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Proteolytic Development and Volatile Compounds Profile of Domiati Cheese under Modified Atmosphere Packaging

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Abstract: This study explored the impacts of modified atmosphere packaging (MAP) treatment on the proteolytic development and volatile compounds of Domiati cheese during storage. Domiati cheese samples were kept for 75 days at refrigerator temperature, under aerobic packaging (C1) or vacuum (C2). In parallel, other Domiati cheese samples were kept under MAP, at different levels of CO_2 and N_2, as follows: 10% CO_2/90% N_2 (D1), 15% CO_2/85% N_2 (D2), 25% CO_2/75% N_2 (D3), 100% CO_2 (D4), and 100% N_2 (D5). The normal control (C1) treatment showed the highest reduction in pH from 6.64 at zero time to 6.23 and 6.01 after 40 and 75 days of storage, respectively. On the other hand, the under-vacuum samples (C2) showed the lowest reduction in pH, from 6.64 at zero time to 6.49 and 6.28 after 40 and 75 days of storage, respectively. Proteolysis during cheese storage was lower in MAP of cheeses than in the C1 treatment. Total free amino acids (FAAs) were higher in C1 treatment than other cheeses during the whole storage period. The lowest level of total FAA was detected in D4 treatment after 75 days of storage. Volatile acids, aldehydes, ketones, and esters compounds were detected in all treatments during storage, but particularly higher in aerobic packaging than the other treatments after 75 days. The level of each acid compound increased with storage period, and the increases were particularly clear in pentanoic acid, hexanoic acid, heptanoic acid, benzoic acid, and *n*-decanoic acid. The normal control (C1) showed high contents of the different volatile ketone compounds. However, the samples packaged under 100% N_2 (D5) showed the significantly highest levels of all the volatile ketones after 75 days of storage, particularly 2-pentanone, acetoin, methyl isobutyl ketone, 2-heptanone, 2-nonanone, and 2-undecanone. Some important compounds contributing to the good flavor of the cheese are acetic acid, butanoic acid, pentanal, benzaldehyde, acetoin, and 2,3-butanedione. The CO_2 and N_2 treatments exerted significant changes in all groups during the storage of cheese. All cheese samples showed gradual increases in CO_2 co-occurring with parallel decreases in N_2 during refrigerated storage periods, except for D4 treatment (100% CO_2), which showed a decrease. A significant decrease in O_2 level occurred in C1 treatment during cold storage.

Keywords: Domiati cheese; modified atmosphere packaging; proteolytic development; volatile compounds

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1. Introduction

Domiati cheese is the most prevailing white soft cheese in Egypt. It is consumed as fresh or matured. The microbiological properties and safety of Domiati cheese are the main

areas of concern for consumers and producers. Domiati cheese is made of cows' or buffalos' milk or mixtures of them. Its shelf-life is usually limited to a few days due to the product exposure to the atmosphere before packaging. Moreover, due to its high moisture content and the initial pH of 6.0, Domiati cheese is very susceptible to spoilage by yeasts, molds, and coliform bacteria. Thus, along with other soft cheeses, they could be a significant dairy safety concern, showing a large incidence and potential for survival/growth of pathogens and spoilage organisms [1]. They depend on the microorganisms' type introduced from the raw milk, the processing efficiency, and the hygienic practice applied in the dairy plant. The processing of cheese making plays a vital role in microorganisms' reproduction, thus making the product unsuitable for human consumption [2].

Different modified atmosphere packaging (MAP) techniques have created interest in the MAP of dairy and food products to extend the shelf-life and promote quality. These techniques undertake many basic roles, such as improving the product image, reducing microbial growth, preventing chemical degradation, and avoiding the use of preservatives. Moreover, MAP in plastic bags protects against weight loss and dehydration and absorption of undesirable tastes and odors from the atmosphere [3]. Bags are usually packaged with different modified atmospheres (low in O_2 and high in CO_2 and N_2) to minimize microbial growth and retard enzymatic activities [3]. Carbon dioxide gas has a fungistatic and bacteriostatic effect, while nitrogen is primarily an inert gas to prevent food packages from collapsing [3]. However, MAP techniques were shown to affect beneficial microbes involved in sensory characteristics of dairy and food products during storage.

Volatile compounds isolated from Domiati cheese after three months were 1-propanol, 2-butanol, 2-butanone, and acrolein, as well as high amounts in esters. Volatile compounds identified from Feta cheese with the head space technique included relatively high amounts of 2-butanol, 1-propaneol, ethanol, and 2-butanone, as well as low levels of 2-propanol, 2-methyl 1-propanol, pentane, ethyl acetate, and ethyl butyrate [4]. Moreover, various sulfur volatile compounds contribute to taste and odor; most of the flavor compounds appeared to develop during two months of ripening [5]. Various volatile compounds in Domiati cheese originated from the breakdown of proteins, lactose, and lipids by many factors. It is obvious that several interactions between microbes and enzymes occur in this cheese in relation to volatile compound formation [6].

