

Technological quality, bioactive features, and glycemic index of gluten-free cakes formulated with lyophilized wild *Prunus spinosa* fruit

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Abstract

This study evaluated the effects of rice flour cakes enriched with wild fruit *Prunus spinosa* powder (PS) on techno-functional, nutrition, and eating quality. PS increases the cakes' ash, protein, and moisture values. Total phenolic content increased from 35.91 ± 0.25 to 73.39 ± 0.58 mg GAE g⁻¹, and gluten-free cakes' antioxidant capacity markedly increased with the increasing level of PS powder addition ($p < 0.05$). The increase in PS decreased the glycemic index (eGI), and the lowest eGI was determined as 69.38 ± 1.21 in cake samples containing 4% PS. Except for the 1% PS added sample, the increase in PS caused a decrease in specific volume, % porosity, and increase in hardness, including four days of storage. The addition of PS increased the redness (a^*) and decreased the yellow (b^*) and brightness (L^*) values. The flow behavior index (n) of cake batters containing 1% and 2% PS were similar to the control group. Cakes containing 1% PS received the highest scores in sensory analysis. Although the increase in PS amount negatively affects the textural properties of gluten-free rice flour cakes, it is crucial to increase its bioactive properties and nutritional values and positively contribute to health by reducing the glycemic index.

Keywords: antioxidant; gluten-free cake; glycemic index; phenolic; *Prunus spinosa*; rice flour

Introduction

Bakery products have an essential place in terms of consumption on a global scale (Salehi *et al.*, 2016). Cakes are among these products; in recent years, the increase in consumers who cannot consume gluten for health-related reasons (celiac, wheat allergy, gluten sensitivity) (Di Gioia *et al.*, 2017; Gagnet *et al.*, 2021; Kaur *et al.*, 2022; Sciarini *et al.*, 2010; Singh *et al.*, 2015) led to a direction towards gluten-free products with higher nutrition values and paved the way for the evolution of traditional cake structures. For this purpose, efforts that add gluten-free flours, micro and bioactive component sources, mineral substances, and rich dietary

fibers to the formulations of cakes to increase nutritional quality have accelerated (Jeong & Chung, 2019; Kaur *et al.*, 2022). Rice flour does not contain gluten due to its structure and contains low allergens, but it is insufficient in terms of proteins, dietary fibers, vitamins, and bioactive components (Gadallah, 2017; Helm & Burks, 1996; Sivaramakrishnan *et al.*, 2004; Turabi *et al.*, 2008). Another problem with gluten-free dough is its insufficient flexibility and extensibility, resulting in low-quality cake products (Jeong *et al.*, 2021; Kaur *et al.*, 2022). Therefore, replacing wheat flour and producing high-quality gluten-free cakes simultaneously is difficult. For this purpose, it is seen as a strategic instrument to add functional food features to gluten-free flours by

including fruit, vegetables, and waste (shell, core, pulp) and concurrently to increase the nutritional quality of the cake through micro and macro components such as antioxidants, dietary fibers, vitamins, proteins, amino acids, and bioactive compounds (Majzoobi *et al.*, 2016; Saeidi *et al.*, 2018). *Prunus spinosa* L., known by different names such as gorse, sloe, güvem (name of Turkey), and prune, is a dark blue fruit found on shrubby dwarf trees. It naturally grows widely in Europe, North America, northern parts of Africa, Turkey, Caucasus, and New Zealand (Capek & Košťálová, 2022; Ozcan & Baycu, 2008). It has mature fruits, bluish-purple smoky skin, greenish flesh, large seeds, and a sour taste. Studies have determined that it is a valuable fruit containing bioactive compounds such as coumarins, flavonols, and proanthocyanidins, as well as a high amounts of mineral substances and crude fiber content. It has been determined that it contains high amounts and a variety of phenolic and antioxidants, and therefore, it is a precious wild fruit (Backes *et al.*, 2020; Guimarães *et al.*, 2014; Guimarães *et al.*, 2013). The fact that people do not prefer it due to its sour taste, and the studies on it are insufficient to have limited its use in the food industry to the production of jam and liquor on a local scale (Morales *et al.*, 2013; Pinacho *et al.*, 2015). It is a perennial plant that grows quickly in wild areas, which is essential for the industry (Capek & Košťálová, 2022; Marakoglu *et al.*, 2005; Pinacho, 2015). Including gluten-free products as a healthy product can positively affect health. Despite the well-known benefits of PS, there is limited information on its potential nutritional quality, phenolic antioxidant, and glycemic index activity in bakery products. Therefore, in this study, we produced a functional gluten-free rice cake using PS, a source of many micronutrients and bioactive compounds. However, the effects on cakes' nutritional, technological, and sensory properties were investigated. To our knowledge, no previous research has been done on adding PS to create functional bakery products. The introduction should briefly place the study in a broad context and highlight its importance. It should define the purpose of the work and its significance. The current state of the research field should be carefully reviewed, and key publications cited. Please highlight controversial and diverging hypotheses when necessary. Finally, briefly mention the main aim of the work and highlight the principal conclusions. As far as possible, please keep the introduction comprehensible to scientists outside your particular field of research. References should be numbered in order of appearance and indicated by journal guidelines.

