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The mallow, *Malva aegyptiaca* L. (Malvaceae): Phytochemistry analysis and effects on wheat dough performance and bread quality

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ABSTRACT

The effects of *M. aegyptiaca* leaves supplementation (1, 2, 3 and 5%) in wheat bread formulation were examined in order to obtain an antioxidant-enriched product. Mallow powder contained relatively high levels of dietary fiber and flavonoids. LC-HRESIMS analysis of the mallow ethanolic extract allowed the identification of 7 flavonoids, 6 terpenoids and 3 fatty acids. The (P/L) ratio of the dough increased with the increment of mallow powder level, whereas the deformation energy (W) decreased. At the highest substitution level, a reduction in bread specific volume and springiness, and an increase in hardness and chewiness were observed. Rising levels of mallow powder into bread increased flavonoids content and antioxidant capacity, and decreased crust colour parameters (L*, a* and b*). Sensory evaluation showed that mallow supplementation at 3% level remained acceptable. All these data suggest that *M. aegyptiaca* may be useful as a candidate for improving nutraceutical quality of bread.

1. Introduction

Nowadays, there is an increasing consumer demand for foods fortified with natural antioxidants in order to improve their general state of health and to reduce the risk of chronic diseases of oxidative stress origin. In fact, foods rich in antioxidants play an essential role in the prevention of cardiovascular diseases, cancers, inflammation and problems caused by cutaneous aging (Fan, Zhang, Yu, & Ma, 2006). Plants are an inexhaustible source of functional and bioactive compounds and they have been used for thousands of years for their preservative properties and also to enhance the flavour and colour of food. Currently, their incorporation into the formulation of conventional foods is becoming an interesting approach, which may significantly improve their nutraceutical potential and therefore they could become a "preventive model" for disease prevention (Zouari, 2015).

Bread made from white flour is considered as refined product with a low antioxidant capacity. Due to its widespread consumption throughout the world, it could be considered as an interesting vehicle for functional supplements. Dziki, Rózyło, Gawlik-dziki, and Świeca (2014) reviewed the current trends in the enhancement of antioxidant activity of wheat bread by the addition of raw materials rich in phenolic antioxidants, such as other cereals, spices, herbs, fruits and waste products from the food industry. However, few studies focused on the bread enrichment with green parts of vegetables. Moreover, many studies did not take into consideration the consumer acceptance of enriched breads (Dziki et al., 2014).

Malva aegyptiaca L. is a wild plant of the Malvaceae family, widespread in North Africa and edible by the local population. The macerate of *M. aegyptiaca* leaves was used in the treatment of dysentery, constipation and fevers. Besides, the cataplasm of leaves is also used in treating wounds, painful and skin-eruptions (Boual, Kemassi, Ould El Hadj Khelil, Michaud, & Ould El Hadj, 2011). It was reported that this vegetable contains several important nutrients such as essential minerals, n-3 and n-6 fatty acids, and various terpene compounds with valuable biological properties. Furthermore, this vegetable possesses an important antioxidant activity

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and thus has the potential to be used as a cheap natural source for reducing cellular oxidative damage (Zouari et al., 2011).

In the present work, *M. aegyptiaca* (common name: khoubbiza) leaves were used as a new functional ingredient for enhancing nutraceutical properties of wheat bread. Mallow leaves were characterized in terms of chemical and techno-functional characteristics. In fact, they were analyzed by Liquid Chromatography-High Resolution Electrospray Ionization Mass Spectrometry (LC-HRESIMS) technique in order to identify the phenolic compounds frequently associated with antioxidant activity. Then, the effects of mallow leaves powder substitution to wheat flour on alveographic properties of dough, and texture, antioxidant and sensory characteristics of bread were assessed.

2. Material and methods

2.1. Plant material and mallow powder preparation

The wild *M. aegyptiaca* was collected, in January 2016, from the area of Maknassy (Sidi Bouzid, Tunisia) characterized by a mean rainfall of 200–300 mm/year. After harvest, the leaves were separated and shade-dried for 20 days. Then, they were ground in a spice grinder (Black & Decker CBG100S Smartgrind, Maryland, USA), sieved through 250 μm sieve and the obtained powder was stored at 4 °C until use.

2.2. Chemical analysis and functional characteristics of mallow powder

Dietary fiber content was determined according to the gravimetric enzymatic method (Prosky, Asp, Schweizer, DeVries, & Furda, 1988). Total chlorophyll and β-carotene contents, water holding capacity (WHC) and fat absorption capacity (FAC) were determined by the methods described by Ayadi, Abdelmaksoud, Ennouri, and Attia (2009). WHC and FAC were expressed as g of water bound per 100 g of mallow powder and as g of oil bound per 100 g of mallow powder, respectively.

