Effect of pumpkin seed flour, chestnut flour, and rosehip flour on wheat bread staling rate

Rosen Chochkov[1], Denka Zlateva[2], Dana Stefanova[2], Petya Ivanova[1]

1 – University of Food Technologies, Plovdiv, Bulgaria
2 – University of Economics, Varna, Bulgaria

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Corresponding author:

Rosen Chochkov
E-mail:
rosen4o4kov@abv.bg

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Abstract

Introduction. The aim of this study was to analyse the impact of chestnut flour, rosehip flour, and pumpkin seed flour (in the amount of 5% or 10% on the basis of wheat flour) on the staling rate of wheat bread.

Materials and methods. The bread was made from type 500 wheat flour with the addition of pumpkin seed, chestnut or rosehip flour (5% or 10%). Bread staling was determined by measuring the deformation characteristics of the bread crumb. The total, plastic and elastic deformation of the bread crumb in penetrometric units (p. u.) was measured on 3; 24; 48; 72, and 96 h after baking.

Results and discussion. It was found that as the storage time increases, the values characterizing the total, plastic, and elastic deformation of the bread crumb decrease for all the samples. After three hours after baking, the highest total deformation was determined in the control sample (144 p. u.). At longer storage times (72 h and 96 h), the crumb of the wheat bread hardened progressively, and the samples with 5% rosehip flour, as well as 5% and 10% pumpkin seed flour, preserved their softness.

The duration of storage affects differently depending on the type and quantity of the supplement. At the end of storage, the sample with 10% rosehip flour and with 10% pumpkin seed flour had the lowest rate of change of plastic deformation (32% and 37%, respectively). The elastic properties of the bread crumb deteriorated at significantly slower rates in the enriched samples. In the case of bread with 5% rosehip flour after storage for 96 h, the elastic deformation decreased by 38%, and in the case of bread with 10% pumpkin seed flour – by 35.5%, while for the control sample, the elastic deformation decreased by 70%.

Conclusions. The appropriate dosing of the chestnut, rosehip and pumpkin seed flour in the wheat bread formulation results in preserving the softness and elasticity of the crumb and can retard bread staling. The most significant delay in staling was achieved when using 5% rosehip flour and 10% pumpkin seed flour.

Introduction

Staling of bakery products is accompanied by changes in the physical, chemical, and sensory properties of bread, it reflects on consumer preferences and willingness to buy and consume the product and causes economic losses because of the waste of large quantities of bread (Demirkesen et al., 2014). Typically, dough formulations include different components, each of which undergoes changes during the baking process of the bread, as well as during storage of the bread, making staleness an extremely difficult phenomenon to describe (Gray and BeMiller, 2003).

Bread staling is a process that occurs during storage and involves redistribution of moisture, change in the structural properties of the crumb and the crust and deterioration of the taste, smell and other sensory characteristics of bread. It has been clearly proven that staling is a result of complex reasons, such as changes in the starch fraction (more specifically – amylopectin retrogradation), migration and redistribution of moisture, and changes in gluten structure (Amigo et al., 2019). It is believed that approximately 3% of global bakery products fail to be realised on the market because of the staling process (Gómez et al., 2008).

Even though this process has been first studied and described as early as in 1852 (Boussingault, 1852), a number of authors have tried to clarify its mechanism at a later stage. Some of them (Goesaert et al., 2009; Hallberg and Chinachoti, 2002) highlight that amylopectin retrogradation is the main reason for the changes in the physical and chemical properties of the bread crumb during storage. The process of retrogradation starts when the amylopectin chains transform, and the amorphous molecules turn into into a denser and stable crystal form (Slade and Levine, 1991). By using light microscopy, Hug-Iten et al. (1999) studied the microstructure of starch in bread that has undergone staling. According to the authors, air gaps and partial separation of the starch from the protein phase are observed, which is the reason for the crumbliness of the bread crumb.

