

EFFECT OF MODIFIED ATMOSPHERE PACKAGING (MAP) ON THE SHELF LIFE AND QUALITY OF GLUTEN-FREE VEGAN CARROT CAKE

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The objective of this study was to investigate the effect of different packaging gas compositions: air (AIR), 100 % N₂ (inert atmosphere, INE), and 30 % CO₂/70 % N₂ (modified atmosphere, MAP), on the quality of gluten-free vegan carrot cake stored at room temperature (20 ± 2 °C) for 35 days. Physicochemical properties, microbiological stability, and sensory characteristics of the cakes were assessed. Among the tested conditions, the MAP treatment showed the most favorable overall performance. Sensory evaluation indicated that MAP-packaged samples achieved the highest scores (4.5 out of 5) even after 35 days of storage. Microbiological stability remained within acceptable limits (< 3 log colony-forming units, cfu/g sample) under both MAP and INE, while AIR-packaged samples exhibited faster spoilage. Moisture loss was significantly reduced under nitrogen packaging (12.21–15.51 %) compared with air-packaged samples (10–25 %), further indicating improved product stability. These results demonstrate that the composition of packaging gases plays a crucial role in maintaining product quality and stability. This study underscores the significant potential of MAP for the development of new plant-based, gluten-free bakery products with extended shelf life and preserved quality.

Keywords: gluten-free vegan cake; moisture; texture; color values; microbiological status; sensory acceptance

ВЛИЈАНИЕ НА ПАКУВАЊЕТО ВО МОДИФИЦИРАНА АТМОСФЕРА (МАР) ВРЗ РОКОТ НА ТРАЕЊЕ И КВАЛИТЕТОТ НА БЕЗГЛУТЕНСКИ ВЕГАНСКИ КОЛАЧ ОД МОРКОВ

Целта на ова истражување беше да се испита ефектот на различните состави на гасовите за пакување: воздух (AIR), 100 % N₂ (инертна атмосфера, INE) и 30 % CO₂/70 % N₂ (модифицирана атмосфера, MAP), врз квалитетот на веганскиот колач од моркови без глутен, складиран на собна температура (20 ± 2 °C) во тек на 35 дена. Беа оценети физичкохемиските својства, микробиолошката стабилност и сензорните карактеристики на колачите. Меѓу тестираните услови, третманот со MAP се покажа како најповолен. Сензорното оценување покажа дека примероците спакувани со MAP постигнаа највисоки резултати (4,5 од 5) дури и по 35 дена складирање. Микробиолошката стабилност остана во рамките на прифатливите граници (< 3 log колонии-формирачки единици, cfu/g примерок) и под MAP и под INE, додека примероците спакувани со AIR покажаа побрзо расипување. Губиток на влага беше значително намален во услови на пакување со N₂ (12,21–15,51 %) во споредба со примероците спакувани со воздух (10–25 %), што дополнително

укажува на подобрена стабилност на производот. Овие резултати покажуваат дека составот на гасовите за пакување игра клучна улога во одржувањето на квалитетот и стабилноста на производот. Оваа студија го нагласува значајниот потенцијал на MAP за развој на нови пекарски производи без глутен на растителна основа со продолжен рок на траење и зачуван квалитет.

Клучни зборови: вегански колач без глутен; влага; текстура; вредности на боја; микробиолошки статус; сензорна прифатливост

1. INTRODUCTION

Consumer demand for gluten-free and vegan foods has increased markedly in recent years, extending beyond individuals with medically diagnosed gluten-related disorders to a broader population motivated by health, ethical, and sustainability considerations.^{1,2} Consequently, the global "free-from" food market continues to expand rapidly, creating strong incentives for the development of innovative gluten-free bakery products.³ The global market for gluten-free cakes and pastries is projected to grow at a compound annual growth rate (CAGR) of 7.5 % from 2024 to 2030.^{4,5} Despite this demand, gluten-free baked goods remain technologically challenging to produce. The absence of gluten compromises structure, elasticity, and moisture retention, often resulting in inferior texture and accelerated staling compared with conventional products.^{6,7} These limitations are particularly pronounced in cakes, where high moisture content and starch-rich formulations contribute to rapid quality deterioration and reduced shelf life.⁸ The shelf life of gluten-free bakery products is strongly influenced by formulation, water activity, packaging conditions, and storage environment.⁹ In response to consumer demand for clean-label foods, increasing attention has been given to natural preservation strategies, including the use of essential oils with antimicrobial and antioxidant properties.¹⁰ When combined with modified atmosphere packaging (MAP), these natural compounds may provide synergistic protection against microbial spoilage while maintaining sensory quality.¹¹

This study investigated a novel gluten-free carrot cake formulated without refined sugar, eggs, or dairy. This aim was to evaluate the effectiveness of different packaging gas compositions (air, 30 % carbon dioxide (CO₂)/70 % nitrogen (N₂), and 100 % N₂) in extending shelf life to at least 30 days at ambient temperature (20 ± 2 °C), to assess physicochemical stability, including moisture and texture changes, as well as to determine the microbiological safety and sensory acceptability of the packaged cakes throughout the storage period. These findings advance

research into plant-based bakery products and support the effective use of MAP to extend shelf life and preserve the quality of clean-label gluten-free cakes.