2.3. Antioxidant activity and LC-HRESIMS analysis of mallow powder extract

2.3.1. Ethanolic extract preparation

The mallow powder (25 g) was extracted by maceration using 250 ml of ethanol during 24 h. The solvent was then evaporated under vacuum and the residual solvent was removed by flushing with nitrogen.

2.3.2. Flavonoids content and antioxidant activity

Flavonoids content was measured in ethanolic extract of mallow powder as previously described (Dewanto, Wu, Adom, & Liu, 2002) and expressed as mg quercetin equivalent (QE)/100 g of mallow powder. DPPH· radical-scavenging and reducing power assays of ethanolic extract were measured as was described by Yildirim, Mavi, and Kara (2001). DPPH· radical-scavenging activity was presented by IC_{50} value, which was defined as the extract concentration needed to scavenge 50% of DPPH·. In the reducing power assay, the presence of antioxidants in the sample would result in the reduction of Fe^{3+} to Fe^{2+}, which can be monitored by measuring the formation of Perl's Prussian blue ($Fe_4[Fe(CN)_6]3$) at 700 nm. The increase in absorbance at 700 nm indicated an increase in reductive capacity. The extract concentration (EC_{50}) providing 0.5 of absorbance at 700 nm was presented.

2.3.3. LC-HRESIMS analysis

One hundred mg of mallow powder extract was dissolved in 100 ml of 10% methanol, filtered and then 1 ml was transferred into LC-MS vials. Reversed-phase column (Pursuit XRs ULTRA 2.8, C18, 100 × 2 mm, Agilent Technologies, UK) was used to carry out HPLC analyses. Twenty μl of the sample was injected at a column temperature set at 30 °C. Mobile phases consisted of 0.1% formic acid in water (A) and 0.1% formic acid in methanol (B). A gradient program was used for separation at a flow rate of 1 ml/min. Mobile phases consisted of an initial composition of 100% solvent A, with a gradient of 100% solvent B over 20 min, hold at 100% solvent B for 5 min and 100% solvent A for 25 min. Drying gas flow rate was 1 ml/min at 320 °C. MS was operated in the positive ion mode in a mass range of 100–2000 m/z. High resolution mass spectral data were obtained on a Thermo Instruments ESI-MS system (LTQ XL/LTQ Orbitrap Discovery, UK) connected to a Thermo Instruments HPLC system (Accela PDA detector, Accela PDA autosampler and Accela Pump).

2.4. Alveographic properties of dough

Alveograph measurements were performed using the MA 82 Alveograph (Chopin, Tripette et Renaud, Villeneuve La Garenne, France) following the standard method (A.A.C.C. 2000). The studied samples were wheat flour (control) and blends containing a mixture of wheat flour and mallow powder in the ratios (m/m): 99/1 (formulation 1: F1), 98/2 (formulation 2: F2), 97/3 (formulation 3: F3), and 95/5 (formulation 4: F4). The following alveographic parameters (P, L and W) were automatically recorded by a computer software program. The maximum overpressure (P) needed to blow the dough bubble indicated the dough resistance to deformation or its tenacity. This parameter was related to the quality and quantity of gluten as well as to their ability to absorb water. The average abscissa (L) at bubble rupture indicated the dough extensibility or the ability of the gluten to hold the gas. The configuration ratio (P/L) indicated the balance between the tenacity and the extensibility of the dough. The deformation energy (W) represented an index of dough strength.

2.5. Bread elaboration

Flat bread was prepared in a local pastry industry (Société Patisserie-Masmoudi, Sfax, Tunisia). The loaf had a circular form with an average thickness of 1 cm and a diameter of 4 cm. The standard bread formulation is consisted of: 1 kg wheat flour, 433 g water, 100 g olive oil, 33.33 g whole egg, 23.33 g sodium chloride, 8.33 g dried yeast and 1.66 g sodium bicarbonate. The chemical composition of wheat flour (g/100 g) was as follows: starch, 71.74; water, 13.40; proteins, 10.27; total fiber, 0.93 and fat, 0.77 as determined by a multipurpose analyzer (MPA) spectrometer (Bruker Optics, Wissembourg, France). Breads with variable levels of mallow powder were made from wheat flour (control) and blends containing 1 g, 2 g, 3 g and 5 g of mallow powder per 100 g wheat flour substitution basis (F1, F2, F3 and F4). The yeast was dissolved in warm water (35 °C) and the resulting solution was added to the dry ingredients and finally the olive oil was added. The mixture was blended manually for 10 min and the resulting dough was fermented for 90 min at 30 °C. The dough circles were shaped and placed on proofing trays for 2 h before baking, which was conducted at 180 °C for 10 min. Finally, the flat breads were cooled to room temperature and then stored at −18 °C in plastic bags. Prior to analysis, the breads were thawed for 24 h at 4 °C, and then equilibrated to room temperature for 4 h.

