Pan bread quality as affected by some nano and fermented-nano food industries by-products

El-Sayed I. Yousif 1, Attia A. Yaseen 2, Abdel-Fatah A. Abdel-Fatah 1, Abdel-Hafeez A. Shouk 2, Mohamed G. Gadlla 1 and Ayman A. Mohammad 2*

Abstract

Background: Rapid development of nanotechnology is expected to transform many areas of food technology with increasing investment and market share. Also, phytochemical-rich foods have attracted consumer’s attention due to their ability to promote benefits for human health. So, in this study, the suitability of some food industry by-products [nano-wheat bran (NWB), nano-wheat germ (NWG), fermented nano-rice bran (FNRB), fermented nano-carrot pomace (FNCP), and fermented nano-pomegranate peel (FNPP)] as supplements for pan bread was investigated.

Results: Loaf volume significantly (p ≤ 0.05) decreased, while loaf weight increased by increasing the level of all tested materials as compared to control bread. Hardness and chewiness of bread samples increased, while springiness and cohesiveness decreased by increasing the level of replacement of all nano-materials. The control bread had the lowest value of alkaline water retention capacity (AWRC) being 326, 292, 265, and 237%, respectively for 3, 24, 48, and 72 h of storage time at room temperature. At all levels of replacing, noticeable increase of AWRC was detected as well as retarding staling rate of bread during storage.

Conclusion: The sensory evaluation results of bread indicated that functional pan bread with acceptable quality could be prepared from NWB, NWG, and FNRB up to 15% and FNCP and FNPP up to 5% replacement level.

Keywords: Food by-products, Solid-state fermentation, Nanotechnology, Pan bread

Background

In the last decades, consumer demands in the field of food production have changed considerably. Consumers more and more believe that foods contribute directly to their health (Mollet and Rowland 2002). Today foods are not intended to only satisfy hunger and to provide necessary nutrients for humans but also to prevent nutrition-related diseases and improve physical and mental well-being of the consumers (Roberfroid 2000; Menrad 2003). In this regard, functional foods play an outstanding role; the increasing demand on such foods can be explained by the increasing cost of healthcare, the steady increase in life expectancy, and the desire of older people for improved quality of their later years (Siro et al. 2008).

In recent years, attention has turned to plant foods as a source of dietary fiber and phytochemicals with biological activity due to their ability to promote benefits for human health, such as reduction in the incidence of some neurodegenerative diseases, reduction in the occurrence of factors linked to cardiovascular disease, and antioxidant, antimutagenic, anti-allergenic, anti-inflammatory, and antimicrobial activities (Tripoli et al. 2007; Martins et al. 2011). Furthermore, increased consumption of dietary fiber improves immune function (Zhao et al. 2015) and colon cancer (Hu et al. 2009).
Polyphenols or polyphenol-rich food by-products have been widely incorporated in dough to prepare various baked food products such as breads, cakes, muffins, and cookies to improve their functional properties and reduce the content of foodborne toxins (Sabally et al. 2016; Martins et al. 2017; Yeh et al. 2017; Gomez and Martinez 2018; Zamora and Hidalgo 2018; Ou et al. 2019).

The accessibility of the bioactive compounds with health benefits may also be limited, as they are trapped into rigid cell structures (Hemery et al. 2007). So there is a need for a new processing technology that can efficiently produce novel ingredients with optimized techno-functional and nutritional attributes. Consequently, several strategies have been suggested including the use of by-product pretreatments such as fermentation and enzymatic or heat treatments that may enhance the bioaccessibility of phenolics (De Kock et al. 1999; Salmenkallio-Marttila et al. 2001; Penella et al. 2008). Also, micro and nanotechnology are emerging technologies which show great potential in nutraceuticals and functional foods for human health and improving food quality and safety (Chen et al. 2006; Esfanjani and Jafari 2016; Rodrigus et al. 2017). So, different processing methods can be employed to modify the bran structures and composition to enhance the bioaccessibility by wet-fractionation processes (enzymatic treatments and fermentation) (Katina et al. 2005; Moore et al. 2006) or dry-fractionation processes (ultra-fine grinding, air-classification, and electro-static separation) (Antoine et al. 2004; Hemery et al. 2007).

Solid-state yeast treatments significantly increased the radical scavenging ability of cereal brans due to increasing the releasable phenolic compounds (Moore et al. 2007; Prabhu et al. 2014). Their data supports the fact that fermented bran can be utilized for enhancement of functional properties, giving way to further studies for fermented bran to be incorporated into food. On the other hand, Hemery et al. (2010) evaluated the potential of using ultrafine grinding and electrostatic separation methods to improve the bioaccessibility of phenolic acids in wheat bran-rich breads. It was observed that the finer the bran particles in the bran-rich breads, the more bioaccessible the phenolic acids.