More recent studies show that gluten, non-gluten proteins, lipids and pentosans also play an important role in the staling process (Blaszczak et al., 2004; Hallberg et al., 2002; Smith and Johansson, 2004). According to other authors, the migration of water molecules plays a significant role in bread staling (Baik and Chinachoti, 2000; He and Hoseney, 1990). Water migrates from gluten to starch, which results in a decrease in the crumb plasticity. On the other hand, water is redistributed and incorporated in the amylopectin crystal formed during retrogradation and causes hardening of the crumb during storage.

It could be suggested that all components of the bread's chemical composition (more or less) have an impact on the speed of the process. Actually, the kinetics of staling depends on a complex set of factors: the raw materials used (the ingredients of the dough, yeast, enzymes), the technological process (kneading, fermentation, baking, cooling) and the storage conditions (humidity and temperature).

Bread staling has a negative impact on the quality of the product and significantly reduces its consumer acceptability, while causing great economic losses. Finding possibilities for retarding the process is the subject of many studies. Various attempts have been made to improve the shelf life of bakery products through additives, formulation changes (Curti et al., 2016); changes in the technological process (Bosmans et al., 2013) or in storage conditions (Rasminssen and Hansen, 2001), as well as the type of packaging (Akhtar et al., 2008). In this regard, the use of various additives to improve shelf life is a widespread approach in the bakery industry.

Different researchers have focused their interest on the selection of additives that could delay bread staling (Fadda et al., 2014; Tebben et al., 2018). In this context, efforts focus on the use of additives of natural origin with high nutritional value, which also include flours from alternative raw materials.

There are only some isolated studies on the impact of chestnut, rosehip and pumpkin seed flour on the structural properties of bread crumb and wheat bread staling. Dall'Asta et al. (2013) replaced 50% of wheat flour with chestnut flour in the bread formulation and found that the staling process was delayed in the enriched bread. Man et al. (2012) found that the input of 5%, 10% or 15% chestnut flour had a clear favourable effect on the elasticity of the bread crumb, with a well-defined linear correlation ($r = 0.7980$). Addition to the high dietary fibre content, pumpkin seeds are also distinguished by the fact that they are a very good source of protein, which makes them suitable for inclusion in the bread formulation (Costa et al., 2018; Stabnikova et al., 2023). The authors highlight that the elasticity of bread enriched with 30% pumpkin seed flour is lower as compared to the control sample (made of wheat flour only), however, the differences are not statistically significant. Gomez et al. (2004) explained this by the increased fibre content and the reduced gas-retention capability of dough. The increased quantity of dietary fibres and proteins in the enriched samples of bread resulted in increased hardness of the crumb (9.64 N for the enriched sample as compared to 6.07 N for the control sample).

Halvorsen et al. (2002) pointed out that rosehip had the highest antioxidant activity compared to other berries in the *Rosaceae* family. Rosehip powder has been used in bread formulation to enhance bioactivity of bread, to increase total phenolic content and dietary fibre content (Zhou et al., 2023). However, data on the effect of rosehip flour on bread staling are scarce.

The objective of this study was to analyse the impact of replacement of 5% or 10% of wheat flour with chestnut flour, rosehip flour and pumpkin seed flour on the staling rate of wheat bread.

Materials and methods

Materials

The bread samples were made of the following raw materials:

- **Wheat flour –** with average chemical composition, as follows: moisture – 12.36%, acidity – 2.10 °H, ash content – 0.50% of dry matter, wet gluten – 27.60%, fat 0.9 g/100 g, of which saturated fat 0.3 g; carbohydrates 70.3 g/100 g, of which sugars 3.4 g, dietary fibres 4.0 g/100 g; proteins 10.8 g/100 g;
- **Water** – meeting the requirements of Regulation No. 9 of the Ministry of Health of 16.03.2001 on the quality of water intended for drinking and household purposes (SG, issue No. 30 of 2001);
- **Bread yeast, compressed –** Lesafmaya (Lesaffre) according to the manufacturer's technological documentation;
- **Salt** – meeting the requirements of Regulation (Decree of the Council of Ministers No. 23/2001) about the requirements for the composition and characteristic of food grade salt (SG, issue No. 11/2001).