2.6. Colour measurement

Colour measurement parameters (lightness L*, redness a* and

yellowness b*) were carried out using a colour flex spectrocolorimeter (Hunter Associates Laboratory Inc., Reston, VA). L* value indicates the lightness, 0–100 representing dark to light, a* value gives the degree of the green–red colour, with a higher positive a* value indicating more red. The b* value indicates the degree of the blue–yellow colour, with a higher positive b* value indicating more yellow.

2.7. Instrumental texture and specific volume of bread

Hardness (N), springiness (mm) and chewiness (N × mm) of bread was measured using a texturometer (Lloyd Instruments Ltd., West Sussex, UK) as previously described by Ayadi et al. (2009). Bread volume (cm^3) was determined by the rapeseed displacement method and bread specific volume (cm^3/g) was measured as bread volume divided by bread mass.

2.8. Flavonoids content and antioxidant activity of bread

Flavonoids content, DPPH· radical-scavenging activity (%) and Fe^{3+} reducing power (A_{700}, absorbance at 700 nm) were determined in an ethanolic extract of bread as previously described (Dewanto et al., 2002; Yildirim et al., 2001). For this reason, bread sample was converted into fine powder by using a pestle and mortar. Then, 10 g of powdered sample were homogenized with 20 ml ethanol for 24 h at ambient temperature using an orbital shaker at stirring speed of 200 rpm. After filtration, the obtained extract was recovered and kept at 4 °C until further analysis.

2.9. Sensory evaluation

The sensory properties (colour, odour, taste, texture and overall acceptability) of fresh prepared breads were evaluated according to the method of Murray, Delahunty, and Baxter (2001) by sixty panelists. A seven-point hedonic scale was used, where 7: like very much, 6: like moderately, 5: like slightly, 4: neither like nor dislike, 3: dislike slightly, 2: dislike moderately and 1: dislike very much for each attribute.

2.10. Statistical analysis

All analytical determinations were performed in duplicate for three samples ($n = 3$). One-way analysis of variance was conducted using the SPSS software for Windows™ (version 17, SPSS Inc., Chicago, IL, USA). Duncan's multiple range test ($p < 0.05$) was used to compare the average responses between treatments.

3. Results and discussion

3.1. Phytochemical and functional characteristics of mallow powder

The results for the chemical and functional characteristics of the mallow powder were presented in Table 1. In our previous work, carbohydrates (78.80 g/100 g DM) followed by proteins (8.70 g/100 g DM) were found to be the most abundant macronutrients for *M. aegyptiaca* leaves, whereas the fat was the least available one (3.68 g/100 g DM). In addition, the ash content was found to be 8.82 g/100 g DM with potassium (2.11 g/100 g DM), calcium (1.73 g/100 g DM) and magnesium (0.55 g/100 g DM) as the most concentrated minerals (Zouari et al., 2011). In the present study, soluble dietary fiber (21.50 g/100 g mallow powder) was found to be much higher than insoluble dietary fiber (9.65 g/100 g mallow powder). Mallow powder presented an important water holding capacity (WHC) of 511 g water/100 g that was related to the hydrophilic constituents. Alongside its hydration property, mallow powder possessed the capacity to hold oil with fat absorption capacity (FAC) of 319.1 g oil/100 g mallow powder that was related mainly to the surface properties of mallow macromolecules. WHC and FAC suggest some possibilities about the use of such substances as functional ingredients in food products: e.g., dietary fiber with high WHC can be used to avoid syneresis and to modify the viscosity and texture of some formulated foods. By contrast, dietary fiber with important FAC could stabilize emulsions and high fat food products (Elleuch et al., 2011). It's well known that the mallow family is characterized by the presence of mucilaginous cells that store polysaccharides, allowing the retention of large amounts of water (Boual et al., 2011). In addition to their technological functionality, polysaccharides play an important role in many physiological processes and in the prevention of some diseases. In fact, mucilages are one of the major components responsible for the therapeutic effects of *Malva*, mainly due to their anti-complementary and cough suppression activities (Gasparetto, Martins, Hayashi, Otuky, & Pontarolo, 2011).

