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# Physico-chemical and Sensory Properties of Bread Enriched with Lemon Pomace Fiber

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**Abstract**

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Lemon fiber (LF) was extracted from lemon pomace and incorporated into bread. The results showed that the hardness of bread increased with the increase of LF substitution (0–9%), but the cohesiveness, springiness, and specific volume decreased with the fiber substitution. Both Peleg-Normand and Wiechert models fitted well the stress relaxation data of bread. The parameters ($k_1$ and $k_2$, as well as $P_0$–$P_2$ and $\lambda_2$) in two models were followed significantly affected by the amount of LF. The breads enriched with LF revealed higher total phenolic contents and DPPH radical scavenging capacities than the control bread. Compared with water, methanol was a better solvent for extracting total phenolic compounds of bread. These results suggest that the addition of 3% LF can produce healthy and acceptable bread with higher phytochemicals and antioxidant capacities.

**Keywords**: texture; rheology; stress relaxation; polyphenol; antioxidant

The consumption of dietary fiber (DF) offers health benefits including lower risk of coronary heart diseases, cancer, and diabetes (Anderson *et al*. 2009). Hence DF has received tremendous attention from the food scientists and industry. Bread is a popular and important staple food in the world, and can be enriched with various DFs sources, such as wheat bran (Ugarčić-Hardi *et al*. 2009), apple pomace (Masoodi & Chauhan 1998), and pomegranate peel powder (Altunkaya *et al*. 2013). To the best of our knowledge, there are no reports on the use of lemon pomace fiber as a source of DF in bread making.

DF incorporated into wheat flour interacts directly with gluten proteins and affects the formation of gluten-starch matrix. Excess amounts of insoluble dietary fibers had an adverse effect on the formation of gluten network and reduced the quality of bread due to the gluten dilution effect or gluten-fiber interaction (Masoodi & Chauhan 1998; Wang *et al*. 2003; Kaack *et al*. 2006). Potato fiber containing a high level of insoluble DF led to increases in the hardness and gumminess of bread (Kaack *et al*. 2006). However, recent studies showed that both total phenolic and antioxidant content of bread could be enhanced by the addition of fibers, such as grape seed flour (Hoye & Ross 2011) and pomegranate peel powder (Altunkaya *et al*. 2013).

Lemon is the third most important *Citrus* species in the world and is mainly processed into fruit juice and tea drinks. Since its pomace accounts for 50–65% of the whole fruit mass, there is a great interest in utilising the biomass, instead of discarding it. The pomace contains many phytochemicals, such as fiber, phenolics, flavonoids, and carotenoids (Marin *et al*. 2007; Wang *et al*. 2008). Hence, lemon pomace is a good phytochemical source for the food enrichment. The aim of this study was to investigate the rheological, antioxidative, and sensory properties of bread enriched with lemon pomace fiber.

## MATERIAL AND METHODS

***Materials***. Wheat flour containing 13.64% of crude protein was obtained from Chia-Fha Enterprise Co. Ltd. (Taichung, Taiwan). Commercial instant dried

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yeast was from S.I. Lesaffre Co. (France). Food-grade sucrose, salt and shortening were from Taiwan Sugar Co. (Taiwan). All the chemicals used in food analysis were of analytical grade.

Freshly pressed lemon (*Citrus limon* cv. Eureka) pomace was obtained from a local citrus juice industry in Pingtung county, Taiwan. Lemon fiber (LF) was prepared from the pomace, according to the method reported by Fu *et al.* (2014). The proximate compositions (AACC 2000) and total, insoluble, and soluble dietary fibers (AOAC 2000) of LF produced were determined. Water holding capacity of the fiber was measured by the method of Chau and Huang (2003).

***Preparation of bread***. Basic dough formula on 340 g flour basis consisted of water (187 g), instant dried yeast (4.04 g), sucrose (20.4 g), shortening (17.0 g), and salt (5.1 g). Bread was produced by using an automatic Kaiser breadmaker (Model BM-812; Wilson Sources Inc., Kowloon, China). Identical baking program (menu 1) was performed for all the breads in this study. The ending point of baking was set as "Light Crust". The total baking time was 3 h and 11 min, including two-stage kneading (25 min), rest (5 min), shaping (20 s), three-stage fermentation (115 min), and baking (46 min). Apart from the basic dough, 3–9% fiber-enriched breads were prepared with wheat flour partially substituted by 10.2–30.6 g LF, respectively. After the production, the breads were cooled for 90 min at room temperature.