Materials and Methods

Materials

The *Prunus spinosa* was purchased from Kirklareli (Turkey) local market and stored at 4°C. The fruits were washed and kept in a petri dish (100x20 mm²) at -80°C

overnight. The freeze-drying of fruits was performed using a freeze dryer (Martin Christ, Beta 1-8 LSC plus, Osterode am Harz, Germany). The drying process of lyophilized PS was carried out according to the standard fruit drying process with primary drying and secondary drying parameters set to 68 h at -50°C with 0.1 hPa and 2 h at 20°C with 0.1 hPa, respectively. Then, they were pulverized with a grinder, placed in a glass bottle, wrapped with aluminum foil, and protected from light. It was stored in a package that would not absorb moisture at +4°C until the cake was produced. Rice flour (Doja-Yenimahalle, Ankara, Turkey), shortening, and sucrose were also purchased from local markets. Xanthan gum was purchased from Metro Chef. Folin-Ciocalteu reagent, 2,2-diphenyl-1-picrylhydrazyl (DPPH), and gallic acid was purchased from Sigma-Aldrich (St Louis, MO, USA).

Preparation of rice cakes

The cake samples were manufactured by using the method of Dhen et.al (2016) with some modifications. Cake batter consisting of 100% rice flour (white rice), 20% sucrose, 20% eggs (from the whole egg), 25% milk (3.3% fat milk), 5% shortening, and a total of 2% baking powder and vanilla (powder), and 0.1% gum (Xanthan gum) (all percentages are given based on flour weight) was prepared. Powdered PS was replaced with flour at the rate of 1, 2, and 4%. The egg was whipped for 4 min at speed 6 with a mixer (Kitchen Aid, UK). Sucrose, shortening, and milk were mixed at speed 3 for 2 min. Finally, dry ingredients (flour, baking powder, vanilla, gum, powdered fruit) were added and mixed for 1 min at speed 2. Cake batter (80g) was poured into glass cake molds and baked at 160°C for 35 min in an electric oven (Fimak, Türkiye). It was left to cool for 2 h and placed into plastic pouches to prevent drying. Two batches of each formulation containing five cake samples were prepared.

Cake analyses

Moisture, ash, and protein analysis, specific volume

The moisture content, ash, protein, and specific volume of cakes were determined according to the AACC method 44-15.02, AACC 08-01 method, and AACC 46-12 method (AACC, 2000; Quiles *et al.*, 2018).

Hardness

The hardness values of a whole cake sample were determined with a 36 mm diameter cylindrical compression probe and a 5 kg weight load (SMS TA.XT2 Plus, England) and calculated by the equipment software according to the methods of Bozdogan *et al.* (2019) and Gualarte *et al.* (2012). (Bozdogan *et al.*, 2019; Gualarte *et al.*, 2012).

Porosity (%)

The pictures prepared in JPEG file format for the image analysis of the crumb structure (distribution of air cells) were analyzed with the Image-J2 image analysis program (Rueden *et al.*, 2017). Porosity was calculated as the percentage of pores in the whole measured area (Aranibar *et al.*, 2019; Rueden *et al.*, 2017).

Color analysis

Color measurements of cake samples were conducted as crumb and crust (Chroma Meter CR-400, Konica-Minolta Sensing Inc., Osaka, Japan) using Hunter color measurement device (L^* , a^* and b^*).

The batter viscosity

The batter viscosity was measured using a rheometer with a parallel plate configuration (Anton Paar MCR 302, Austria). PP25 probe was used at 25°C with a 1 mm gap interval. The apparent viscosity was determined as a function of the shear rate. The results were fitted to the Power Law (Eq. 1).

$$\tau = K \cdot \gamma'^n \quad (1)$$

where τ is shear stress (Pa), γ' shear rate (1/s), K is the consistency index (Pa.sⁿ), and n is the flow behavior index (Shevkani *et al.*, 2015).

Total phenolics and antioxidant assays

Samples extraction

Degreased, dried and powdered cake samples and lyophilized PS were mixed with ethanol solution (70%) (1:10 w/w) and homogenized with ultraturax for 5 min. Bioactive components of samples were extracted by keeping them at room temperature in the dark for 2 h and centrifuged at 6000 rpm for 10 min. The supernatant was taken and stored for analysis at +4°C (Kaur & Kaur, 2018).

