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Antioxidant activities of chicory (*Cichorium intybus* L.) and purslane (*Portulaca oleracea* L.) leaves powder and their applications for preservation of cupcakes

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PAPER

Abstract

Since the advantages of a healthy diet on one's quality of life are widely recognized, the food industry must come up with new food products that have particular health-improving qualities. Using water and ethanolic extraction, the current study assessed the antioxidant capacities of purslane and chicory leaves extracts. The study assessed how MCI (*Chicory* (*Cichorium intybus*) leaf powder) (cupcake flour replaced with 5% and 10% chicory leaf powder) and MPO (*Purslane (Portulaca oleracea)* leaf powder) (cupcake flour replaced with 5% and 10% purslane leaf powder) affected the physical traits, textural qualities, nutritional content, lipid oxidation, and consumer acceptability of cupcakes. Total phenols, flavonoids, and 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity were evaluated. The texture, color, and nutritional content of cupcake samples from MCI and MPO were assessed and compared to control samples. According to the findings, raising the MPO and MCI by 10% reduced the specific volume, yellowness, and lightness of crumb and crust while enhancing the redness. After the storage period, both cakes (MCI and MPO) had higher nutritional values than the control. A nine-point hedonic scale was used to rate the acceptability of cupcakes. As MPO and MCI concentrations increased, darker colors predominated ($p < 0.05$). However, the overall consumer acceptance of cupcake samples declined with increase in MCI and MPO concentrations, and compared to MCI cupcake samples, MPO cupcake samples were substantially more acceptable ($p < 0.05$). The results of sensory evaluation also showed that the sensory qualities of the product did not change significantly by the addition of up to 5% of MPO and MCI to cupcake recipe. Hence, up to 5% MPO and MCI can be used as a flour substitute in cupcake recipes to increase the nutritional value and functioning of cakes without degrading the quality.

Keywords: acceptability; chicory; cupcakes; fortification; leaf powder; nutritional composition; purslane

Introduction

As the benefits of a balanced diet on the quality of life are widely recognized, there is an increasing demand for healthy food options. This increasing demand has put significant pressure on the food industry to deliver nourishing, convenient, and health-promoting products. One of the critical components of a healthy diet is antioxidant activity, which plays a fundamental role in dietary function. In response to aging population, rising healthcare costs, growing consumer interest in nutrition, and advancements in food technology, foods with high antioxidant activity are developed (Al Jumayi *et al.*, 2022). These multifunctional foods, rich in nutraceutical ingredients, are often derived from various plant sources and offer numerous health benefits. Dietary fiber, in particular, is well known for its vital role in maintaining optimal health (Lee and Lucey, 2004).

Baked goods are globally enjoyed and serve as an ideal medium for incorporating bioactive compounds and dietary fibers due to their widespread consumption (Astrup and Monteiro, 2022; Kadam and Prabhasankar, 2010; Siddiqui *et al.*, 2022). Numerous studies have explored the enrichment of wheat-based cereal products with dietary fiber, showcasing its potential to improve nutritional profiles (Santos *et al.*, 2022; Wiedemair *et al.*, 2022). The inclusion of dietary fiber, alongside enhanced nutritional and functional properties, is therefore essential for developing innovative fiber-enriched foods like baked goods.

Chicory (*Cichorium intybus* L.) stands out as a nutrient-dense ingredient, offering carbohydrates, essential fatty acids, amino acids, and a spectrum of vitamins, including A, B6, K, tocopherol, carotene, and zeaxanthin. Additionally, chicory is a rich source of critical minerals such as phosphorus (P), potassium (K), zinc (Zn), copper (Cu), and iron (Fe) (Das *et al.*, 2016; Jancic *et al.*, 2017; Perović *et al.*, 2021). Its leaves are particularly abundant in phenolics and total flavonoids (Rohman and Man, 2010).

Moreover, chicory contains diverse bioactive compounds like inulin, sesquiterpene lactones, caffeic acid derivatives, hydroxycoumarins, flavonoids, alkaloids, steroids, terpenoids, and volatile compounds. These compounds collectively contribute to its notable physiological properties, including antioxidant and antimicrobial effects (Denev *et al.*, 2014; Peña-Espinoza *et al.*, 2018; Perović *et al.*, 2021).

Similarly, purslane (*Portulaca oleracea* L.) contains a wide variety of nutraceutical components, such as flavonoids, alkaloids, terpenoids, organic acids, essential fatty acids, polysaccharides, vitamins, sterols, proteins, and minerals (Kumar *et al.*, 2021; Petropoulos *et al.*, 2016). Purslane is particularly notable for its superior nutritional content, compared to other green leafy vegetables (Srivastava *et al.*, 2021).

The current pharmacological studies have shown that *P. oleracea* has several bioactive properties, such as antibacterial and antioxidant properties (Kumar, *et al.*, 2022). *P. oleracea* extracts have hepatoprotective, neuroprotective, and antimicrobial properties (Chugh *et al.*, 2019). Cakes, being a popular snack, serve as an effective vehicle for delivering phenolic antioxidants and functional plant-based compounds. High concentrations of bioactive chemicals in edible plants enhance the overall quality of various food products, including processed wheat items, by minimizing chemical reactions and inhibiting microbial growth (Admassu and Kebede, 2019b; Javanmardi *et al.*, 2019; Li *et al.*, 2014). Despite the potential benefits, there is a notable lack of scientific literature regarding the bioactive compounds in *C. intybus* and *P. oleracea*. Therefore, this study aims to evaluate the antioxidant activities of graded amounts of chicory (*C. intybus* L.) and purslane (*P. oleracea* L.) powder in cupcakes, focusing on their effects on the products' appearance, texture, flavor, and the overall acceptability.

Materials and Methods

Plan of study

An overview of this study's methodology is presented in Figure 1.

Plant collection and identification

Chicory (*C. intybus* L.) and purslane (*P. oleracea* L.) were harvested from a nearby field in Menoufia, Egypt. Botanists from the Department of Botany at Menoufia University's Faculty of Agriculture identified plant leaves. By following the methods outlined by Wadood *et al.* (2013), fresh leaves were thoroughly washed, dried, and crushed.

Plant extracts

Weighing 50 g of each plant powder, 1 L of each of the following was added, resulting in extracts with various concentrations of each plant powder:

- Water (100% concentration);
- mixture of ethanol and water (25:75, v/v);
- mixture of ethanol and water (50:50, v/v);
- ethanol (100% concentration).

Then, using an overhead stirrer, the stirring process was carried out for 12 h at room temperature (25°C) to obtain plant extracts, which were then processed into powders according to the method demonstrated by Al Jumayi *et al.* (2022).

Total phenolic content (TPC), total flavonoid content (TFC), and antioxidant activity of plant extracts

Total phenolic content

In order to determine TPC by using the Folin–Ciocalteu method, each plant extract was diluted with 70% (v/v)

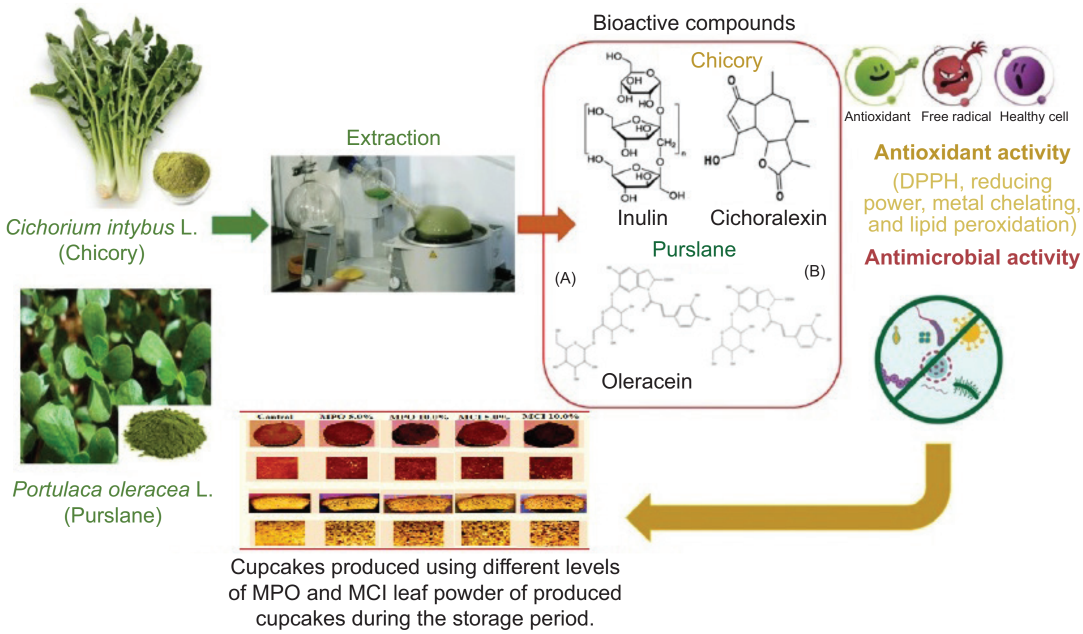

Figure 1. Conceptual framework of the methodology.

ethanol (Pangestuti *et al.*, 2019). For every 100 mL of the diluted sample and gallic acid standard solution, 2 mL of 2% Na_2CO_3 was added. After allowing the mixtures to settle down for 5 min at 22°C, 100 mL of Folin–Ciocalteu reagent was added, and the mixtures were mixed thoroughly in the dark. After an incubation period of 30 min, absorbance of the solutions was measured at 760 nm using a spectrophotometer (SCHOTT Instruments, UviLine 9400, EU). Phenolic content in dried leaves was expressed in milligrams of gallic acid equivalents (mg GAEs).

Total flavonoid content

Total flavonoid content was calculated using the method as described by Ghane *et al.* (2018). In a 10-mL volumetric flask, 0.5 mL of each plant extract, 5 mL of distilled water, and 0.3 mL of sodium nitrite ($NaNO_2$) were combined in a 1:20 ratio. After 5 min, 3 mL of aluminum chloride (1:10) was added. After 6 min, 2 mL of 4% NaOH was added, and the final volume was adjusted to 10 mL with distilled water. The absorbance of the prepared solutions was measured at 510 nm using a spectrophotometer (SCHOTT Instruments) against a blank. TFC was expressed as milligrams of quercetin equivalents (mg QEs) per gram of dried leaves.

DPPH radical scavenging activity

The free radical scavenging activity was measured with DPPH radical assay, as described by Ebrahimzadeh *et al.* (2016). Different concentrations of plant extract (100, 250, 500, 750, and 1,000 mg/L [w/v]) were individually mixed with 2 mL of 100-μM DPPH solution. Then the absorbance was measured at 517 nm using a spectrophotometer (SCHOTT Instruments). The results were expressed as half-maximal inhibitory concentration (IC50) values, which represent the amount of plant extract required to scavenge 50% of DPPH radicals.

Active compounds by gas chromatography–mass spectrometry (GC-MS)

Chromatographic analyses of the samples were performed with an Agilent system gas chromatograph and Agilent 7683 mass spectrometer detector. Samples were measured by using a column of HP-5 Ms. Helium (99.999%) was used as gas flow at a rate of 1.0 mL/min, and the injection volume was 1 L. Initial column temperatures were maintained at 90°C for three cycles, then increased to 230°C in 15°C/min increments, and finally from 230 to 310°C, with a 5-min maximum holding period. The Spectroscopic Library of the United States (NIST) was also used as a source of information. the energy of electron ionization was 65 eV or such value. Three copies of each study were prepared.