2. EXPERIMENTAL

2.1. Materials

Most of the ingredients used in the gluten-free carrot cake were supplied by the company Aronija Zdrava Hrana DOOEL (Biocosmos, Veles, North Macedonia). Lemons were sourced from Fresh&Co (Gevgelija, North Macedonia). Cinnamon essential oil (*Cinnamomum cassia*) was purchased from Fitofarm (Skopje, North Macedonia). Clove essential oil, a blend of clove oil, clove bud oil, and sunflower oil, marketed under the BioVitals brand, was sourced from Alfazen Koz. ve Gıda San. Tic. Ltd. Şti. (Bağcılar, Istanbul, Türkiye). Carbon dioxide gas (CO₂, 99.7 %) and nitrogen gas (N₂, 99.99 %) were supplied by TSG Technical Gases AD in Skopje, North Macedonia. Polyethylene terephthalate (PET) trays and 0.04 mm two-layer biaxially oriented polypropylene (BOPP) films were purchased from Kiro Dandarо (Bitola, North Macedonia).

2.2. Preparation of gluten-free vegan carrot cake

The formulation of the gluten-free carrot cake is shown in Table 1. Raw hazelnuts and dried carrots were ground to fine particles (about 1–2 mm) using a high-speed blender (NutriBullet®, Homeland Housewares LLC, Los Angeles, CA, USA, 20.000 rpm). Fresh lemon juice was extracted from lemons with an electric citrus press (Philips HR2737/70, Amsterdam, Netherlands). The dry ingredients (gluten-free flour blend, ground hazelnuts, ground dried carrots, psyllium husk, baking soda, baking powder, and salt) and the liquid ingredients (agave syrup, apple purée, sunflower oil, and lemon juice) were prepared separately. The liquid phase was gradually added to the dry mixture using a kitchen mixer (Vivax HM-200W, Shenzhen, China), set to speed setting 3, mixed continuously for 4 minutes. The cinnamon and clove essential oils were added

to the batter during mixing to ensure uniform distribution. The prepared batter was poured into a 33 cm × 53 cm × 2 cm stainless steel baking mold in 1540 g portions. Cakes were baked in a preheated industrial

convection oven with forced-air ventilation (UNOX XVC705E, UNOX S.p.A., Cadoneghe, Italy) at 160 °C for 24 min.

Table 1

Ingredients of gluten-free vegan carrot cake

Ingredient	w/%
Organic light agave syrup	29.65
Organic gluten-free flour mix ¹	25.23
Unsweetened apple purée	17.67
Refined sunflower oil	11.98
Raw hazelnuts	8.2
Dried carrot	3.15
Organic psyllium husk	1.89
Lemon juice	0.88
Organic baking soda	0.88
Baking powder	0.38
Cinnamon essential oil	0.05
Clove essential oil	0.03
Salt	0.01

¹Gluten-free flour mix composition: organic unmodified corn starch (43.5 %), organic unmodified potato starch (35.7 %), organic rice flour (14.0 %), organic buckwheat flour (6.2 %), xanthan gum (0.6 %).

2.3. Packaging and gluten-free vegan carrot cake

After baking and cooling at room temperature (20 ± 2 °C) for 120 min, the carrot cakes were cut into 9 cm × 2 cm × 3 cm pieces (± 5 %) and placed in PET trays measuring 13 cm × 3.5 cm × 3.5 cm. These trays were sealed with 52 ± 3 μm BOPP film using a gas-injection packaging machine (DXDZ-320B series, Hualian Machinery, Wenzhou, China), creating a pillow-type package. Three packaging conditions were used: (i) ambient air (control, AIR); (ii) 30 %

CO₂/70 % N₂ (modified atmosphere, MAP); and (iii) 100 % N₂ (inert atmosphere, INE). The product-to-headspace ratio was maintained at 1:2. The sealed samples were stored in a dark chamber at 20 ± 2 °C and 45–55% relative humidity, simulating typical ambient storage for packaged bakery products. The visual appearance of the gluten-free vegan carrot cake on top view, side cross-section of cake samples in PET trays, and sealed packaged cake samples, is presented in Figure 1. Samples were stored for 35 days at ambient temperature (20 ± 2 °C).