The present study was carried out to develop sustainable methods such as solid state fermentation and superfine grinding (nano size) as well as the combination between them in order to modify the structure of tested food by-products (wheat bran, wheat germ, carrot pomace, rice bran, and pomegranate pomace). Moreover, wheat flour was substituted using the obtained nano and fermented nano-food by-products at the levels of 5, 15, and 25% to prepare functional pan bread. Baking quality, organoleptic characteristics, texture properties, staling, and color attributes of pan bread were evaluated.

Materials and methods

Raw materials
Wheat flour (72%), wheat flour (82%), whole-meal wheat, wheat bran, and wheat germ were obtained from the North Cairo Flour Mills Company, Egypt (2013). Rice bran was obtained from the Rice Research and Training Centre, Sakha, Kafr El-Sheikh, Egypt (2013). Carrot (Daucus carota) and pomegranate fruits (Punica granatum L.) were purchased from the local market, Giza, Egypt (2013). Active dry yeast (Saccharomyces cerevisiae) was obtained from the Egyptian Sugar and Integrated Industries Company (ESIIC), Chemicals Factory, El-Hawamdia City, Giza, Egypt (2014). Salt, shortening, and sugar were purchased from the local market, Egypt (2015).

Stabilization of wheat germ and rice bran
Wheat germ and rice bran were stabilized in an air-oven at a temperature of 120 ± 2 °C for 1 min according to Younas et al. (2011). The stabilized wheat germ and rice bran were ground using a Moulinex grinder and passed through a 40-mesh (420 μm) and packed in polyethylene bags and stored at −30 °C until use.

Preparation of carrot pomace and pomegranate peel
Carrot pomace and pomegranate peel obtained after juice extraction were dried in an air-oven at 50 ± 1 °C for 16 h. The dried sample was ground using a Moulinex grinder and passed through a 40-mesh sieve and packed in polyethylene bags.

Solid-state yeast fermentation
Yeast strain (Saccharomyces cerevisiae FC-620) was obtained from Microbial Chemistry Dept. collection, National Research Centre, Dokki, Cairo, Egypt. The yeast cells were activated; a loopful of the culture was transferred to 250-mL Erlenmeyer flask containing 50 mL broth medium (0.3% yeast extract, 0.3% malt extract, 0.5% peptone, and 5% sucrose) and incubated for 24 h at 32 °C under shaking condition. Solid-state yeast treatment was carried out according to the method of Moore et al. (2007) as follows: 50 mL of yeast preparation (1380 cfu/mL) was mixed with 100 g sample in a sterile conical flask. Flasks were sealed with cotton seals and incubated at 32 °C for 48 h. All treated samples were dried at 50 ± 1 °C for 16 h and stored in polyethylene bags at −30 °C until use.

Preparation of nano and fermented nano materials
Wheat bran and wheat germ as well as fermented rice bran, fermented carrot pomace, and fermented pomegranate peels were ground using PM 100 Planetary Ball-mill (Fig. 1), Retsch, Germany, as described by Zhu et al. (2010) with some modifications. In brief, samples...
(150 g) were put in a zirconium oxide bowl (Fig. 1a) with zirconium oxide balls (Fig. 1b). The bowls were fixed in their bowl's tray (Fig. 1c); then, the ball-mill was closed and adjusted at 30-Hz frequency for 60 min at room temperature.

Transmission electron microscopy (TEM)
All ground samples were examined with a JEOL JX 1230 technique with micro analyzer probe, Japan. This technique was used to determine the particle size of the investigated samples according to Casuccio et al. (2004). The TEM images show that the particle size of WB, WG, RB, CP, and PP was distributed in a range of $10^{10} - 21$, $7^{10} - 19$, $15^{10} - 47$, $8^{10} - 58$, and $21^{10} - 35$ nm, respectively, which indicated that they are in the nano scale.

Preparation of functional formulas
Nano-wheat bran, nano-wheat germ, fermented nano-rice bran, fermented nano-carrot pomace, and fermented nano-pomegranate peel were well blended with wheat flour (72% extraction) replacement at the levels of 5, 15, and 25%. Fifteen blends were prepared, and three control samples were made with 100% wheat flour 72%, wheat flour 82%, and whole-meal wheat flour for comparison.

Baking quality of pan bread
Pan bread was processed according to procedure described by Lazaridou et al. (2007). Weight, volume, and specific volume of the produced bread loaves were determined as described in AACC (2000). Volume of loaves was measured using rapeseeds displacement method.

Color measurements
The color of bread samples was measured using Hunter Lab scan XE with the CIE color scale. This instrument was standardized against the white tile of Hunter Lab color standard (LX No.16379): $X = 77.26$, $Y = 81.94$, and $Z = 88.14$. The $L^*$, $a^*$, and $b^*$ values were reported. Total color difference ($\Delta E$) was calculated as:

$$\Delta E = [(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2]^{0.5}$$

Sensory evaluation of pan bread
Sensory evaluation was conducted for the freshly baked breads by 10 semi-trained panelists from the staff (male and female) aged from 25 to 60 years old from the Food Industries and Nutrition Division, National Research Centre, Egypt. The sensory evaluation was conducted in a laboratory under ambient temperature (25°C) as described by Kulp et al. (1985) for symmetry of shape (5), crust color (10), break and shred (10), crumb texture (15), crumb color (10), aroma (20), taste (20), and mouth feel (10).