Preparation of cupcakes

Raw materials

Baking powder, vanilla, sugar, oil, and plastic bags were obtained from a local market in Shibin El Kom, Menoufia, Egypt. Wheat flour was bought from a confectionery. In order to maintain product's freshness, eggs were purchased daily. In Shibin El Kom, Menoufia, Egypt, we bought leaf powder of *C. intybus* and *P. oleracea* from a local farmer. A conventional cupcake recipe was used to make cupcakes (Kim and Shin, 2014). The sugar batter method was used to prepare cupcake batter in a Kenwood mixer (Model KM 400; Kenwood, UK). In a cupcake, 5% and 10% substitution levels of *C. intybus* and *P. oleracea* leaves powder were used to replace partially heated flour. Standard cupcake varieties were used as controls (0%).

Treatments

Five cupcake formulations were prepared as follows:

- **Control**: Cupcake formula without antioxidant or antibacterial agents and no addition of *C. intybus* or *P. oleracea* leaf powders.
- **MPO 5.0%**: Cupcake formula with 5% substitution of *P. oleracea* (purslane) leaf powder in place of flour.
- **MPO 10.0%**: Cupcake formula with 10% substitution of *P. oleracea* (purslane) leaf powder in place of flour.
- **MCI 5.0%**: Cupcake formula with 5% substitution of *C. intybus* (chicory) leaf powder in place of flour.
- **MCI 10.0%**: Cupcake formula with 10% substitution of *C. intybus* (chicory) leaf powder in place of flour.

The concentrations of *C. intybus* and *P. oleracea* were selected based on the earlier experiments conducted in our laboratory. A homogenous mixture was produced by combining manually each formulation. Shortening and creamed flour were combined with foam made of sugar–egg, which was then combined with vegetable oil. Shortening and sugar were creamed in a mixer for 10 min at 180 rpm. The eggs were stirred for 6 min after addition. For each type of cupcake, 12 batters each weighing 50 g were placed in paper baking cups in an aluminum muffin pan. The cupcakes took 30 min to bake in an oven at 180°C (Thermawatt TG103, Athens, Greece). After being brought to room temperature, cupcakes were wrapped in polyethylene film and kept at 4°C for 16 days. After preparation of cupcakes, their properties were evaluated. Sensory analysis was conducted after microbial characterization of the product.

Color measurement

Instrumental color (L*, a*, and b*) analysis of samples was conducted using a scale color spectrophotometer (machine colors Tristimulus) with CIE Lab colorimeter (Hunter, Lab Scan XEReston, VA, USA) according to the method described by Al Jumayi *et al.* (2022).

Nutritional Composition

Methods provided by Association of Official Analytical Chemists (Method 990.03; AOAC, 2005) were used to determine the chemical composition of cupcakes, including those for moisture, total ash, crude fiber, total fat, and total nitrogen. N-free extract was estimated by difference, and protein was determined as N 5.7. Zinc (Zn), calcium (Ca), and iron (Fe) were estimated using the official method of AOAC. Samples were dried for 2 h before being washed in a furnace at around 510°C. The same process was carried out twice to extract minerals into the solution after adding HCl to ash and boiling it in a water bath until dry. The contents of Zn, Ca, and Fe were calculated using an atomic absorption spectrophotometer (Model UV-VIS-2802PC; Shimadzu, Kyoto, Japan).

Assessment of lipid oxidation (thiobarbituric acid reactive substances [TBARS])

According to Peretti *et al.* (2006), the TBARS values (mg of malonaldehyde per kg of oil sample) of cupcake samples were calculated spectrophotometrically. In order to measure absorbance, a spectrophotometer was used at 530 nm (model UV-VIS-2802PC; USA).

Sensory evaluation

Fifty-five students and faculty members of Department of Food Science and Technology, Faculty of Agriculture, Menoufia University were recruited as participants in the study. Pilot research with 10 participants was conducted before the main trial to determine whether the recipes were acceptable and to adjust methodology as necessary. In the pilot trial, replacements of 0%, 5%, 10%, 15%, and 20% (w/w) were used; however, 15% and 20% replacements were not well-received. As a result, the primary research changed the substitution levels for *P. oleracea* and *C. intybus* to 0%, 5%, and 10% (w/w). The main trial was closed to the participants who participated in the pilot study. To stop participants from talking to each other, they were divided into different cubicles. Each of the six samples received a distinct three-digit code from a database of random numbers, and the serving order was determined by tables of random permutations of nine. On a hedonic scale of 1–9 points (1: dislike very much, and 9: like), 45 regular customers (63% females, 37% males, aged 18–50 years) evaluated cupcake samples for their organoleptic qualities (taste, flavor, odor, texture, and the overall acceptability). Purchasing intention (1: definitely wouldn't buy, and 5: definitely would buy) was evaluated on a hedonic scale of 1–5 points. The acceptability index (AI) was calculated by multiplying the average and maximum scores on a hedonic scale (1–9 points) by 100. Each participant got around half a slice from each of the six cupcake samples on a plastic plate. Each participant received a cup of water to cleanse their mouth in-between tasting of each cupcake sample and answering the questions related to sensory evaluation. To avoid bias, participants only received a restricted amount of information about *C. intybus* and *P. oleracea*. If necessary, research assistants helped panelists during sensory assessment.

Statistical analysis

Suitable statistical techniques, such as the Bonferroni test and Tukey's test, were used to assess the data. The production optimization of the MPO and MCI-enriched cakes was carried out by utilizing the Design-Expert software 11.1.2.0. The experiment was conducted thrice, and each time three measurements were reproduced. The effect was determined as statistically significant by using the least significant difference ($p < 0.05$) technique. A two-way analysis of variance was carried out for statistical comparisons involving different variables. The significance of group differences was assessed using Duncan's analysis ($p < 0.05$). The central composite design (CCD) was employed for designing the experiments. MPO and MCI concentrations (X1) and storage time (X2) at three distinct coded levels—low (1), medium (0), and high (+1)—were examined as independent variables.

Results

Total phenolic content and total flavonoid content

Figures 2 and 3 display TPC and TFC results, respectively. The TPC and TFC values of each plant extract changed considerably ($p < 0.05$) between 100% ethanol and 100% aqueous extracts. The TPC values of *C. intybus*'s and *P. oleracea*'s 100% water extracts were 48.55 mg GAE/g dry samples and 47.34 mg GAE/g dry samples, respectively. The TPC values of *C. intybus*'s and *P. oleracea*'s 100% ethanolic extracts were 47.29 mg GAE/g dry samples and 47.02 mg GAE/g dry samples, respectively. Regarding TFC, 0.63 mg QE/g sample and 0.70 mg QE/g sample were present in the aquatic extracts (100% water) of *C. intybus* and *P. oleracea*, respectively, while *C. intybus*'s and *P. oleracea*'s extracts with 100% ethanol had TFC values of 0.46 mg QE/g sample and 0.50 mg QE/g sample, respectively.

Concentrations of phenolic acids in chicory and purslane extracts

Table 1 lists phenolic acids quantified by high-performance liquid chromatography (HPLC) in purslane and chicory extracts. A significant proportion of phenolic acids, 90.6% in chicory and 86.5% in purslane, was found to be bounded. The primary phenolic acids in chicory, with contents exceeding 100 μg/g, included ferulic acid, p-coumaric acid, protocatechuic acid, catechin hydrate, p-hydroxybenzoic acid, and gallic acid. In purslane, the dominant phenolic acids were p-coumaric acid, ferulic acid, vanillic acid, chlorogenic acid, protocatechuic acid, catechin hydrate, naringin, and syringic acid. Various studies reported individual and total phenolic acid contents in

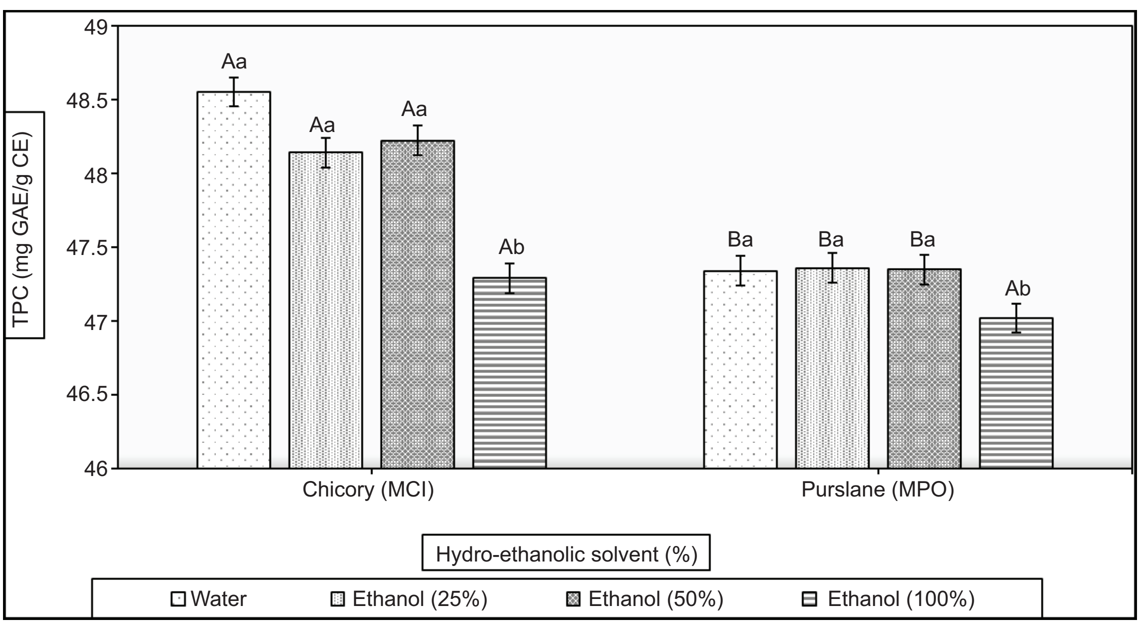

Figure 2. TPC values of different hydroethanolic extracts were obtained for chicory and purslane leaves. Results are expressed as mean values ± standard deviation and significance ($p \leq 0.05$). All measurements were done in triplicate. Histograms with different lowercase letters indicate a statistically significant difference ($p \leq 0.05$) in TPC because of different extraction conditions, while different capital letters indicate a statistically significant difference ($p \leq 0.05$) between chicory and purslane for the same extraction condition. TPC: total phenolic content; GAE: gallic acid equivalent; CE: crude extracts.