***Stress relaxation of bread***. Stress relaxation of bread was measured by a texture analyser (TA) (TA-XT2i; Stable Micro Systems, Surrey, UK) according to the method proposed by Wu *et al.* (2012) with some modifications. In brief, the cool bread was cut into slices (12 mm in thickness) by toast slicer machine. One of the central slices was taken and the determination of the stress relaxation (SR) was performed using the TA equipped with a cylindrical probe of P20 (20 mm diameter). The residual force of the sliced bread sample was continuously recorded for 240 s at 20% strain.

The stress relaxation data were analysed using a Peleg-Normand model (Eq. 1), proposed by Peleg and Normand (1983).

$$\frac{F_0 t}{F_0 - F(t)} = k_1 + k_2 t \quad (1)$$

where: $F_0$ – initial force; $F(t)$ – momentary force at time $t$; $k_1$, $k_2$ – constants

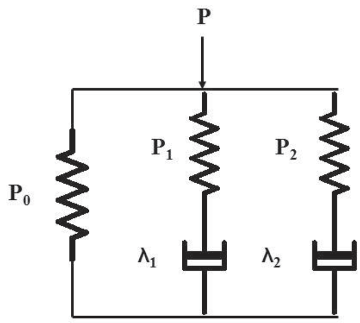


Figure 1. Wiechert model consisting of a single spring ($P_0$) in parallel with two Maxwell models composed of spring ($P_1$ and $P_2$) and dashpot ($\lambda_1$ and $\lambda_2$) in series

Equation (2) shows a Wiechert model (Bellido & Hatcher 2009) consisting of one single spring in parallel with two Maxwell elements (Figure 1).

$$P(t) = P_0 + P_1 \exp(-t/\lambda_1) + P_2 \exp(-t/\lambda_2) \quad (2)$$

where: $P(t)$ – actual stress as a function of time in the stress relaxation test; $\lambda_1$, $\lambda_2$ – time constants (relaxation times); $P_0$, $P_1$, $P_2$ – spring constants (relaxation moduli)

The experimental data were modelled by using non-linear regression in SigmaPlot 8.0 software (Systat Software Inc., San Jose, USA).

***Texture and specific volume of bread***. Texture profile analysis (TPA) of the sliced bread samples was carried out by the TA equipped with a P20 adapter moving at a rate of 2 mm/s, and the penetration depth into the bread sample was 20 mm. The hardness (N), cohesiveness, and springiness were calculated using the TPA curve. The cooled bread was weighed and its volume was determined by the seed displacement in a loaf volume meter. Specific volume was expressed as ml/g.

## *Total phenolic content (TPC) and antioxidant activity of bread*

*Extraction of total phenolic compounds.* Total phenolic compounds of bread were extracted according to the method reported by Ragaee *et al.* (2011) with slight modifications. Briefly, ground freeze-dried bread (500 mg) was placed in a 50-ml centrifuge tube and 10 ml of 80% methanol or distilled water solvent was added. The mixture was agitated for 30 min using a platform shaker and subsequently centrifuged at 6000 *g* for 10 minutes. The supernatant was removed, and the extraction was repeated once. The combined supernatants of both 80% methanol

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and distilled water solvents were individually used to measure the TPC and antioxidant capacity.

***Determination of TPC***. TPC was determined by Folin-Ciocalteau method (Cilliers *et al*. 1990) using ferulic acid as a standard. Briefly, samples (1.0 ml) were introduced into test cuvettes, and 0.25 ml of Folin-Ciocalteu's reagent, 0.25 ml of sodium carbonate (20% w/v), and 2.5 ml distilled water were added. After standing for 60 min at room temperature, the absorbance at 725 nm was measured. The results were expressed as mg ferulic acid equivalent per g bread.

***Determination of antioxidant activity as DPPH scavenging capacity***. Radical 2,2-diphenyl-1-picrylhydrazyl (DPPH) scavenging capacity of the extract of total phenolic compounds was measured according to the method of Liyana-Pathlrana and Shahidi (2007) with some modifications, and by using Trolox or ascorbic acid as a standard for 80% methanol or water extracts, respectively. Briefly, an aliquot of 1ml of the test sample was mixed with 1ml of 0.1 mM DPPH in methanol. The mixture was shaken vigorously and left to stand for 30 min at 25°C. The absorbance of the resulting solution was determined at 517 nm against methanol or water blank. The results were expressed as mg Trolox equivalent or ascorbic acid equivalent per g bread.