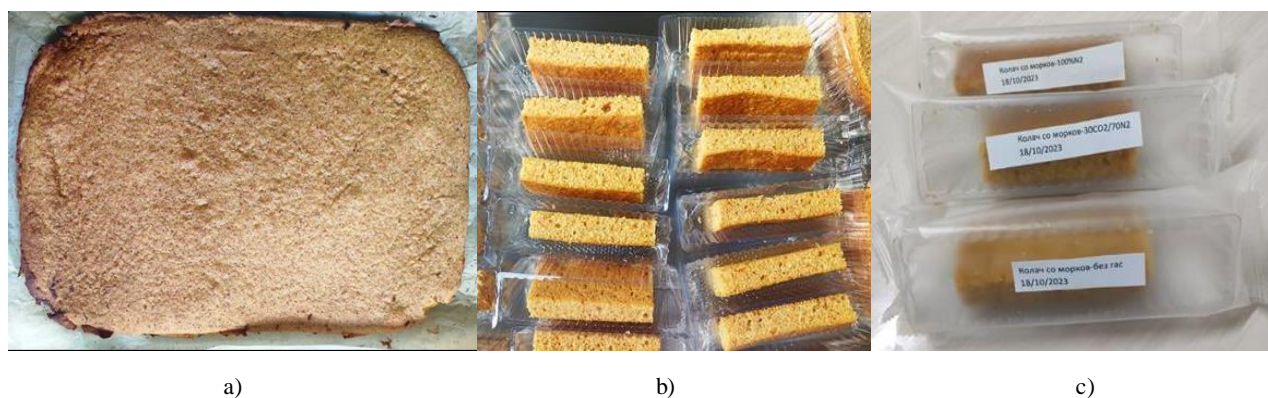


Fig. 1. Gluten-free vegan carrot cake: a) top, b) side-cross section of the cake placed in PET trays, and c) sealed and labelled packed cake samples

2.4. Gluten determination

Gluten assessment in the cakes was performed in triplicate ($n = 3$) using the GlutenTox Home rapid test (Biomedal S. L., Seville, Spain), with detection thresholds of 5 mg/kg and 20 mg/kg, corresponding to the regulatory limits for gluten-free and low-gluten products, respectively.

2.5. Determination of water activity (a_w) and moisture content

A portable water activity meter (WA-160A, Amittari Instruments Co., Shenzhen, China) was used to measure a_w at a temperature of 25 °C. Moisture content was determined according to the AOAC 925.10 standard method¹² by drying at 105 °C to constant mass. Measurements were performed in triplicate ($n = 3$).

2.6. Texture measurement

Texture properties (firmness, N; springiness, %) were determined by using a TA.XT Plus texture analyzer equipped with software Exponent v6.1 software (Stable Micro Systems, Godalming, UK). Cakes cut into cubes (9 cm × 2 cm × 3 cm) were compressed into four replicates per sample ($n = 4$) with a P/20 cylindrical probe at a constant deformation speed of 1.0 mm/s to a penetration depth of 10 mm (50 % sample compression) and 5 s relaxation time.

2.7. Color measurement

Color was measured in the Commission Internationale de l'Eclairage (CIE) $L^*a^*b^*$ color space with a tristimulus colorimeter (Dr. Lange Spectracolor, Washington, USA) at 8 mm aperture, D65 illumination, and a standard observation angle of 2°. The instrument was calibrated using a standard white plate ($L^* = 95.93$, $a^* = -0.19$, $b^* = 3.12$). Surface color was determined at five different spots on each side, resulting in a total of ten repetitions ($n = 10$). Based on L^* , a^* , and b^* values, hue angle (h^*), chroma (C^*), and browning index (BI) were calculated according to Equations (1), (2), and (3), respectively. The color difference of the cake surface between the 1st and 35th days of storage was calculated according to Equation (4).

$$h^* = \tan^{-1} \frac{b^*}{a^*} \quad (1)$$

$$C^* = \sqrt{((a^*)^2 + (b^*)^2)} \quad (2)$$

$$BI = 100 * \frac{x - 0.31}{0.172}$$

$$\text{where } x = \frac{a^* + 1.75L^*}{5.645L^* + a^* - 3.012b^*} \quad (3)$$

$$\Delta E^*_{ab} = \sqrt{((L^*_1 - L^*_2)^2 + (a^*_1 - a^*_2)^2 + (b^*_1 - b^*_2)^2)} \quad (4)$$

2.8. Microbiological assessment

Microbiological quality was evaluated by determining the levels of aerobic mesophilic bacteria (MKC EN ISO 4833-1:2013),¹³ molds and yeasts (MKC EN ISO 21527-2:2008),¹⁴ and the presence of *Salmonella* spp. (MKC EN ISO 6579-1:2017)¹⁵ and *Listeria monocytogenes* (MKC EN ISO 11290-1:2017).¹⁶ Microbiological analyses were performed in triplicate ($n = 3$) in an accredited laboratory (FoodLab, Skopje, North Macedonia). Results were expressed as log cfu (colony-forming units)/g of sample.

2.9. Sensory evaluation

A sensory evaluation was conducted with 45 untrained and trained panelists (36 % men, 64 % women, aged 20–60 years). The panel consisted of students (40 %, aged 20–25) and staff members (30 %, aged 30–60) from the Faculty of Technology and Technical Sciences (Veles, North Macedonia) and the Faculty of Technology and Metallurgy (Skopje, North Macedonia), as well as employees (30 %, aged 30–60) of Biocosmos (Veles, North Macedonia). Cake samples were served on white disposable plates and coded with three-digit random numbers, together with water for palate cleansing between samples. A five-point hedonic scale (1 = dislike very much, 5 = like very much) was used to evaluate appearance, odor, color, taste, texture, mouthfeel, and overall acceptability. Different assessor groups were used on days 1 and 35 to ensure independent evaluations and to minimize bias.

