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The Effect of Packaging Covers and Gaseous Composition on Maintaining the Postharvest Quality of Marigold

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Abstract

Marigold (*Calendula officinalis* L.) is also known as one of the common edible flowers. In this study, the improvement of nutritional value and shelf life of marigolds under the covers of packaging and gaseous composition was investigated. Marigold flowers in plastic packages made of polyethylene, polyamide, and the combination of these two covers with gas composition (1: 5% O_2 + 5% CO_2; 2: 5% O_2 + 10% CO_2; 3: 5% O_2 + 20% CO_2) or without gas combination were placed along with the control (without packaging). These packages were transferred to the cold store at 4°C, and sampling was done to evaluate the desired traits on days 0, 5, 10, and 15. The results showed that with the increased storage time in all edible marigold flowers, the amount of weight loss, ion leakage, and decay increased, and the relative water content (RWC), total phenol, flavonoids, and carotenoids decreased. At the same time, using polyethylene and polyamide packaging treatment, along with gaseous composition, improved the visual and nutritional quality of flowers. Among all the treatments, the combination treatment of 5% O_2 + 10% CO_2 and polyethylene + polyamide coating had better effects in maintaining the quality and increasing the shelf life of the edible marigold flowers. As a result, considering the nutritional and medicinal value of the marigold flowers, this treatment can be used as an effective and safe method to increase the shelf life and maintain the appearance quality and biochemical compounds of this plant.

Keywords: Bioactive compounds, Edible flowers, Modified atmosphere packaging, Plastic cover, Visual quality.

INTRODUCTION

Edible flowers include dozens of flowers with different shapes, colors, and sizes, which are used worldwide to improve the sensory and nutritional quality of food (Aquino-Bolaños *et al*., 2013). They are used in sauces, jellies, syrups, alcoholic beverages, vinegar, honey, oils, canned flowers, salad, tea, and other drinks and desserts (Koike *et al*. ., 2015). These edible flowers provide a range of nutrients, including amino acids, vitamins, minerals, proteins, and antioxidant compounds (Fernandes *et al*., 2017). However, the knowledge about people's attitudes towards the consumption of edible flowers is limited. In addition, edible flowers are highly perishable, and their high perishability requires attention to safe and healthy storage methods to improve their shelf life after harvest (Marchioni *et al*., 2022). Various ways are used by the food industry to increase the shelf life of food products and to ensure their quality and safety, which is essential to ensure public health (Koike *et al*., 2015; Pêgo *et al*., 2022).

Marigold (*Calendula officinalis* L.) is one of the common edible flowers. The flowers and leaves of this plant are edible. Its fresh and chopped petals are added to a salad or used as a seasoning. Dried petals have a more intense flavor and are used as a seasoning in soups, cakes, drinks, and baked goods (Roberts, 2000). In the petals of this plant, there are compounds such as carotenoids, phenolic compounds, coumarins, and flavonoids, and the presence of these compounds has made marigolds a rich source of antioxidant compounds. (Wiktorowska *et al*., 2010; Khalid and da Silva, 2012).

Short shelf life is the main limiting factor in the trade and marketing of edible flowers, especially marigolds, and these products have a shallow shelf life due to high respiration (Kelley *et al*., 2003). Packaging is important for the quality preservation of vegetables, flowers, and fruits during storage and sale. Meanwhile, modified atmosphere packaging (MAP), by reducing the concentration of O_2 and increasing the concentration of CO_2 compared to standard air, is an effective technology for delaying the postharvest decay of horticultural products (Yang *et al*., 2022). MAP is the modification of gas composition produced from respiration (passive MAP) or by adding and removing gases from food packages (active MAP) to manipulate O_2 and CO_2 levels. A decrease in O_2 levels and an increase in CO_2 levels can reduce respiration and ethylene production, delay ripening and tissue softening, and reduce compositional changes associated with maturation, thus increasing shelf life (Daş et al., 2006; Fagundes *et al*., 2015). Kou *et al*. (2012) reported that all edible carnation flowers packaged with the MAP system had significantly reduced water loss and higher overall quality than those commercially packaged in plastic clamshell containers. Storage of edible squash flowers under a controlled atmosphere (CA) in four different gas combinations caused all flowers to show better preservation of total sugars, total soluble solids, pH, titratable acidity, and lower physiological weight loss. Combining O_2 5%, CO_2 10%, and N_2 showed the highest amount of ascorbic acid, polyphenols, and carotenoids (Aquino-Bolaños *et al*., 2013). In marigolds (*Tagetes erecta* L.) stored at 23˚C, low-density polyethylene (LDPE) bags significantly reduced weight loss and maintained color and overall appearance for up to 8 days of storage (Pal *et al*., 2016). The effectiveness of modified passive atmosphere packaging (MAP) on the chemical and qualitative properties of marigolds during 10 days of storage at 5˚C showed that weight loss in flowers wrapped with continuous and micro-perforated, macro-perforated, and control were 3, 7 and 30%, respectively. The total phenol concentration of packaged flowers was significantly higher than the control and similar to the initial value. Also, at the end of storage, unpackaged flowers were considered unsalable, with severe wilting and shriveling, while all packaged flowers were fresh and salabl (Fadda *et al*., 2020). The effects of two kinds of modified atmosphere packaging on the post-harvest quality of fresh mulberry leaf vegetables stored at 4˚C were shown that the respiration rate of the fresh leaves in the modified polyethylene packages was lower than the normal polyethylene packages. Also, the content of total soluble solids, soluble protein, and total polyphenol in modified polyethylene packages had less change compared to normal polyethylene packages, and the content of vitamin C was also higher (Yang *et al*., 2022).