Table 1
Chemical and functional characteristics of *M. aegyptiaca* leaf powder.

Parameters	
Soluble dietary fiber[a]	21.50 ± 1.15
Insoluble dietary fiber[a]	9.65 ± 0.60
Total chlorophyll[b]	83.20 ± 0.60
β-Carotene[b]	50.0 ± 0.10
Flavonoids[c]	1.20 ± 0.05
DPPH· scavenging activity (IC_{50}, mg/ml)	0.35 ± 0.06
Reducing power (EC_{50}, mg/ml)	0.16 ± 0.02
Water holding capacity[a]	511 ± 5
Fat absorption capacity[a]	319.10 ± 13.50

Data presented as the mean ± standard deviation ($n = 3$).
[a] g/100 g of mallow powder.
[b] mg/100 g of mallow powder.
[c] mg quercetin equivalents (QE)/100 g of mallow powder.

The antioxidant effects of the plant leaf alcoholic extracts are linked to the presence of antioxidant substances, such as tocopherols, carotenoids and flavonoids. In the present study, Table 1 shows β-carotene and flavonoids contents to be 83.2 mg/100 g mallow powder and 1.2 g QE/100 g mallow powder, respectively. As compared to *Malva sylvestris* (Gasparetto et al., 2011), the leaves of *M. aegyptiaca* contained high level of flavonoids, which are correlated with their powerful antioxidant potential in 2,2-diphenyl-1-picrylhydrazyl DPPH· radical-scavenging (IC_{50}: 0.35 mg/ml) and Fe^{3+} reducing (EC_{50}: 0.16 mg/ml) assays (Table 1). It is well known that diets rich in vegetables and eventually in antioxidant compounds are associated with a reduced risk of many diseases including coronary heart disease and cancers (Duthie, Duthie, & Kyle, 2000). Therefore, it is important to identify compounds that contribute to good health in the diet. To the best of our knowledge, there are no studies on the identification of flavonoids in *M. aegyptiaca*. Liquid chromatography-high resolution electrospray ionization mass spectrometry (LC-HRESIMS) allowed the identification of 16 compounds in the ethanolic extract of *M. aegyptiaca* leaves (Table 2). These compounds are divided into 7 flavonoids, 6 triterpenoids and 3 fatty acids. A survey of the literature shows that most of the identified flavonoids had potent antioxidant potential. In fact, the IC_{50} values relative to the DPPH· radical-scavenging activities of quercetin-3-*O*-rutinoside, isorhamnetin, quercetin-3-*O*-glucoside and isorhamnetin-7-*O*-glucoside were 5.54, 5.22, 1.74 and 0.83 μg/ml, respectively (Leu, Li, Yao, & Wu, 2006; Yokozawa et al., 1998; Zhao, Dou, Wu, & Aisa, 2013). Thus, the consumption of mallow included in food products has the potential to confer antioxidant properties with health benefits. The identified triterpenoids from *M. aegyptiaca* leaves were malvasterone, 5α-

Table 2
Liquid Chromatography-High Resolution Electrospray Ionization Mass Spectrometry (LC-HRESIMS) analysis of the ethanolic extract of *M. aegyptiaca* leaves.

Suggested compounds[a]	Accurate mass	Molecular formula[b]
Flavonoids		
Isorhamnetin	317.06418	$C_{16}H_{12}O_7$
Isorhamnetin-7-*O*-glucoside	479.11646	$C_{22}H_{22}O_{12}$
Quercetin-3-*O*-rutinoside	611.16096	$C_{27}H_{30}O_{16}$
Quercetin-3-*O*-glucoside	465.10235	$C_{21}H_{20}O_{12}$
Kaempferol 3-*O*-glucoside	449.10794	$C_{21}H_{20}O_{11}$
Kaempferol-3-*O*-rutinoside	595.16545	$C_{27}H_{30}O_{15}$
Malvidin-3-*O*-glucoside	493.13445	$C_{23}H_{25}O_{12}$
Triterpenoides		
Picrasinoside F	557.29486	$C_{28}H_{44}O_{11}$
Dichrostachine F	621.30524	$C_{36}H_{44}O_9$
Lupenone	425.37749	$C_{30}H_{48}O$
Lupenol	427.39324	$C_{30}H_{50}O$
5α-Stigmast-9(11)-en-3β-ol	415.39324	$C_{29}H_{50}O$
Malvasterone	413.37739	$C_{29}H_{48}O$
Fatty acids		
Malvalic acid	281.24758	$C_{18}H_{32}O_2$
Linolenic Acid	279.23176	$C_{18}H_{30}O_2$
Palmitic acid	257.24781	$C_{16}H_{32}O_2$