2.10. Statistical analysis

All experiments were performed at least in triplicate. The results are expressed as mean ± standard deviation (SD). Statistical analysis was conducted using one-way analysis of variance (ANOVA), followed by Tukey's post hoc test to determine significant differences among the samples at a significance level of $p < 0.05$. Minitab

v 17.1 Statistics Software (Minitab LLC, State College, PA, USA)¹⁷ was used.

3. RESULTS AND DISCUSSION

3.1. Gluten content

According to European Union Commission Regulation (EC) No 41/2009,¹⁸ food products containing less than 20 mg/kg of gluten may be labelled as "gluten-free", while those with a gluten level between 20 and 100 mg/kg may be classified as "very low gluten". In the analyzed vegan carrot cake samples, gluten was not detected at either of the tested concentrations (5 mg/kg and 20 mg/kg).

These findings confirm compliance with gluten-free labelling requirements and suggest that no cross-contamination occurred during production.

3.2. Water activity and moisture content

Water activity (a_w) and moisture content are key physicochemical parameters that significantly influence the quality of bakery products, particularly in terms of texture, microbial stability, and shelf life. Their changes during storage reflect complex interactions among water redistribution, starch retrogradation, protein–water interactions, and packaging permeability.^{19,20}

Table 2

Water activity (a_w) and moisture content in gluten-free vegan carrot cake packaged in different gas atmosphere conditions

Storage (days)	Packaging conditions					
	AIR	MAP	INE	AIR	MAP	INE
	a_w			Moisture content (%)		
1	(0.71±0.01) ^{aC}	(0.71±0.01) ^{aB}	(0.71±0.00) ^{aC}	(22.67±0.38) ^{aA}	(22.67±0.38) ^{aA}	(22.67±0.38) ^{aA}
7	(0.76±0.01) ^{abB}	(0.78±0.02) ^{aA}	(0.74±0.01) ^{bB}	(20.40±0.35) ^{aB}	(20.30±0.36) ^{aB}	(19.90±0.26) ^{aB}
15	(0.77±0.00) ^{aB}	(0.77±0.01) ^{aA}	(0.74±0.01) ^{bB}	(18.50±0.10) ^{aC}	(20.00±0.10) ^{bB}	(19.80±0.30) ^{cB}
25	(0.79±0.01) ^{aA}	(0.77±0.01) ^{bA}	(0.78±0.01) ^{abA}	17.37±0.21) ^{aD}	(19.27±0.32) ^{aC}	19.70±0.53) ^{bB}
35	(0.77±0.01) ^{aB}	(0.75±0.01) ^{abA}	(0.76±0.01) ^{bB}	(17.00±0.20) ^{aD}	(19.00±0.36) ^{bC}	(19.15±0.09) ^{cB}

Mean value ($n = 3$) ± standard deviation (SD). Lowercase letters show significant differences in a row among the packaging type (Tukey test, $p < 0.05$). Uppercase letters show differences in a column across the storage time (Tukey test, $p < 0.05$). AIR – atmosphere condition. MAP – 30 % CO₂/70 % N₂. INE – 100 % N₂.

The effects of storage time and packaging conditions on a_w and moisture content in vegan carrot cake samples are shown in Table 2. During storage, a_w increased significantly ($p < 0.05$) in all packaging treatments, whereas total moisture content decreased progressively. This inverse relationship suggested internal water redistribution and structural relaxation phenomena within the product matrix. In gluten-free matrices, the water immobilization capacity was reduced due to the absence of a continuous gluten network. Amylopectin recrystallization and matrix consolidation reduced the availability of hydrogen-bonding sites, leading to the release of previously bound water into more mobile fractions.^{8,21}

On day 1, no significant differences ($p > 0.05$) in a_w (0.71) or moisture content (22.67 %) were observed among AIR, MAP, and INE treatments. MAP-packaged samples showed the highest a_w (0.78) on day 7. This increase was attributed to the dissolution of CO₂ into the aqueous phase of the product, resulting in the formation of carbonic acid

and a subsequent reduction in pH. The acidification of the matrix may have altered protein–polysaccharide interactions and weakened water-binding capacity, thereby facilitating redistribution of water from bound to more mobile states.²²

Samples packaged under INE maintained significantly lower a_w values (0.74) up to day 15, followed by an increase to 0.78 on day 25. In contrast, AIR-packaged samples demonstrated significantly higher a_w values than MAP and INE treatments during prolonged storage, reaching a maximum value of 0.79 on day 25. These results indicated enhanced water migration under aerobic conditions, accompanied by accelerated structural deterioration and product ageing.

For AIR-packaged cake samples, water content decreased significantly ($p < 0.05$) from day 1 to day 25, while no significant difference ($p > 0.05$) was observed between days 25 and 35. MAP samples exhibited significant changes ($p < 0.05$) only up to day 15, with no further statistically significant differences

in moisture through day 35. In INE samples, a significant decrease ($p < 0.05$) occurred between days 1 and 7, followed by no further significant changes ($p > 0.05$) to day 35 of storage.