Since marigold can be used as an edible flower due to its compounds, such as carotenoids and flavonoids, and high antioxidant properties, it seems necessary to evaluate its performance and storage capacity for transportation and marketing. On the other hand, due to the lack of specific guidelines for the maintenance of edible flowers and also the lack of use of postharvest growth factors of cut flowers (such as silver nitrate, hydroxyquinoline sulfate, etc.) due to their edible nature, this research was conducted to develop the shelf life and preserve the bioactive compounds of edible marigolds by using different packaging covers and gaseous composition.

MATERIALS AND METHODS

Plant materials and experimental treatments

This research was carried out in 2020-2021 in the laboratories and cold storages of the department of horticultural sciences and food packaging laboratory of the Department of Food Industry, Faculty of Agriculture and Natural Resources, University of Tehran, to study the effect of packaging covers and gas composition on preserving the postharvest quality of marigold edible flowers. The marigolds were harvested at the full blooming stage and quickly transferred to the cold room at 2˚C for cooling. At first, damaged and deformed flowers were removed. Then the flowers were separated from the peduncle part of the stem, and 10 to 15 flowers were weighed and placed in plastic bags made of polyethylene, polyamide, and the combination of these two covers with or without gaseous composition. The gas composition used includes 1) 5% O_2 + 5% CO_2; 2) 5% O_2 + 10% CO_2; 3) 5% O_2 + 20% CO_2. For the control sample, no packaging was done. These packages were transferred to the cold store at 4˚C, and sampling was done to evaluate the desired traits on days 0, 5, 10, and 15.

Visual quality

The visual quality of the flowers was evaluated after storage for days 0, 5, 10, and 15. Visual quality with a 9-point hedonic scale where 9: Like extremely; 7: Like moderately; 5: Neither like nor dislike; 3: Dislike moderately, and 1: Dislike extremely (Meilgaard *et al*., 1991).

Decay

The degree of decay of marigolds was evaluated by a 3-member trained panel according to the method of Loaiza and Cantwell (1997). Decay in a 5-point scale based on visual observation of the degree of decay, where 0: no decay; 1: The beginning of decay; 2: Less than a quarter of the surface area decayed; 3: The decay of a quarter to a half of the surface; 4: Decay was more than half of the surface, done.

Analysis of O_2 and CO_2 gases inside the packages

The concentration of O_2 and CO_2 inside the packages containing flowers was measured during the sampling days using a portable gas analyzer (model Oxybaby 6.0, WITT, Denmark), previously calibrated by sampling the atmospheric air. O_2 and CO_2 values were expressed in units of kilopascal (kPa).

Weight loss

To measure the amount of weight loss according to the Conte *et al*. (2009) method, first, the weight of the flowers was measured at zero time (W1) in all treatments, then the secondary weight (W2) was measured on the 5, 10 and 15 days of storage and the percentage of weight loss was calculated using the following formula: Weight loss = (W1-W2) / W1 × 100 (%).

Relative water content and electrolyte leakage

The relative water content (RWC) was measured by the method of Weatherley *et al*. (1950) and was measured by weight in distilled water and finally expressed as a percentage. Ion leakage was also measured using an electrical conductivity meter on sampling days and was calculated as a percentage (Hong *et al*., 2000).