[a] The suggested compounds was according to Dictionary of Natural Products (DNP 23.1, 2015 on DVD) and characteristic fragmentation pattern.
[b] The formulas deduction was from the quasimolecular ion peak $[M+H]^+$. Three samples were analyzed ($n = 3$).

stigmast-9(11)-en-3β-ol, lupenone, lupenol, picrasinoside F and dichrostachine F (Table 2). During the last years, there has been an unprecedented escalation of interest in triterpenoids, also known as phytosterols, due to their cholesterol-lowering properties (Saleem, 2009). The sterol malvasterone (5α-stigmast-9(11)-en-3-one) is reported for the first time from ethanolic extract of the roots of *Malva parviflora* (Sharma & Ali, 1999). Besides, 5α-stigmast-9(11)-en-3β-ol is also a sterol isolated from *Costus speciosus* roots (Gupta, Lal, & Shukla, 1981). Lupenone and lupenol are pentacyclic triterpenoids. Lupenol was reported as a therapeutic and chemo-preventive agent for the treatment of inflammation and cancer (Saleem, 2009). Picrasinoside F is a quassinoid glucoside, which represented highly oxygenated triterpene and dichrostachine F represented a meroterpene derivative or terpenophenolic compound. Dichrostachines A-R were isolated from the root and stem barks of *Dichrostachys cinerea* and most of the compounds inhibited the enzyme protein farnesyl transferase, which suggest their use as potential anticancer drugs (Long et al., 2009).

3.2. Alveographic characteristics of dough

The mallow powder contained a relatively high content of dietary fiber so that its incorporation into wheat flour could modify the rheological parameters of dough and therefore the physical properties of bread. Fig. 1 shows that a partial substitution of wheat flour by the mallow powder induced important modifications on the rheological characteristics of the dough (P, L and W). In fact, when the substitution level increased, the tenacity of the dough (P) significantly ($p < 0.05$) rose from 82.33 mm H_2O (control sample) to 127 mm H_2O at 5% substitution level (F4), which might be a consequence of poor hydration of gluten. Nevertheless, a significant ($p < 0.05$) decrease in the extensibility (L) and the deformation energy (W) was observed, indicating that high substitution level opposed to the dough development. By comparing the control sample and formulation 4, (L) and (W) values decreased from 68 mm to 35 mm and from 193.66 10^{-4} J to 170.66 10^{-4} J, respectively. The (P/L) ratio gave information about the elastic resistance and extensibility balance of flour dough. The observed effect on (P) and (L) parameters became evident in the (P/L) ratio whose increase indicated an inextensible dough (Fig. 1). Our findings recall those reported by Wang, Rosell, Benedito, and de Barber (2002) and Borchani et al. (2011), who showed an increase in (P/L) ratio and a reduction in deformation energy (W) of the dough containing pea and date flesh fiber, respectively. The obtained results could be mainly explained by the high water retention capacity of mallow powder (511 g/100 g DM), which could prevent an optimal hydration of gluten. Furthermore, interactions between fibrous material and gluten could affect the gluten network structure, which was responsible for the dough quality. Consequently, gluten proteins were not optimally developed in order to form a viscoelastic network able to retain the fermentation gas.