Compared to day 1, the moisture losses after 35 days were highest in AIR samples (10–25 %), followed by MAP (10.44–16.88 %), and lowest in INE (12.21–15.51 %). This comparison confirmed that INE packaging provided the greatest overall moisture stability after the initial storage period, demonstrating improved water retention compared with MAP and AIR packaging systems.

Similar trends regarding the decrease in moisture during storage have been reported by Saeidi et al.²³ in gluten-free muffins enriched with sugar beet fiber and Wu et al.²⁴ in rice cakes stored for 21 days. Li et al.⁸ also reported the highest moisture retention in nitrogen-packaged low-gluten sponge cake, which aligned with the present findings. However, unlike the current results, these authors observed a gradual decrease in a_w during storage. This discrepancy may

have been attributed to differences in formulation, particularly between gluten-containing and gluten-free systems, as well as variations in fiber content, porosity, and packaging permeability. Gluten-free products enriched with plant fibers exhibited distinct water-binding behavior and internal water redistribution mechanisms, which may have contributed to increasing a_w values despite an overall reduction in moisture content.^{25,26}

3.3. Texture

The textural profile of bakery products is a crucial quality attribute that strongly influences consumer acceptance, particularly in taste perception.²⁷ The evolution of texture parameters during storage clearly demonstrated that the packaging atmosphere significantly influenced the crumb-firming kinetics and structural resilience of the gluten-free vegan carrot cake (Table 3).

Table 3

Firmness and springiness of gluten-free vegan carrot cake packaged in different gas atmosphere conditions

Storage (days)	Packaging conditions					
	AIR		MAP		INE	
	Firmness (N)			Springiness (%)		
1	(1526.32±127.75) ^{aA}	(1358.60±155.86) ^{abA}	(1236.05±77.31) ^{bB}	(28.22±0.58) ^{bA}	(27.22±1.85) ^{bA}	(26.29±2.21) ^{aA}
7	(1759.62±257.28) ^{aA}	(1587.90±120.76) ^{aA}	(1953.19±396.77) ^{aAB}	(29.77±0.52) ^{aA}	(26.48±2.27) ^{aAB}	(27.37±2.02) ^{aAB}
15	(1810.12±162.73) ^{aA}	(1997.88±421.68) ^{aA}	(1846.57±292.54) ^{aA}	(31.19±0.94) ^{aA}	(27.30±1.42) ^{aAB}	(26.02±2.37) ^{aAB}
25	(2239.51±212.59) ^{aB}	(2595.70±316.08) ^{aB}	(2243.79±265.08) ^{aA}	23.90±2.44) ^{aAB}	(24.38±0.30) ^{aB}	(25.29±2.41) ^{aB}
35	(2450.68±148.68) ^{aB}	(2589.41±101.71) ^{aB}	(2338.22±368.68) ^{aA}	(21.22±3.47) ^{aB}	(24.68±1.40) ^{aB}	(26.16±4.18) ^{aAB}

Mean value ($n = 4$) ± standard deviation (SD). Lowercase letters show significant differences in a row among the packaging type (Tukey test, $p < 0.05$). Uppercase letters show differences in a column across the storage time (Tukey test, $p < 0.05$). AIR – atmosphere condition. MAP – 30 % CO₂/70 % N₂. INE – 100 % N₂.

Firmness of the gluten-free vegan cake increased significantly during storage across all packaging treatments. The firmness increased significantly ($p < 0.05$) in AIR- and MAP-packaged samples from day 1 to day 25, and to day 15 in INE samples, followed by no further significant changes until day 35.

On day 1, packaging type significantly affected firmness ($p < 0.05$). The highest firmness was determined in AIR (1526.32 ± 127.75 N), followed by MAP (1358.60 ± 155.86 N) and INE (1236.05 ± 77.31 N). No significant differences among AIR, MAP, and INE samples were observed from day 7 onward ($p > 0.05$).

By day 35, all samples reached similarly high firmness values, AIR (2450.68 ± 148.68 N), MAP (2589.41 ± 101.71 N), and INE (2338.22 ± 368.68 N), indicating that packaging type had no significant long-term effect on textural evolution.

Springiness decreased overall during storage ($p < 0.05$). In AIR-packaged samples, springiness increased slightly from day 1 to day 15, then declined significantly at days 25 and 35 ($p < 0.05$). MAP samples remained relatively stable through day 15 but showed significantly lower springiness at days 25 and 35 compared with day 1 ($p < 0.05$). INE-packaged samples exhibited the highest stability, with no significant changes in springiness throughout storage ($p > 0.05$).

On day 1, springiness values were significantly higher ($p < 0.05$) in AIR (28.22%) and MAP (27.22 %) samples than in INE-packaged samples (26.16 %). After 7 days of storage, no significant differences among packaging treatments were observed ($p > 0.05$). By day 35, springiness decreased to 21.22 % in AIR, 24.68 % in MAP, and 26.16% in INE-packaged samples.