Determination of total phenolic content (TPC)

The total phenolic content (TPC) of cake samples was determined using the Folin Ciocalteu (FC) reagent according to the method of Singleton *et al.*, (1999).

Antioxidant activity assays

The antioxidant activity of all cake samples was assessed following three different methods, each based on a different radical reduction principle: DPPH (2,2-diphenyl-1-picrylhydrazyl). The scavenging capacity of cake samples against the DPPH radical was performed according to the method described by Brand-Williams *et al.*, (1995). ABTS (2,2'-azino-bis-3 ethylbenzothiazoline-6-sulphonic acid) radical scavenging activity was conducted according to the method of Re *et al.*, (1999). With some modifications,

ferric reducing antioxidant power (FRAP) was applied as described by Benzie & Strain, (1996).

Glycemic index analysis

The in vitro method was used to determine the glycemic index properties of cake samples, with some minor modifications (Englyst *et al.*, 1999). The enzymes and all other solutions necessary for digestion and analysis were prepared as described by Yaman *et al.*, (2019). Total starch (TS) determination and estimated glycemic index (eGI) analysis were the two stages of the glycemic index analysis.

Total starch determination

Megazyme Total Starch Assay Kit" was used to determine the total starch content of the samples. Total starch (TS) analysis was carried out as described by Bekiroglu *et al.* (2022), with slight adjustments to the method developed by Goñi *et al.* (1996). All procedures in the analysis were applied to the white bread determined as the reference and cake samples. Finally, GOPOD reagent was added to the samples and absorbances were measured with a UV spectrophotometer (UV-1280, Shimadzu) set to 510nm. The following equation (Eq.1) was used to calculate TS values, with the dilution factor 50 and the glucose coefficient was used as 0.9.

$$\text{Total starch} = \frac{A_{TS} \times F \times 0.9 \times 100}{W} \quad (1)$$

A_{TS} : Total starch absorbance of samples, F: Dilution factor (50), W: Weight of samples (mg)

Estimated glycemic index (eGI) determination

Estimated GI values were determined using the "Megazyme D-Glucose (GOPOD Format Megazyme International Ireland Ltd.) Assay Kit. The analysis described by Englyst *et al.* (1999) and Yaman *et al.* (2019) was modified by making minor changes to the methods and applied to powdered white bread and cake samples. The *in vitro* digestion process was applied to the samples for 180 min. The hydrolysis index (HI) was calculated for each sample. Then, the eGI values of the samples were calculated by using the equation (Eq.2).

$$GI = 39.71 + 0.549 \times HI \quad (2)$$

Sensory analysis

Sensory evaluations of the cakes were rated for taste, crumb and crust color, pore structure, texture, and overall acceptability by 30 semi-trained panelists (15 women and 15 men aged between 25 and 35) familiar with the cakes. A 7-point hedonic scale was used, where 1 and 7 represented "extremely disliked" and "highly liked," respectively (Oliveira *et al.*, 2016).

Statistical analysis

All experiments were performed in triplicates. The data were subjected to one-way analysis of variance. The significant differences between samples were calculated with Tukey test ($p < 0.05$). SPSS for Windows version 16 (SPSS Inc., Chicago, IL, USA) was used for all data analysis.

Results and Discussion

Moisture, ash, and protein content cake samples

Moisture, ash and protein values of cakes containing different proportions of PS were given in Table 1. Depending on the increase in the amount of PS in the formulations, the ash and protein content increased ($p < 0.05$). The moisture content of the samples containing PS was lower than the control. The ash content of the cakes ranged from $0.55 \pm 0.02\%$ to $1.54 \pm 0.05\%$ and increased significantly with the incorporation level of PS (1-4%). It was determined that proteins increased from $8.38 \pm 0.38\%$ to $8.99 \pm 0.41\%$, depending on the amount of *Prunus spinosa* in cakes. The mineral content of blackthorn fruits was reported between 0.3 and 0.8% which is generally accepted for fruits (Sikora et al., 2013). PS which is also known as blackthorn fruit had the ash content between 0.55% and 1.54% in this study. PS mainly contains K, Ca, P, Mg, S, Na, B, and Al minerals (Marakoglu et al., 2005). Therefore, it was observed that there was a significant increase ($P < 0.05$) in the amount of ash, that is, mineral matter, in parallel with the PS ratio in the formulation of the cakes. Kavaz Yuksel (2015) investigated the addition of PS as an enrichment agent in ice cream production and reported that the amount of mineral matter, which was 1.02 (%) in the control sample, increased to 1.25 (%) with 5% PS. It was determined that proteins increased from $8.38 \pm 0.38\%$ to $8.99 \pm 0.41\%$ depending on the amount of PS in cakes. The protein content, which can vary greatly depending on the soil and climate conditions of the region where it is harvested, is approximately 3.4% in PS fruit (Marakoglu et al., 2005). PS fruit, with its high protein content compared to stone fruits such as peaches, cherries, and apricots, not only strengthens its phenolic and antioxidant capacities but also enriches the foods with protein. To improve the functional quality of gluten-free bakery products, proteins obtained from different sources and enriching additives that can increase the protein content have been suggested (Storck et al., 2013).