Types of flour, unconventional for breadmaking:

Pumpkin seed flour (Figure 1) with average chemical composition: fat 10 g/100 g, of which saturated fat 1 g; carbohydrates 34 g/100 g, of which sugars 0 g, dietary fibres 16 g/100; proteins 56 g/100 g;

Figure 1. Pumpkin seed flour

Chestnut flour with average chemical composition: fat 3.7 g/100 g, of which saturated fat 0.7 g; carbohydrates 70.9 g/100 g, of which sugars 29.5 g, dietary fibres 10.8 g/100 g; proteins 6.4 g/100 g (Figure 2).

Figure 2. Chestnut flour

Rosehip flour with average chemical composition: fat 0 g/100 g, of which saturated fat 0 g; carbohydrates 38 g/100 g, of which sugars 3 g, dietary fibres 24 g/100; proteins 2 g/100 g (Figure 3).

The types of flour so described are available in the retail network in Bulgaria.

Figure 3. Rosehip flour

Preparation of the bread samples

To identify the impact of pumpkin seed, chestnut and rosehip flour on wheat bread staling, the following samples of bread have been studied:

- Control bread sample made of wheat flour (CS);
- Bread sample containing 5% pumpkin seed flour (PSF 5%);
- Bread sample containing 10% pumpkin seed flour (PSF 10%);
- Bread sample containing 5% chestnut flour (CF 5%);
- Bread sample containing 10% chestnut flour (CF 10%);
- Bread sample containing 5% rosehip flour (RF 5%);
- Bread sample containing 10% rosehip flour (RF 10%).

The formulations of the bread samples described above are presented in Table 1.

Table 1

Formulation of the bread samples

Raw materials	**Bread samples**						
	CS	**PSF 5%**	**PSF 10%**	**CF 5%**	**CF 10%**	**RF 5%**	**RF 10%**
Wheat flour (type 500),%	100	95	90	95	90	95	90
Water,%	56.0	56.0	56.0	56.0	56.0	56.0	56.0
Compressed yeast,%	2.0	2.0	2.0	2.0	2.0	2.0	2.0
Salt,%	1.5	1.5	1.5	1.5	1.5	1.5	1.5
Pumpkin seed flour,%	–	5	10	–	–	–	–
Chestnut flour,%	–	–	–	5	10	–	–
Rosehip flour,%	–	–	–	–	–	5	10

The bread was made by type 500 wheat flour based on a two-stage method of dough making. Initially, dough was made with 40% of the total quantity of flour and water (flour to water ratio of 1:1) and the entire quantity of yeast in a kneading machine (Labomix 1000, Hungary). The control sample was made of wheat flour only and the additional ingredients indicated in Table 1. For the enriched samples, 5% or 10% (on the basis of wheat flour) unconventional for the baking industry flours (pumpkin seed flour, chestnut flour or rosehip flour) were added to the water for kneading and were homogenised very well, followed by adding the necessary quantity of wheat flour. The dough was fermented for 60 min at a temperature of 33 °C. This was followed by kneading the bread dough with the remaining raw materials (salt and the remaining quantity of the flour and water) until homogenised. The bread dough was divided, shaped and subjected to final fermentation for 60 – 70 min at a temperature of 35 °C (Tecnopast CRN 45–12, Novacel ROVIMPEX Novaledo, Italy). After the final fermentation, the dough was baked in an electrical oven (Salva E-25, Spain), pre-heated to 220 – 230 °C. Baking time was 24 min. After baking, the bread was left to cool down for 3 hours at room temperature.

Methods

Bread staling was determined by measuring the deformation characteristics of the crumb. An automated penetrometer type AP-4/2 was used. The total, plastic and elastic deformation of the bread crumb (in p. u.) was measured 3; 24; 48; 72 and 96 h after baking. The measurement was made on a piece of the bread crumb (from the central part of the loaf) with a thickness of 40 mm. The deformation characteristics were identified in the following way: on a flat surface, the bread crumb was subjected to the impact of a body with a certain mass, which was left to fall freely for a certain time (5 s). This caused penetration of the body and immersing in the bread crumb and the degree of its immersing in the crumb determined its compression or total deformation (Dt). Then, the immersing system was removed and the bread crumb partially recovered its height as a result of its elasticity properties, based on which the plastic deformation was determined (Dp). The difference between total and plastic deformation constitutes the elastic deformation (De), based on which the bread freshness is identified. Depending on the size of the studied piece, the measurement was performed on three (or five) spots at least 30 mm away from the crust.