***Sensory evaluation.*** The sensory evaluation of cooled breads with 0–9% LF substitutions was performed according to the method reported by Fu *et al*. (2014). The colour, odour, texture, and overall preference of the bread samples were evaluated by a 7-point hedonic scale; which was 1 – very unacceptable, 2 – unacceptable, 3 – mildly unacceptable, 4 – neither unacceptable nor acceptable, 5 – mildly acceptable, 6 – acceptable, and 7 – very acceptable.

***Statistical analysis***. The data in triplicate, unless stated otherwise, for different treatments were analysed by one-way ANOVA and Duncan's new multiple range tests to determine the statistical significance of differences between the values by using the SPSS v. 13.0 software (SPSS, Chicago, USA).

## RESULTS AND DISCUSSION

***Proximate composition and water holding capacity***. Proximate composition of the LF powder prepared was 8.32% moisture (wet basis), 4.41% ash, 6.90% crude protein, 0.55% crude fat, and 63.85% total dietary fiber content (dry basis). The contents of insoluble and soluble dietary fibers were 51.44 and 12.41%, respectively. This indicated that the insoluble and soluble DFs were the major constituents in the LF powder. Marin *et al*. (2007) reported that dried lemon juice by-product contained 17.1–23.3% soluble DF and 54.0–64.5% insoluble DF, depending on the method of preparation. Water holding capacity (WHC) of wheat flour and LF was 1.2 ± 0.1 and 11.04 ± 0.9 ml/g, respectively. The results indicated that LF had obviously a higher WHC than wheat flour.

***Stress relaxation of bread***. The fundamental viscoelastic properties of foods can be measured by stress relaxation. In Table 1, the stress relaxation data of bread fitted well ($R^2$ > 0.99) to the Peleg-

Table 1. Effect of lemon fibre (LF) amount on stress relaxation parameters of bread

| Model | Parameter | LF (%) | | | |
|---|---|---|---|---|---|
| | | 0 | 3 | 6 | 9 |
| Peleg-Normand | $F_0$ (N) | 1.42 ± 0.06$^d$ | 2.55 ± 0.37$^c$ | 4.69 ± 0.35$^b$ | 8.28 ± 0.16$^a$ |
| | $k_1$ (s) | 23.99 ± 1.10$^a$ | 19.62 ± 5.18$^b$ | 17.22 ± 1.09$^b$ | 16.13 ± 2.36$^b$ |
| | $k_2$ | 2.09 ± 0.04$^a$ | 1.92 ± 0.23$^b$ | 1.81 ± 0.04$^c$ | 1.75 ± 0.09$^c$ |
| | $R^2$ | 0.999 | 0.999 | 0.999 | 0.999 |
| Wiechert | $P_0$ (kPa) | 2.42 ± 0.48$^d$ | 4.18 ± 0.67$^c$ | 6.95 ± 0.49$^b$ | 11.12 ± 1.00$^a$ |
| | $P_1$ (kPa) | 1.05 ± 0.23$^d$ | 2.01 ± 0.38$^c$ | 4.15 ± 0.32$^b$ | 7.96 ± 0.87$^a$ |
| | $P_2$ (kPa) | 0.76 ± 0.15$^d$ | 1.38 ± 0.24$^c$ | 2.68 ± 0.18$^b$ | 4.86 ± 0.54$^a$ |
| | $\lambda_1$ (s) | 4.56 ± 0.24$^a$ | 4.23 ± 0.35$^a$ | 4.03 ± 0.10$^a$ | 4.05 ± 0.36$^a$ |
| | $\lambda_2$ (s) | 72.7 ± 2.75$^a$ | 67.3 ± 2.71$^b$ | 63.7 ± 1.48$^{bc}$ | 60.7 ± 0.21$^c$ |
| | $R^2$ | 0.994 | 0.995 | 0.995 | 0.994 |

All data are the means ± SD of three replicates; means with the different letters within a row are significantly different ($P$ < 0.05); $F_0$ – initial force; $k_1$, $k_2$ – constants; $\lambda_1$, $\lambda_2$ – time constants (relaxation times); $P_0$, $P_1$, $P_2$ – spring constants (relaxation moduli); $R^2$ – coefficient of determination