Proteolysis is the primary interaction that takes place throughout the cheese of ripening [7,8], and it is the key factor to promote the diversity in body and flavor [9]. During the primary interaction, residual rennet enzymes, together with milk proteases, hydrolyze casein and, thus, produce big or medium-size peptides. Then, in the secondary interaction of proteolysis, proteins and big peptides are gradually hydrolyzed into lower peptides and amino acids with the aid of intracellular and extracellular enzymes of starter bacteria or other cheese microbials [10]. Such a breakdown of protein networks plays a significant role in textural properties and in the release of FAAs with a key role in production of sapid compounds [11].

Recently, the packaging methods may be a suitable replacement for cheese preservation based on reducing the level of O_2. MAP modifies the atmosphere surrounding the cheese, retarding microorganisms' growth without affecting the cheeses. This research is a continuation of the project to develop the production of Domiati cheese by using MAP methods. Hence, the present study's goal was to investigate the proteolytic development and volatile compounds profile of the Domiati cheese when packaged under vacuum and modified atmosphere during storage at 5 °C for different periods.

2. Materials and Methods

2.1. Materials

Cow and buffalo milk were provided by the herds of the Faculty of Agriculture, Benha University, Egypt, during the seasons of 2020 and 2021. The calf rennet (150 International Milk Clotting Units (IMCU)/mL) used for making cheese was prepared in the laboratory of the Dairy Science Department, Faculty of Agriculture, Benha University. A commercial fine-

grade table salt (NaCl) was provided from El-Nasr Saline's Company, Egypt. Calcium chloride ($CaCl_2$) was obtained from El-Gomhoria Co., Cairo, Egypt. Polyamide/polyethylene (PA/PE; 40/30 microns) was provided from El-Nasr Plastic Company, Cairo, Egypt. Carbon dioxide and nitrogen gases were obtained from Egyptian Gases Co., Cairo, Egypt. In addition, high-purity standard volatile compounds were provided from Sigma (USA): hydroxyacetic acid, acetic acid, butanoic acid, pentanoic acid, hexanoic acid, heptanoic acid, benzoic acid, *n*-decanoic acid, acetaldehyde, pentanal, hexanal, heptanal, benzaldehyde, 2-methylundecanal, nonanal, 2,3-butanedione, 2-pentanone, 3-methyl-2-butanone, acetoin, methyl isobutyl ketone, 4-methyl-2-hexanone, 2-heptanone, acetophenone, 2-undecanone, formic acid ethenyl ester, acetic acid ethenyl ester, ethyl hexanoate, δ-nonalactone, ethyl decanoate, and heptane. All chemicals, solvents, and reagents (analytical grade) were provided from Sigma (St. Louis, MI, USA). The USA supplied the solid-phase microextraction (SPME) fibers from Supelco Inc. (Bellefonte, PA, USA).

2.2. Domiati Cheese Manufacturing

Cheeses were made from pasteurized milk, according to the method noted by Abou-Donia [12], with the following modifications. A fresh milk mixture (cow milk and buffalo milk, 1:1) was heated to 75 °C for 15 min and then cooled to 40 °C. NaCl (4%; w/w) and $CaCl_2$ (0.02%; w/w) were incorporated into the cooled milk before adding the rennet and incubation at 40 °C until complete coagulation. The cheese curd was put in molds (cylindrical stainless steel; about $\frac{1}{2}$ kg capacity) and left overnight to drain whey by gravity. On the second day, the molds were turned upside down and left for 4 hours. The molds were then taken off, and the resultant cheese was stored at 5 °C. The same procedure was typically used for the two control and 5 MAP treatments. The results obtained in this study represented the means of 3 replicated experiments (3 batches).

2.3. Packaging Treatments

All cheese samples were packed in PA/PE oxygen permeability 0.001 $cm^2/m^2/d$; water vapor permeability was zero and divided in groups, with each bag containing 250 g cheeses and each group having 25 bags. Five different MAP conditions were carried out by using different ratios (v/v) of CO_2 and N_2, as presented in Table 1. Gas mixtures were produced by using a gas mixer (Gas mixer with gas model KM100-3ME on stainless steel receiver, Italy) and applied to packages by injection. The packages were connected to the vacuum sealer (model Boss No. 48, Bad Homburg, Germany), and the pouches were heat-sealed by using of a sealing machine (R 200, Repack, Italy) connected to the gas mixer. Control treatments were packaged under ordinary atmosphere (packaged in air) (C1) and under vacuum (C2). All the seven groups were stored at 5 °C for 75 days. Determinations of the volatile compounds and headspace gas compositions were carried out on cheese representative samples during storage periods [13].