Texture properties of bread crumb
Texture parameters (hardness, adhesiveness, springiness, cohesiveness, gumminess, and chewiness of bread samples) were measured objectively by using a texture analyzer TA-CT3 (Brookfield, USA) as adopted by the standard method by AACC, method 74-09 (AACC 2000).

Freshness of pan bread
Loaves freshness of each packed sample was tested at room temperature (25°C) during storage for 3, 24, 48, and 72 h by alkaline water retention capacity (AWRC) according to the method of Yamazaki (1953), as modified by Kitterman and Rubenthaler (1971).

Statistical analysis
All samples were analyzed in triplicates, and the results were expressed as means ± standard error. Analysis was assessed using the Statistical Analysis System software System for Windows (SAS System for Windows (Statistical Analysis System), 2008). The significant difference between the mean values were determined by using the analysis of variance (ANOVA), and Duncan’s multiple range test was conducted at a significance level of $p < 0.05$. 
Results

Baking quality of pan bread

Loaf weight, volume, and specific volume reduction of the produced pan bread are shown in Table 1 and Fig. 2. Incorporation of tested materials to wheat flour significantly \((p < 0.05)\) decreased loaf volume of final products. Maximum reduction of loaf volume was recorded to bread made from wheat flour incorporated with FNCP and FNPP at 25% and wheat flour 100% \((202, 211, \text{ and } 214 \text{ cm}^3)\) with about 53.27, 51.71, and 51.09% specific volume reduction, respectively compared to those made from wheat flour 72% \((432.50 \text{ cm}^3)\).

In contrast, loaf weight of bread made from wheat flour incorporated with fiber materials significantly \((p < 0.05)\) increased compared to control sample. Maximum increase was recorded to FNWB at 25% \((138.25 \text{ g})\) compared to 134.75 g for the control sample. Similar findings were found in the calculated specific loaf volumes. On the other hand, quality characteristics of bread samples incorporated with nano and fermented nano materials were improved when compared to whole-meal bread. For instance, loaf volume property rated all tested sample higher than whole-meal sample except those containing FNPP at 25% \((211 \text{ cm}^3)\).

Organoleptic characteristics of pan bread

Table 2 presents the results obtained in the subjective evaluation of organoleptic characteristics of the control and the fiber-enriched bread samples conducted with 10 panelists. The bread made from wheat flour (72%) had the highest scores for all measured characteristics, whereas minimum scores were given to the bread prepared from wheat flour incorporated with 25% FNPP and FNCP. It was possible to observe that increasing the level of NWG, FNCP, and FNPP caused an increase in darkness (Fig. 2). Moreover, samples incorporated with FNRB, FNCP, and FNPP at the level of 25% were given the lowest score for break and shred \((5.9–4.9)\), crumb texture \((9.5–7.9)\), and mouth feel \((4.9–4.1)\) compared to 9.2, 14.2, and 9.0 for the control sample. The statistical analysis showed no significant differences between the crust color, break and shred, crumb texture, and mouth feel.

| Sample | Weight (g) | Volume (cm³) | Specific volume reduction (%) |
|--------|------------|--------------|------------------------------|
| Pan bread made from wheat flours (control samples) | | | |
| 72% extraction | 134.75 EF ± 0.25 | 432.50 A ± 2.50 | 0.00 ± 0.00 |
| 82% extraction | 135.25 EF ± 0.25 | 338.00 E ± 1.00 | 22.12 ± 0.68 |
| 100% extraction | 136.00 CDEF ± 0.50 | 214.00 K ± 0.150 | 51.09 B ± 0.36 |
| Pan bread made from wheat flour72% incorporated with | | | |
| Nano-wheat bran (%) | | | |
| 5 | 136.00 CDEF ± 0.50 | 373.50 B ± 1.50 | 14.33 L ± 0.71 |
| 15 | 137.25 ABC ± 0.25 | 342.50 D ± 2.50 | 22.12 ID ± 0.52 |
| 25 | 136.50 BCDE ± 0.50 | 271.50 I ± 1.50 | 38.01 DI ± 0.66 |
| Nano-wheat germ (%) | | | |
| 5 | 136.50 BCDE ± 0.50 | 349.00 D ± 1.00 | 20.25 J ± 1.18 |
| 15 | 136.75 BCDE ± 0.25 | 341.00 E ± 1.00 | 22.12 I ± 0.42 |
| 25 | 137.00 ABCD ± 0.01 | 302.50 G ± 2.50 | 31.15 F ± 0.58 |
| Fermented nano-rice bran (%) | | | |
| 5 | 137.50 AB ± 0.01 | 359.00 F ± 1.00 | 18.69 D ± 0.41 |
| 15 | 136.50 BCDE ± 1.0 | 305.00 G ± 5.00 | 30.53 F ± 0.36 |
| 25 | 138.25 A ± 0.25 | 288.00 I ± 2.00 | 35.20 E ± 0.49 |
| Fermented nano-carrot pomace (%) | | | |
| 5 | 137.25 ABC ± 0.25 | 318.00 F ± 2.00 | 27.75 G ± 0.45 |
| 15 | 136.75 BCDE ± 0.75 | 252.50 I ± 2.50 | 42.37 F ± 1.01 |
| 25 | 135.50 EF ± 0.01 | 202.00 H ± 2.00 | 53.27 A ± 0.82 |
| Fermented nano-pomegranate peel (%) | | | |
| 5 | 137.75 ABCD ± 0.25 | 337.00 F ± 3.00 | 32.36 H ± 0.94 |
| 15 | 135.75 DEF ± 0.25 | 267.50 I ± 2.50 | 38.32 D ± 0.48 |
| 25 | 136.00 CDEF ± 0.01 | 211.00 K ± 1.00 | 51.71 B ± 0.81 |