Batter viscosity

The viscosity of cake batters is important to determine its correlation with cakes' micro and macro properties

Table 1. Quality characteristics of cake samples.

Samples Name	Moisture %	Ash %	Protein %	SV (mL/g)	Hardness (g)	Porosity (%)	Crumb Color			Crust Color		
							L*	a*	b*	L*	a*	b*
Control	34.34 ± 0.12^a	0.55 ± 0.02^d	8.38 ± 0.38^d	2.12 ± 0.12^a	1622.7 ± 9.1^d	6.21 ± 0.25^d	67.36 ± 0.02^a	2.46 ± 0.3^d	35.36 ± 0.09^a	67.28 ± 0.22^a	0.07 ± 0.02^d	25.52 ± 0.31^a
PS1	31.73 ± 0.01^c	0.76 ± 0.01^c	8.5 ± 0.39^c	2.10 ± 0.04^a	1844.2 ± 12.1^c	10.19 ± 0.13^c	59.77 ± 0.39^b	4.36 ± 0.08^c	26.31 ± 1.13^b	58.91 ± 0.21^b	2.38 ± 0.22^c	17.93 ± 0.62^b
PS2	31.26 ± 0.09^c	1.02 ± 0.03^b	8.85 ± 0.4^b	1.89 ± 0.04^b	1905.5 ± 5.1^b	13.80 ± 0.54^b	56.02 ± 0.60^c	6.68 ± 0.31^b	23.64 ± 0.60^c	57.61 ± 0.83^c	3.74 ± 0.44^b	15.67 ± 1.19^c
PS4	32.31 ± 0.29^b	1.54 ± 0.05^a	8.99 ± 0.41^a	1.61 ± 0.01^c	2072.4 ± 11.6^a	24.89 ± 0.33^a	51.25 ± 0.01^d	8.80 ± 0.008^a	21.62 ± 0.22^d	53.84 ± 0.71^d	4.73 ± 0.22^a	12.30 ± 0.13^d
Samples with different letters in the same column differ significantly ($P < 0.05$). PS1: 1% <i>Prunus spinosa</i> addition to cake, PS2: 2% <i>Prunus spinosa</i> addition to cake, PS4: 4% <i>Prunus spinosa</i> addition to cake.												

(Saghafi *et al.*, 2019). Power law constants of gluten-free cake batters, including PS, were shown in Table 2. The Power Law model provided a good fit to the shear stress vs shear rate data of the cake batters ($R^2=0.93-0.99$). The consistency index (K), an indicator of the viscous nature of the system, ranged from 8.02 to 283.21 Pa sⁿ in this study (Table 2). No significant differences were observed between the control and samples containing 1% and 2% PS addition. The highest amount of PS addition (4%) led to a significant K -value increment compared to other samples with PS. This can be due to the stronger structure of the cake sample containing 4% PS addition. This increment in the high-level addition of carrot pomace powders into gluten-free cake batter was also observed in a study by Kirbaş *et al.* (2019). This is explained by the higher water binding capacity of pomace powder depending on the addition level (Kirbaş *et al.*, 2019).

The flow index (n) values of cake batters are between 0.08 and 0.57. According to Fischer *et al.* (2009), the n value can be equal to or lesser than or greater than 1, which refers to the Newtonian flow, shear-thinning, or shear-thickening flow behavior, respectively (Fischer *et al.*, 2009). The n values of all cake batters in the current

study are lower than 1, showing shear-thinning (pseudoplastic) behavior. The lowest n value indicates high viscosity at the low shear rate (Kirbaş *et al.*, 2019). An increase in apparent viscosity with the addition of PS was observed, and the apparent viscosity of all cake batters decreased with the applied shear rate (Figure 1).

The control sample showed the lowest viscosity, while PS4 had the highest. This may be attributed to the increased dietary fiber (DF) content of the cake formulation due to the addition of PS. DFs are known to have high water holding capacity (WHC). When the WHC of cake batters increases due to the addition of DF sources such as fruits, the apparent viscosity increases (Ronda *et al.*, 2011)

Volume, hardness, porosity, and color quality characteristics of cake samples

Values for specific volume (SV), hardness, porosity, crumb, and crust color (L^* (brightness), a^* (+ red, – green), and b^* (+ yellow, – blue)) characteristics are presented in Table 1. The inclusion of more than 1% of PS causes a significant reduction in the SV of cakes ($P<0.05$). No difference was observed between the control and PS1 cakes ($P>0.05$). This can be related to the apparent viscosity of cake batters. Kirbaş *et al.* (2019) reported that there was a negative correlation between volume and apparent viscosity. As observed in Figure 1, the apparent viscosity of the control sample was the lowest, while the PS4 sample showed the highest viscosity. This caused a dramatic reduction in the SV values of cake samples. When the viscosity of cake batters increases, the air bubbles in the system cannot expand sufficiently during baking, thus reducing the SV of cakes observed (Hedayati & Mazaheri Tehrani, 2018; Kirbaş *et al.*, 2019). It was determined that the crumb porosity of the cakes from 6.21 to

Table 2. Rheological data of cake batters determined by the Power law model.