2.4. pH Values

The pH values were measured by a pH meter JENCO Model 1671, USA [13].

Table 1. Experimental modified atmosphere packaging designs of cheeses.

Treatments	Codes	Designs	
Control	C1	Air atmosphere packaging	
	C2	Under vacuum packaging	
MAP *		CO_2 % (v/v)	N_2 % (v/v)
	D1	10	90
	D2	15	85
	D3	25	75
	D4	100	
	D5		100

* Modified atmosphere packaging.

2.5. Assessment of Proteolysis

The extracts of total free amino acids (FAAs) were carried out according to Kuchroo and Fox [14], and total FAAs were estimated by using the Cd-ninhydrin technique of Folkertsma and Fox [15] and expressed as μM leucine equivalents, using a standard curve.

2.6. Identification and Quantification of Volatile Compounds

Volatile compounds were isolated from the cheese samples and extracted with the headspace SPME method [16], using 50/30 μm divinylbenzene/carboxen/ polydimethylsiloxane fibers. A cheese sample (10 g) from each treatment was put into glass vials (30 mL) and mixed at 55 °C for 60 min to allow for equilibrium. The fiber was inserted into the injection port of the Agilent 7890B gas chromatograph (GC; Agilent Technologies Inc., Palo Alto, CA, USA), held for 5 min for preconditioning, and then inserted into the vial and placed into the headspace for 60 min (under the above conditions). After absorbing the volatile compounds, the fibers were put into the gas chromatography–mass spectrometry (GC–MS) injector port for desorption at 270 °C for 3 min to desorb volatile compounds into the GC. The carrier gas (helium at 1 mL/min) pushed the volatile compounds absorbed onto the SPME fiber through HP-5MS column (30 m length, 0.25 mm inside diameter, and 0.25 μm film thicknesses; Agilent Technologies Inc., Palo Alto, CA, USA). The GC temperature was kept at 35 °C for 5 min, then increased at a rate of 4 °C/min to 140 °C for 5 min, and finally gradually increased at a rate of 10 °C/min to 250 °C for 5 min. The transfer lines' temperature was set to 250 °C. The mass detector was worked at 150 °C in electron impact mode, at a voltage of 70 eV and an ion source temperature of 230 °C. Mass spectra of various treated samples were observed with a mass range of 40 to 400 m/z, with 5 scans and no solvent delay [17].

Volatile compounds were identified by comparing the mass spectra with those in the library of mass spectra profiles in the data of a standard database (NIST version 11 mass spectral database: Agilent Technologies Inc., Palo Alto, CA, USA). The retention indices (RIs) were computerized in isolated materials by the NIST 11 database, using the similar capillary column, and a progression of *n*-alkanes C_3–C_{25} (AccuStandard Inc., New Haven, CT, USA) were run under similar chromatographic conditions [17]. Specific chemical standards were used to confirm the identifications, i.e., hydroxyacetic acid, acetic acid, butanoic acid, pentanoic acid, hexanoic acid, heptanoic acid, benzoic acid, *n*-decanoic acid, acetaldehyde, pentanal, hexanal, heptanal, benzaldehyde, 2-methylundecanal, nonanal, 2,3-butanedione, 2-pentanone, 3-methyl-2-butanone, acetoin, methyl isobutyl ketone, 4-methyl-2-hexanone, 2-heptanone, acetophenone, 2-undecanone, formic acid ethenyl ester, acetic acid ethenyl ester, ethyl hexanoate, δ-nonalactone, ethyl decanoate, and heptane. Data were expressed as milligrams of volatile compounds per 100 g of cheese (mg 100 g^{-1}).

2.7. Headspace Gas Analysis

The levels of O_2 and CO_2 were determined before volatile compounds to verify packaging atmospheres for each group. Before opening the Domiati cheese bags, headspace gas analysis was measured by using an oxygen analyzer (Witt oxy baby headspace gas analyzer (O_2/CO_2), model PCO_2 Plus/100, Gas Data Ltd., Coventry, UK), according to the method reported by Garabal et al. [18]. Determinations were performed at nine different timings, following initiation of treatments. The results obtained from this study represented value means from 3 replicates experiments in which all analyses were carried out on triplicate samples.

2.8. Statistical Analysis

Results corresponding to treatments determined during storage periods were subjected to ANOVA (two-way ANOVA). The statistical variances between the reported data were analyzed by Duncan's test. Variances between treatments were significant at $p < 0.05$. Statistical analyses of three separate experiments were applied, and analyses were determined in triplicate. Data analysis was carried out by using PROC GLM in SAS (version

6.12, SAS Institute Inc. Cary, CA, USA) and expressed as the means and standard error (means ± SE). Quantitative PCR datasets were analyzed by using PROC GLM of SAS [19]. The applied static model is as follows:

$$Y_{ij} = \mu + T_i + e_{ij} \quad (1)$$

where Y_{ij} is the dependent variable, μ is the overall mean, T_i is the effect of treatment ($i = 1, \ldots, 7$), and e_{ij} is the residual standard error.