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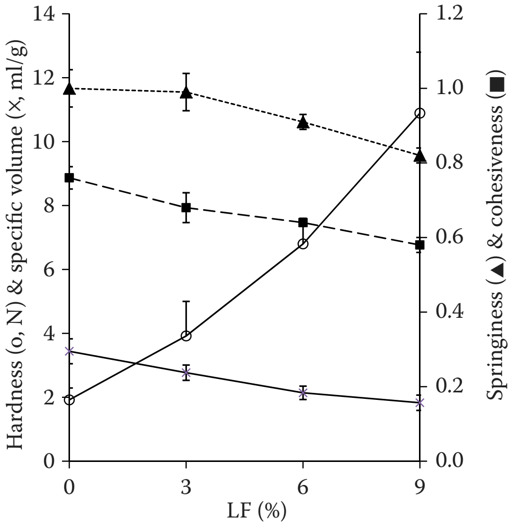


Figure 2. Effect of lemon fiber (LF) amount on the texture and specific volume of bread

Normand model. $F_0$ (initial force) values of breads increased with the increasing the LF substitution. However, both $k_1$ and $k_2$ parameters of bread enriched with 3–9% LF were significantly lower than those of control bread. Thus, the bread enriched with a high level of LF was more rigid and less elastic than the control bread, due to the adverse effect of LF on the formation of gluten network by fiber. The result of stress relaxation in this study is consistent with the report of Wu *et al.* (2012) stating that increasing the substitution of flour by wheat bran resulted in a lower elasticity of the steamed bread.

Since bread is a typical viscoelastic solid food, we found that the stress relaxation data of bread could be fitted well ($R^2 > 0.99$) to the Wiechert model with two-Maxwell elements (Eq. (2)). Wiechert models with three-Maxwell elements were found adequate for demonstrating the stress relaxation data of noodle (Bellido & Hatcher 2009). In Table 1, relaxation moduli ($P_0$, $P_1$, and $P_2$) of bread significantly increased with the increasing substitution of LF. Although $\lambda_1$ value (relaxation time) of bread was not significantly affected by LF, breads enriched with 3–9% LF had obviously lower $\lambda_2$ values than the control bread. The higher relaxation modulus and shorter relaxation time of bread with a high LF content indicated that the bread had a more rigid and less elastic behaviour, corresponding to the results from the Peleg-Normand model. Figueroa *et al.* (2013) reported that a slower (longer) relaxation time of wheat dough and gluten was associated with good baking quality.

***Texture and specific volume of bread***. The hardness of bread generally increased with increasing LF substitution, but the cohesiveness, springiness, and specific volume decreased with the substitution (Figure 2). However, no significant difference was observed in hardness, springiness, and specific volume between 3% LF-enriched and control breads.

In this study water absorption in dough making was fixed at 55%, while wheat flour would compete with LF for the water uptake during the preparation of dough. Since LF had obviously a higher WHC than wheat flour, it was not easy for wheat flour to absorb sufficient water for the full development of the gluten protein network, especially at a high LF level. Hence, the LF-enriched bread had a higher hardness due to the rigid nature of fiber or the competition for water absorption between LF and flour components. Since insoluble DF is the major DF in LF, LF added may interfere with the formation of gluten network. This led to the lower cohesiveness, springiness and specific volume of the LF-enriched bread. The results shown in Figure 2 are comparable to those in the studies of Hoye and Ross