	K (Pa s ⁿ)	n	R^2
Control	8.02±0.58 ^b	0.57±0.01 ^a	0.99
PS1	21.24±1.18 ^b	0.36±0.01 ^b	0.95
PS2	18.18±3.25 ^b	0.43±0.05 ^b	0.97
PS4	283.21±69.74 ^a	0.08±0.02 ^c	0.93

Samples with different letters in the same column differ significantly ($P<0.05$). K : consistency index, n : flow behavior index. PS1: 1% *Prunus spinosa* addition to cake, PS2: 2% *Prunus spinosa* addition to cake, PS4: 4% *Prunus spinosa* addition to cake.

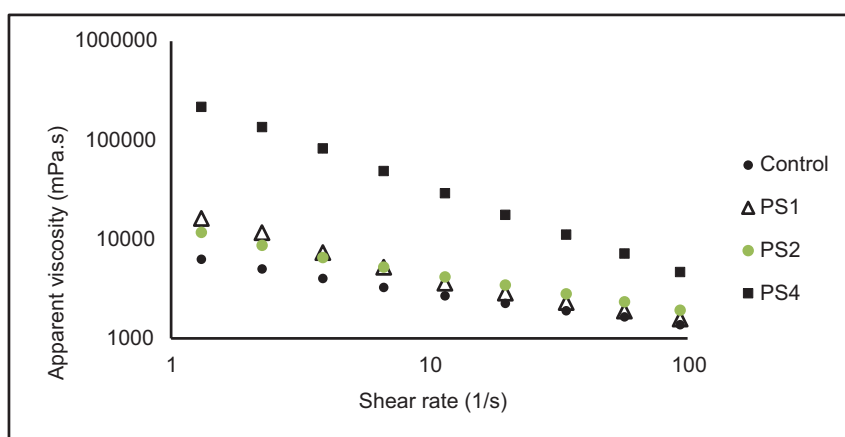


Figure 1. Effects of PS substitution on the viscosity of cake batter. PS1: 1% *Prunus spinosa* addition to cake, PS2: 2% *Prunus spinosa* addition to cake, PS4: 4% *Prunus spinosa* addition to cake.