Antioxidant capacity

The antioxidant capacity of the flowers was determined based on the chemical neutralization reaction of 1, 1 diphenyl-2-picrylhydrazine (DPPH) free radical using a spectrophotometer at 517 nm and presented as % (Brand-Williams *et al*., 1995).

The content of phenols, flavonoids, and carotenoids

The total phenol content was measured using the methanolic extract of petals and Folin-Ciocaltio reagent, and the absorbance of the samples was recorded using a spectrometer at 750 nm and finally expressed in mg g^{-1} fresh weight of the petals (Singleton and Rossi, 1965). The total flavonoid content was measured using the methanolic extract of the petals according to the method of Chang *et al*. (2002). The absorbance of the samples was recorded at 415 nm, and finally, it was expressed in mg g^{-1} fresh weight of the petals. To measure carotenoids, pigments were extracted by dimethyl sulfoxide (DMSO). Then, the absorbance of the obtained solution was recorded using a spectrophotometer at 480 and 510 nm. The total carotenoid content of the petals was expressed in mg g^{-1} fresh weight of the petals (Soroori *et al*., 2021).

Statistical analysis

This research was carried out factorially in a completely randomized design with four levels of packaging kind, four levels of gas composition, and three levels of storage duration in three replications. SAS software (version 9.1) was used to analyze the variance and compare the mean of the measured traits. Means were compared using Duncan's Multiple Range Test at the 5% probability level.

RESULTS AND DISCUSSION

Visual quality and decay

The effects of treatments (packaging with or without gaseous composition), storage time, and their interaction were significant at the 1% probability level ($P < 0.01$) on the visual quality and decay of edible marigold flowers (Table 1). As the storage time increased, the visual quality of marigolds decreased, and the decay rate increased. The highest visual quality was related to the treatment of the combination of polyethylene and polyamide with a gaseous composition of 5% O_2 and 10% CO_2, which improved the visual quality by 57% compared to the control (without packaging and gaseous composition) (Table 2). The lowest percentage of decay of edible marigold flowers was also obtained during 15 days of storage in the treatment of polyethylene and polyamide combination with a gas composition of 5% O_2 and 10% CO_2, which caused a 74% reduction in decay compared to the control (Table 2). The use of polymer coatings, regardless of the type of gaseous composition used, by preventing the weight loss of the product, maintaining the organoleptic and visual characteristics, while increasing the storage time, preserves the appearance quality, and reduces the decay of edible marigolds (Fadda *et al*., 2020). Packaging isolates the product from the external environment and helps to preserve the product in non-sterile conditions, and reduces the contact of the product with pathogens and contaminants (Yang *et al*., 2022). The increase of CO_2 and the decrease of O_2 by reducing the activity of microorganisms, including fungi, prevent the product's decay and increase the quality of horticultural products after harvesting. In the present study, the combined use of polyethylene and polyamide coating and the MAP maintained the appearance quality and prevented excessive decay of the marigold edible flowers. In accordance with our results, in carnation, MAP reduced decay incidence by avoiding weight loss and maintaining visual quality for 7 days of storage at 5°C (Kou *et al*., 2012). Also, Pal *et al*. (2016) showed in marigolds that low-density polyethylene (LDPE) bags significantly preserved the color and general appearance of flowers by preventing weight loss until the 8 days of storage.

O_2 and CO_2 gases concentration on the packages

The effect of treatments (packaging with or without gaseous composition), storage time, and their interaction on CO_2 and O_2 concentration of packages containing edible marigold flowers were significant (Table 1). The packages of marigolds containing a high concentration of CO_2 had more CO_2 at the end of the storage period. The concentration of this gas in the flowers covered with polyethylene coating was lower than that of polyethylene and polyethylene + polyamide coatings, and the highest CO_2 percentage was 3.45% in polyethylene + polyamide treatment with 20% CO_2 and 5% O_2 on the 15th day of storage (Table 3). Unlike the CO_2 concentration, the O_2 concentration in the polyethylene treatment was higher than the polyethylene and polyethylene + polyamide treatment. The highest O_2 concentration (18.27%) on the 15th day of storage was related to the flowers packed in polyethylene cover without gaseous composition (Table 3). The proper balance between CO_2 and O_2 levels can be very different depending on the product, storage conditions, and the characteristics of the packaging films used (Fadda *et al*., 2020). Our results showed that with the increase of CO_2 concentration inside the packages, the amount of O_2 decreased, so that on the last day of storage, the amount of O_2 reached below 14% in some treatments, and it can harm the appearance and nutritional quality of the flower. The decrease in O_2 level and increase in CO_2 level despite the reduction in respiration, ethylene, and delay in the softening and ripening of the product can also cause the activity of anaerobic microorganisms (Fagundes *et al*., 2015). Therefore, these results highlight the need to adjust MAP conditions in terms of O_2 and CO_2 concentration for each product. Paying attention to the concentration of gases used in the storage conditions of products under MAP has been reported in the studies of Aquino-Bolaños *et al*. (2013) and Palma *et al*. (2023).