Statistical analysis

Analysis of variance (ANOVA) was performed to determine whether storage time and bread formulations significantly ($p \leq 0.05$) affected staling of breads.

Results and discussion

Impact of pumpkin seed, chestnut and rosehip flour on total deformation of bread crumb

The results regarding the impact of pumpkin seed, chestnut and rosehip flour on the total deformation of the bread crumb during storage are presented in Figure 4.

It was found that with the increase in storage time, the values characterising total deformation decreased in all samples studied, i.e. the crumb became harder. Three hours after baking, the values in the bread samples containing 5% non-traditional flours were close each to the other and varied from 131 to 138 p.u. The highest total deformation was measured in the control sample (144 p.u.) indicating that the wheat bread had the softest crumb.

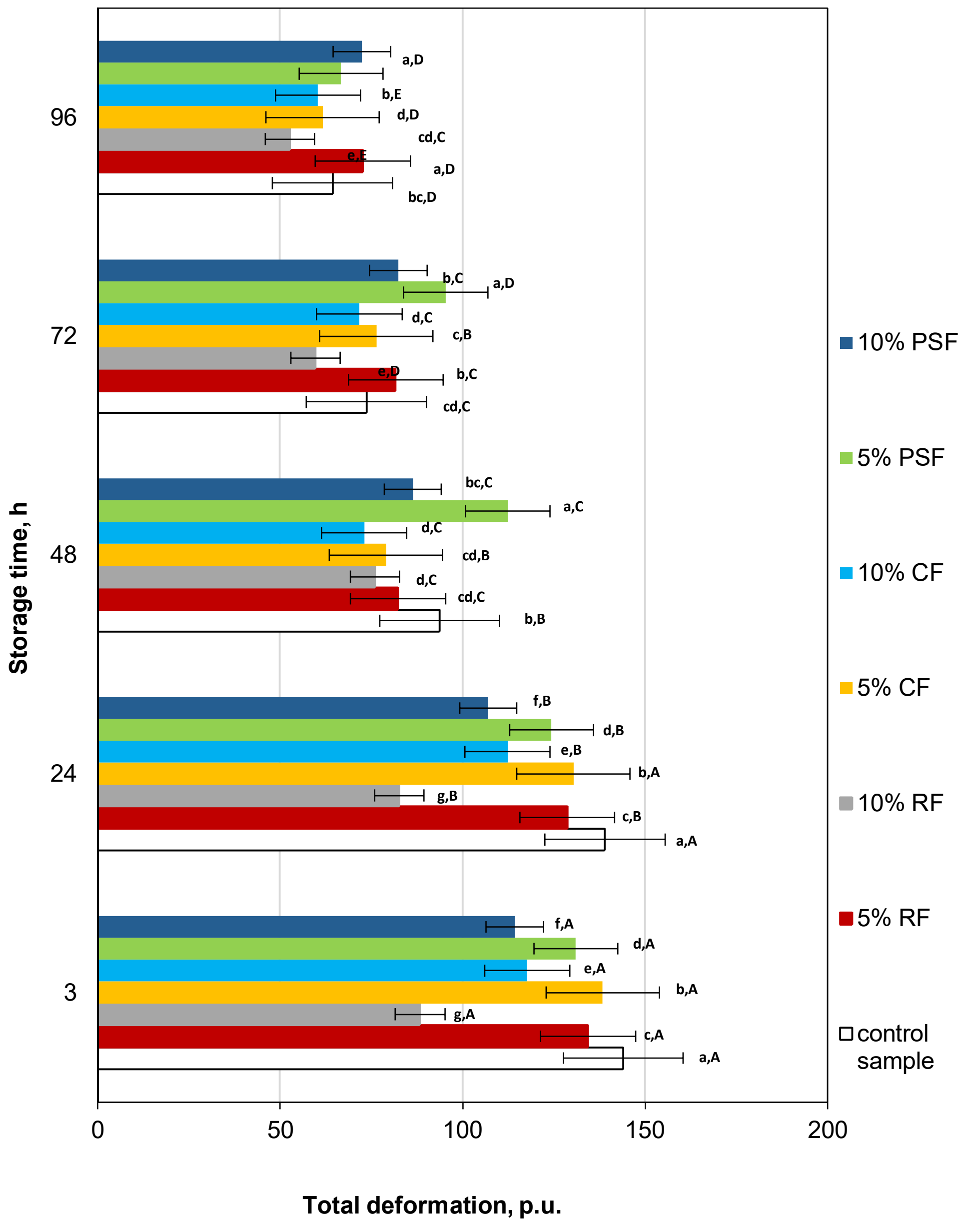

Figure 4. Effect of pumpkin seed flour, chestnut flour and rosehip flour on the total deformation of wheat bread crumb during storage:
[a–g] Means of the seven samples at the corresponding time of the measurement without a common letter differ significantly (p < 0.05).
[A–E] Means of one sample measured at 3; 24; 48; 72 and 96 h after baking without a common letter differ significantly (p < 0.05)