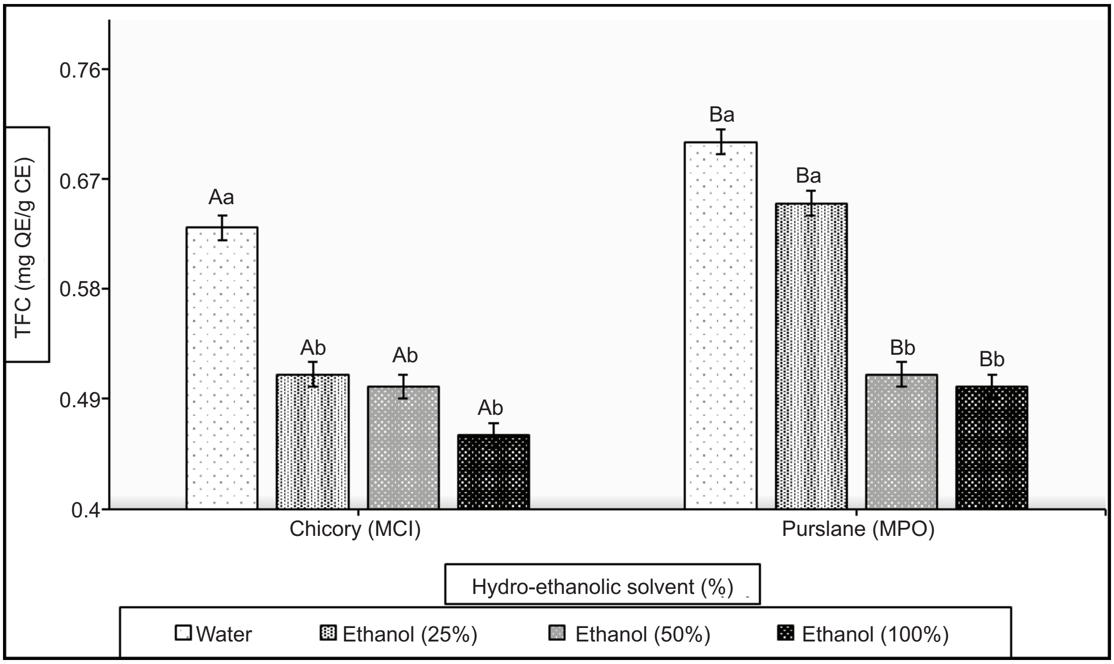

Figure 3. TFC values of different hydroethanolic extracts were obtained for chicory and purslane leaves. All measurements were done in triplicate (±SD). Different lowercase letters indicate a statistically significant difference ($p \leq 0.05$) in TPC because of different extraction conditions for the same plant. Different uppercase letters indicate a statistically significant difference ($p \leq 0.05$) in TPC between chicory and purslane for the same extraction condition. TFC: total flavonoid content; QE: quercetin equivalent.

chicory and purslane extracts; however, these values varied significantly due to differences in environmental factors, analytical techniques, and extraction methods (Dykes and Rooney, 2007; Fernandez-Orozco *et al.*, 2010; Janda *et al.*, 2021b; Menga *et al.*, 2010; Stalikas, 2007).

Chemical composition of cupcake samples

Table 2 illustrates the effects of various treatments and storage times on the chemical composition of MPO and MCI cupcakes. The results indicated that the addition of MPO and MCI leaves powder significantly altered the moisture, lipid, ash, fiber, and protein contents of cupcakes. The control samples exhibited the lowest concentrations: moisture (3.28 ± 0.09%), fat (23.27 ± 0.02%), ash (0.45 ± 0.008%), fiber (0.14 ± 0.006%), and protein (6.59 ± 0.02%). Conversely, the MPO 10% cupcakes (with 10% *P. oleracea* L. leaf powder) showed the highest levels of moisture (3.83 ± 0.04%), fat (23.33 ± 0.01%), ash (0.51 ± 0.010%), fiber (0.72 ± 0.015%), and protein (8.09 ± 0.02%). The MCI 10% cupcakes (with 10% *C. intybus* leaf powder) displayed moisture (3.54 ± 0.03%), fat (23.30 ± 0.02%), and ash (0.48 ± 0.01%). Regarding nitrogen-free extract (NFE), the control samples had the highest concentration of 66.28 ± 0.10%, while the MPO and MCI 10% samples had the lowest concentrations at 63.52 ± 0.05% and 64.90 ± 0.01%, respectively.

Color

The L*, a*, and b* values, representing lightness, redness, and yellowness, respectively, were used to characterize the color properties of cupcakes (Table 3). Color tests indicated no significant differences between cupcakes containing 5% and 10% of *C. intybus* and *P. oleracea*, compared to the control samples ($p > 0.05$). However, addition of *C. intybus* and *P. oleracea* powder significantly decreases the lightness ($p < 0.05$). Cupcakes with *C. intybus* were darker than those with *P. oleracea* (Table 3). Additionally, the texture of cupcakes remained unaffected by the addition of *C. intybus* and *P. oleracea* powder at various substitution levels. The substitution of MPO and MCI leaves powder for flour considerably ($p \leq 0.05$) changed the color of cake samples' crust and crumbs (Figures 4A and 4B). The L*, a*, and b* values of the crust decreased with increasing MPO and MCI levels, indicating darker crusts. However, the a* value of the crumb increased, signifying enhanced redness, while the L* and b* values of the crumb decreased, resulting in darker and less yellow crumbs. The crust and crumb of the cupcake samples followed this trend, becoming darker with higher levels of MPO and MCI. The control sample exhibited a light brown color, whereas the MPO and MCI leaf powders were darker in color. Consequently, incorporating MPO and MCI leaf powders into the cupcake formulations resulted in darker crusts and crumbs.

Table 1. Free, bound, and total phenolic acids in chicory and purslane extracts (µg/g, dry basis).

Sample	p-Coumaric acid	Ferulic acid	p-Hydroxybenzoic acid	Vanillic acid	Caffeic acid	Chlorogenic acid	Protocatechuic acid	Gallic acid	Catechin hydrate	Naringin	Syringic acid	Total phenolics
Chicory (*C. intybus*)												
Free	5.0^{a}	20.3^{a}	1.0^{a}	ND	4.8^{a}	ND	72.7^{a}	ND	288.2^{a}	ND	12.6^{a}	404.5^{a}
Total	258.5^{a}	463.4^{a}	7.4^{a}	15.4^{a}	9.2^{a}	ND	126.5^{a}	123.4^{a}	324.8^{a}	35.0^{a}	21.6^{a}	1,285.1^{a}
Bound*	258.5^{a}	483.7^{a}	7.4^{a}	15.4^{a}	13.9^{a}	ND	199.1^{a}	123.4^{a}	613.0^{a}	35.0^{a}	34.1^{a}	1,783.6^{a}
Purslane (*P. oleracea*)												
Free	15.5^{b}	37.5^{b}	ND	5.2^{b}	ND	17.92^{b}	20.1^{b}	ND	155.3^{b}	42.9^{b}	21.5^{b}	315.7^{b}
Total	568.5^{b}	228.1^{b}	11.6^{b}	10.2^{b}	24.4^{b}	59.99^{b}	27.0^{b}	116.5^{b}	393.0^{b}	489.3^{b}	86.8^{b}	2,015.3^{b}
Bound	584.0^{b}	265.5^{b}	11.6^{b}	15.4^{a}	24.4^{b}	77.91^{b}	47.0^{b}	116.5^{b}	548.2^{b}	532.2^{b}	108.4^{b}	2,331.0^{b}

Different superscripted lowercase letters (e.g., "a" for chicory and "b" for purslane) indicate a statistically significant difference ($p \leq 0.05$) between phenolic acid content in chicory and purslane within each form (free, bound, or total); ND: not detectable.

The texture of cupcake samples remained unaffected by the addition of *C. intybus* and *P. oleracea* powder at various substitution levels (Dykes and Rooney, 2007). The substitution of MPO and MCI leaves powder for flour significantly altered the color of cake samples' crust and crumbs ($p \leq 0.05$), as illustrated in Figures 4A,4B (Menga *et al.*, 2010). Increasing the levels of MPO and MCI resulted in decreased L*, a*, and b* values of crust, while there was an increase in the a* value of crumb despite decrease in L* and b*. This trend indicated that both crust and crumb of cake samples became darker with higher substitutions of MPO and MCI (Stalikas, 2007). The control sample exhibited a light brown color whereas the samples containing MPO and MCI leaves powder appeared darker (Ebrahimzadeh *et al.*, 2016). Consequently, the incorporation of MPO and MCI leaves powder into cupcake formulations resulted in a darker crust and crumb.

Figure 5 displays images of crumbs and crusts of cupcakes produced with varying levels of MPO and MCI leaves powder. It has been reported that the incorporation of MPO and MCI leaves powder resulted in increased a* and b* values with a decrease in the L* values of crumb (Gül and Şen, 2017). Additionally, past studies showed that bread fortified with grape seed powder exhibited a darker color, compared to the control sample (Pecˇivová *et al.*, 2014). Further research indicated that color alterations in baked goods enriched with plant-based powders could be attributed to phenolic compounds and other phytochemicals present in plant material, which influenced Maillard reaction during baking (Janda *et al.*, 2021a). Additionally, studies on muffins enriched with various plant extracts reported similar findings, showing that the incorporation of such ingredients not only affects color but also enhances antioxidant activity (Ertürk *et al.*, 2018). The darker color observed in cupcakes with MPO and MCI powder contributed to the overall sensory appeal and perceived quality of the product, as color is a crucial factor in consumer acceptance (Kumar *et al.*, 2020). The darker color observed in baked cupcakes could be due to the presence of polyphenolic compounds and carotenoids found in both MPO and MCI leaves. These compounds participate in Maillard reaction and other browning reactions during baking, contributing to a deeper color and enhanced flavor profiles (Janda *et al.*, 2021a). Additionally, a study conducted by Shyu *et al.* (2019) found that the use of green tea powder in muffins resulted in a significant increase in both a* and b* values while decrease in L* value, corroborating the trend observed with MPO and MCI powders.

Furthermore, research showed that the addition of vegetable powders not only altered color but also enhanced the nutritional profile of baked products. For instance, bread enriched with spinach powder was found to have

Table 2. Chemical composition of chicory and purslane leaves-supplemented cupcakes.