Table 1. The results of variance analysis of characteristics of edible marigolds under the effect of packaging treatments and gaseous composition during the postharvest period.

S.o.V	df	MS										
		Visual quality	Decay	CO_2	O_2	Weight loss	Electrolyte leakage	RWC	Antioxidant capacity	Phenols	Flavonoids	Carotenoids
Treatment (T)	12	0.53^{**}	121.66^{**}	1.54^{**}	128.07^{**}	47.09^{**}	44.38^{**}	1140.38^{**}	5.36^{**}	0.005^{**}	11.98^{**}	0.01^{**}
Storage time (S)	3	11.57^{**}	1471.4^{**}	4.35^{**}	148.82^{**}	756.9^{**}	184.66^{**}	1053.5^{**}	137.72^{**}	0.41^{**}	297.38^{**}	0.86^{**}
T× S	36	0.12^{**}	$26..18^{**}$	0.25^{**}	3.11^{*}	11.04^{**}	13.07^{**}	35.42^{ns}	1.46^{ns}	0.009^{ns}	2.64^{ns}	0.017^{ns}
Error	104	0.01	3.88	0.09	0.21	2.23	0.711	52.202	0.285	0.17	0.73	0.0005
CV (%)	-	5.27	4.45	5.81	8.08	4.7	5.94	7.73	8.04	7.82	4.23	1.12

*, ** and ^{ns}: Significant at $P < 0.05$, $P < 0.01$ and insignificant based on the Duncan's multiple-range test, respectively.

Table 2. Comparison of the average visual quality and decay of edible marigolds under the effect of packaging treatments and gaseous composition during the post-harvest period.

Treatments / Storage time (day)	Visual quality				Decay			
	0	5	10	15	0	5	10	15
Control	5.09^{a}	3.90^{defghi}	2.52^{m}	1.52^{n}	0^{q}	14.61^{gh}	35.09^{b}	51.27^{a}
Polyethylene	5.09^{a}	4.31^{bcdef}	3.53^{ijkl}	3.04^{lm}	0^{q}	5.58^{lm}	12.85^{i}	19.68^{cd}
Polyamide	5.09^{a}	4.14^{cdefg}	3.53^{ijkl}	3.14^{kl}	0^{q}	5.83^{l}	12.43^{i}	19.3^{cd}
Polyethylene + Polyamide	5.09^{a}	4.42^{bcde}	3.64^{hijkl}	3.24^{jkl}	0^{q}	4.77^{lmn}	12.56^{i}	19^{cde}
Polyethylene + CO_2 5% + O_2 5%	5.09^{a}	4.62^{abc}	3.53^{ijkl}	3.34^{ijkl}	0^{q}	4.32^{n}	10.05^{jk}	18.06^{ef}
Polyamide + CO_2 5% + O_2 5%	5.09^{a}	4.73^{ab}	3.84^{fghij}	3.55^{hijkl}	0^{q}	4.26^{no}	10.43^{j}	16.96^{f}
Polyethylene + Polyamide + CO_2 5% + O_2 5%	5.09^{a}	4.62^{abc}	3.64^{hijkl}	3.45^{ijkl}	0^{q}	4.42^{n}	10.26^{j}	17^{f}
Polyethylene + CO_2 10% + O_2 5%	5.09^{a}	4.62^{abc}	4.04^{defgh}	3.85^{efghij}	0^{q}	3.05^{p}	9.88^{jk}	15.2^{g}
Polyamide + CO_2 10% + O_2 5%	5.09^{a}	4.52^{bcd}	4.14^{cdefg}	3.95^{defghi}	0^{q}	3.19^{op}	9.9^{jk}	15.16^{g}
Polyethylene + Polyamide + CO_2 10% + O_2 5%	5.09^{a}	4.42^{bcde}	4.24^{bcdef}	3.95^{defghi}	0^{q}	3.85^{nop}	9.4^{k}	14.2^{gh}
Polyethylene + CO_2 20% + O_2 5%	5.09^{a}	4.31^{bcdef}	4.04^{defgh}	3.75^{fghijk}	0^{q}	4.32^{n}	12.12^{i}	19.82^{c}
Polyamide + CO_2 20% + O_2 5%	5.09^{a}	4.73^{ab}	4.24^{bcdef}	3.65^{ghijkl}	0^{q}	4.66^{mn}	13.76^{h}	19.19^{cd}
Polyethylene + Polyamide + CO_2 20% + O_2 5%	5.09^{a}	4.62^{abc}	4.04^{defgh}	3.55^{hijkl}	0^{q}	4.53^{mn}	13.99^{h}	18.6^{de}