3. Results

3.1. pH Values

The pH values are depending on the packaging methods and the storage periods (Figure 1). At day one, the pH value varied between 6.50 ± 0.15 and 6.64 ± 0.10 in all treatments. At day 75, the pH value decreased during storage varying from 6.40 ± 0.10 to 6.01 ± 0.15 in all samples ($p < 0.05$). The normal control (C1) treatment showed the highest reduction in pH value, compared with the other treatments in day one, and decreased during storage periods. This may be due to the normal lactic acid bacteria leading to acid production. On the other hand, the under-vacuum samples (C2) showed the lowest reduction in pH, i.e., from 6.64 at zero time to 6.49 and 6.28 after 40 and 75 days of storage, respectively, due to reducing the oxygen which limits the chances for bacterial growth or activity. A similar result was noticed in the samples preserved with 100% N_2. The spread of N_2 in the atmosphere surrounding the preserved samples may also limit the availability of oxygen. Replacing N_2 by different proportions of CO_2 resulted in lower pH values in the range 6.21–6.23 after 75 days in comparison to C2, D1, D2, and D5 treatments. These slight reductions in pH value may be due to the dissolution of CO_2 in water forming H_2CO_3.

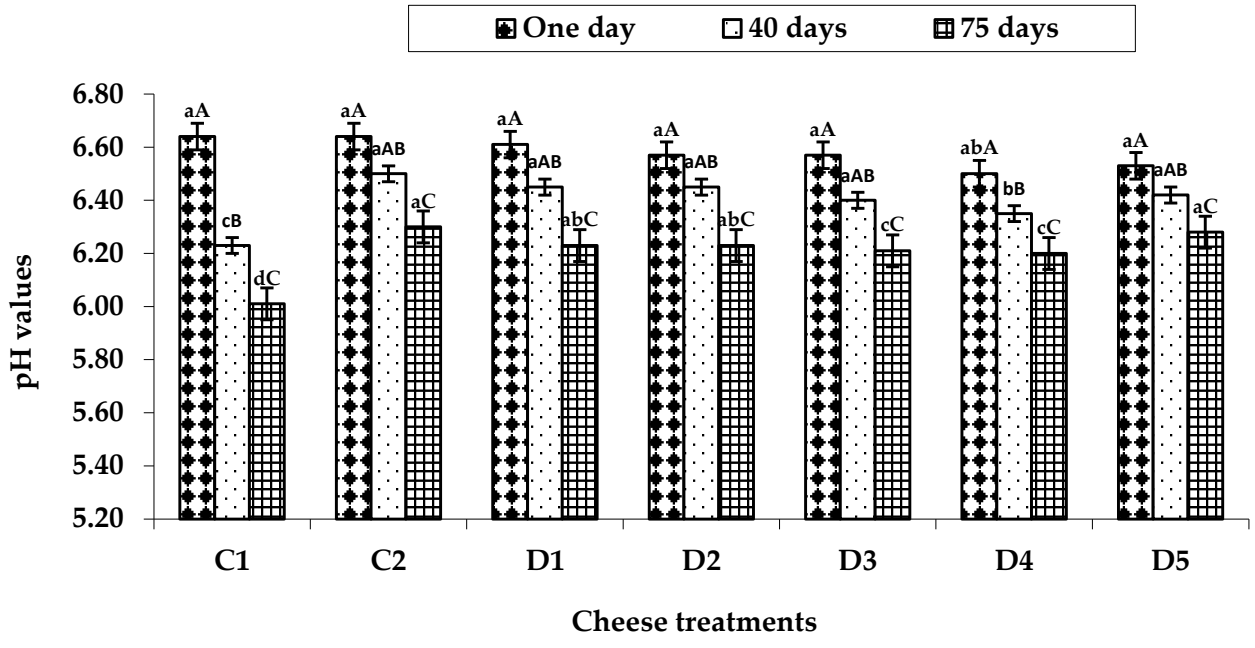


Figure 1. The pH values of the Domiati cheese packaged under different modified atmospheres. C1, control packaged under aerobically atmosphere; C2, control packaged under vacuum; D1, 10% CO_2/90% N_2; D2, 15% CO_2/85% N_2; D3, 25% CO_2/75% N_2; D4, 100% CO_2; and D5, 100% N_2; $^{a–d}$ bars within the same storage day (treatments) not sharing a common letter are significantly different ($p < 0.05$); $^{A–C}$ bars not sharing a common letter during the storage periods are significantly different ($p < 0.05$).