Changes in firmness and springiness in the studied vegan gluten-free carrot cake were comparable to those reported by Wyrwiz et al.²⁸ for high β -glucan gluten-free cakes packaged in PET under modified atmosphere conditions with different gas compositions, including 100 % N₂, 60 % CO₂/40 % N₂, and approximately 78 % N₂/21 % O₂/0.04 % CO₂, during 14 days of storage.

Wei et al.²⁹ reported a significant negative correlation ($p < 0.05$) between moisture content and firmness in rice cakes packaged in PET under ambient air conditions and stored for 28 days at 25 °C, which is comparable to our results for moisture content (Table 2) and firmness (Table 3).

The increase in firmness and reduction in springiness during storage, generally characteristic of gluten-free high-moisture bakery products, are mainly associated with starch retrogradation, moisture redistribution, and weakening of the starch-hydrocolloid network. These effects are more pronounced in gluten-free systems because structural stability depends primarily on interactions among

starch, hydrocolloids, and dietary fiber rather than gluten viscoelasticity.³⁰

The greater increase in firmness and reduction in springiness observed in MAP-packaged samples during prolonged storage may be associated with partial dissolution of CO₂ in the aqueous phase, resulting in pH changes, altered protein-starch interactions, and enhanced moisture migration within the crumb structure.²² In contrast, samples packaged under 100 % N₂ maintained lower firmness and higher springiness retention, likely due to reduced oxidative reactions and better preservation of moisture-binding capacity and crumb plasticity.^{31,32}

3.4. Color parameters

The color characteristics of the gluten-free vegan carrot cake throughout storage are shown in Table 4.

A significant decrease in L* values was observed in all packaging treatments during storage ($p < 0.05$), indicating progressive darkening of the cake surface. On day 1, MAP-packaged samples showed significantly lower L* values than INE samples ($p < 0.05$), while AIR samples did not differ significantly ($p > 0.05$). By day 35, the highest L* value was retained in MAP-packaged samples (50.90), followed by INE (49.42) and AIR samples (47.22), suggesting better lightness retention under modified atmosphere conditions.

Table 4

Colour parameters of gluten-free vegan carrot cake packaged in different gas atmosphere conditions at 1st and 35th day storage

Colour parameters	Packaging conditions						ΔE^*		
	AIR	MAP	INE	AIR	MAP	INE	AIR	MAP	INE
	Storage (day)								
1 st			35 th						
L*	(57.94±0.89) ^{ab}	(55.74±2.52) ^b	(59.50±1.41) ^a	(47.22±1.73) ^b	(50.90±1.01) ^a	(49.42±1.63) ^{ab}			
a*	(16.70±0.30) ^a	(17.54±1.58) ^a	(16.56±0.95) ^a	(14.25±1.26) ^a	(10.93±0.33) ^b	(11.04±0.40) ^b			
b*	(45.58±6.32) ^a	(46.75±1.14) ^a	(44.67±7.56) ^a	(32.33±1.92) ^a	(24.17±2.88) ^b	(26.26±2.00) ^b	17.22	24.02	21.70
h*	(1.22±0.04) ^a	(1.21±0.03) ^a	(1.21±0.08) ^a	(1.15±0.05) ^a	(1.14±0.04) ^a	(1.17±0.02) ^a			
C*	(48.57±6.00) ^a	(49.96±1.08) ^a	(47.74±6.76) ^a	(35.37±1.43) ^a	(26.55±2.70) ^b	(28.49±1.97) ^b			
BI	(162.68±32.53) ^a	(177.47±15.62) ^a	(153.43±35.38) ^a	(134.44±3.78) ^a	(84.41±12.29) ^b	(95.48±9.22) ^b			

Mean value ($n = 10$) \pm standard deviation (SD). Lowercase letters show significant differences in a row among the packaging type (Tukey test, $p < 0.05$). AIR – atmosphere condition. MAP – 30 % CO₂/70 % N₂. INE – 100 % N₂. *The colour difference of the cake surface between the 1st and 35th days of storage.

No significant differences in a^* , b^* , h^* , C^* , and BI values were observed among packaging treatments on day 1 ($p > 0.05$). During storage, significant decreases ($p < 0.05$) in a^* , b^* , C^* , and BI values were observed in MAP- and INE-packaged samples, whereas changes in AIR-packaged samples were not significant ($p > 0.05$). The reduction in a^* and b^* values indicated decreased redness and yellowness, while lower C^* values reflected reduced color saturation during storage. In contrast, h^* values remained stable throughout storage in all treatments ($p > 0.05$), showing preservation of the characteristic orange–brown hue of the carrot cake.

The browning index (BI) associated with Maillard-derived brown pigments decreased significantly in MAP- and INE-packaged samples by day 35 ($p < 0.05$). No significant changes in BI were determined in AIR-packaged samples ($p > 0.05$).