*In each column, means with similar letter(s) are not significantly different ($P < 0.05$) using the Duncan's multiple range test.

Table 3. Comparison of the average CO_2 and O_2 concentration of edible marigolds under the effect of packaging treatments and gaseous composition during the post-harvest period.

	CO_2				O_2			
Treatments / Storage time (day)	**0**	**5**	**10**	**15**	**0**	**5**	**10**	**15**
Control	0^{q}	0^{q}	0^{q}	0^{q}	0^{m}	0^{m}	0^{m}	0^{m}
Polyethylene	0.61^{nop}	0.82^{lm}	1.1^{ij}	1.31^{gh}	20.88^{abcde}	19.64^{abcdef}	18.41^{cdefg}	18.27^{cdefg}
Polyamide	0.5^{p}	0.72^{mn}	1.01^{jk}	1.11^{ij}	21.48^{abc}	18.42^{bcdefg}	17.65^{efgh}	16.57^{fghij}
Polyethylene + Polyamide	0.61^{nop}	0.61^{nop}	0.91^{kl}	1.01^{jk}	20.49^{abcde}	17.65^{efghi}	17.71^{efgh}	16.9^{fghij}
Polyethylene + CO_2 5% + O_2 5%	0.5^{p}	0.82^{lm}	1.1^{ij}	1.42^{fg}	21.31^{abc}	16.45^{fghijk}	17.62^{fghi}	18.34^{cdefg}
Polyamide + CO_2 5% + O_2 5%	0.7^{mn}	0.72^{mn}	0.91^{kl}	1.01^{jk}	21.21^{abc}	17.9^{defgh}	17.62^{fghi}	15.46^{ghijkl}
Polyethylene + Polyamide + CO_2 5% + O_2 5%	0.66^{no}	0.61^{nop}	0.81^{lm}	0.91^{kl}	20.94^{abcd}	14.8^{ghijkl}	16.26^{ghijk}	14.69^{ghijkl}
Polyethylene + CO_2 10% + O_2 5%	0.55^{op}	0.82^{lm}	1.21^{hi}	1.52^{f}	20.88^{abcde}	15.42^{ghijkl}	15.37^{ghijkl}	15.12^{ghijkl}
Polyamide + CO_2 10% + O_2 5%	0.7^{mn}	0.71^{mn}	1.01^{jk}	1.72^{e}	21.69^{ab}	14.44^{hijkl}	14.39^{ijkl}	14.52^{jkl}
Polyethylene + Polyamide + CO_2 10% + O_2 5%	0.61^{nop}	0.82^{lm}	1.21^{hi}	2.13^{d}	20.59^{abcde}	14.02^{ijkl}	14.52^{hijkl}	14.9^{ijkl}
Polyethylene + CO_2 20% + O_2 5%	0.66^{no}	1.23^{hi}	2.12^{d}	3.04^{b}	21.01^{abcd}	12.79^{kl}	13.42^{jkl}	14.72^{ghijkl}
Polyamide + CO_2 20% + O_2 5%	0.55^{op}	1.43^{fg}	2.22^{d}	3.14^{b}	21.85^{a}	$13.\ 9^{ijkl}$	13.76^{jkl}	13.47^{l}
Polyethylene + Polyamide + CO_2 20% + O_2 5%	0.5^{p}	1.54^{f}	2.65^{c}	3.45^{a}	21.09^{abcd}	13.67^{jkl}	13.14^{kl}	13.99^{kl}

*In each column, means with similar letter(s) are not significantly different ($P < 0.05$) using the Duncan's multiple-range test ($P < 0.05$).