Values in the same column followed by different letters are significantly different \((p < 0.05)\)
crumb texture, and mouth feel of the control sample and those containing 5 and 15% NWB and 5% NWG. Also, there were no significant differences between crumb color of the control sample and those containing 5% NWB.

Regarding taste and aroma acceptance, all tested samples were acceptable for the panelists except the sample containing 25% FNPP. Moreover, there were no significant differences ($p < 0.05$) between the taste of control sample and those made from wheat flour (82%) and

Fig. 2 Photographs of pan bread as affected by addition of nano and fermented nano materials. NWB, nano wheat bran; NWG, nano wheat germ; FNRB, fermented nano-rice bran; FNCP, fermented nano-carrot pomace; and FNPP, fermented nano-pomegranate peel.
wheat flour incorporated with NWB at the levels of 5 and 15% (Table 2). Also, aroma of the samples containing 5 and 15% NWB did not differ significantly compared to the control sample. In general, the sensory test results suggested that addition of NWB, NWG, and FNRB up to 15% in bread formulations would not in general interfere with bread acceptability, while addition of FNCP and FNPP could not add more than 5%.

Texture profile analysis of pan bread
The results of the texture profile analysis (TPA) of bread samples are shown in Table 3. As can be seen, addition of fiber materials in formulation of bread samples significantly \((p<0.05)\) affected the textural properties of the produced bread. Hardness of bread samples were significantly increased by increasing the level of addition of all tested materials as compared to bread made from wheat flour 72% (white control sample), while the hardness of bread samples incorporated with all fiber materials, except FNPP at 25%, rated them lower than those made from wheat flour 100% extraction (control sample containing normal wheat bran).

There were no significant differences between springiness and cohesiveness values of control bread samples made from wheat flour 72, 82%, and whole-meal flour. Addition of both NWB and NWG up to 15% and FNCP up to 5% had no significant effects on the springiness and cohesiveness values, while addition of FNRB and FNPP at all tested levels significantly decreased the springiness and cohesiveness values of the produced bread. Bread samples containing FNCP at 15 and 25% recorded the lowest springiness values. Bread samples made from wheat flour incorporated with NWB at 25% showed higher cohesiveness value (1.00) (Table 3). All other tested materials at all tested levels negatively affected cohesiveness properties of the produced bread. In general, all tested samples showed higher chewiness values compared to the control sample. The highest chewiness value was determined in the whole-meal bread sample. The crumb chewiness of bread