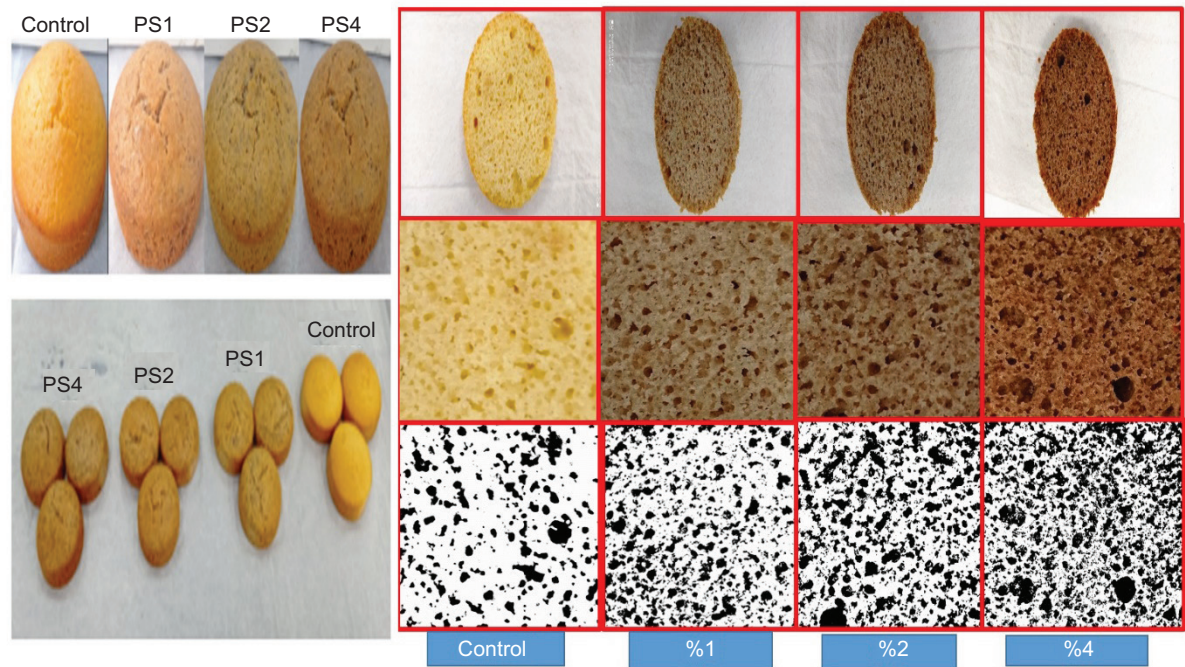


Figure 2. The crust and crumb appearance and binarized image of muffins Codes: Control formulation (control), formulation fortified with 1, 2, and 4% *Prunus spinosa* (PS1, PS2, and PS4, respectively).

24.89% increased significantly with the increase in PS when compared to the control sample (Table 1, Figure 2).

The increase in viscosity supports this situation. In addition, some studies suggested that the high amount of protein, ash, and dietary fiber added to cakes enriched with different ingredients led to a significant increase in pores (Gökşen & Ekiz, 2021; Grasso *et al.*, 2020; Marchetti *et al.*, 2018; Quiles *et al.*, 2018). The cake's hardness significantly increased when the amount of PS addition increased (1622.7–2072.4 g) ($p<0.05$). Figure 3 illustrates the effect of PS incorporation on the cake's hardness during storage. Significant increases were observed during 4 days of storage as expected.

Gradual increases were obtained for the samples PS2 and PS4 during storage. However, PS1 samples gave a similar trend to the control sample. A dramatic effect was observed for these samples on day 3 of storage. The crust and crumb color characteristics of cakes enriched with PS were shown in Table 1. It was determined that L^* values of cake crust decreased significantly ($p<0.05$) from 67.28 to 53.84, and b^* values of cake crust decreased from 25.52 to 12.30 depending on the increase in PS. On the other hand, it was observed that the a^* values for cake crust increased from 0.07 to 4.73. While the increase in PS increased the a^* value of the crust and crumb colors of the cake samples, it caused the L^* and b^* values to decrease. PS addition suppressed the yellow color by making the cake red and dull.

Total phenolic content (TPC) and antioxidant capacity (FRAP, ABTS, and DPPH)

The phenolic content of the samples was shown in Table 3. Phenolic content increased with the addition of PS and was determined in the cakes with the highest PS ratio (4%). The antioxidant capacity results determined using three different methods, ABTS, FRAP, and DPPH, are shown in Table 3. As it can be observed from the results, the amount of PS added increased the cake's antioxidant capacity. For the FRAP technique, an antioxidant method based on the iron reduction principle and the highest radical reducing power was determined in the cake sample with 4% PS (73.1 ± 0.43 mg TEAC/100g sample). At the same time, it remained at the lowest level (17.8 ± 1.14) in the control sample. The FRAP values of other samples were found to be 31.45 ± 0.85 for PS1 and 47.51 ± 1.42 mg TEAC /100 g for PS2 ($p<0.05$). ABTS, which is another extensively used approach for assessing the antioxidant capacity of foods, is a technique based on the principle of reduction of TPTZ- Fe^{+3} to TPTZ- Fe^{+2} (Karamać *et al.*, 2018). Although the scavenging of the ABTS radical was lowest in the control cake sample (76.46 ± 0.28 mg TEAC /100g sample), an increase to 88.64 ± 0.31 for PS1, 93.92 ± 1.26 for PS2, and 154.91 ± 0.17 mg TEAC /100 g sample for PS4, was observed ($p<0.05$). The reduction of DPPH radical, was calculated as % inhibition, and the reduction values varied between $3.44\pm0.07\%$ (Control) and $18.85\pm0.14\%$ (PS4). Compared to the control cake, the addition of PS contributed