Weight loss and electrolyte leakage

The effects of treatments (packaging with or without gaseous composition), storage time, and their interaction were significant at the 1% probability level on weight loss and electrolyte leakage of edible marigold flowers (Table 1). Along with increasing storage time, the percentage of weight loss and electrolyte leakage of marigolds risen significantly in all treatments. The lowest weight loss was related to the treatment of polyethylene + polyamide with a gas combination of 5% O_2 and 10% CO_2, which compared to the control, kept 70% of the weight of edible marigolds during 15 days of storage (Table 4). The highest percent of electrolyte leakage of edible marigolds on the 15th day of storage was 34% in the control treatment. In comparison, the percent of electrolyte leakage at the end of storage was 9.18% related to the treatment of polyethylene + polyamide with a gas composition of 5% O_2 and 10% CO_2 (Table 4).

Table 4. Comparison of the average weight loss and electrolyte leakage of edible marigolds under the effect of packaging treatments and gaseous composition during the post-harvest period.

	Weight loss				**Electrolyte leakage**			
Treatments / Storage time (day)	**0**	**5**	**10**	**15**	**0**	**5**	**10**	**15**
Control	0^{u}	9.8^{i}	22.27^{b}	34.37^{a}	5.29^{t}	11.51^{efg}	22.76^{b}	34.00^{a}
Polyethylene	0^{u}	5.27^{pq}	8.96^{j}	17.93^{c}	6.77^{qrs}	6.98^{o-s}	9.40^{hij}	14.35^{d}
Polyamide	0^{u}	6.69^{lm}	9.43^{ij}	17.5^{c}	5.89^{rst}	7.82^{k-q}	10.5^{fg}	17.6^{c}
Polyethylene + Polyamide	0^{u}	4.96^{p-s}	8.85^{j}	15.2^{d}	5.88^{rst}	7.12^{n-r}	8.45^{j-m}	16.42^{c}
Polyethylene + CO_2 5% + O_2 5%	0^{u}	4.36^{rst}	8.23^{k}	15.16^{d}	6.37^{qrs}	7.20^{m-r}	8.44^{j-m}	11.79^{ef}
Polyamide + CO_2 5% + O_2 5%	0^{u}	5.77^{nop}	7.10^{l}	14.2^{e}	6.21^{rst}	8.22^{k-o}	8.29^{k-o}	12.18^{e}
Polyethylene + Polyamide + CO_2 5% + O_2 5%	0^{u}	4.75^{qrst}	7.01^{l}	13.53^{ef}	6.91^{p-s}	7.56^{l-q}	8.44^{j-m}	10.37^{fgh}
Polyethylene + CO_2 10% + O_2 5%	0^{u}	4.03^{t}	6.11^{mno}	13.18^{f}	6.49^{qrs}	6.96^{p-s}	9.26^{hij}	11.00^{efg}
Polyamide + CO_2 10% + O_2 5%	0^{u}	4.26st	6.15^{mno}	11.82^{gh}	5.29^{t}	6.94^{p-s}	8.85^{i-l}	12.00^{e}
Polyethylene + Polyamide + CO_2 10% + O_2 5%	0^{u}	4.32st	5.38^{pq}	11.2^{h}	6.06^{rst}	6.9^{p-s}	8.34^{j-n}	9.18^{h-k}
Polyethylene + CO_2 20% + O_2 5%	0^{u}	4.007^{t}	5.76^{op}	12.2^{g}	6.37^{q-t}	6.37^{r-q}	8.02^{k-p}	10.15^{ghi}
Polyamide + CO_2 20% + O_2 5%	0^{u}	4.16st	6.04^{mno}	13.14^{f}	6.15^{rst}	6.68^{qrs}	7.58^{l-q}	9.07^{h-l}
Polyethylene + Polyamide + CO_2 20% + O_2 5%	0^{u}	5.21^{pqr}	6.37^{mn}	13.2^{f}	5.79st	6.88^{p-s}	8.03^{k-p}	10.2^{gh}

*In each column, means with similar letter(s) are not significantly different ($P < 0.05$) using the Duncan's multiple-range test ($P < 0.05$).

In general, evaporation, transpiration, and respiration of products in the post-harvest period lead to weight loss, and in addition, the longer the storage period of the product passes, the more weight loss increases. The low percentage of weight loss in the packaged samples is due to the lower permeability of the packaging cover, which provides a suitably modified atmosphere for the protection of flowers (Daş *et al*., 2006). Also, the lower weight loss in the gaseous composition with more CO_2 is due to the lower respiration rate of the packaged flowers in the atmosphere with high CO_2 and less O_2 (Pal *et al*., 2016). The difference between polyethylene and polyamide coatings also depends on the permeability of these coatings to water vapor, O_2, and CO_2. In this regard, Yang *et al*. (2022) reported that the respiration rate of samples in modified polyethylene packages was lower than that of normal polyethylene packages. Most of the physiological disorders of fresh products during storage are related to the cell membrane. Electrolyte leakage is an indicator of membrane permeability. Oxidation and peroxidation reactions of membrane lipids increase ion leakage (Kelley *et al*., 2003). In this research, the amount of electrolyte leakage increased with the increase in storage time. Still, the packaging of the products, especially the packaging with the modified atmosphere, could reduce this increase. The effect of modified atmosphere and packaging in reducing electrolyte leakage of horticultural products after harvest has been reported by Lee *et al*. (2023) in apple fruit and Hamzeh-Kalkenari *et al*. (2021) in button mushrooms.