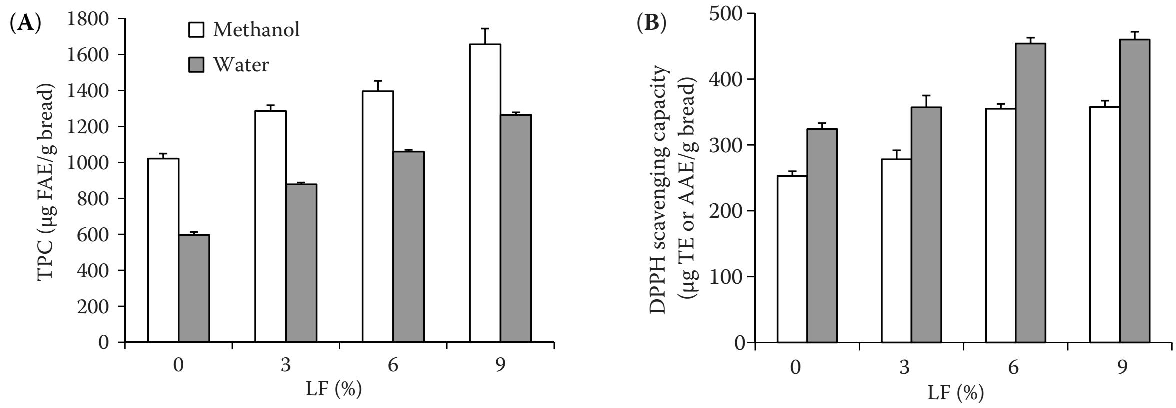


Figure 3. Effect of lemon fiber (LF) amount on total phenolic content (**A**) and DPPH radical scavenging capacity (**B**) of bread

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Table 2. Effect of lemon fibre (LF) amount on the sensory evaluation of bread

| LF (%) | Colour | Flavour | Texture | Overall |
|---|---|---|---|---|
| 0 | 5.20 ± 1.10[a] | 4.67 ± 1.32[a] | 5.20 ± 1.16[a] | 5.10 ± 1.24[a] |
| 3 | 5.13 ± 0.79[a] | 4.67 ± 1.37[a] | 4.93 ± 1.20[a] | 5.00 ± 1.29[a] |
| 6 | 5.03 ± 0.96[a] | 3.77 ± 1.10[b] | 4.23 ± 1.31[b] | 4.13 ± 1.11[b] |
| 9 | 4.90 ± 1.47[a] | 3.17 ± 1.46[b] | 3.57 ± 1.41[c] | 3.40 ± 1.19[c] |

All data are the means ± SD ($n$ = 30); means with the different letters within a column are significantly different ($P$ < 0.05)

(2011), who reported that the replacement with above 5% grape seed flour significantly increased the bread hardness and decreased the bread loaf volume.

***TPC and antioxidant capacity of bread***. Lemon peel was rich in phenolic acids, flavonoids, and carotenoids, which contributed to a good antioxidant capacity (Wang *et al*. 2008). The result given in Figure 3A indicated that TPC of bread in both 80% methanol and water extracts increased with the increase of LF level. When LF substitution was increased from 0% to 9%, TPC of the bread was improved by 25.9–47.3, 36.7–77.9, and 62.2–111.9%, respectively. Moreover, TPC of methanol extract was higher than that of water extract at the same LF level. This showed that methanol was a better solvent for extracting total phenolic compounds of bread.

In Figure 3B, breads enriched with 3–9% LF had significantly higher DPPH scavenging capacity than the control bread. The antioxidant activity of TPC extract from LF-enriched bread was increased by 10.0% to 42.0% as compared to the control bread. The correlation coefficients between TPC and DPPH scavenging capacity for methanol and water extracts were 0.890 and 0.938, respectively. This showed that TPC of bread had a good linear relationship with DPPH scavenging capacity.

Hoye and Ross (2011) indicated that breads with 10% grape seed flour had a higher TPC than the control bread. Altunkaya *et al*. (2013) reported that the increasing replacement of flour by pomegranate peel powder (0–10%) led to higher antioxidant levels in ethanolic extracts.

***Sensory evaluation of bread***. The bread enriched by a high substitution (6 and 9% LF) had significantly low sensory scores in the flavour, texture, and overall acceptability (Table 2). But none sensory score of 3% LF-enriched bread showed any significant difference between those of the control bread. Therefore, we suggest that the fiber-enriched bread can be prepared with 3% LF in order to increase the intake of phytochemicals and maintain the sensory acceptability for the consumers.

## CONCLUSIONS

LF can be supplied as a potential functional food ingredient containing abundant dietary fiber and phytochemicals. The amount of LF significantly affects the rheology, total phenolic content, and antioxidant capacity of bread. The increases in the hardness of bread enriched with LF may relate to the decrease of free water in dough or the rigid nature of fiber. Both Peleg-Normand and Wiechert models fitted well the stress relaxation data of bread. High substitution of LF would result in an adverse effect on the formation of gluten network, and a led to lower cohesiveness, elasticity, and specific volume of the LF-enriched bread. The addition of LF could elevate TPC and antioxidant capacity of the fiber-enriched bread. In conclusion, this study demonstrated that the fiber-enriched bread with good sensory quality, phytochemicals, and antioxidant capacity could be produced by the substitution of wheat flour by 3% LF.

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