Treatment	Storage period (days)			
	0	5	10	15
Moisture (%)				
Control	2.98 ± 0.09^{A,n}	3.03 ± 0.03^{A,m,n}	3.19 ± 0.03^{A,l,m}	3.25 ± 0.04^{A,k,l}
MPO 5.0%	3.20 ± 0.04^{A,B,l}	3.25 ± 0.03^{A,B,k,l}	3.30 ± 0.03^{A,B,j,k}	3.46 ± 0.04^{A,B,i}
MPO 10.0%	3.34 ± 0.03^{B,i,j}	3.49 ± 0.03^{A,B,h,i}	3.55 ± 0.06^{A,g,h}	3.51 ± 0.03^{A,f}
MCI 5.0%	3.48 ± 0.03^{B,C,f,g}	3.53 ± 0.03^{B,e,f}	3.69 ± 0.04^{A,d,e}	3.75 ± 0.02^{A,c,d}
MCI 10.0%	3.63 ± 0.04^{B,d}	3.70 ± 0.03^{A,B,b,c}	3.76 ± 0.03^{A,B,b}	3.93 ± 0.03^{A,a}
Crude protein (%)				
Control	5.59 ± 0.02^{e}	5.59 ± 0.02^{e}	5.58 ± 0.01^{e}	5.60 ± 0.02^{A,e}
MPO 5.0%	6.81 ± 0.03^{d}	6.81 ± 0.03^{d}	6.88 ± 0.02^{d}	6.90 ± 0.02^{A,d}
MPO 10.0%	7.25 ± 0.01^{A,B,c}	7.45 ± 0.01^{A,c}	7.49 ± 0.02^{A,c}	7.33 ± 0.01^{A,B,c}
MCI 5.0%	7.58 ± 0.02^{B,b}	7.77 ± 0.02^{A,B,b}	7.77 ± 0.01^{A,B,b}	7.76 ± 0.02^{A,B,b}
MCI 10.0%	7.99 ± 0.02^{A,B,a}	8.09 ± 0.02^{A,a}	8.08 ± 0.01^{A,a}	8.07 ± 0.02^{A,a}
Crude fat (%)				
Control	23.27 ± 0.02^{A,e,f}	23.27 ± 0.02^{A,e,f}	23.27 ± 0.01^{A,e,f}	23.26 ± 0.04^{A,f}
MPO 5.0%	23.29 ± 0.01^{A,c,d,e}	23.29 ± 0.01^{A,c,d,e}	23.28 ± 0.01^{A,d,e,f}	23.28 ± 0.01^{A,d,e,f}
MPO 10.0%	23.30 ± 0.02^{A,a,b,c,d}	23.30 ± 0.02^{A,a,b,c,d}	23.29 ± 0.02^{A,d,e,f}	23.29 ± 0.02^{A,d,e,f}
MCI 5.0%	23.31 ± 0.02^{A,a,b,c,d}	23.31 ± 0.02^{A,a,b,c,d}	23.31 ± 0.01^{A,a,b,c,d}	23.29 ± 0.03^{A,b,c,d,e}
MCI 10.0%	23.33 ± 0.01^{A,a}	23.33 ± 0.01^{A,a}	23.32 ± 0.02^{A,a,b}	23.32 ± 0.02^{A,a,b,c}
Crude fiber (%)				
Control	0.12 ± 0.006^{A,e}	0.14 ± 0.006^{A,e}	0.14 ± 0.006^{A,e}	0.13 ± 0.010^{A,e}
MPO 5.0%	0.22 ± 0.010^{A,d}	0.27 ± 0.010^{A,d}	0.24 ± 0.010^{A,d}	0.26 ± 0.006^{A,d}
MPO 10.0%	0.31 ± 0.015^{A,c}	0.33 ± 0.015^{A,c}	0.34 ± 0.010^{A,c}	0.32 ± 0.015^{A,c}
MCI 5.0%	0.52 ± 0.010^{A,b}	0.57 ± 0.010^{A,b}	0.56 ± 0.015^{A,b}	0.56 ± 0.021^{A,b}
MCI 10.0%	0.61 ± 0.015^{A,a}	0.72 ± 0.015^{A,a}	0.71 ± 0.021^{A,a}	0.71 ± 0.017^{A,a}
Ash (%)				
Control	0.45 ± 0.008^{A,f,g}	0.45 ± 0.008^{A,f,g}	0.44 ± 0.011^{A,g}	0.44 ± 0.011^{A,g}
MPO 5.0%	0.47 ± 0.010^{A,d,e}	0.47 ± 0.010^{A,d,e}	0.47 ± 0.012^{A,d,e}	0.46 ± 0.015^{A,e,f}
MPO 10.0%	0.48 ± 0.006^{A,b,c,d}	0.48 ± 0.010^{A,c,d,e}	0.48 ± 0.012^{A,c,d,e}	0.47 ± 0.015^{A,d,e}
MCI 5.0%	0.50 ± 0.012^{A,a}	0.50 ± 0.012^{A,a}	0.50 ± 0.010^{A,a,b}	0.49 ± 0.006^{A,a,b,c}
MCI 10.0%	0.51 ± 0.010^{A,a}	0.51 ± 0.010^{A,a}	0.51 ± 0.015^{A,a}	0.50 ± 0.015^{A,a}
NFE				
Control	66.28 ± 0.10^{A,a}	66.24 ± 0.04^{A,a}	66.22 ± 0.02^{A,a}	66.22 ± 0.04^{A,a}
MPO 5.0%	65.76 ± 0.04^{A,b}	65.74 ± 0.06^{A,b}	65.73 ± 0.08^{A,b}	65.71 ± 0.02^{A,b}
MPO 10.0%	64.90 ± 0.01^{A,c}	64.87 ± 0.04^{A,c}	64.83 ± 0.07^{A,c}	64.82 ± 0.06^{A,c}
MCI 5.0%	64.15 ± 0.08^{A,d}	64.14 ± 0.09^{A,d}	64.11 ± 0.02^{A,d}	64.07 ± 0.03^{A,d}
MCI 10.0%	63.52 ± 0.05^{A,e}	63.49 ± 0.07^{A,e}	63.46 ± 0.09^{A,e}	63.42 ± 0.10^{A,e}

Notes. Mean values followed by different superscripted lower case letters in the same column represent significant difference ($p < 0.05$) in treatments, while different superscripted capital letters in the same row represent significant difference ($p < 0.05$) in time.
Control sample: cupcakes without MPO and MCI (control); MPO 5.0%: cupcakes with 5.0% MPO; MPO 10.0%: cupcakes with 10.0% MPO; MCI 5.0%: cupcakes with 5.0% MCI; and MCI 10.0%: cupcakes with 10.0% MCI; NFE: nitrogen-free extract.

increased levels of vitamins A and C along with a higher antioxidant capacity (Khan *et al.*, 2020). Similar findings were reported in cupcakes containing beetroot powder, which enhanced both color and nutritional benefits (Zhao *et al.*, 2020). The sensory attributes of color significantly influenced consumer perceptions and preferences. A study done by Luhova *et al.* (2021) emphasized that products with vibrant colors, attributed to natural additives, were perceived as healthier and more appealing. This suggested that the darker color obtained

Table 3. Color profile of chicory and purslane leaves-supplemented cupcakes.

Treatment	Storage period			
	0 day	5 days	10 days	15 days
L^*				
Control	$46.79 \pm 0.14^{A,B,a}$	$47.78 \pm 0.14^{A,a}$	$47.77 \pm 0.14^{A,a}$	$47.77 \pm 0.14^{A,a}$
MPO 5.0%	$44.98 \pm 0.13^{A,b}$	$44.97 \pm 0.12^{A,b}$	$44.96 \pm 0.12^{A,b}$	$44.96 \pm 0.11^{A,b}$
MPO 10.0%	$39.14 \pm 0.18^{A,c}$	$39.14 \pm 0.18^{A,c}$	$39.13 \pm 0.19^{A,c}$	$39.12 \pm 0.19^{A,c}$
MCI 5.0%	$36.48 \pm 0.22^{A,d}$	$36.48 \pm 0.23^{A,d}$	$46.47 \pm 0.24^{A,d}$	$36.46 \pm 0.25^{A,d}$
MCI 10.0%	$32.64 \pm 0.25^{A,e}$	$32.64 \pm 0.25^{A,e}$	$42.63 \pm 0.26^{A,e}$	$32.62 \pm 0.27^{A,e}$
a^*				
Control	$10.82 \pm 0.09^{A,a}$	$10.81 \pm 0.10^{A,a}$	$10.81 \pm 0.09^{A,a}$	$10.80 \pm 0.10^{A,a}$
MPO 5.0%	$6.44 \pm 0.05^{A,b}$	$6.43 \pm 0.06^{A,b}$	$6.43 \pm 0.05^{A,b}$	$6.42 \pm 0.06^{A,b}$
MPO 10.0%	$4.78 \pm 0.07^{A,c}$	$4.77 \pm 0.06^{A,c}$	$4.76 \pm 0.06^{A,c}$	$4.76 \pm 0.06^{A,c}$
MCI 5.0%	$2.55 \pm 0.08^{A,d}$	$2.54 \pm 0.08^{A,d}$	$2.54 \pm 0.08^{A,d}$	$2.53 \pm 0.08^{A,d}$
MCI 10.0%	$1.37 \pm 0.04^{A,e}$	$1.37 \pm 0.04^{A,e}$	$1.36 \pm 0.04^{A,e}$	$1.36 \pm 0.05^{A,e}$
b^*				
Control	$22.29 \pm 0.13^{A,a}$	$22.29 \pm 0.12^{A,a}$	$22.28 \pm 0.12^{A,a}$	$22.27 \pm 0.13^{A,a}$
MPO 5.0%	$19.18 \pm 0.12^{B,b}$	$19.18 \pm 0.11^{B,b}$	$19.17 \pm 0.11^{B,b}$	$21.16 \pm 0.12^{A,b}$
MPO 10.0%	$17.83 \pm 0.14^{A,c}$	$17.82 \pm 0.15^{A,c}$	$17.82 \pm 0.14^{A,c}$	$16.81 \pm 0.16^{B,c}$
MCI 5.0%	$16.37 \pm 0.12^{A,d}$	$16.36 \pm 0.13^{A,d}$	$16.35 \pm 0.12^{A,d}$	$16.35 \pm 0.13^{A,d}$
MCI 10.0%	$15.82 \pm 0.17^{A,e}$	$15.82 \pm 0.17^{A,e}$	$15.81 \pm 0.16^{A,e}$	$15.80 \pm 0.15^{A,e}$

Mean values followed by different lower case superscripted letters in the same column represent significant difference ($p < 0.05$) in treatments, while different superscripted capital letters in the same row represent significant difference ($p < 0.05$) in time. Control sample: cupcakes without MPO and MCI (control); MPO 5.0%: cupcakes with 5.0% MPO; MPO 10.0%: cupcakes with 10.0% MPO; MCI 5.0%: cupcakes with 5.0% MCI; and MCI 10.0%: cupcakes with 10.0% MCI.

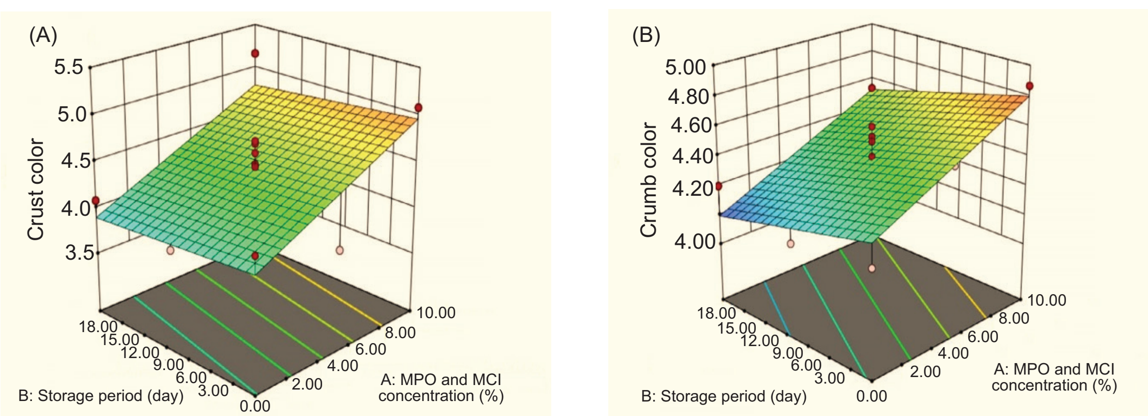

Figure 4. 3D surface plot of the effect of different addition levels of MPO and MCI leaves powder on (A) crust color and (B) crumb color of produced cupcakes during the storage period.

by the incorporation of MPO and MCI could enhance the marketability of these cupcakes, aligning with consumer trends favoring functional foods. Hence, the incorporation of MPO and MCI leaves powder into cupcake formulations not only affected their color properties but also potentially improved their nutritional value and consumer appeal.