RWC and antioxidant capacity

The effect of treatments (packaging with or without gaseous composition) and storage time was significant at the 1% probability level on RWC and antioxidant capacity of edible marigolds. Still, the interaction effect of treatment and storage time was not significant (Table 1). As the storage time progressed, the RWC and antioxidant capacity of the edible marigolds decreased significantly (Table 5). The highest percentage of RWC in the entire storage period was related to the treatment of polyethylene + polyamide with a gaseous composition of 5% O_2 and 10% CO_2, which compared to the control and the treatment polyethylene + polyamide without gas composition, 12% and 4% RWC of marigolds were preserved, respectively (Table 5). The highest antioxidant capacity (67.15%) of marigold flowers was also obtained in the polyethylene + polyamide treatment with a gas composition of 5% O_2 and 10% CO_2. In comparison, the lowest amount of antioxidant capacity (53.22 %) was observed in the control treatment (Table 5).

Table 5. Comparison of the average RWC, antioxidant capacity, total phenol, flavonoids, and carotenoids of edible marigolds under the effect of packaging treatments and gaseous composition during the postharvest period.

Treatments	RWC	Antioxidant capacity	Total phenol	Flavonoids	Carotenoids
Control	46.88^{e}	53.22^{d}	43.95^{d}	9.75^{d}	0.92^{d}
Polyethylene	53.5^{d}	59.67^{c}	52.45^{c}	11.95^{cd}	1.01^{b}
Polyamide	55.82^{dc}	61.92^{c}	54.45^{c}	11.87^{cd}	0.97^{c}
Polyethylene + Polyamide	56.52^{bcd}	63.7^{bc}	55.5^{c}	12.2^{c}	0.99^{a}
Polyethylene + CO_2 5% + O_2 5%	58.05^{abcd}	65.28^{abc}	56.15^{bc}	12.62^{bc}	1.04^{a}
Polyamide + CO_2 5% + O_2 5%	58.91^{abc}	65.37^{abc}	56.97^{bc}	12.8^{b}	1.02^{ab}
Polyethylene + Polyamide + CO_2 5% + O_2 5%	58.19^{a-d}	65.55^{abc}	57.9^{bc}	12.92^{ab}	1.02^{ab}
Polyethylene + CO_2 10% + O_2 5%	61.26^{ab}	65.02^{ab}	60.27^{a}	13.25^{a}	1.04^{a}
Polyamide + CO_2 10% + O_2 5%	62.01^{a}	66.05^{ab}	60.9^{a}	13.3^{a}	1.05^{a}
Polyethylene + Polyamide + CO_2 10% + O_2 5%	63.12^{a}	67.15^{a}	61.2^{a}	13.68^{a}	1.02^{ab}
Polyethylene + CO_2 20% + O_2 5%	60.61^{abc}	65.8^{ab}	58.97^{a}	13.03^{a}	1^{b}
Polyamide + CO_2 20% + O_2 5%	59.4^{abc}	65^{abc}	59.02^{ab}	13.08^{a}	1.02^{b}
Polyethylene + Polyamide + CO_2 20% + O_2 5%	59.4^{abc}	65.45^{abc}	58.95^{abc}	12.94^{ab}	1.01^{b}
Storage time (day)					
0	72.51^{a}	71.53^{a}	64.63^{a}	15.2^{a}	1.15^{a}
5	70.89^{ab}	66.69^{b}	59.31^{b}	14.06^{ab}	1.09^{ab}
10	68.91^{b}	62.13^{c}	53.9^{c}	12.1^{b}	0.98^{b}
15	62.28^{c}	54.76^{d}	48.83^{d}	8.9^{c}	0.81^{c}

*In each column, means with similar letter(s) are not significantly different ($P < 0.05$) using the Duncan's multiple-range test ($P < 0.05$).