### Table 2 Organeptic characteristics of pan bread as affected by addition of nano and fermented nano materials

| Sample | Symmetry of shape (5) | Crust color (10) | Break and shred (10) | Crumb texture (15) | Crumb color (10) | Aroma (20) | Taste (20) | Mouth feel (10) |
|--------|-----------------------|------------------|----------------------|-------------------|------------------|------------|------------|----------------|
| Pan bread made from wheat flours (control samples) | | | | | | | | |
| 72% extraction | 8.5 | 9.3 | 9.2 | 14.2 | 9.6 | 18.9 | 18.3 | 9.0 |
| 82% extraction | 5 | 7 | 6.9 | 10.8 | 7 | 15.2 | 16.1 | 7.8 |
| 100% extraction | 7 | 6.2 | 6.3 | 9.9 | 6.1 | 14.3 | 15.1 | 6.8 |
| Pan bread made from wheat flour 72% incorporated with | | | | | | | | |
| Nano-wheat bran (%) | | | | | | | | |
| 5 | 4.4 | 8.8 | 8.6 | 13.4 | 8.8 | 18.2 | 18.2 | 8.8 |
| 15 | 3.8 | 7.3 | 8.1 | 12.8 | 7.6 | 16.6 | 16.2 | 7.8 |
| 25 | 3.4 | 7.0 | 6.8 | 11.8 | 7.0 | 15.2 | 14.6 | 7.1 |
| Nano-wheat germ (%) | | | | | | | | |
| 5 | 3.8 | 7.6 | 7.9 | 12.7 | 7.8 | 16.1 | 15.3 | 7.8 |
| 15 | 3.1 | 5.8 | 7.0 | 12.1 | 6.6 | 14.4 | 12.5 | 6.6 |
| 25 | 2.5 | 4.4 | 5.4 | 9.6 | 5.6 | 13.4 | 11.5 | 5.9 |
| Fermented nano-rice bran (%) | | | | | | | | |
| 5 | 4.0 | 7.3 | 7.1 | 10.8 | 6.9 | 13.9 | 13.4 | 6.8 |
| 15 | 3.6 | 6.1 | 6.4 | 10.1 | 6.8 | 12.5 | 11.4 | 5.4 |
| 25 | 2.8 | 5.8 | 5.9 | 9.5 | 5.8 | 11.2 | 9.9 | 4.6 |
| Fermented nano-carrot pomace (%) | | | | | | | | |
| 5 | 3.1 | 6.8 | 6.6 | 10.3 | 6.6 | 14.3 | 13.8 | 6.8 |
| 15 | 2.5 | 4.9 | 6.0 | 9.2 | 5.3 | 11.4 | 12.1 | 5.3 |
| 25 | 1.8 | 4.0 | 5.0 | 7.9 | 4.2 | 11.9 | 12.2 | 4.1 |
| Fermented nano-pomegranate peel (%) | | | | | | | | |
| 5 | 2.7 | 5.4 | 5.7 | 9.3 | 6.0 | 12.9 | 11.9 | 5.2 |
| 15 | 2.3 | 4.5 | 5.2 | 8.0 | 4.5 | 11.9 | 11.3 | 5.3 |
| 25 | 1.7 | 3.7 | 4.9 | 7.9 | 3.8 | 9.1 | 7.8 | 4.9 |

Values in the same column followed by different letters are significantly different \((p<0.05)\)
incorporated with fiber materials changed in different ways. Addition of NWB, NWG, FNCP, and FNPP significantly increased chewiness value gradually with increasing the level of addition. While in the case of FNRB, the chewiness values gradually decreased (Table 3).

Color attributes of pan bread
The color characteristics ($L^*$, $a^*$, $b^*$, and $\Delta E$) of bread crust are given in Table 4. The crust of bread samples partially substituted with fiber materials had significantly ($p < 0.05$) lower lightness ($L^*$) value compared to control sample. On the other hand, composite breads which contained fiber materials tended to be lighter than whole bread sample except the samples containing FNCP and FNPP at 15 and 25%. Redness ($a^*$) values of the crust of bread samples were significantly different and showed different trends. Addition of NWB, FNRB, and FNCP at 25% significantly decreased $a^*$ values compared to control sample. On contrary, addition of NWG at all tested levels significantly increased $a^*$ value. The highest $a^*$ value was recorded to bread incorporated with 25% NWG (12.23). Yellowness ($b^*$) value of all bread crusts was significantly ($p < 0.05$) decreased compared to control sample. Whole-meal bread crust had lower $b^*$ value than the bread samples containing NWB and FNRB up to 25%, NWG up to 15%, and FNCP and FNPP up to 5%. The minimum $b^*$ value was measured in bread containing 25% FNCP.

Total color differences ($\Delta E$) between the crusts of tested bread samples compared to control sample are presented in Table 4. Generally, it was found that all tested materials gradually increased $\Delta E$ value of the produced bread as the addition level increased. Bread incorporated with 25% FNCP was the most dissimilar in color (41.39), whereas bread sample incorporated with 5% NWB or FNRB were the closest in color to control bread (7.89 and 8.33, respectively). Moreover, addition of both materials up to 25% gained lower $\Delta E$ values (10.74 and 15.56, respectively) compared to those manufactured using 82% and whole meal flours (14.84 and 21.96, respectively).