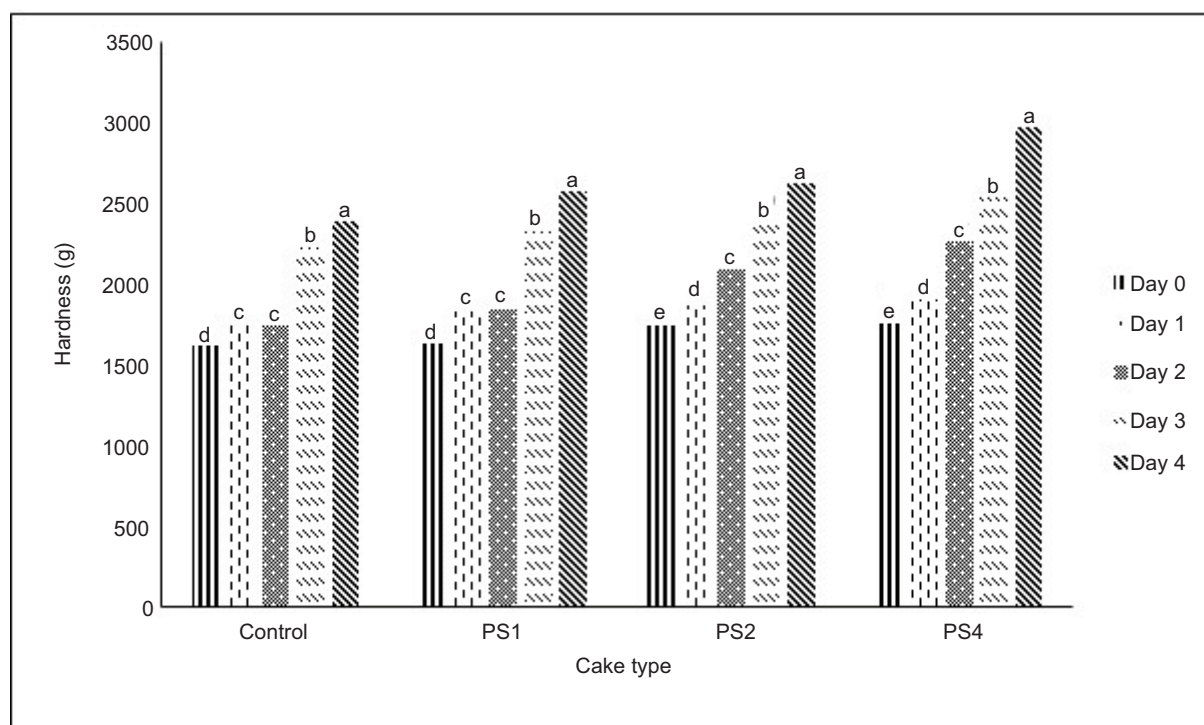


Figure 3. Effects of PS addition on the hardness value of the cakes during 4 days of storage. Samples with different letters in the same cake type differ significantly ($P<0.05$).

Table 3. Phenolic content, antioxidant activities, hydrolysis index (HI), and predicted estimated glycemic index (eGI) of the cake samples.

Sample	TPC mg GAE g ⁻¹	FRAP (mg TEAC /100g sample)	ABTS (mg TEAC /100g sample)	DPPH (Inhibition %)	HI	eGI
PS	566.93±2.71 ^a	396.94±1.67 ^a	892.0±7.95 ^a	67.19±0.07 ^a		
Control	35.91±0.25 ^e	17.8±1.14 ^e	76.46±0.28 ^c	3.44±0.07 ^e	91.09±2.88 ^a	89.08±1.47 ^a
PS1	42.95±0.29 ^d	31.45±0.85 ^d	88.64±0.31 ^c	6.23±0.78 ^d	87.86±0.35 ^a	86.30±1.04 ^a
PS2	51.61±1.45 ^c	47.51±1.42 ^c	93.92±1.26 ^c	9.37±0.28 ^c	77.94±0.83 ^b	75.05±1.16 ^b
PS4	73.39±0.58 ^b	73.1±0.43 ^b	154.91±0.17 ^b	18.85±0.14 ^b	70.75±0.39 ^c	69.38±1.21 ^c

Samples with different letters in the same column differ significantly ($P<0.05$). PS1: 1% *Prunus spinosa* addition to cake, PS2: 2% *Prunus spinosa* addition to cake, PS4: 4% *Prunus spinosa* addition to cake. The data are expressed as average±standard deviation in three replicates. Samples with different letters in the same column differ significantly ($P<0.05$).

considerably to the % inhibition of the DPPH radical and, finally, the strongest inhibition at the highest usage rate.

As the concentration of PS increased, its antioxidant capacity and phenolic content were strengthened. Levent *et al.* (2021) observed similar findings in antioxidant and phenolic substance contents in gluten-free cake samples enriched with grape seeds, pomegranate seeds, poppy seeds, cinnamon, and turmeric (Levent *et al.*, 2022). Another study reported that using persimmon powder in gluten-free cake significantly improved its antioxidant capacity (DPPH, FRAP) and phenolic content, paralleling the rising substituted ratio (Yeşilkanat & Savlak, 2021).

The antioxidant and phenolic content of rice flour, frequently preferred in gluten-free bakery products, is quite low compared to other gluten-free flours such as chickpea, lentil, and oat (Rocchetti *et al.*, 2017). PS can be considered a rich source of phenolics and antioxidants, as it mainly contains quercetin and kaempferol phenols, neochlorogenic and caffeic-derived phenolic acids, coumarin derivatives such as aesculetin, umbelliferone, and anthocyanins in substantial quantities (Pinacho *et al.*, 2015). In this context, adding PS, a potential phenolic and antioxidant source, can strengthen the bioactive properties of gluten-free cakes, making them much more functional foods.