3.2. Proteolysis

The release of total FAAs in Domiati cheese samples that were packaged under different MAP conditions is shown in Table 2. Total FAAs values were significantly affected by packaging treatments and the storage period ($p < 0.05$) in all cheese samples. The total FAAs levels increased during the storage, reaching the highest levels in all samples at 75 days. The normal air-atmosphere sample (C1) showed the highest levels of total FAA after 40 and 75 days of storage, i.e., 191 and 356 µM leucine equivalents, respectively. Low levels of total FAAs were detected in the cheese samples packaged under vacuum (C2) and those packaged under 100% N_2. The other samples showed low reductions in total FAAs when replacing N_2 by different proportions of CO_2. However, the lowest level of total FAAs was associated with the samples packaged under 100% CO_2 (D4) after 75 days of storage. This particularly low total FAAs content may be due to the reduced pH incurred by the high level of CO_2, reducing the pH and making the medium less optimum for proteolytic activities. The combined gas packaging systems (D1–D3) kept the level of total FAAs at intermediate levels ranging between 250 and 271 µM leucine equivalent, which is lower than that for the atmosphere air control (C1) treatment, recording 356 µM leucine equivalent, and higher than the vacuum control (C2) treatment and the single gas packaging system (D4 and D5 treatments), in the range of 204–222 µM leucine equivalent.

Table 2. Total free amino acids levels (means ± SE) in cheese packaged under different modified atmospheres (CO_2 and N_2) during storage.

Treatments		Total Free Amino Acids (µM Leucine Equivalents)		
	CO_2/N_2 (%; *v*/*v*)	**One Day**	**40 Days**	**75 Days**
C1		43 ± 0.004^{aC}	191 ± 0.003^{aB}	356 ± 0.005^{aA}
C2		24 ± 0.002^{cC}	135 ± 0.004^{dB}	221 ± 0.006^{dA}
D1	10/90	31 ± 0.002^{bC}	168 ± 0.002^{bB}	271 ± 0.007^{bA}
D2	15/85	29 ± 0.001^{bcC}	160 ± 0.003^{cB}	254 ± 0.005^{bcA}
D3	25/75	29 ± 0.001^{bcC}	160 ± 0.004^{cB}	250 ± 0.002^{cA}
D4	100/0	23 ± 0.002^{cC}	119 ± 0.004^{eB}	204 ± 0.005^{eA}
D5	0/100	31 ± 0.001^{bC}	133 ± 0.003^{dB}	222 ± 0.004^{dA}

C1, control packed under aerobically atmosphere; C2, control packed under vacuum; D1, 10% CO_2/90% N_2; D2, 15% CO_2/85% N_2; D3, 25% CO_2/75% N_2; D4, 100% CO_2; and D5, 100% N_2; $^{a–e}$ there is a significant difference ($p < 0.05$) between any two means, within the same column, have the different superscript letters; $^{A–C}$ there is a significant difference ($p < 0.05$) between any two means for the same attribute, within the same row, have the different superscript letter.

3.3. Volatile Acid and Aldehyde Compounds

Data of volatile acid and aldehyde compounds for all samples are shown in Table 3. Eight acid compounds were detected in the volatile extract of different cheese samples. Acid compounds' levels in all treatments showed significant changes with time and according to treatment. The level of each acid compound was increased with storage period and the increases were particularly clear for pentanoic acid, hexanoic acid, heptanoic acid, benzoic acid, and *n*-decanoic acid. These volatile acids contribute to good taste and odor of cheese. The highest level of acid compounds ($p < 0.05$) was detected in C1 (air atmosphere) treatment after 75 days of storage, while the lowest ones were associated with D5 (100% N_2) samples. The under-vacuum control samples came in the second order, and the 100% CO_2 treatment (D4) came in the third order. Other treatments (D1–D3) combining CO_2 and N_2 recorded values between the limits of the previous high-level samples (C1, C2, D4, and D5). Seven aldehyde compounds were observed in all treatments (Table 3). Aldehyde compounds showed significant increases ($p < 0.05$) in all treatments with storage of all treatments. The air-atmosphere control showed the highest levels ($p < 0.05$) of all the aldehyde compounds than the MAP samples after 75 days of storage. Hexanal, heptanal, and benzaldehyde were the major aldehyde compounds identified, accounting for 80% of

the total aldehyde's compounds. Benzaldehyde was the only cyclic compound detected in all treatments. The levels of benzaldehyde in all treatments ranged from 17.74 to 26.13 mg 100 g^{-1} after 75 days.