The total color difference (ΔE^*) values between day 1 and day 35 were above 3 in all packaging treatments, indicating color changes clearly perceptible to the human eye. The highest ΔE^* value was observed in MAP-packaged samples (24.02), followed by INE (21.70) and AIR (17.22). Comparable findings regarding decreasing chromatic parameters during 8 days of storage of gluten-free cakes supplemented with carrot flour were reported by Halim et al.³³

The decreased values of surface color parameters may be related to carotenoid degradation, moisture redistribution, and structural changes that affect light scattering in the crumb matrix. Primarily, oxygen promotes pigment degradation, while dissolved CO_2 in MAP systems may affect pH and moisture distribution, thereby accelerating non-enzymatic browning.

Gas composition optimization alone may not be sufficient to preserve the long-term visual quality of gluten-free vegan baked goods; therefore, additional strategies, such as incorporating antioxidants, may be required.^{34–36}

3.5. Microbiological quality

In Table 5, the results of the microbiological analysis of gluten-free vegan cake samples packaged under AIR, MAP, and INE conditions are presented. *Salmonella* spp. and *Listeria monocytogenes* were not detected in any of the samples,

while yeast and mold counts remained below 1 log cfu/g throughout the 35-day storage period. On day 1, total bacterial counts were below the detection limits (< 1 log cfu/g). A slight increase in total bacterial counts was observed on day 7, highest in MAP-packaged samples (1.45 log cfu/g), followed by AIR-packaged (1.04 log cfu/g) and INE-packaged (1.05 log cfu/g) samples.

The total bacterial counts in AIR-packaged samples increased significantly ($p < 0.05$) from 2.25 log cfu/g on day 15 to 3.48 log cfu/g on day 35 of storage, indicating that oxygen availability promoted microbial proliferation. For MAP-packaged samples, total bacterial counts increased significantly ($p < 0.05$) from day 7 to day 25 (2.40 log cfu/g). An insignificant difference ($p > 0.05$) was observed between days 25 and 35 (2.45 log cfu/g), suggesting that CO_2 -enriched atmospheres delay microbial growth during prolonged storage.

The findings for MAP-packaged samples are comparable to those reported by Djenane et al.³⁶ who suggested that microbial growth may be supported by residual O_2 trapped within the numerous pores of the cake matrix, despite the extremely low O_2 concentration in the package headspace (approx. 0.01%).

In INE-packaged samples, total bacterial counts changed significantly throughout storage ($p < 0.05$), reaching the highest value at day 15 (2.49 log cfu/g), followed by a gradual decrease to 1.66 log cfu/g on day 35. Nitrogen, an inert gas, restricts oxygen availability, and inhibits the growth of aerobic microorganisms, thus maintaining the freshness of products.²⁶ The total bacterial counts remained within acceptable limits for bakery products during most of the storage period (< 3 log CFU/g), according to the Rulebook on Special Requirements Related to Microbiological Criteria for Food,³⁷ indicating satisfactory microbiological quality and safety of the gluten-free vegan carrot cake under the applied packaging conditions.

The obtained data on microbial status align with previous studies showing that oxygen-restricted environments inhibit bacterial and fungal growth in gluten-free cakes and bakery products, highlighting the importance of packaging atmosphere for extending freshness and shelf life.^{29,37}

Table 5

Microbiology in gluten-free vegan carrot cake packaged in different gas atmosphere conditions

Storage (days)	Packaging conditions					
	AIR	MAP	INE	AIR	MAP	INE
	Total bacterial count (log cfu/g)			Moulds and yeasts (log cfu/g)		
				Salmonella (ND in 25 g)		
				Listeria monocytogenes (ND in 25 g)		
1	(< 1)	(< 1)	(< 1)			
7	(1.04 ± 0.04) ^{aA}	(1.45 ± 0.15) ^{bA}	(1.05 ± 0.06) ^{aA}	(< 1)	(< 1)	(< 1)
15	(2.25 ± 0.05) ^{bB}	(1.99 ± 0.04) ^{cB}	(2.49 ± 0.03) ^{aB}	ND	ND	ND
25	(2.77 ± 0.02) ^{cC}	(2.40 ± 0.03) ^{bC}	(2.26 ± 0.02) ^{cC}	ND	ND	ND
35	(3.48 ± 0.04) ^{dD}	(2.45 ± 0.02) ^{bC}	(1.66 ± 0.05) ^{cD}			

Mean value ($n = 3$) ± standard deviation (SD). Lowercase letters show significant differences in a row among the packaging type (Tukey test, $p < 0.05$). Uppercase letters show differences in a column across the storage time (Tukey test, $p < 0.05$). AIR – atmosphere condition. MAP – 30 % CO₂/70 % N₂. INE – 100 % N₂. ND-not detected.

3.6. Sensory attributes

Sensory evaluation of the gluten-free vegan carrot cake was conducted for the AIR-packaged sample on day 1 and for MAP and INE samples after 35 days of storage. The AIR-packaged sample on day 35 was excluded due to increased aerobic mesophilic bacterial counts above acceptable limits.³⁷ Figure 2 presents the sensory evaluation

results for appearance, color, aroma, taste, texture, mouthfeel, and overall acceptability.

The sensory evaluation showed that the AIR-packaged cake at day 1 exhibited the highest scores across almost all attributes, with appearance (4.3), texture (4.4), color (4.5), aroma (4.4), taste (4.3), mouthfeel (4.5), and overall acceptability (4.5), reflecting the expected sensory quality of a freshly baked product.