Table 3 Texture profile parameters of pan bread as affected by addition of nano and fermented nano materials

| Sample                                | Hardness (N) | Springiness (mm) | Cohesiveness | Chewiness |
|---------------------------------------|--------------|------------------|--------------|-----------|
| Pan bread made from wheat flours (control samples) |              |                  |              |           |
| 72% extraction                        | 4.87$^A$     | 7.51$^A$         | 0.986$^A$    | 36.05$^N$ |
| 82% extraction                        | 7.89$^b$     | 7.35$^A$         | 0.972$^A$    | 56.38$^M$ |
| 100% extraction                       | 37.10$^d$    | 7.33$^A$         | 0.907$^b$    | 246.77$^A$|
| Pan bread made from wheat flour 72% incorporated with |              |                  |              |           |
| Nano-wheat bran (%)                   |              |                  |              |           |
| 5                                    | 9.47$^k$     | 7.14$^{BCD}$     | 0.978$^A$    | 59.36$^M$ |
| 15                                   | 9.81$^k$     | 7.40$^{AB}$      | 0.912$^b$    | 66.24$^L$ |
| 25                                   | 15.03$^d$    | 6.96$^{DEF}$     | 1.000$^A$    | 104.61$^G$|
| Nano-wheat germ (%)                  |              |                  |              |           |
| 5                                    | 13.65$^d$    | 7.42$^{AB}$      | 0.901$^b$    | 91.28$^I$ |
| 15                                   | 14.32$^e$    | 7.26$^{ABC}$     | 0.902$^b$    | 93.79$^I$ |
| 25                                   | 26.03$^d$    | 6.90$^{DEF}$     | 0.868$^c$    | 155.92$^C$|
| Fermented nano-rice bran (%)         |              |                  |              |           |
| 5                                    | 17.24$^i$    | 7.04$^{CDE}$     | 0.822$^D$    | 99.81$^H$ |
| 15                                   | 17.70$^i$    | 6.82$^{EF}$      | 0.795$^O$    | 95.97$^A$ |
| 25                                   | 29.73$^c$    | 6.40$^G$         | 0.679$^E$    | 126.06$^C$|
| Fermented nano-carrot pomace (%)     |              |                  |              |           |
| 5                                    | 17.80$^i$    | 7.31$^{ABC}$     | 0.882$^{BC}$ | 114.80$^J$|
| 15                                   | 23.76$^e$    | 6.15$^{GH}$      | 0.802$^D$    | 117.19$^f$|
| 25                                   | 29.75$^c$    | 6.09$^H$         | 0.689$^!$    | 126.06$^C$|
| Fermented nano-pomegranate peel (%)  |              |                  |              |           |
| 5                                    | 13.83$^j$    | 7.36$^{AB}$      | 0.815$^O$    | 82.94$^K$ |
| 15                                   | 24.84$^d$    | 6.72$^E$         | 0.801$^D$    | 133.66$^!$|
| 25                                   | 39.16$^e$    | 6.74$^f$         | 0.618$^F$    | 163.22$^E$|

Values in the same column followed by different letters are significantly different ($p < 0.05$)
Freshness properties of pan bread

Alkaline water retention capacity (AWRC) of pan bread loaves could be considered as an indication for staling and freshness. Therefore, it was estimated for each sample at zero time and after storage periods (24, 48, and 72 h.) as shown in Table 5. Moreover, loss of freshness during storage is shown in Fig. 3. Results showed that there was a gradual decrease in swelling power of pan bread after baking. Also, data presented in the same table showed significant negative effects of adding fiber materials on the swelling power of pan bread samples at zero time. Bread samples containing NWB, FNCP, and FNPP had higher AWRC values than those containing NWG and FNRB. For instance, pan bread incorporated with FNRB at 25% had the lowest AWRC value being 222.34%.

Data in Fig. 3 illustrates that the pan bread containing 5% NWB, 5% FNPP, and control samples had the highest staling rate after 24 h (14.06, 10.80, and 10.51%, respectively), while pan bread containing 5 and 15% NWG had the lowest staling rate at the same time (8.2 and 0.17%, respectively). Also bread samples containing FNRB at 5 and 15% had low staling rate (1.07 and 1.09%, respectively). All tested materials showed positive effects on retarding staling of the baked pan bread during 48 and 72 h of storage at room temperature. Also, it was noticed that as the levels of NWB, FNCP, and FNPP increased, the rate of staling was decreased. But in the case of NWG and FNRB, this trend was up to 15%, while addition of both materials at 25% increased the staling rate of produced bread being 12.64 and 26.44%, respectively, after 72 h of storage compared to 8.25 and 5.75% for breads containing 15% NWG and FNRB, respectively after 72 h of storage.

Discussion

Incorporation of tested materials to wheat flour significantly (p < 0.05) decreased loaf volume of final products. Maximum reduction of loaf volume was recorded to bread made from wheat flour incorporated with FNCP and FNPP at 25% and wheat flour 100%. It is widely accepted that the gluten network is mainly responsible for
the technological properties of baked goods. This explains the almost linear relation between physical properties of bread and flour gluten content. Loaf volume and crumb texture are the main quality characteristics of bread (Katina et al. 2006). Loaf volume is the term used to measure the increase in the volume of fermented dough during oven spring process. This increase in loaf volume is mainly due to expansion of gases which are formed during fermentation process (Bender 2005).