Effect of addition levels of *C. intybus* and *P. oleracea* on the nutritional composition of cupcakes

Tables 4 and 5 display the caloric value of cupcakes with *C. intybus* and *P. oleracea* leaves powder. The addition of MCI to cupcakes significantly ($p \leq 0.05$) boosted the overall mineral content, as accessed by Tukey's test. The

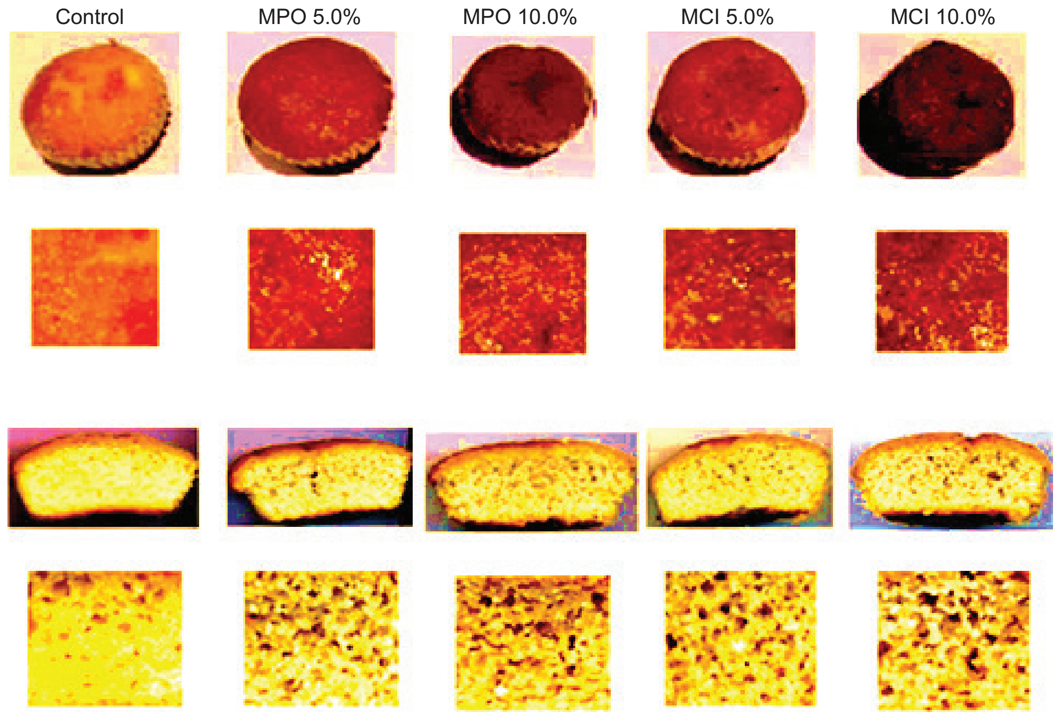

Figure 5. Crust and crumb of cupcakes produced using different levels of MPO and MCI leaves powder during storage period.

Table 4. Nutritional composition of cupcakes with different levels of MPO and MCI leaves powder on dry weight (DW) basis.

Cupcake samples	Moisture (g/100 g)	Ash (g/100 g)	Fat (g/100 g)	NDF (g/100 g)	Protein (g/100 g)
***C. intybus* cupcake (MCI)**					
0% (control)	35.18 ± 1.32^{a}	1.97 ± 0.06^{a}	3.12 ± 0.11^{a}	13.75 ± 0.10^{a}	13.68 ± 0.01^{c}
MCI 5%	31.84 ± 0.25^{a}	2.40 ± 0.00^{b,c}	3.58 ± 0.06^{a}	19.02 ± 2.73^{a}	13.96 ± 0.00^{d}
MCI 10%	32.39 ± 0.40^{a}	2.54 ± 0.15^{b,c}	3.33 ± 0.32^{a}	14.77 ± 2.94^{a}	14.59 ± 0.01^{e}
***P. oleracea* cupcake (MPO)**					
0% (control)	33.56 ± 0.40^{a}	2.28 ± 0.01^{ab}	2.98 ± 0.42^{a}	23.87 ± 4.86^{a}	13.07 ± 0.06^{a}
MPO 5%	38.93 ± 6.93^{a}	2.46 ± 0.06^{b,c}	2.78 ± 0.02^{a}	20.60 ± 1.20^{a}	13.16 ± 0.01^{a,b}
MPO 10%	33.60 ± 0.10^{a}	2.62 ± 0.04^{c}	3.36 ± 0.13^{a}	17.75 ± 1.14^{a}	13.34 ± 0.10^{b}

Mean values followed by different superscripted lower case letters in the same column represent significant difference ($p < 0.05$) in treatments.

overall mineral content of cupcakes increased significantly after addition of 10% MPO leaf powder. Protein content in cupcakes also increased significantly by addition of 5% and 10% *C. intybus* leaf powder ($p \leq 0.05$).

Control sample: cupcakes without MPO and MCI (control); MPO 5.0%: cupcakes with 5.0% MPO; MPO 10.0%: cupcakes with 10.0% MPO; MCI 5.0%: cupcakes with 5.0% MCI; and MCI 10.0%: cupcakes with 10.0% MCI; NDF: neutral detergent fiber.

Lipid oxidation

According to Ferreira et al. (2019), lipid oxidation is a significant contributor to the reduction of sensory quality and shelf life in food products. The TBARS assay of beef samples treated with chicory (C. intybus L.) and purslane (P. oleracea L.) leaf powder extracts during storage at 4°C corroborated this finding (Figure 6). Both the addition of chicory and purslane leaf powders and the duration of the storage period significantly influenced the TBARS levels in the meat samples ($p \leq 0.05$). Adding a high concentration of MPO and MCI leaves powder (10%) to cupcake samples did not decrease TBARS levels, compared to the addition of 5% MPO and MCI leaves powder for a storage period of 15 days ($p > 0.05$). TBARS values ranged from 0.25 ± 0.11 to 0.76 ± 0.12 mg MDA/kg at the initiation of storage period (day 0), and MPO and MCI leaves powder resulted in high ($p \leq 0.05$) TBARS values compared to the control. Chicory and purslane leaf powders, known for their use in enhancing the nutritional profile of various food products,

Table 5. Mineral content of cupcakes with different levels of MPO and MCI leaves powder on dry weight (DW) basis.

Cupcake samples	Calcium (mg/kg)	Zinc (mg/kg)	Iron (mg/kg)
***C. intybus* cupcake (MCI)**			
0% (control)	0.04 ± 0.00	3.00 ± 0.00[a]	6.50 ± 2.83[a]
MCI 5%	0.10 ± 0.00	3.50 ± 9.90[a]	7.60 ± 2.83[a]
MCI 10%	0.15 ± 0.00	2.95 ± 0.71[a]	16.55 ± 2.12[b]
***P. oleracea* cupcake (MPO)**			
0% (control)	0.03 ± 0.00	3.20 ± 0.00[a]	16.50 ± 2.83[b]
MPO 5%	0.05 ± 0.00	3.05 ± 0.71[a]	23.20 ± 2.83[c]
MPO 10%	0.09 ± 0.00	3.10 ± 0.00[a]	30.95 ± 9.19[d]

Mean values followed by different superscripted lower case letters in the same column represent significant difference ($p < 0.05$) in treatments. Control sample: cupcakes without MPO and MCI (control); MPO 5.0%: cupcakes with 5.0% MPO; MPO 10.0%: cupcakes with 10.0% MPO; MCI 5.0%: cupcakes with 5.0% MCI; and MCI 10.0%: cupcakes with 10.0% MCI.

may act as pro-oxidants in treated cupcake samples due to their high chlorophyll content (Gorji *et al.*, 2019). However, further analysis of cereal processing and related products is necessary to fully understand their role and effects.

Texture properties

Hardness, height, and volume of cupcake samples were evaluated based on texture qualities (Figures 7A–C). The addition of *C. intybus* (CI) and *P. oleracea* (PO) leaves powder significantly affected these parameters ($p < 0.05$). Specifically, cupcakes with 1.0% MPO and MCI leaf powders exhibited lower hardness, height, and volume compared to those with 0.5% MPO, 0.5% MCI, and the control samples ($p < 0.05$). These differences were statistically significant. for 0.5% MPO, 0.5% MCI, and control samples. Volume is a critical physical characteristic of cupcakes, as consumers generally prefer products with higher volume. Addition of 5% MPO and 5% MCI enhanced the volume of cupcakes; however, increasing leaf powder concentration to 10% adversely affected the volume of cupcakes ($p < 0.05$).

Phenolic components present in *C. intybus* and *P. oleracea* could compromise their emulsifying properties, leading to softer baked products with higher concentrations of MPO and MCI extracts (Jongberg *et al.*, 2015). The detrimental effects on emulsification were evident only with the use of 1.0% MPO and MCI leaves powder in cupcake formulations ($p < 0.05$). Interestingly, cupcakes with 1.0% leaves powder exhibited higher cohesiveness, compared to those with 0.5% leaves powder and control samples (both negative and positive) ($p < 0.05$). This suggested that while higher concentrations of MPO and MCI negatively impacted volume and texture, they could also enhance the cohesiveness of the final product, potentially contributing to improved structural integrity. Interestingly, cupcakes with 1.0% leaves powder exhibited higher cohesiveness compared to those with 0.5% leaves powder and control samples (both negative and positive) ($p < 0.05$).

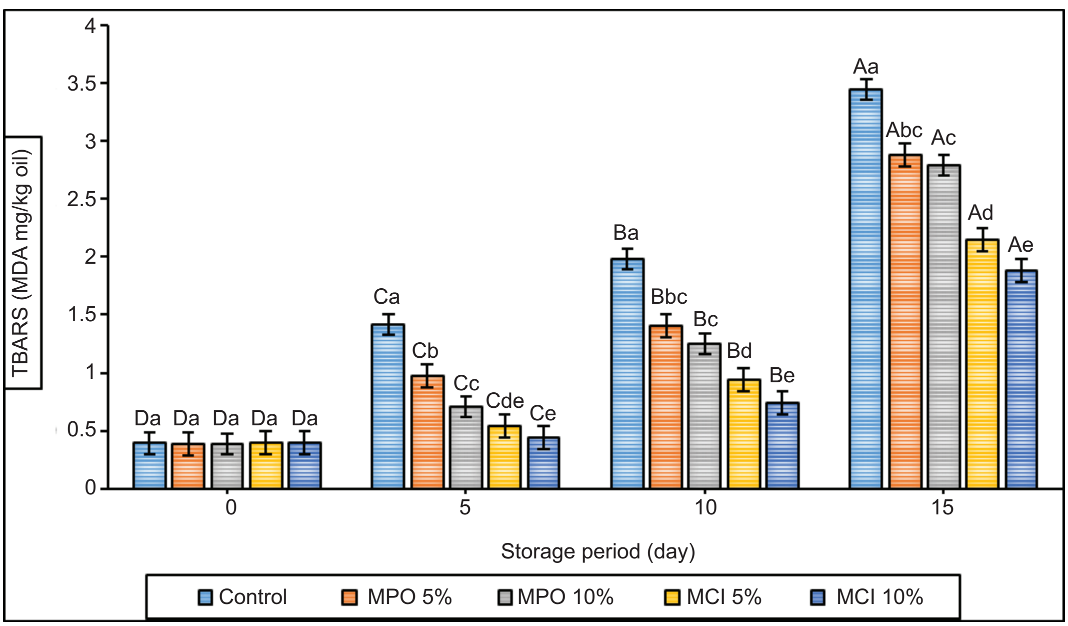

Figure 6. Histograms with different lower case letters represent significant difference ($p < 0.05$) in treatments, while different capital letters represent significant difference ($p < 0.05$) in time. The TBARS of cupcakes produced using different levels of MPO and MCI leaves powder during the storage period.