3.3. Effects of mallow powder enrichment on bread quality

3.3.1. Instrumental texture, specific volume and colour properties

Data on instrumental texture and specific volume of bread were presented in Table 3. A significant ($p < 0.05$) decrease in bread specific volume was noted at the highest substitution level (F4), which was expected since the extensibility (L) of the dough was considerably reduced. Similar studies also reported a significant ($p < 0.05$) reduction in specific volume of bread enriched by 2–8% turmeric powder or 3–9% lemon fiber (Fu, Chang, & Shiau, 2015; Lim, Park, Ghafoor, Hwang, & Park, 2011). However, Borchani et al. (2011) showed that specific bread volume was not significantly ($p > 0.05$) affected by 3% supplementation of date flesh fiber. Table 3 also shows that bread enrichment with mallow powder resulted in a significant increase ($p < 0.05$) of hardness and chewiness of bread as well as a significant decrease ($p < 0.05$) of its springiness. Obtained results were also comparable to the studies on bread supplemented with lemon fiber or turmeric powder (Fu et al., 2015; Lim et al., 2011). Thus, the densification of these breads could be mainly attributed to the interactions between gluten and fibrous material. In fact, dietary fiber contained in mallow powder could alter the formation of a continuous gluten network and exhibited a destabilizing effect at the interfaces of gas cells of the dough (Noort, Van Haaster, Hemery, Schols, & Hamer, 2010). The colour of bread crust is an important parameter to determine its acceptability. Therefore, the effects of mallow powder addition on the bread crust colour were presented in Table 4. The prepared mallow powder showed a high lightness value (L^*: 55.38) and a low intensity of yellow (b^*: 13.39) and green (a^*: −7.02) colours (Table 4). Mallow powder has comparable colour parameters with cladodes powder from prickly pear, which was used as a functional ingredient in cookies formulation (Msaddak et al., 2015). Table 4 shows that there was a significant ($p < 0.05$) difference between control bread and samples enriched with mallow powder. In fact, mallow-enriched bread was found to be darker than the control, as shown by lower L^* value. Indeed, a decrease in L^* value from 64.75 (control product) to 46.31 in bread prepared with 5% level of mallow powder was observed. In addition, a^* and b^* values decreased with increasing the substitution level, which indicate that yellow colour declined toward a greenish colour. This could be explained mainly by the richness of mallow powder in chlorophyll (Table 1) and to other pigments such as the anthocyanin malvidin-3-*O*-glucoside (Table 2).

Although addition of mallow powder resulted in a certain variation in hardness and colour of bread, the mallow-enriched bread could provide added nutritional value, resulted in its enrichment with beneficial phytochemicals. Therefore, we also investigated flavonoids content, antioxidant activity and sensory evaluation of formulated bread.

3.3.2. Flavonoids content and antioxidant activity

In order to evaluate the mallow powder contribution to the

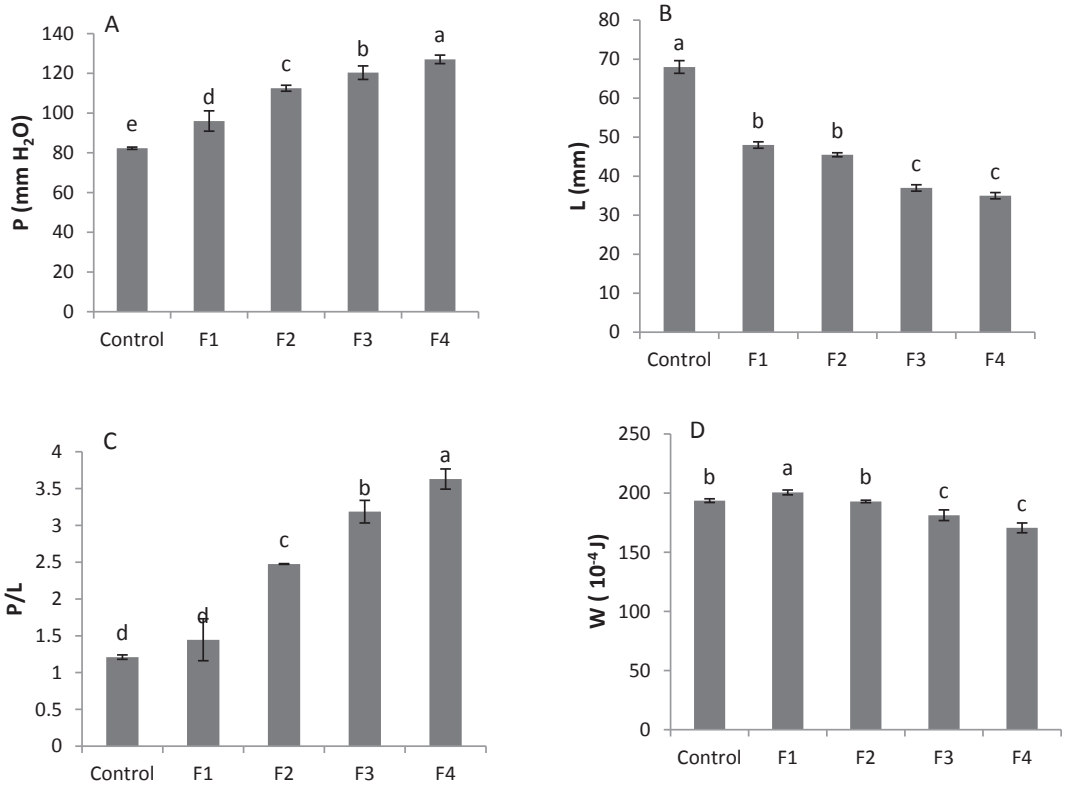

Fig. 1. Effect of *M. aegyptiaca* leaves on the tenacity P (A), extensibility L (B), P/L ratio (C) and deformation energy W (D) of dough.
[a,b,c,d,e]Different letters above the bars indicate significant differences ($p < 0.05$).