In general, food industry by-products incorporation in wheat flour often resulted in a decrease on volume and specific volume (Belghith et al., 2016; Bhol et al. 2016; Seczyk et al. 2017). They suggested that the decreased loaf volume of bread samples incorporated with fiber materials may be due to diluting the gluten matrix of dough, which impairs carbon dioxide retention. Other researchers also observed decreasing volume of different bakery products such as bread, cookies, and rolls after the addition of wheat bran (Gomez et al. 2011), rice bran (Maria et al. 2019), and carrot pomace powder (Kohajdova et al. 2012). According to Wu and Shiau (2015), lower volume can also be related to the addition of fine particle size, as observed with pineapple peel.

Texture profile of bread crumb could be summarized in hardness, springiness, cohesiveness, and chewiness parameters. The hardness is an important factor since it is strongly correlated with consumers’ perception of bread freshness (Onyango et al. 2010). Hardness is mostly attributed to the amylose and amylopectin matrix, which contribute to overall bread texture (Hoseney, 1994; Schiralde and Fessas 2000). Moreover, as described by Gomez et al. (2003), bread hardness also resulted from interactions between gluten and fibrous materials. Cohesiveness, springiness, and chewiness (product of both cohesiveness and springiness) are influenced by the interactions between gelatinized starch and gluten dough that can create elastic dough and form bread sponge structure after heating (Plyer 1988; Hoseney 1994). Chewiness is one of the texture parameters easily correlated with sensory analyses (Gomez et al., 2007). It is

| Table 5 Alkaline water retention capacity of pan bread as affected by addition of nano and fermented nano materials (%) |
| Sample | Storage time (h) | 3 | 24 | 48 | 72 |
| --- | --- | --- | --- | --- | --- |
| Pan bread made from wheat flours (control samples) | 72% extraction | 325.81 ± 0.28 | 291.58 ± 0.61 | 265.62 ± 0.80 | 236.91 ± 1.05 |
| | 82% extraction | 291.51 ± 0.68 | 276.33 ± 0.46 | 254.25 ± 0.62 | 228.74 ± 0.39 |
| | 100% extraction | 266.12 ± 0.47 | 244.57 ± 0.35 | 227.63 ± 0.72 | 207.95 ± 0.46 |
| Pan bread made from wheat flour 72% incorporated with | Nano-wheat bran (%) | 5 | 286.62 ± 0.64 | 246.33 ± 1.68 | 241.39 ± 1.00 | 234.03 ± 1.07 |
| Nano-wheat bran (%) | 15 | 251.52 ± 0.79 | 246.42 ± 0.44 | 236.33 ± 1.06 | 228.34 ± 0.27 |
| Nano-wheat bran (%) | 25 | 230.76 ± 0.87 | 224.69 ± 1.13 | 219.05 ± 1.58 | 201.04 ± 0.12 |
| Nano-wheat germ (%) | 5 | 257.93 ± 1.06 | 255.81 ± 0.56 | 252.55 ± 0.68 | 233.81 ± 1.88 |
| fermented nano-rice bran (%) | 15 | 246.99 ± 0.74 | 246.51 ± 0.81 | 242.61 ± 0.45 | 226.60 ± 1.04 |
| Fermented nano-rice bran (%) | 25 | 240.57 ± 0.54 | 230.21 ± 0.43 | 219.56 ± 1.08 | 210.16 ± 0.92 |
| Fermented nano-carrot pomace (%) | 5 | 229.05 ± 1.07 | 226.60 ± 0.86 | 220.92 ± 0.30 | 216.39 ± 0.72 |
| Fermented nano-carrot pomace (%) | 15 | 224.70 ± 0.52 | 222.25 ± 0.4 | 216.88 ± 1.03 | 211.78 ± 1.19 |
| fermented nano-pomegranate peel (%) | 25 | 222.34 ± 0.38 | 209.19 ± 1.14 | 189.91 ± 0.54 | 163.55 ± 2.17 |
| Fermented nano-pomegranate peel (%) | 5 | 262.13 ± 1.24 | 249.03 ± 0.81 | 229.30 ± 1.30 | 221.04 ± 1.61 |
| Fermented nano-pomegranate peel (%) | 15 | 255.78 ± 0.69 | 245.28 ± 0.45 | 226.71 ± 0.42 | 209.63 ± 1.10 |
| Fermented nano-pomegranate peel (%) | 25 | 244.52 ± 0.89 | 239.39 ± 0.65 | 224.68 ± 0.78 | 201.54 ± 1.09 |
| Values in the same column followed by different letters are significantly different (p < 0.05) |
related to the work needed to chew a solid sample such as bread to a steady state of swallowing. Crumb chewiness is a product of crumb hardness, cohesiveness, and springiness but it is more dependent on firmness.