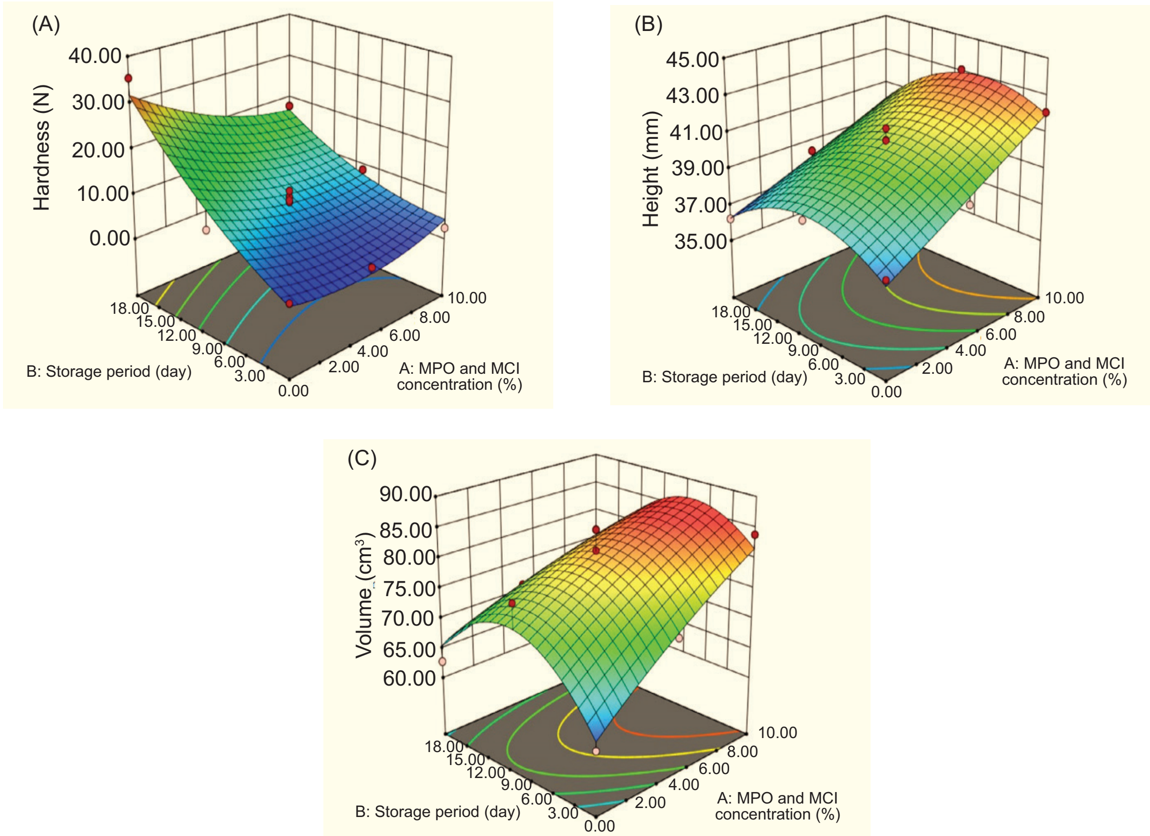

Figure 7. Changes in the (A) hardness, (B) height, and (C) volume of produced cupcakes by the addition of MPO and MCI during storage.

This suggests that while higher concentrations of MPO and MCI can negatively impact volume and texture, they may also enhance cohesiveness of the final product, potentially contributing to improved structural integrity. Studies demonstrated that incorporation of certain plant extracts could positively affect textural properties, such as increasing cohesiveness and elasticity, critical for consumer acceptance (Almeida *et al.*, 2021). Moreover, research done by Jafari *et al.* (2019) demonstrated that the incorporation of dietary fibers from leafy greens not only affected texture but also enhanced nutritional content, indicating a beneficial trade-off between health benefits and textural quality.

Effect of adding MPO and MCI on sensory acceptability of cupcakes

Based on the results of sensory evaluation, the effects of *C. intybus* and *P. oleracea* on the overall acceptability of cupcakes are shown in Tables 6 and 7. As shown in Table 6, the overall acceptability of a cupcake was diminished by the inclusion of different concentrations of *C. intybus* and *P. oleracea* (5% and 10%). In terms of general acceptability, however, *P. oleracea* had a less detrimental effect on cupcakes than chicory *C. intybus*, and *P. olercea* significantly reduced the acceptability of the taste of cupcakes. The overall acceptance of cupcakes varied significantly based on the type and concentration of leaf powder added. Notably, in terms of flavor acceptability, cupcakes containing MCI (*C. intybus*) did not differ significantly from those made with *P. oleracea* if 5% or 10% of *C. intybus* was added. However, incorporation of *C. intybus* leaf powder resulted in a marked decrease in color acceptability. In case 5% MPO was added to cupcakes, no significant difference in color acceptance was observed, compared to the control ($p > 0.05$).

In contrast, the addition of 10% MPO led to a notable decline in color acceptability. The color acceptability for the cupcakes made with 5% *P. oleracea* was significantly higher than that of the cupcakes made with 5% or 10% *C. intybus* as well as 10% *P. oleracea* cupcakes. The increased concentration of *C. intybus* was found to significantly reduce the overall acceptability ($p < 0.05$) of cupcakes. The general acceptance of 5% MPO cupcakes was significantly higher than that of other formulations, including 10% MPO and both 5% and 10% MCI cupcakes. These findings

Table 6. Effect of *C. intybus* and *P. oleracea* on taste, aroma, texture, appearance, color, and the overall acceptability of cupcakes.

	Sensory acceptability					
	Taste	Color	Odor	Texture	Appearance	Overall acceptability
***C. intybus* cupcake (MCI)**						
0% (control)	6.38 ± 2.26^{c,d}	7.16 ± 1.50^{c}	7.17 ± 1.62^{c}	6.42 ± 2.14^{b}	7.31 ± 1.59^{c}	6.97 ± 1.89^{c,d}
MCI 5%	4.67 ± 2.17^{a}	5.16 ± 2.39^{a,b}	4.80 ± 2.41^{a,b}	5.21 ± 2.44^{a}	5.51 ± 2.31^{b}	5.28 ± 2.17^{a,b}
MCI 10%	3.93 ± 2.35^{a}	4.19 ± 2.58^{a}	4.17 ± 2.36^{a}	5.21 ± 2.22^{a}	4.22 ± 2.70^{a}	4.31 ± 2.33^{a}
***P. oleracea* cupcake (MPO)**						
0% (control)	7.17 ± 1.56^{d}	7.67 ± 1.21^{c}	7.34 ± 1.46^{c}	6.95 ± 1.99^{b}	7.61 ± 1.33^{c}	7.72 ± 1.33^{d}
MPO 5%	5.91 ± 2.20^{b,c}	6.66 ± 1.96^{c}	5.84 ± 1.99^{b}	6.16 ± 2.11^{a,b}	6.52 ± 2.07^{b,c}	6.54 ± 1.97^{c}
MPO 10%	4.88 ± 2.16^{a,b}	5.54 ± 2.17^{b}	4.95 ± 1.98^{a,b}	5.97 ± 1.92^{a,b}	5.81 ± 2.06^{b}	5.43 ± 1.96^{b}

Mean values followed by different superscripted lower case letters in the same column represent significant difference ($p < 0.05$) in treatments. Control sample: cupcakes without MPO and MCI (control); MPO 5.0%: cupcakes with 5.0% MPO; MPO 10.0%: cupcakes with 10.0% MPO; MCI 5.0%: cupcakes with 5.0% MCI; and MCI 10.0%: cupcakes with 10.0% MCI.

were consistent with the research that indicated that both flavor and color were crucial determinants of consumer acceptability of baked products (Ragaert *et al.*, 2016). The negative impact of higher concentrations of *C. intybus* on acceptability could be attributed to the leaf powder's strong flavor profile, which could dominate the sweetness and richness typically expected in cupcakes (Kumar *et al.*, 2020). Moreover, a study conducted by Aydin *et al.* (2021) noted that the color of baked goods significantly influenced consumer preferences, with darker products often receiving lower acceptability scores.

Discussion

The findings of the current study support the findings of Munswamy *et al.* (2013), who found that *P. acidus* leaf extract contains a high concentration of flavonoids and phenols. The present research also confirmed the findings of Dailey and Vuong (2015) that the number of bioactive compounds recovered from plant materials is significantly impacted by extraction solvents, concluding that aqueous solvent (100% water) is the most effective solvent for extracting biologically active compounds. These findings were corroborated by the findings of Ash and Fraioli (2016). , In plant extracts, TPC and TFC are normally used parameters to quantify the concentration of phenolic compounds and flavonoids, respectively. These compounds are known for their antioxidant properties and potential health benefits. Differences in TPC and TFC values of ethanol and aqueous extracts vary, depending on several factors. Ethanol and water have different solvent properties, and certain phenolic compounds and flavonoids may be more soluble in one solvent compared to another. For example, some polar phenolic compounds may dissolve better in aqueous solutions whereas others may have better solubility in ethanol.

The choice of solvent affects the efficiency of extraction. Ethanol is generally considered a better solvent for extracting a wide range of phenolic compounds and flavonoids because of its ability to dissolve both polar and non-polar compounds. However, certain compounds may have higher extraction yields in aqueous solutions, especially if they are more polar. Different solvents can selectively extract different compounds from plant materials. Ethanol may selectively extract certain phenolic compounds and flavonoids, while aqueous extraction may favor other compounds. The choice of solvent and extraction method should be optimized based on target compounds. The solvent used for extraction can influence the chemical composition of extract. Ethanol may facilitate the extraction of certain compounds through dissolution or chemical interactions, leading to differences in TPC and TFC values compared to aqueous extracts. The composition of phenolic compounds and flavonoids can vary greatly depending on plant species and part of the plant used for extraction. Certain plants may contain compounds that are more readily extracted with ethanol, while others may yield higher concentrations in aqueous extracts. Factors such as temperature, time, and solvent-to-sample ratio can influence extraction efficiency and composition of the extract. Optimization of these parameters for each solvent can maximize the extraction of phenolic compounds and flavonoids. Overall, differences in TPC and TFC values of ethanol and aqueous extracts highlight the importance of solvent selection and extraction optimization in obtaining accurate and meaningful results in phytochemical analysis.