3.4. Volatile Ketone Compounds

Table 4 shows the contents of the volatile ketone compounds in the samples' headspace of all treatments. Ten ketones were detected in the different cheese samples, showing significant changes. Ketone compounds increased during storage between treatments. The normal control (C1) treatment showed high contents of the different volatile ketone compounds. However, the samples packaged under 100% N_2 (D5) showed the significantly ($p < 0.05$) highest levels of all the volatile ketones after 75 days of storage, particularly 2-pentanone, acetoin, methyl isobutyl ketone, 2-heptanone, 2-nonanone, and 2-undecanone. Conversely, the sample packaged under 100% CO_2 (D4) recorded the significantly ($p < 0.05$) lowest levels of the detected volatile ketones after 75 days. Compared to the normal control (C1), the levels of D5 were significantly higher, while those of D4 treatment were significantly lower ($p < 0.05$). The samples of the combined treatments (CO_2/N_2) showed values higher than D4 but lower than D5. The sample (D3, 25% CO_2/75% N_2) was the best combined sample, showing significantly ($p < 0.05$) higher levels than the other combined samples (D1 and D2) or the 100% CO_2 sample (D4), but it remained in the same significant level of normal control (C1).

3.5. Volatile Ester and Hydrocarbon Compounds

Table 5 shows the volatile ester and hydrocarbon compounds in the headspace of all treatments' samples. Very low levels of volatile esters were detected on day one and increased greatly after 75 days of storage. The normal control (C1) treatment exposed to the atmospheric air showed the highest significant increases from zero-day levels. All other treatments after 75 days were significantly higher than for day zero but significantly lower than the normal control (C1). Nevertheless, the treatment D5 (100% N_2) was the closest to the normal control but still significantly lower. Compatibility between the general hydrophobic/hydrophilic nature of this class of components and the atmospheric air may explain the distinction of the normal control. Treatment D4 (100% CO_2) significantly ($p < 0.05$) showed the least levels of these compounds, while the combined treatments showed logical values between these two extremes depending on their hydrophobic/hydrophilic characteristics. This trend may be similar to what has been observed with the volatile ketones. A total of four volatile hydrocarbon compounds were detected in all extracts for samples (Table 5). The level of these hydrocarbon compounds was low in all treatments. Hydrocarbon compounds increased ($p < 0.05$) in all treatments for up to 75 days of storage. These compounds were not significantly ($p > 0.05$) different between all treatments after 75 days. Among these components, heptane (varying from 6.49 to 6.57 mg 100 g^{-1}) was the most prominent in all treatments after 75 days of storage.

Table 3. Volatile acid and aldehyde compounds identified in cheese packaged under modified atmospheres (means ± SE).