Fermentation was meant for making the dough lighter and spongier by the action of proteolytic enzymes, organic and inorganic acids, alcohol, and the acidic environment (Pyler and Gorton 2009). In that respect, the negative effect of fibrous materials on both loaf volume and crumb texture of bread samples was possibly a result of the fiber weakening or crippling dough structure and reducing CO₂ gas retention, compared to the control sample manufactured using wheat flour 72% extraction. On the other hand, the cruel effect of FNPP on the texture profiles of bread sample could be due to the fact that long fermentation and acidic condition of dough system incorporated with fiber may release bound phenolic acids (Katina et al. 2005), which could disturb the gluten network development via inhibition of disulfide bond formation (Koh and Ng 2008; Han and Koh, 2011) by altering the protein molecular structure via inducing the conformation-helix protein complexes to form antiparallel-β-sheet structures (Nawrocka et al. 2016). Moreover, appreciable amounts of water could have strongly bound to the added fiber during baked goods making, so less water was available for the development of the starch-gluten network, causing an underdeveloped gluten network and reduced loaf volume (Sivam et al. 2010), and this could explain the high loaf weight of bread samples incorporated with fiber materials.

FNPP causes a larger adverse effect compared to other materials. This can be explained by the higher phenolic content of the pomegranate peels. From physical behavior, it has been previously concluded (Wang et al., 2003, 2004) that bound phenolic compounds can bind to gluten proteins, resulting in interactions between cell wall chains and gluten proteins, which is reflected in gluten’s ability to agglomerate. The increase in particle surface area, in nano-materials, is likely to favor these phenolic-mediated cell wall-gluten interactions, by making more chains accessible.

Rosell and Santos (2010) reported that insoluble fibers prevent the free expansion of dough during fermentation, and also, they can induce an early fixing of the structure due to their higher water binding capacity. Also, Wang et al. (2004) reported that not only the amount of gluten is reduced, but also the properties of gluten are affected. The resulting gluten becomes stiffer and less extensible. This could also lead to a lower ability of the dough to retain gas. Therefore, the lower springiness values could be due to dilution of gluten, which causes lower ability to hold gases consequently bread elasticity reduction. Also, the reduction of cohesiveness parameter indicates that the bread formulated with fiber materials have low ability to resist before the bread texture deformed under the teeth.

On the other hand, the crust color of bread is known to be associated with Maillard reaction, thus containing more protein can increase the Maillard reaction and browner color (Gomez et al. 2003). So, the high $a^*$ values of bread containing NWG could be due to its high protein content (Mohammad et al. 2015), while the low $a^*$ values of bread containing FNCP and FNPP could be due to decrease in protein content of composite breads which can affect the Maillard reaction in the

![Fig 3: Loss of freshness in pan bread during storage as affected by addition of nano and fermented nano materials. WF, wheat flour; NWB, nano-wheat bran; NWG, nano-wheat germ; FNCP, fermented nano-carrot pomace; FNPP, fermented nano-pomegranate peel.](image-url)
crust. But the darkness resulting from the addition of FNCP and FNPP could be due to the dark color of their powders (Amir et al. 2013).

Freshness properties of bread samples were negatively affected by adding fiber materials at zero time. This could be due to the lower swelling power of these materials compared to wheat flour. The obtained results were in agreement with those obtained by Payne (2000); Khorshid et al. (2011); and Mehder (2013), while all tested materials showed positive effects on retarding staling of the baked pan bread during 48 and 72 h of storage at room temperature. Similar results have been published by Somaie et al. (2019), as they reported that fermented sterilized wheat bran showed improving effect regarding freshness properties of bread.

From the rheological point of view, the theory of Hug-Iten et al. (1999) attributed the staling to the starch modifications after the baking process. Starch retrogradation occurs during the cooling period after baking, in which the amylose and amyllopectin chains aggregate forming crystalline double helices stabilized by hydrogen bonds, leading to bread hardening. The incorporated fibers increased the breakdown values of the dough, consequently increased the amylolytic activity and decreased the retrogradation process compared to the control sample insuring the obtained data.

On the other hand, Feili et al. (2013) postulated that the antistaling effect may be attributed to the already demonstrated water-binding capacity of fiber, which in turn reduced water loss during storage, as well as the probable interaction between fiber and starch, resulting in the delay of starch retrogradation. They also stated that the best effect in delaying the bread staling was noticed after 2 days by using a short-length fiber. Also, the antistaling effect of NWG and FNRB could be attributed to the high fat content of these materials (Mohammad et al. 2015).

Conclusion
Size reduction of fibrous raw and fermented to nano scale significantly altered the functional and physical properties compared to coarse particles. Application of nano and fermented nano-powders in food product might make such foods more appealing to consumers than coarse materials. Also, the fine powders can be potentially used in greater amounts to enrich food products such as bread with less sensorial implications. Utilization of the various materials in food products highlighted can potentially increase their nutritional value. Enriching these foods with fine powders may reduce gritty taste that consumers associate with coarse-enriched products. It could be concluded from this subjective evaluation that functional pan bread with acceptable quality could be prepared from NWB, NWG, and FNRB up till 15% and FNCP and FNPP up to 5% replacement level.