Table 7. Effect of *C. intybus* and *P. oleracea* leaves powder on the overall acceptability of cupcakes as shown by the overall sensory evaluation scores.

Cupcake samples	Dislike extremely (1)	Dislike very much (2)	Dislike moderately (3)	Dislike slightly (4)	Neither likes nor dislikes (5)	Like slightly (6)	Like moderately (7)	Like very much (8)	Like extremely (9)	Overall liking (6–9)	Overall disliking (1–4)
***C. intybus* cupcake (MCI)**											
0% (control)	1^{A} (1.9)^{B}	1 (1.9	0 (0)	1 (1.9)	7 (13.0)	10 (18.5)	9 (16.7)	13 (24.1)	12 (22.2)	44 (81.5)	3 (5.6)
MCI 5%	3 (5.6)	4 (7.4)	4 (7.4)	8 (14.8)	10 (18.5)	9 (16.7)	9 (16.7)	5 (9.3)	2 (3.7)	25 (46.3)	19 (35.2)
MCI 10%	7 (13.0)	6 (11.1)	8 (14.8)	12 (22.2)	7 (13.0)	5 (9.3)	4 (7.4)	2 (3.7)	3 (5.6)	14 (25.9)	33 (61.1)
***P. oleracea* cupcake (MPO)**											
0% (control)	0 (0)	0 (0)	0 (0)	0 (0)	3 (5.6)	9 (16.7)	10 (18.5)	16 (29.6)	16 (29.6)	51 (94.4)	0 (0)
MPO 5%	1 (1.9)	2 (3.7)	1 (1.9)	3 (5.6)	10 (18.5)	7 (13.0)	13 (24.1)	8 (14.8)	9 (16.7)	37 (68.5)	7 (13.0)
MPO 10%	2 (3.7)	4 (7.4)	2 (3.7)	7 (13.0)	14 (25.9)	9 (16.7)	10 (18.5)	3 (5.6)	3 (5.6)	25 (46.3)	15 (27.8)

[a]Scores imparted by different panelists; [b]percentage of consumers panel (N = 54) that imparted scores. Control sample: cupcakes without MPO and MCI (control); MPO 5.0%: cupcakes with 5.0% MPO; MPO 10.0%: cupcakes with 10.0% MPO; MCI 5.0%: cupcakes with 5.0% MCI; and MCI 10.0%: cupcakes with 10.0% MCI.

The levels of proteins, moisture, fiber, and ash changed between 6.68% and 11.28%, 4.43% and 4.55%, 0.22% and 6.34%, and 2.54% and 5.63%, respectively, with an increase in leaf extract concentration. The chemical compositions of cupcakes, such as moisture, lipid (fat), ash, fiber, protein, and nitrogen-free extract (NFE), can potentially change with the addition of purslane and chicory leaf powder. The moisture content of cupcakes may change depending on the water content of leaves powder and their interaction with other ingredients. If the leaves powder has a higher moisture content, then they would increase the overall moisture content of cupcakes. Purslane and chicory leaves powders typically have low-fat content, so adding them to cupcakes is unlikely to significantly affect the lipid content unless they are mixed with ingredients containing high levels of fat, such as butter or oil. Ash content represents the mineral content of cupcakes. Purslane and chicory leaves powder may contribute minerals to cupcakes, potentially increasing the ash content. Purslane and chicory leaves powder are known to be rich in dietary fiber, which contributes to increased fiber content in cupcakes. This can have potential health benefits, such as improved digestion and satiety. Purslane and chicory leaves powder are also sources of protein, so their addition may increase the protein content of cupcakes. This provides additional nutritional value and may be beneficial for individuals looking to increase their protein intake. NFE primarily represents carbohydrate content of cupcakes, excluding fiber, protein, and fat. The addition of purslane and chicory leaves powder, which contain carbohydrates, may affect the NFE content of cupcakes, depending on the proportion of carbohydrates in leaves powder relative to other ingredients.

Overall, specific changes caused in the chemical composition of cupcakes with addition of purslane and chicory leaves powder depend on the factors such as the quantity of leaves powder added, the composition of other ingredients in cupcakes, and the processing conditions. Conducting detailed compositional analysis before and after adding leaves powder can provide precise information of these changes.

However, in baking experiments, variations in ingredients, processing methods, and storage conditions can lead to differences in the chemical composition and the overall characteristics of the final product. Factors such as ingredient proportions, baking temperature, humidity, and storage duration also influence the chemical composition and quality of cupcakes.

A comparative analysis of the chemical composition of cupcakes is needed to identify significant differences in moisture, lipid, ash, fiber, protein, and NFE of control samples and those with 10% purslane and chicory leaves powder. We obtained the moisture, lipid, ash, fiber, protein, and NFE content data for both control samples and cupcakes with 10% purslane and chicory leaves powder. It was ensured that the data were collected using standardized methods and techniques, and statistical tests were performed to compare the mean values of each component between control and experimental samples (cupcakes with 10% purslane and chicory leaves powder). For continuous variables (such as moisture, lipid, ash, fiber, protein, and NFE), independent sample t-tests or analysis of variance (ANOVA) were used to determine significant differences between groups. Results of statistical analyses were examined to identify significant differences in moisture, lipid, ash, fiber, protein, and NFE of control samples and cupcakes with 10% purslane and chicory leaves powder.

We considered the magnitude of differences as well as statistical significance while interpreting the results and discussed the implications of the observed differences in terms of nutritional composition, sensory attributes, and potential health benefits. We considered the factors, such as ingredient interactions, processing methods, and sample variability, that could influence the results. Finally, we summarized significant differences in moisture, lipid, ash, fiber, protein, and NFE of control samples and those with 10% purslane and chicory leaves powder and provided insights into the potential impact of incorporating purslane and chicory leaves powder on the nutritional profile of cupcakes. The relevance of these findings in the context of product development, consumer preferences, and dietary considerations was also discussed. Any significant differences in the chemical compositions of cupcakes of control samples and those with 10% purslane and chicory leaves powder are identified by conducting a systematic analysis and interpretation of data. Sengev *et al.* (2013) discovered that adding more moringa leaf powder (MLP) significantly increased the protein (9.07–13.97%), ash (1.10–1.65%), and fiber (2.10–3.28%) contents of wheat bread. MPO and MCI with higher concentrations of moisture, fat, protein, ash, and fiber are most likely to alter chemical composition (Sahin *et al.*, 2021; Yamashita *et al.*, 2018). Moisture increased significantly ($p \leq 0.05$) during the 15-day storage, but levels of crude protein, crude fat, crude fiber, and ash did not increase significantly (for all treatments; Table 1). The ability of fiber in wheat flour and leaves powder of MCI and MPO to absorb more water can increase moisture content after storage. The hydrophilic nature of cupcakes and the oxidation of polyunsaturated fatty acids are connected to reduce other properties. These results were similar to those of Nadarajah and Mahendran (2015). Additionally, the protein content increased along with the concentration of *C. intybus* in cupcakes ($p \leq 0.05$). A cupcake with 5% MPO had a little higher protein content than the one with 10% MPO; however, the difference was significant ($p \leq 0.05$). Comparing the protein composition of two cupcakes, the one with *C. intybus* leaf powder contains more protein than the one with *P. oleracea* leaf powder.

Nitrogen-free extract is a nutritional measure that represents the carbohydrate content of food, which includes sugars, starches, and fiber. Following factors contribute to the lower concentration of NFE in cupcakes with 10% purslane and chicory leaves powder, compared to control samples:

Dilution effect: Adding leaf powder to cupcake batter increases the overall volume of ingredients without necessarily adding carbohydrates in the same proportion. This dilution effect can lead to a lower concentration of carbohydrates, including NFE, in the final product.

Fiber content: Purslane and chicory leaves powder may contain higher levels of dietary fiber, compared to other cupcake ingredients. Dietary fiber, although technically a carbohydrate, is classified separately from NFE because it can't be digested by humans. Therefore, the inclusion of fiber-rich leaf powder can reduce the proportion of digestible carbohydrates (NFE) in cupcakes.

Reduced sugar content: Depending on the formulation and composition of leaf powder, there might be a lower concentration of sugars in the cupcakes with 10% purslane and chicory leaves powder. Leaves powder may have inherently lower sugar content, compared to other cupcake ingredients, such as sugar or fruit puree, leading to a decrease in NFE.

Starch content: Purslane and chicory leaves powder might not contribute significantly to the starch content of cupcakes. Starch is a major component of NFE, and if the leaves powder are low in starch or if they interfere with starch gelatinization during baking, it could result to lower NFE concentrations in the final product.

Nutrient interactions: Some components present in leaves powder may interact with carbohydrates or affect their digestibility. For example, certain compounds in leaves powder could inhibit carbohydrate-digesting enzymes, leading to reduced availability of carbohydrates used for absorption.

Baking process: The baking process itself can affect the composition of carbohydrates in the final product. High temperature and prolonged baking period could lead to caramelization or Maillard reaction, which can alter the carbohydrate profile of cupcakes, potentially reducing NFE content.

Analytical variation: Variations in analytical methods used to determine NFE concentrations could also contribute to differences observed between control samples and samples with leaves powder. Differences in sample preparation, extraction techniques, or analytical instruments could lead to discrepancies in final values.

A combination of factors related to ingredient composition, baking process, and analytical considerations could lead to a lower concentration of NFE in cupcakes with 10% purslane and chicory leaves powder, compared to control samples.

Iron levels in two types of cupcakes would be increased considerably ($p \leq 0.05$) by addition of 10% MCI or 5% MPO.

The choice of minerals for analysis, specifically Fe, Zn, and Ca, was justified based on several factors, including their physiological importance, prevalence in food sources, and relevance to the target population or research objectives. According to this experiment, prooxidants, such as chicory and purslane leaves powder, were active at the start of the cupcake storage period. Vegetable juice components were analyzed by Gazzani et al. (1998) and initially showed pro-oxidant activity in vitro. However, over time, this activity diminished, and the juice components eventually exhibited natural antioxidant properties. The antioxidant activity of chicory and purslane leaves powder changes during storage because of various factors, such as exposure to light, air, moisture, and temperature fluctuations. Generally, the antioxidant activity of plant materials tends to decrease over time because of the degradation of antioxidants or oxidation reactions. The significance of the initial pro-oxidant activity of these leaves powder exists in their potential to generate reactive oxygen species (ROS) under certain conditions. Pro-oxidants are substances that promote oxidation reactions, leading to the production of ROS, which can be harmful to cells and tissues. However, it is important to note that certain pro-oxidants, when present at moderate levels, can also stimulate the body's antioxidant defense mechanisms, leading to beneficial effects. In the context of chicory and purslane leaves powder, understanding their initial pro-oxidant activity can help in optimizing their storage conditions and processing methods to minimize oxidative degradation and maximize their antioxidant potential. By managing factors such as exposure to light, air, and temperature, it could be possible to mitigate pro-oxidant activity and preserve the antioxidant properties of these leaves powder over an extended storage period. Monitoring changes in antioxidant activity over time could provide valuable insights into the stability and shelf-life of chicory and purslane leaves powder, helping to ensure their efficacy and quality for various applications, such as food additives, dietary supplements, or functional ingredients in pharmaceuticals. Therefore, components of chicory and purslane leaves powder used in this study had a pro-oxidant effect on day 0 of storage. However, in the current study, chicory and purslane leaves powder acted as an antioxidant in cupcakes samples that were treated for 4 days or more up to the end of the storage period. The lowest TBARS values (0.33 ± 0.05 and 0.22 ± 0.14 mg MDA/kg, respectively) were observed in samples containing 5% and 10% chicory and purslane leaves powder on day 5 of storage ($p \leq 0.05$). The control sample's TBARS value was 0.83 ± 0.33 mg MDA/kg on day 5 of storage, and nonsignificant fluctuations ($p > 0.05$) persisted up to the end of the storage period (15th day). On day 10 (0.99 0.18–0.82 0.15 mg MDA/kg), the lipid oxidation of chicory and purslane leaves powder-treated cupcake samples was significantly greater ($p \leq 0.05$). Otherwise, the treated samples' (10% MPO and MCI) lipid oxidation rate lowered significantly up to the end of the storage period. The most likely cause of this decrease in TBARS levels was either further oxidation of MDA into other organic lipid oxidation products that were not detected by the thiobarbituric acid reaction or MDA degradation by bacteria that specifically utilized the carbonyl group (Georgantelis *et al.*, 2007). Plant extracts' phenolic components are good at minimizing the effects of oxidation. Estévez and Cava (2006) observed that oxidative rancidity in food products could not be prevented or delayed by high amounts of antioxidants or preservatives. According to the findings, the natural antioxidant content of chicory and purslane leaves powder significantly increases fat oxidation during storage period. The extracts of chicory and purslane contain essential oils, in addition to a high concentration of phenols, which aid in regulating and inhibiting lipid oxidation.