The packaging of the marigold edible flowers in polyethylene and polyamide covers, along with the MAP system, preserves the RWC of the flowers and increases their antioxidant capacity by preventing weight loss, respiration, and stress resulting from aging (Fadda *et al*., 2020). The increase in antioxidant capacity increases the inhibition of active oxygen species (ROS) and reduces the browning index, and improves the appearance quality of flowers. Kou *et al*. (2012) reported that all edible flowers packaged with the MAP system had a significantly lower water loss and higher overall quality than those commercially packaged in collapsible plastic containers. However, the effect of MAP on antioxidant capacity is inconsistent and species-dependent. In edible squash flowers, antioxidant activity significantly decreased with storage (Aquino Bolaños *et al*., 2013), while in broccoli sprouts, an increase in total antioxidant activity as well as total phenol content was observed during storage at 2°C (Leja *et al*. 2001). In addition, the total antioxidant activity reported by these authors was calculated based on fresh weight, but during storage, vegetables and, especially, flowers are leading in water loss, which strongly affects the estimation of antioxidant activity and total phenol concentration.

Total phenol, flavonoids, and carotenoids

The effect of treatments (packaging with or without gaseous composition) and storage time was significant at the 1% probability level on total phenol, flavonoids, and carotenoids of edible marigolds. Still, the interaction effect of treatment and storage time was not significant (Table 1). As the storage time progressed, the content of total phenols, flavonoids, and carotenoids in edible marigolds decreased significantly (Table 5). The highest content of total phenol (61.02 mg g^{-1} petal fresh weight) and flavonoids (13.68 mg g^{-1} petal fresh weight) was related to the polyethylene + polyamide treatment with 5% O_2 and 10% CO_2 gas composition, which compared to the control, 39% and 40% of total phenol and flavonoids contents of marigolds were preserved during the storage period, respectively (Table 5). The highest content of carotenoids (1.05 mg g^{-1} petal fresh weight) of marigolds was also obtained in the polyamide treatment with a gaseous composition of 5% O_2 and 10% CO_2. In comparison, the lowest content of carotenoids (0.93 mg g^{-1} petal fresh weight) was observed in the control treatment (Table 5). Phenolic compounds and flavonoids represent bioactive molecules in edible flowers. The total phenolic content of flowers is highly variable and species-dependent (Barros *et al*., 2020). Our results showed that in all flowers, the concentration of total phenols and flavonoids decreased significantly during the storage period. Still, at each sampling time, the concentration of phenols and flavonoids in packed flowers was always higher than in the control flowers, while the difference between packaging treatments mainly was not significant. Also, the use of the MAP system resulted in better preservation of the content of phenols and flavonoids in packaged flowers. Aquino-Bolaños *et al*. (2013) reported that in edible squash flowers, the concentration of phenolic compounds in air-stored flowers decreased more than in modified atmosphere packaging. Fadda *et al*. (2020) showed that the total phenol concentration of packed flowers was significantly higher than the control and similar to the initial value. Also, Yang *et al*. (2022) reported that the content of total polyphenols in modified polyethylene packages had less change compared to conventional polyethylene packages, and the content of vitamin C was also higher. Carotenoids are essential molecules in marigold petals because they provide color and contribute significantly to their high health properties (Barros *et al*., 2020). Our results showed that despite the decrease in the carotenoid content of marigold flowers with the increase in storage time, the packaging of the flowers kept the carotenoid content compared to the control. Similarly, in edible squash flowers, packaged samples retained most of the primary carotenoid content, while a significant reduction occurred in control flowers (Aquino-Bolaños *et al*., 2013). Also, similar results have been reported by Fadda *et al*. (2020). In addition, the MAP system prevents the loss of carotenoids during the storage period by maintaining a high humidity level inside the packages.

CONCLUSION

In general, the use of modified atmosphere packaging technology, regardless of the type of gaseous composition, significantly prevents weight loss, and ionic leakage, maintaining RWC and increasing antioxidant capacity, which preserves the visual and nutritional quality and prevents the decay of the edible marigold flowers stored at 4°C for 15 days. In comparison, the control flowers were facing a severe drop in quality. The results obtained from this research showed that among all the treatments, the combination treatment of O_2 5% + CO_2 10% along with polyethylene + polyamide coating had better effects in maintaining the quality and increasing the shelf life of edible marigold flowers. As a result, considering the nutritional and medicinal value of the edible marigold flowers, this treatment can be used as an effective and safe method to increase the shelf life and maintain the appearance quality and biochemical compounds of this plant.

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