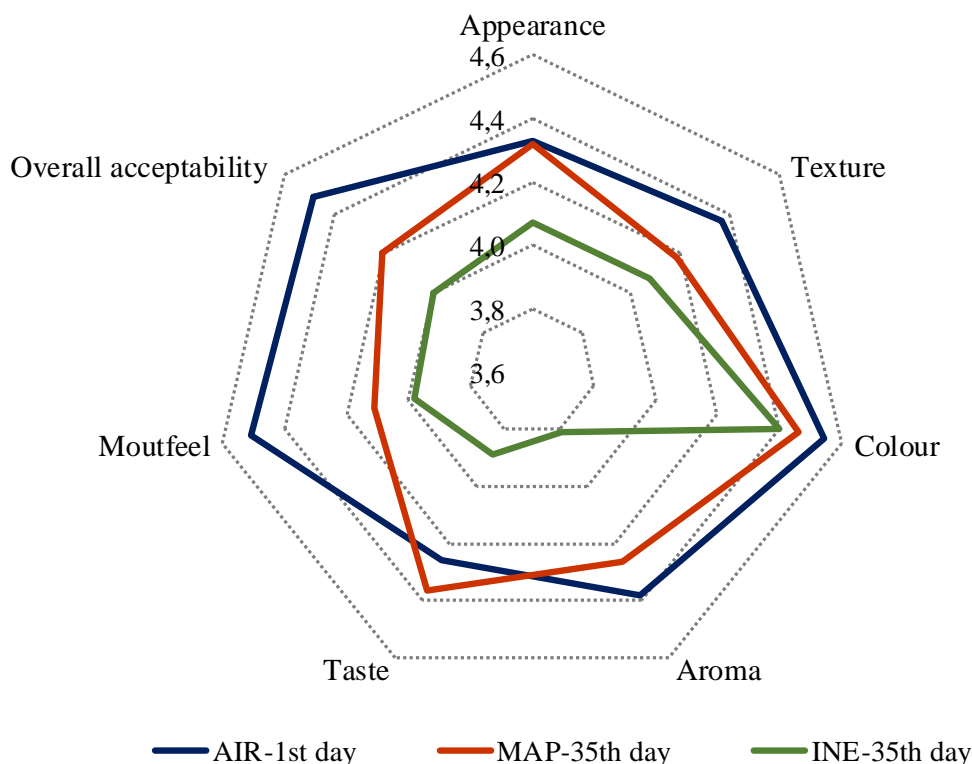


Fig. 2. Radar graph of the sensory evaluation of gluten-free vegan carrot cake packed in AIR (1st day), MAP (35th day) and INE (35th day)

After 35 days of storage, MAP-packaged samples maintained high sensory ratings, with values ranging from 4.2 to 4.5 for most attributes, indicating effective preservation of sensory quality under modified atmosphere conditions. In comparison, INE-packaged samples showed slightly lower but still acceptable scores, ranging from 3.8 to 4.4, suggesting a minor reduction in aroma and taste intensity, while overall acceptability remained at a good level (4.0). Overall, both MAP and INE treatments effectively preserved the sensory quality of the gluten-free vegan carrot cake during 35 days of storage at 20 ± 2 °C, with MAP showing slightly better retention of aroma and taste. The obtained sensory results are comparable to those reported in the literature,^{28,36,38} which observed high consumer acceptability during storage of bakery products packaged under modified atmosphere conditions. Thus, modified atmosphere packaging can be proposed as an effective method for extending the sensory shelf life of gluten-free vegan bakery products by preserving key quality attributes such as texture, flavor, and overall acceptability.

4. CONCLUSION

The application of MAP (30% CO₂/70% N₂) and INE (100% N₂) packaging instead of air significantly extended the shelf life of gluten-free vegan carrot cake during 35 days of storage at room temperature. MAP was more effective at preserving sensory quality and textural properties, while INE packaging provided better moisture retention and microbial stability than to ambient air packaging.

The novelty of the developed gluten-free carrot cake lay in the carefully selected ingredients, each providing specific functional and nutritional properties within the gluten-free matrix. Organic agave syrup and apple purée acted as natural sweeteners and moisture-retaining agents, while psyllium husk improved structure, water binding, and texture in the absence of gluten, dairy, and eggs. Dried carrot and hazelnuts enhanced the nutritional profile in terms of dietary fiber, minerals, and bioactive compounds. Cinnamon and clove essential oils served as natural flavoring agents with potential antioxidant and antimicrobial effects.

The study findings highlighted the potential of combining optimized plant-based formulations with modified atmosphere packaging (MAP) to develop clean-label, shelf-stable bakery products with improved nutritional value, without the use of artificial preservatives. A limitation of this study was the evaluation at a single storage temperature. Future research should investigate the effects of at

least one lower temperature (e.g., 4 °C), which is expected to further extend the shelf life of the vegan gluten-free cake. Furthermore, optimization of the modified atmosphere packaging (MAP) composition is recommended.

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