Owing to a high amount of natural antioxidants and ability to resist lipid oxidation, MPO and MCI (5.0%) are the best treatment options. Additionally, this process often improves the product's uniformity and firmness of the cupcakes. The phenolic components present in chicory and purslane extracts, which are accountable for their antioxidant activity through reductive, free radical scavenging, and lipid oxidation inhibitory actions, may be responsible for the reduced TBARS values of treated cupcake samples. These findings were in agreement with other studies that examined the impact of plant extracts on lipid peroxidation in cupcakes.

When chicory and purslane leaves powder were added to dough, the color of cupcake samples darkened (Table 2 and Figure 2). Owing to high concentrations of chlorophyll and other pigments, chicory and purslane leaves are dark green in color, which is the main cause of the dark color changes of cupcake. Bourekoua *et al.* (2018) reported similar outcomes. A lightening agent can be used to conceal the dark color, making the chicory and purslane leaves powder-containing cupcakes more acceptable. As consumers are more accustomed to cupcakes of a golden-brown hue color, the dark color may reduce their attractiveness; however, white cupcakes are more frequently consumed than brown cupcakes. The application of lightning agents was not examined in this study. Brown cupcakes with 5% MPO and MCI had nearly the same shade as the control, demonstrating that a concentration lower than 5% could provide a lighter cupcake with greater nutritional benefits.

Consumer acceptability: The consumer acceptability of cupcakes with *C. intybus* and *P. oleracea*, compared to the control, would typically be evaluated through sensory analysis. This involves assessing attributes such as taste, texture, aroma, and overall liking by a panel of consumers. The comparison revealed the impact of addition of chicory or purslane on consumer perception, compared to the control cupcakes. Results could vary based on factors such as recipe formulation, concentrations of the chicory or purslane used, and individual preferences of consumers.

Strategies to address darkening of color: Darkening of the product, often referred to as browning, occurs due to various factors, such as enzymatic reactions, Maillard reaction browning, or oxidation of pigments. To address this issue, several strategies are considered: (a) Ingredient selection: using ingredients with lower levels of enzymes or precursors to browning reactions can help mitigate color changes. For example, selecting chicory or purslane varieties with lower enzyme activity or browning potential. (b) pH adjustment: controlling the pH of cupcake batter can influence enzymatic browning reactions. Adjusting of pH to an optimal range for enzyme activity can help prevent excessive browning. (c) Antioxidants: incorporating antioxidants, such as ascorbic acid (vitamin C) or citric acid, can help inhibit oxidation reactions that contribute to browning. (d) Processing methods: Altering processing methods, such as blanching, steaming, or using sulfites, can help deactivate enzymes responsible for browning. (e) Packaging: packaging of cupcakes using materials that provide a barrier to light and oxygen can help minimize color changes during storage. (f) Natural colorants: adding natural colorants, such as beetroot powder or turmeric, can mask or complement any undesirable color changes while also providing additional health benefits. Implementing these strategies can help maintain the visual appeal of cupcakes containing chicory or purslane while preserving their sensory attributes and consumer acceptability.

Despite their unpleasant look, chicory and purslane leaves powder are rich in nutrients, as mentioned previously. Table 3 demonstrates that when chicory and purslane were added to cupcake samples, the protein concentration increased significantly ($p \leq 0.05$) in comparison to the control. The sample of cupcakes with 10% MCI had the highest protein content (15.5 g/100 g). Given the high protein content of *C. intybus* leaves, this was anticipated (Sengev *et al.*, 2013). The results were consistent with the findings of another study, which found that fortified cupcakes containing *C. intybus* and *P. oleracea* had the highest protein content (13.5%), while unfortified cupcakes had the lowest level of proteins (8.5%). Other researchers discovered that the protein content increased gradually if different concentrations of chicory and purslane leaves powder were added. The above-mentioned findings are encouraging because of the growing market for plant proteins.

Choosing the concentration of 5% for fortifying cupcakes with purslane and chicory leaves powder may be based on the following factors:

Optimal balance of flavor and nutrition: Higher concentrations of leaf powder may impart a stronger taste or alter the texture of the cupcakes, potentially affecting consumer acceptance. By selecting a lower concentration like 5%, the cupcakes may retain palatability while providing significant nutritional benefits.

Previous studies or recommendations: Prior research or dietary guidelines may suggest a certain concentration of leaf powder for optimal health benefits or nutritional supplementation. This concentration could have been determined through empirical studies or based on traditional usage.

Nutritional content: The selected concentration likely provides a substantial amount of phenolic compounds, flavonoids, and other bioactive compounds present in purslane and chicory leaves powder, contributing to the desired nutritional enhancement of the cupcakes.

Regarding the impact of these additions on malnutrition interventions, particularly in cases of protein-energy malnutrition (PEM), the inclusion of bioactive plant extracts, such as those from C. intybus and P. oleracea, may offer nutritional benefits by improving the protein and energy content of food products. This could be especially significant in regions where PEM is prevalent, as these plants may enhance the nutritional value of foods and help mitigate the effects of inadequate protein and energy intake in vulnerable populations.:

Nutritional enrichment: Purslane and chicory leaves powder contain various nutrients, such as vitamins, minerals, antioxidants, and dietary fibers. Adding these ingredients increase cupcakes' nutritional value, potentially addressing deficiencies commonly observed in malnourished individuals.

Protein content: While leaves powders may not be a significant source of proteins, they do contribute to the overall nutrient profile. In cases of PEM, any additional protein, even in relatively small quantity, can contribute to meeting its daily requirements.

Antioxidant activity: Phenolic compounds and flavonoids present in purslane and chicory leaves powder possess antioxidant properties that help to combat oxidative stress associated with malnutrition and improve overall health outcomes.

Dietary diversity: Introducing novel ingredients such as purslane and chicory leaves powder into fortified foods promotes dietary diversity, which is crucial for addressing malnutrition. Offering a variety of nutrient-enriched foods increases the likelihood of meeting essential nutrient requirements.

Acceptance and accessibility: Cupcakes are generally a well-liked and easily accessible product, making them a convenient vehicle for delivering nutrients, especially to malnourished individuals. By fortifying cupcakes with purslane and chicory leaves powder, nutrient-rich options are readily available and culturally acceptable. Finally, fortifying cupcakes with purslane and chicory leaves powder at a concentration of 5% provides a balanced approach to enhance nutritional content while considering factors such as flavor, palatability, and potential impact on malnutrition interventions, particularly in cases of PEM.

On the other hand, to avoid a monotonous diet centered on MCI and MPO cupcakes, chicory and purslane could be incorporated into other widely consumed protein-deficient foods. The iron content of cupcakes increased when 10% MPO and MCI were added, while iron concentration increased when 5% MPO and MCI were added. This is anticipated because *P. oleracea* and *C. intybus* contain a significant amount of iron. The iron content found in this study was in line with the prior studies Sengev *et al*. (2013), Gazzani *et al*. (1998) and Georgantelis *et al.* (2007), showing that iron content increased with the addition of *C. intybus* and *P. oleracea*. Further, the findings suggest that MPO- and MCI-containing cupcakes could be a less expensive alternative to mitigate iron deficiency in susceptible populations.

Finally, there was a higher level of consumer acceptability of control cupcake samples (0%), compared to the cupcake samples containing *C. intybus* and *P. oleracea* ($p \leq 0.05$) (Tables 6 and 7). This could be due to bitter flavor and darkened color of the product. These findings were in line with recent studies by Sengev *et al.* (2013), Gazzani *et al*. (1998), and Georgantelis *et al*. (2007), which showed that the bioactive compound levels in *C. intybus* and *P. oleracea* decreased with increasing concentrations. While there was a little difference in texture acceptance of cupcake samples with *P. oleracea* leaf powder, the texture acceptability of cupcake samples containing *C. intybus* decreased significantly ($p \leq 0.05$). Therefore, based on customers' assessment of texture of cupcake samples, *C. intybus* leaf powder appeared to have a negative impact. Cupcakes were harder when *C. intybus* and *P. oleracea* leaves powder were added, probably due to their high fiber content. However, only MIC cupcake showed lessened hardness. Cupcakes made with *P. oleracea* were more acceptable for fortification than cupcakes made with *C. intybus*, because the general acceptance of cupcake samples made with 5% MOP leaf powder was more than that of cupcake samples made with 5% MCI.

Conclusions

The antioxidant activity of chicory and purslane leaves powder was assessed. Purslane and chicory leaves powder used to prepare cupcakes demonstrated the best behavior. The ideal concentration for chicory and purslane leaves powder was identified through preliminary testing. The foods fortified with *C. intybus* and *P. oleracea* require further investigation for including MPO and MCI in known but nutrient-deficient foods. The results of this study suggested that cupcakes fortified with 5% MPO and MCI could be used to enhance current malnutrition interventions, particularly PEM. Although the nutritional value of cakes could be improved by adding MPO and MCI, their effect on product quality must also be taken into account.

MPO improved cake volume, texture, and sensory attributes in contrast to MCI, which decreased such qualities. The investigation found that using MPO and MCI up to 5% boosted cupcakes' quality. Finally, by delaying crumb moisture loss and boosting crumb hardness, the addition of MPO and MCI increased cupcakes' shelf life. This is reflected in the applications and the variety of uses of these plants as well as in the fact that these plants possess crucial characteristics, such as palatability, and are liked by consumers. The findings of this study demonstrate that chicory and purslane could be employed as food additives with a variety of advantages. More research is required to assess the impact of adding chicory and purslane extracts on the physicochemical characteristics of food products.

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Data Availability

Data will be made available on request.

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