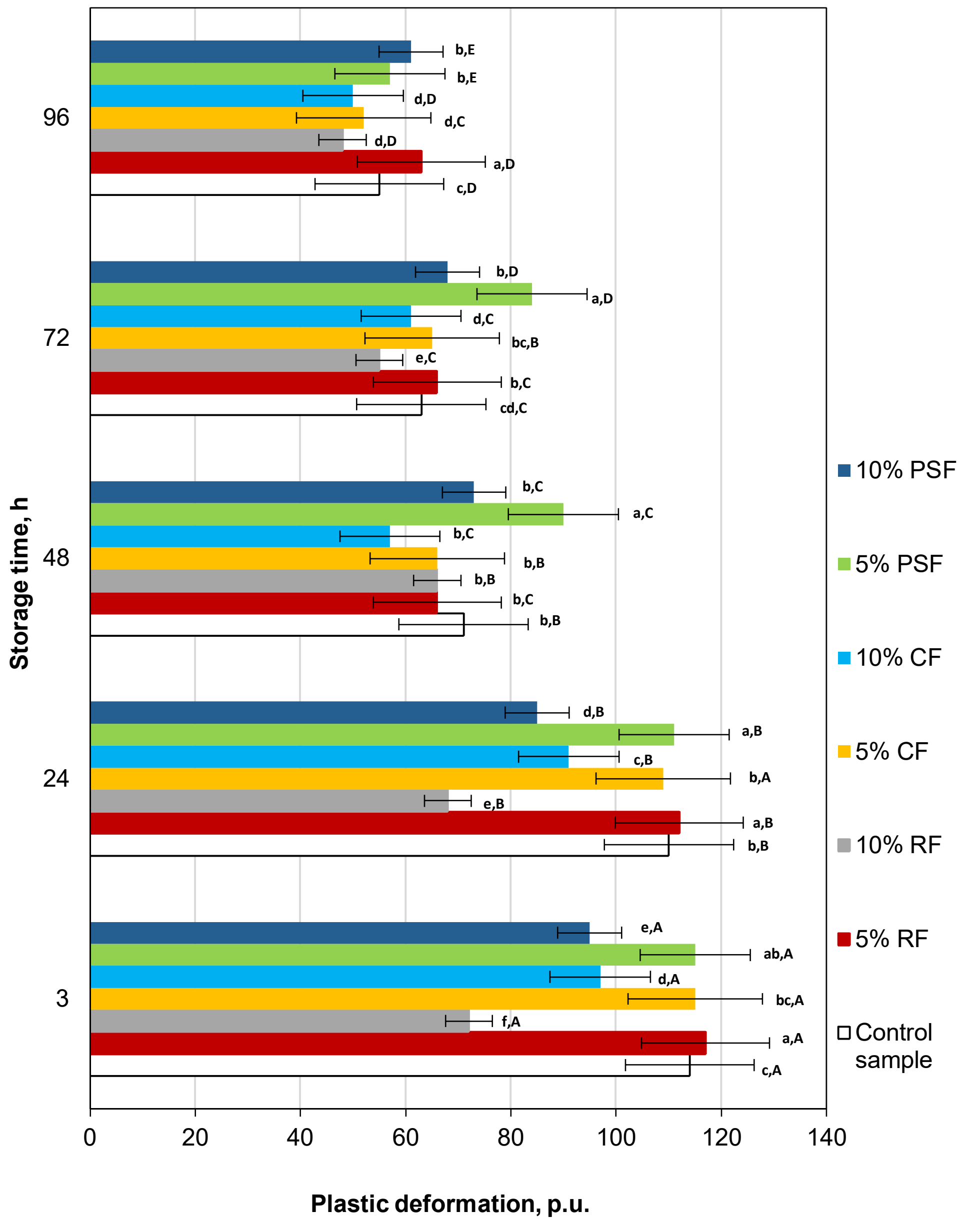

Figure 5. Effect of pumpkin seed flour, chestnut flour and rosehip flour on the plastic deformation of wheat bread crumb during storage:
[a–g] Means of the seven samples at the corresponding time of the measurement without a common letter differ significantly ($p < 0.05$).
[A–E] Means of one sample measured at 3; 24; 48; 72 and 96 h after baking without a common letter differ significantly ($p < 0.05$)

All the samples containing alternative flours in quantities of up to 5% have higher plasticity of the crumb as compared to the bread prepared from wheat flour only. At the beginning of the storage (3 h after baking), the lowest plasticity was observed in the sample with 10% rosehip flour – 72 p. u., whereas the highest values were measured in the samples with 5% chestnut, rosehip and pumpkin seed flour – 117; 115 and 117 p. u., respectively. These exceed, albeit insignificantly, the plastic deformation of the control sample – 114 p. u. After storage for 24 h, the plastic properties of the bread crumb were almost the same to the initial ones in the different samples. After 48-hour storage, the crumb of the bread with 5% pumpkin seed flour retained its high plasticity (the value decreased by 19.3% as compared to the one measured after 3 h), while the other samples showed clear deterioration of the plasticity properties. For instance, this decrease was by 35% as compared to the result after 3 h of storage in the bread with 10% chestnut flour. Rinaldi et al. (2014) also concluded that the input of high quantities of chestnut flour in the formulation of wheat bread (20%) resulted in significantly higher hardness of the crumb as compared to the control sample. The partial replacement of wheat flour with chestnut flour decreases gluten content and hinders the formation of a three-dimensional spatial structure and thus causes increased hardness of the bread (Rodriguez-Sandoval et al, 2017). Demirkesen et al. (2010) also found that adding chestnut flour to the recipe increased the crumb hardness. Authors attributed this to the increased amount of fiber in the enriched samples. Gómez et al. (2003) pointed