Total starch and estimated glycemic index (eGI) determination

Estimated glycemic index (ϵ GI) values and hydrolysis indexes (HI) of samples were given in Table 3. The hydrolysis index, expressed as the digestion of the samples *in vitro*, ranged from 70.75 to 91.08. The HI of the control cake sample was calculated to be 91.08 ± 2.88 . HI values of cakes with 1%, 2%, and 4% PS decreased to 87.85 ± 0.35 , 77.93 ± 0.83 , and 70.75 ± 0.39 , respectively. There was a decrease in all cake samples compared to the control sample in the ϵ GI values shaped by the effectiveness of slow and fast digestible starch and total starch, mainly due to the hydrolysis rate. The ϵ GI value of the cake samples decreased with the increasing PS ratio and was determined as 89.08 ± 1.47 for control, 86.30 ± 1.04 for PS1, 75.05 ± 1.16 for PS2, and 69.38 ± 1.21 for PS4. The ϵ GI value expressed an increase in blood sugar level of the food 2 h after it is consumed is considered a low glycemic index for values less than 55 (ϵ GI<55%). Low glycemic index foods allow the change in blood sugar level to decrease and increase more slowly, depending on the carbohydrate mechanism consumed. In this context, although it is not considered as having a low glycemic index, PS led to an essential contribution to reduce the glycemic index. However, PS addition can be considered as an approach that contributes to develop low GI products that raise blood sugar more slowly.

Sensorial evaluation

The sensory properties of the rice cake samples in terms of taste, crumb-crust color, texture, pore structure, and

general acceptability were shown in Figure 4. Cake samples containing different ratios of *Prunus spinosa* had higher scores than the control sample and were more acceptable. Compared to the control sample, adding more than 2% PS decreased the scores of crumb and crust color, pore structure, taste, texture, and general acceptability. The PS1 cake sample was the most liked cake in all properties, followed by PS2.

Conclusion

This study contributed to the enrichment of gluten-free bakery cakes and expanded consumer options. As a result of this study, it was found that the produced PS-substituted rice flour cakes were significantly higher in total phenolic content, antioxidant activity, and protein amount when compared to the control cake. The cakes containing PS provided additional health benefits by reducing the glycemic index. Adding PS to the cake formulation seems to be a good choice for making a richer functional cake that can be commercialized. However, some challenges remain, such as combining better flavor, texture, and nutritional gluten-free food. Further studies should be designed to overcome such difficulties and increase the sensory perception of gluten-free products among celiac patients.

Author contributions

EC, conceptualization, data curation, methodology, formal analysis, writing – original draft; GO,

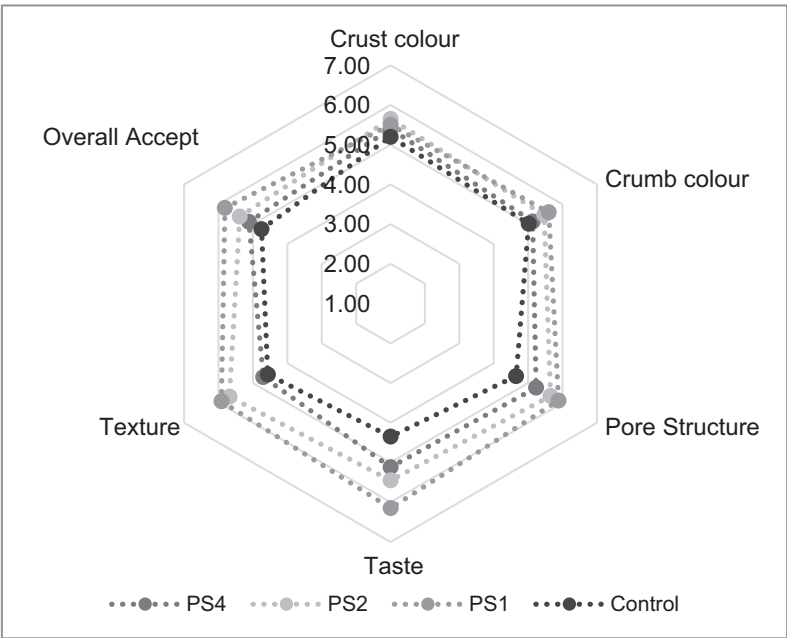


Figure 4. Sensory analysis of the developed cakes.

conceptualization, data curation, methodology, formal analysis, writing – original draft; HB, conceptualization, data curation, methodology, formal analysis, writing – original draft; MA, writing – review & editing, supervision; OS, writing – review & editing, supervision.

Data availability

Data will be made available on request.

Conflict of interest

The authors declare no conflict of interest.

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