Volatile Compounds	Method [1]	RT [2] (min)	RI [3]	Chemical Formula	One Day	Cheese Treatments after 75 Days						
						C1	C2	D1	D2	D3	D4	D5
Acid compounds (mg 100 g^{-1})												
Hydroxyacetic acid	MS, STD	1.05	STD [4]	$C_2H_4O_3$	ND [5]	6.45 ± 1.54 [a]	4.11 ± 0.98 [b]	3.00 ± 0.41 [c]	2.54 ± 0.02 [d]	2.04 ± 0.24 [e]	3.02 ± 0.08 [c]	2.00 ± 0.07 [e]
Acetic acid	MS, STD	3.56	STD	$C_2H_4O_2$	2.08 ± 0.23 [f]	6.78 ± 1.54 [a]	5.98 ± 0.23 [b]	4.55 ± 0.31 [c]	4.01 ± 0.23 [d]	3.89 ± 0.12 [e]	4.88 ± 0.41 [c]	3.81 ± 0.51 [e]
Butanoic acid	MS, RI	5.57	790.66	$C_4H_8O_2$	4.78 ± 0.41 [f]	8.12 ± 2.01 [a]	7.98 ± 1.01 [b]	6.98 ± 0.41 [c]	6.12 ± 0.54 [d]	5.45 ± 0.40 [e]	7.00 ± 0.51 [c]	5.44 ± 071 [e]
Pentanoic acid	MS, RI	9.14	868.78	$C_5H_{10}O_2$	5.47 ± 1.01 [f]	12.12 ± 2.32 [a]	10.23 ± 1.87 [b]	8.97 ± 0.87 [c]	8.04 ± 0.74 [d]	7.35 ± 0.42 [e]	9.02 ± 0.61 [c]	7.20 ± 0.65 [e]
Hexanoic acid	MS, RI	14.18	999.24	$C_6H_{12}O_2$	7.02 ± 1.12 [ff]	16.12 ± 3.21 [a]	15.98 ± 2.00 [b]	13.02 ± 1.02 [c]	12.30 ± 1.01 [d]	11.87 ± 0.99 [e]	13.12 ± 0.85 [c]	11.71 ± 1.02 [e]
Heptanoic acid	MS, RI	17.12	1071.70	$C_7H_{14}O_2$	11.21 ± 2.01 [f]	18.25 ± 3.21 [a]	17.45 ± 2.31 [b]	15.80 ± 1.31 [c]	14.57 ± 1.13 [d]	13.89 ± 1.00 [e]	15.89 ± 1.08 [c]	13.72 ± 1.21 [e]
Benzoic acid	MS, RI	19.61	1159.11	$C_7H_6O_2$	8.97 ± 0.54 [f]	18.45 ± 3.65 [a]	17.45 ± 1.98 [b]	16.08 ± 1.56 [c]	15.01 ± 1.30 [d]	14.47 ± 1.21 [e]	16.21 ± 1.15 [c]	14.33 ± 1.24 [e]
n-Decanoic acid	MS, RI	24.29	1341.01	$C_{10}H_{20}O_2$	10.24 ± 1.47 [f]	24.44 ± 3.65 [a]	24.00 ± 2.45 [b]	22.61 ± 2.61 [c]	21.77 ± 2.31 [d]	20.33 ± 2.07 [e]	22.78 ± 3.08 [c]	20.21 ± 3.21 [e]
Aldehyde compounds (mg 100 g^{-1})												
Acetaldehyde	MS, STD	1.38	STD	C_2H_4O	ND	14.35 ± 2.74 [a]	12.16 ± 1.05 [b]	12.09 ± 1.87 [b]	12.04 ± 1.22 [b]	11.46 ± 1.65 [c]	12.11 ± 1.02 [b]	10.87 ± 1.85 [d]
Pentanal	MS, RI	3.25	664.98	$C_5H_{10}O$	ND	18.37 ± 1.35 [a]	15.77 ± 1.58 [b]	15.57 ± 1.32 [b]	15.51 ± 1.42 [b]	14.87 ± 1.44 [c]	15.63 ± 1.88 [b]	13.09 ± 1.35 [d]
Hexanal	MS, RI	6.45	798.84	$C_6H_{12}O$	ND	21.25 ± 1.70 [a]	19.11 ± 1.99 [b]	19.00 ± 2.07 [b]	18.94 ± 1.79 [b]	17.98 ± 2.05 [c]	19.03 ± 1.84 [b]	15.89 ± 1.69 [d]
Heptanal	MS, RI	9.98	889.78	$C_7H_{14}O$	1.02 ± 0.07 [e]	19.08 ± 2.01 [a]	17.79 ± 1.87 [b]	17.02 ± 1.88 [b]	16.89 ± 1.54 [b]	16.00 ± 1.88 [c]	17.07 ± 2.03 [b]	14.89 ± 1.54 [d]
Benzaldehyde	MS, RI	12.47	977.14	C_7H_6O	1.10 ± 0.09 [e]	26.13 ± 3.04 [a]	20.21 ± 2.14 [b]	19.98 ± 2.03 [b]	19.89 ± 2.01 [b]	19.02 ± 1.98 [c]	20.092.54 [b]	17.74 ± 2.06 [d]
2-Methylundecan al	MS, RI	16.97	1085.05	$C_{12}H_{24}O$	0.79 ± 0.01 [e]	14.22 ± 1.51 [a]	13.02 ± 1.50 [b]	12.94 ± 1.34 [b]	12.90 ± 1.65 [b]	11.45 ± 1.86 [c]	13.00 ± 1.64 [b]	12.04 ± 1.70 [d]
Nonanal	MS, RI	17.86	1114.01	$C_9H_{18}O$	2.11 ± 0.21 [e]	14.77 ± 1.08 [a]	13.04 ± 1.05 [b]	12.90 ± 1.00 [b]	12.84 ± 1.44 [b]	11.78 ± 1.37 [c]	12.99 ± 1.37 [b]	10.97 ± 1.02 [d]

Method [1] RI, agrees with retention index literature; MS, compared with NIST 11 Mass Spectral Database; STD, agrees with mass spectral of standard chemicals; RT [2], retention time; RI [3], retention indices of unknown compounds on HP-5MS column calculated against the GC–MS retention time of *n*-alkanes (C_3–C_{25}); STD [4], agrees with mass spectral of standard chemicals; ND [5], not detected; C1, control packed under aerobically atmosphere; C2, control packed under vacuum; D1, 10% CO_2/90% N_2; D2, 15% CO_2/85% N_2; D3, 25% CO_2/75% N_2; D4, 100% CO_2; and D5, 100% N_2; [a–f]: there is a significant difference ($p < 0.05$) between any two means, within the same row, have the different superscript letters.