out that the addition of fiber increases bread firmness, probably due to the thickening of the walls around the air bubbles in the bread crumb. This probably also explains the low values of plastic deformation, obtained in the sample with 10% rosehip flour. Vartolomei et al. (2021) found that the addition of rosehip flour to wheat flour resulted in a decrease in protein, wet gluten and moisture content. The same results are confirmed by another study, in which the influence of rosehip flour on the composition of wheat mixtures was investigated (Vartolomei et al., 2020).

According to other authors, adding 2% chestnut flour results in improvement of the plastic properties (Wang et al., 2023). In their study, Dall`Asta et al. (2013) found that the crumb hardness of bread with the addition of 20% chestnut flour was lower than that of wheat bread. However, a higher amount of the additive (50%) leads to an increase in hardness, also to a smaller volume and a darker color. This leads to the conclusion that the impact on the plastic properties of bread depends on the amount of additive included in the recipe. In this regard, the ratio between amylose and amylopectin in wheat and chestnut starch also matters. As pointed by Chang et al. (2021) starches isolated from different botanical sources have different ratios of amylose to amylopectin, which has a strong impact on the starch properties. After being plasticized during gelatinization, amylose has a better rearrangement ability than amylopectin, because its linear structure requires relatively little space for rearrangement and resettlement. In contrast, amylopectin has a large number of branches, and its chain distributions are disordered after gelatinization, which makes it much more difficult to rearrange in ordered structure. Amylopectin retrogradation requires a longer time than for amylose (Vamadevan and Bertoft, 2018). Therefore, the short-term retrogradation of starch (from the first few hours to several tens of hours) is generally attributed to the re-arrangement of amylose, while the long-term retrogradation is attributed to the re-arrangement of amylopectin (Chen et al., 2015).

