anticipated due to higher protein contents of SMPI (18.84%, 19.82%, 22.95%, 23.10% and 24.12%) for 0%, 1%, 3%, 6% and 9% respectively, for the wheat flour substituted with SMPI.

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The higher protein content of composite SMPI-wheat flour bread could suggest higher nutritional value compared with control bread (Alu'datt *et al.*, 2012).

Sensory analysis of bread samples: Sensory characteristics of baked products have been widely evaluated over the years. The effect of substitution of SMPI on the sensory properties of wheat bread is presented in Fig. 2. The crumb and crust color, taste and texture scores of composite SMPI-wheat flour bread were tested based on a nine-point hedonic scale, which from the results ranged from 5.16 to 8.23; thus indicating that these bread was reasonably acceptable. The scores of composite SMPI-wheat flour bread dropped slightly with an increase in substitution level of SMPI in the blends. These traits showed a significant difference at 9% substitution level when compared to control (100% wheat flour) bread. Composite bread containing 9% SMPI were much darker in crumb and crust color than other bread samples ($P<0.05$) and this negatively impacted bread acceptability. The variation in crumb and crust color of composite SMPI-wheat flour bread could be due to addition effect caused by increasing the level of sunflower meal protein isolate in the blends. On the other hand, the reduction in crumb and crust color scores of composite SMPI-wheat flour bread could be attributed to thermal non-enzymatic caramelization and maillard reactions between reducing sugars and amino acids, due to increased protein and lysine contents in the composite bread than control bread. Especially given that SMPI samples had higher protein and lysine contents (Chinma *et al.*, 2015). Higher levels of the SMPI addition, particularly 9% SMPI also caused an unpleasant taste and had the lowest appearance, while SMPI addition up to 1% had no significant effect on bread texture. Majority of the panelist's results indicated that a partial replacement of wheat flour in bread with up to 1% SMPI gave satisfactory overall consumer acceptability. However, bread containing 9% SMPI were graded relatively lower, which might be due to excessive amounts of the SMPI negatively affecting color, taste and texture.

Conclusion

From the general results, it could be concluded that the substitution of wheat flour with 0%, 1%, 3%, 6% and 9 % sunflower meal protein isolate in wheat flour influenced the bread quality to a different extent. Substitution of SMPI in bread recipe improved its nutritional value, with only a small reduction in bread quality. Organoleptic properties have shown that the substitution with up to 1% was similar to 100% wheat bread, even though the bread volume was impaired by the addition of SMPI. The protein and amino acid contents of wheat bread were significantly ($P<0.05$) increased due to the addition of SMPI. Bread texture was significantly impaired especially hardness.

Fig. 1. Nutritional value of the sunflower meal protein isolate (SMPI) supplemented bread expressed as its amino acid profile.

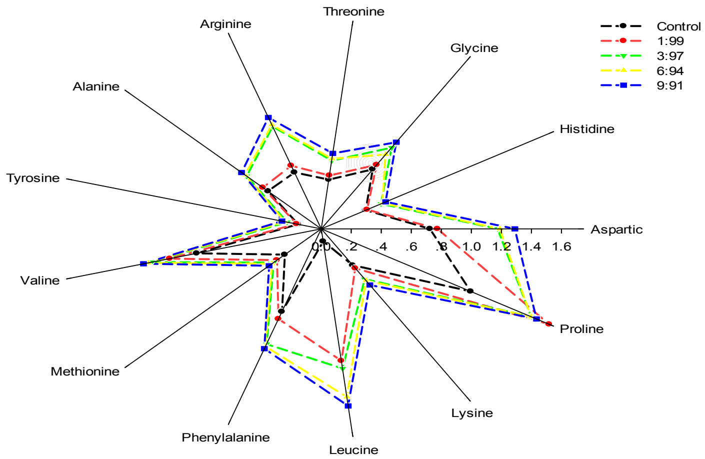
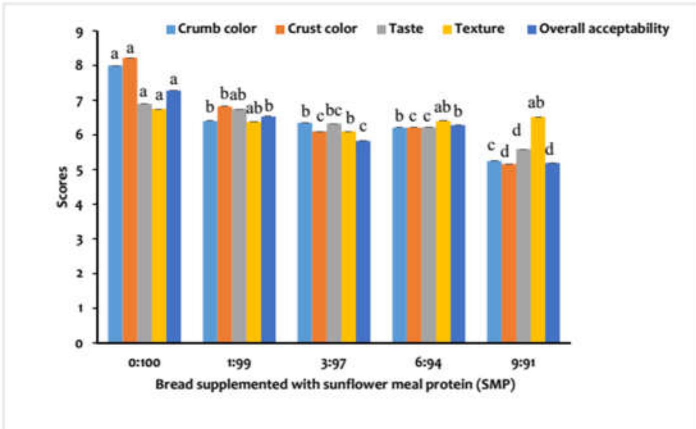


Fig. 2. Hedonic sensory evaluation scores of control bread and bread made from wheat–SMPI composite flour.



Results are expressed as mean ± standard deviation (SD) (n= 20). Same-colored columns with different letters indicate significant difference (P<0.05).

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