Table 3
Effect of mallow powder on the texture and specific volume of bread.

Substitution level (g/100 g of wheat flour)	Hardness (N)	Springiness (mm)	Chewiness (N × mm)	Specific volume (cm^3/g)
0 (control)	6.22 ± 0.29^{d}	3.62 ± 0.07^{a}	8.60 ± 0.44^{c}	1.58 ± 0.09^{a}
1 (F1)	7.09 ± 0.38^{c}	2.07 ± 0.01^{b}	7.93 ± 0.64^{c}	1.60 ± 0.12^{a}
2 (F2)	7.88 ± 0.49^{c}	1.99 ± 0.01^{b}	8.39 ± 0.68^{c}	1.45 ± 0.06^{a}
3 (F3)	10.25 ± 0.26^{b}	1.98 ± 0.03^{b}	11.29 ± 0.71^{b}	1.49 ± 0.01^{a}
5 (F4)	17.91 ± 1.64^{a}	1.83 ± 0.01^{c}	17.75 ± 1.25^{a}	1.32 ± 0.01^{b}

Data presented as the mean ± standard deviation ($n = 3$).
[a,b,c,d] Values with same superscript letters in the same row are non-significant at $p < 0.05$.

Table 4
Colour characteristics of crust of the bread enriched with mallow powder.

Substitution level (g/100 g of wheat flour)	L*	a*	b*
Mallow powder	55.38 ± 0.40	-7.02 ± 0.11	13.39 ± 0.36
0 (control)	64.75 ± 0.04^{a}	-1.84 ± 0.17^{a}	20.37 ± 0.05^{b}
1 (F1)	52.91 ± 0.07^{b}	-1.92 ± 0.02^{a}	14.60 ± 0.09^{a}
2 (F2)	50.62 ± 0.21^{c}	-2.14 ± 0.33^{b}	12.15 ± 0.27^{c}
3 (F3)	48.36 ± 0.61^{d}	-3.93 ± 0.02^{c}	9.10 ± 0.11^{d}
5 (F4)	46.31 ± 0.45^{d}	-4.97 ± 0.19^{d}	5.80 ± 0.06^{e}

Data presented as the mean ± standard deviation ($n = 3$).
[a,b,c,d,e] Values with same superscript letters in the same row are non-significant at $p < 0.05$.

antioxidant properties of the resulting bread, flavonoids content, DPPH· radical-scavenging activity and ferric reducing power were determined (Table 5). Control bread showed the lowest flavonoids content (4.5 mg QE/100 g of bread), DPPH· radical-scavenging activity (5.76%) and reducing power ($A_{700} = 0.36$). Interestingly, mallow powder addition strongly influenced the flavonoids content and the antioxidant activity of bread and the increment of the mallow level substitution further enhanced the antioxidant capacity of the bread. In fact, Table 5 shows that at 5% level of mallow powder substitution, bread exhibited the highest flavonoids content (65.6 mg GAE/100 g of bread), DPPH· radical-scavenging activity (90.3%) and reducing power ($A_{700} = 0.94$). The flavonoids and triterpenoids identified in *M. aegyptiaca* leaves (Table 2) may contribute individually or synergistically to the antioxidant activity observed in the bread enriched with the mallow powder. Similar studies reported that the fortification of

Table 5
Flavonoids and antioxidant activity of bread enriched with mallow powder.