In the control sample plastic deformation after 96 h of storage decreased by more than 50% as compared to the one after 3 h. The slowest rates of change were observed in the sample containing 10% rosehip flour (a decrease of 32%) and 10% pumpkin seed flour (a decrease of 37%).

At the end of the storage time (96 h after baking), the sample with 5% rosehip flour had significantly better plasticity as compared to other samples. Ascorbic acid, normally considered an antioxidant, is used to achieve greater dough stability and bread volume. Enrichment of bread dough with additives rich in ascorbic acid represents a strategy for producing bread with a higher degree of oxidative stability and better overall quality during storage (Osuna et al., 2018). As pointed by Tebben, Shen and Li (2018) ascorbic acid is a reducing agent, but is oxidized to the dehydroascorbic acid during mixing, and this form acts as an oxidant in dough. Authors concluded that reducing compounds from wheat bran may counteract the effects of oxidants in whole wheat dough. Therefore, oxidizing agents are less effective in whole wheat systems compared to those in refined wheat flour (Boz and Karaoglu, 2013) as used in the present study.

Impact of pumpkin seed, chestnut and rosehip flour on elastic deformation of bread crumb

The results regarding the impact of pumpkin seed, chestnut and rosehip flour on the elastic deformation of the bread crumb during storage are presented in Table 2.

Table 2

Effect of pumpkin seed flour, chestnut flour and rosehip flour on the elastic deformation of wheat bread crumb during storage

Samples	Storage time, h				
	3	**24**	**48**	**72**	**96**
Control sample	30.00 ± 0.82^{a,A}	29.67 ± 0.47^{a,A}	20.67 ± 0.47^{a,B}	10.00 ± 0.82^{b,C}	9.00 ± 1.63^{ab,C}
5% RF	16.67 ± 0.94^{c,A}	16.33 ± 0.47^{c,A}	14.00 ± 2.16^{cd,A}	15.00 ± 1.63^{a,A}	10.33 ± 2.05^{ab,B}
10% RF	14.67 ± 1.70^{c,A}	15.00 ± 0.82^{d,A}	10.67 ± 1.70^{ef,B}	3.67 ± 1.25^{c,C}	2.67 ± 0.94^{b,C}
5% CF	23.00 ± 0.00^{b,A}	20.67 ± 0.47^{b,B}	13.00 ± 0.00^{cde,C}	11.00 ± 1.63^{b,CD}	9.33 ± 1.25^{ab,D}
10% CF	21.00 ± 0.82^{b,A}	21.00 ± 0.82^{b,A}	9.67 ± 2.05^{f,B}	9.33 ± 1.25^{b,B}	8.67 ± 1.70^{ab,B}
5% PSF	14.00 ± 1.63^{c,B}	12.67 ± 0.47^{e,BC}	18.00 ± 1.63^{ab,A}	9.67 ± 1.25^{b,CD}	8.00 ± 1.63^{ab,D}
10% PSF	20.67 ± 1.89^{b,A}	20.67 ± 0.47^{b,A}	15.33 ± 2.62^{bc,B}	14.67 ± 1.25^{a,B}	13.33 ± 0.47^{a,B}

$^{a–f}$ Means of the seven samples at the corresponding time of the measurement (at each column) without a common letter differ significantly ($p < 0.05$).