Substitution level (g/100 g of wheat flour)	Flavonoids	Scavenging activity	Reducing power
0 (control)	4.50 ± 1.40^{d}	5.76 ± 1.92^{d}	0.36 ± 0.01^{e}
1 (F1)	18.72 ± 2.72^{c}	49.99 ± 2.0^{c}	0.48 ± 0.01^{d}
2 (F2)	26.88 ± 1.92^{b}	68.26 ± 4.80^{b}	0.57 ± 0.02^{c}
3 (F3)	57.60 ± 3.20^{a}	84.61 ± 2.0^{a}	0.73 ± 0.02^{b}
5 (F4)	65.60 ± 1.60^{a}	90.30 ± 2.25^{a}	0.94 ± 0.03^{a}

Data presented as the mean ± standard deviation ($n = 3$); ^{a,b,c,d,e} Values with same superscript letters in the same column are non-significant at $p < 0.05$; DPPH· radical-scavenging activity (%) and reducing power (A_{700}, absorbance at 700 nm) were determined at 0.5 g of bread/ml of ethanol; Flavonoids are expressed as mg quercetin equivalents (QE)/100 g of bread.

wheat bread with natural raw materials such as grape seed extract, pomegranate peels powder or onion skin enhanced its antioxidant capacity (Altunkaya, Hedegaard, Brimer, Gökmen, & Skibsted, 2013; Gawlik-Dziki et al., 2013; Peng et al., 2010). However, Peng et al. (2010) demonstrated that baking declined by about 30–40% antioxidant activity of bread enriched with grape seed extract. Besides, Holtekjølen, Baevre, Rødbotten, Berg, & Knutsen, (2008) showed that the amount of free phenolics decreased by up to 23.5% during the baking process, while bound phenolics obviously increased up to 6 fold. In general it is accepted that phenolic compounds retain the main of their antioxidant activity after the baking process, which has potential health benefits for consumers (Dziki et al., 2014).

The obtained results suggest that the antioxidant potential of mallow-supplemented bread reinforced their nutritional quality. Thus, bread enrichment with mallow seems to be a very easy and cheap way for improving food quality. However, a compromise between nutritional value and sensory quality should be established, since taste, flavour and texture strongly influenced consumer preferences towards such products.

3.3.3. *Sensory properties*

Sensory analysis was carried out by checking colour, odour, taste, texture and overall acceptability of fresh prepared breads. The current study highlighted significant effect ($p < 0.05$) of mallow powder supplementation on different sensory attributes (Fig. 2). The colour of bread crust is an important parameter to determine its acceptability. The mallow powder gave greenish colour characteristic to finished products, which could be explained mainly by its richness in chlorophyll and other pigments. The highest colour scores were observed in control and then they decreased when the incorporation of mallow powder increased. Likewise, the odour and taste results also showed that there was a decreasing trend of the averages of scores when the incorporation of mallow powder increased. Textural hardness is another important characteristic of bread, since a too hard structure could have a negative effect on sensory quality of the product. Fig. 2 shows a decline in texture score (3.95 in F4 *vs* 6.92 in control), which was expected as was analyzed by the alveographic and the instrumental texture assays. In terms of overall acceptability of the enriched bread, the product containing 3% of mallow powder (F3) remained acceptable since the obtained mean score for the overall acceptability was 5.19 (Fig. 2). A survey of the literature showed that bread enrichment up to 3–5% of functional ingredients (eg. onion skin, green coffee, turmeric powder) gave satisfactory consumer acceptability (Dziki et al., 2014).

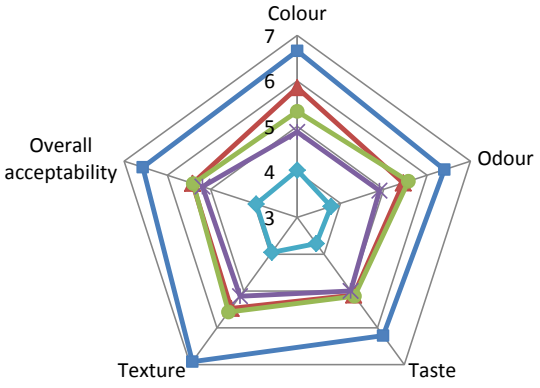

Fig. 2. Sensory evaluation of breads prepared with wheat flour fortified with *M. aegyptiaca* leaves. F1, F2, F3 and F4 represented the formulations containing 1, 2, 3 and 5% of mallow powder, respectively. The control represented the product without enrichment. —control, —F1, —F2, —F3, —F4.

4. Conclusions

Consumption of antioxidant-rich food, in the context of a balanced diet, is associated with the prevention of many degenerative diseases. Consequently, improving the functionality and the sensory properties of traditional food such as bakery products is an interesting approach with respect to the actual consumers demand. The mallow (*Malva aegyptiaca*) was an important medicinal herb and functional food, since it showed richness in bioactive compounds and presented important biological properties. At 3% level substitution, mallow could be used as a functional ingredient to enhance the nutraceutical potential in terms of flavonoids content and antioxidant capacity of wheat bread without altering its sensory acceptability.

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