$^{A–D}$ Means of one sample measured at 3; 24; 48; 72 and 96 h after baking (at each row) without a common letter differ significantly ($p < 0.05$)

The elastic properties of bread crumb are mainly characterised by the quantity and physical properties of gluten, but also by the changes that take place in the bread during staling. At the beginning of storage (3 h after baking), the control sample of bread had significantly better elastic properties of the crumb, which was proven by the highest reported value (30 p. u.). Wheat bread had the highest elasticity even after storage for a period of 24 h (27.33 p. u.) and 48 h (20.67 p. u.), which was followed by sharp deterioration – 96 h after baking, the value characterising elastic deformation decreased by 70%. Crumb elasticity in the enriched samples deteriorated at a significantly slower rate – the elastic deformation decreased by 38% in the bread with 5% rosehip flour and by 35.5% in the sample with 10% pumpkin seed flour. Paciulli et al. (2016) also pointed out that incorporation of chestnut flour in wheat bread formulation results in a delay of staling. According to Černiauskienė et al. (2014) pumpkin seed is a rich source of dietary fiber, including a mixture of plant carbohydrate polymers, both oligosaccharides and polysaccharides (e.g. cellulose, hemicelluloses, pectic substances, gums, resistant starch, inulin), that may be associated with lignin, and other non-carbohydrate components. In five different cultivars *Cucurbita maxima* grown in Lithuania, content of neutral detergent fiber (cellulose, hemicellulose, lignin) ranges from 15.50±0.14 to 26.50±0.00% dry matter. The influence of hemicelluloses on bread staling has been a subject of research in recent years. A penetrometric test revealed that the supplementation of wheat bread recipe of 0.3, 0.5, or 0.7% hemicelluloses increased the penetration depth of the crumb after 72 h of storage, thus delaying crumb hardening, and resulted in bread with a higher specific volume than the control during a 3-days storage period (Hromádková et al., 2007). Jacobs et al. (2008) also confirmed that incorporation of fiber-rich fractions in bread formulation affects bread staling. The authors proposed that the beneficial effects found could be ascribed either to the higher water retention capacity and a possible inhibition of the amylopectin retrogradation, or to the increase of the total area of gas cells (Fadda et al., 2014). Gomez et al. (2003) pointed out that this effect can be attributed to the already mentioned water-binding capacity of the fibers, which in turn reduces the loss of water during storage as well as the possible interaction between fiber and starch, resulting in a delay in starch retrogradation.

After a longer storage period (72 h and 96 h) the elasticity of the enriched samples is higher or close to the one of the control sample. The only exception was the bread with 10% rosehip flour. This was also found by Boz et al. (2013), who observed that adding rosehip flour results in deterioration of the elastic properties and more rapid staling of wheat bread. The reason is the reduced gluten content. Namely gluten forms a viscoelastic network that is responsible for slowing down water migration and retaining gas produced from yeast fermentation (Demirkesen et al., 2013).

In the studied samples, there was more intensive decrease in elasticity at the beginning of storage (3 to 48 h after baking), which was followed by more gradual change. In the control sample, the elastic deformation decreased by 3.33 times. In the bread samples supplemented with 5% alternative types of flour, the change was as follows: for the sample with 5% rosehip flour – 1.61 times, sample with 5% chestnut flour – 2.50 times and sample with 5% pumpkin seed flour – 1.80 times.

Conclusions

1. The staling rate of wheat bread enriched with types of flour, non-traditional for breadmaking (chestnut, rosehip and pumpkin seed flour) depends on both the type and the amount of the additive.
2. Regarding the total deformation – a different intensity of hardening of the bread crumb during storage was found. The most intensive changes were observed in the control sample and the bread with 5% chestnut flour – after 96 hours of storage, the softness decreased by 55%, compared to the value measured after 3 hours of storage. The slowest hardening was observed in the bread with 10% pumpkin seed flour.

3. The plastic deformation of the crumb in wheat bread at the end of the storage (96 h) decreased by more than 50% as compared to the one after 3 h of storage. The slowest rates of change were observed in the sample containing 10% rosehip flour (a decrease of 32%).
4. Up to 48 hours of storage, the control sample of bread had significantly better elastic properties of the crumb, which was followed by sharp deterioration – 96 h after baking, elastic deformation decreased by 70%. Crumb elasticity in the enriched samples deteriorated at a significantly slower rate. The most pronounced delay in the staling process was achieved when using 5% rosehip flour and 10% pumpkin seed flour.
5. The appropriate dosing of rosehip and pumpkin seed flour in the wheat bread formulation resulted in preserving the physical properties (softness, plasticity, elasticity) of the crumb for a longer time and thus – in retarding the wheat bread staling.

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