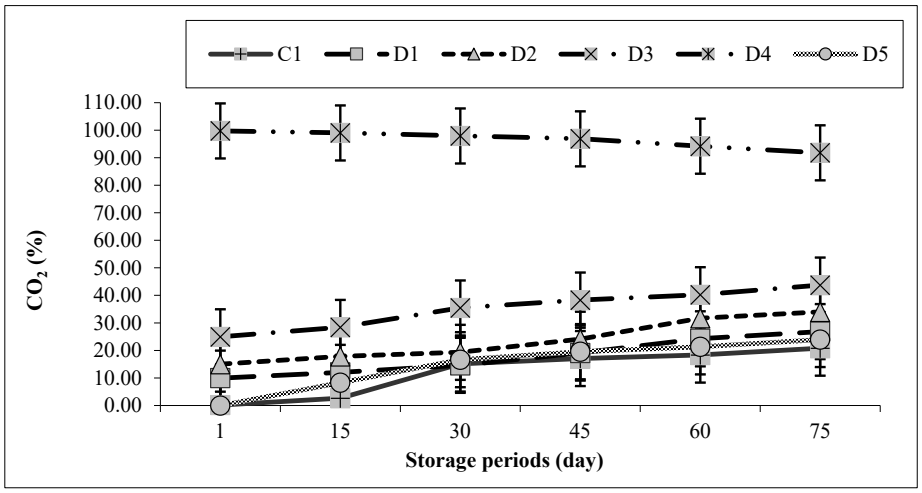
Table 4. Volatile ketone compounds identified in cheese samples packaged under modified atmospheres (means ± SE).

Volatile Compounds	Method [1]	RT [2] (min)	RI [3]	Chemical Formula	One Day	Cheese Treatments after 75 Days						
						C1	C2	D1	D2	D3	D4	D5
Ketone compounds (mg 100 g^{-1})												
2,3-Butanedione	MS, STD	1.99	STD [4]	$C_4H_6O_2$	ND [5]	8.79 ± 1.25 [b]	7.19 ± 1.03 [c]	5.01 ± 0.41 [e]	6.24 ± 0.33 [d]	8.98 ± 0.54 [b]	4.97 ± 0.45 [e]	9.78 ± 0.79 [a]
2-Pentanone	MS, RI	3.05	657.44	$C_5H_{10}O$	ND	17.00 ± 2.38 [b]	16.11 ± 1.04 [c]	13.25 ± 1.23 [e]	15.03 ± 1.34 [d]	17.00 ± 1.43 [b]	13.11 ± 1.36 [e]	17.89 ± 3.01 [a]
3-Methyl-2-butanone	MS, RI	3.08	659.01	$C_5H_{10}O$	ND	8.62 ± 1.65 [b]	7.56 ± 1.00 [c]	5.00 ± 0.31 [e]	6.54 ± 0.22 [d]	8.79 ± 0.69 [b]	4.88 ± 0.31 [e]	9.14 ± 0.23 [a]
Acetoin	MS, RI	4.01	700.04	$C_4H_8O_2$	ND	11.64 ± 2.03 [b]	10.46 ± 0.97 [c]	7.89 ± 0.34 [e]	9.18 ± 0.69 [d]	11.87 ± 1.23 [b]	7.74 ± 0.41 [e]	12.02 ± 1.04 [a]
Methyl isobutyl ketone	MS, RI	4.61	740.10	$C_6H_{12}O$	ND	16.91 ± 2.36 [b]	15.24 ± 1.32 [c]	11.58 ± 0.87 [e]	13.89 ± 0.78 [d]	16.99 ± 1.54 [b]	11.32 ± 1.02 [e]	17.45 ± 3.00 [a]
4-Methyl-2-hexanone	MS, RI	9.45	899.98	$C_7H_{14}O$	ND	14.66 ± 2.00 [b]	13.57 ± 1.24 [c]	9.29 ± 0.89 [e]	12.01 ± 0.95 [d]	14.78 ± 1.69 [b]	9.17 ± 0.84 [e]	15.09 ± 1.08 [a]
2-Heptanone	MS, RI	10.05	912.07	$C_7H_{14}O$	0.87 ± 0.02 [f]	19.00 ± 1.98 [b]	17.84 ± 2.41 [c]	14.21 ± 1.09 [e]	15.98 ± 1.09 [d]	19.01 ± 2.35 [b]	14.09 ± 1.54 [e]	19.78 ± 2.54 [a]
Acetophenone	MS, RI	15.84	1057.77	C_8H_8O	1.01 ± 0.12 [e]	13.76 ± 1.75 [b]	12.34 ± 1.87 [c]	9.87 ± 0.74 [e]	11.37 ± 0.87 [d]	13.89 ± 1.48 [b]	9.79 ± 0.48 [e]	14.45 ± 1.02 [a]
2-Undecanone	MS, RI	22.87	1321.02	$C_{11}H_{22}O$	0.78 ± 0.01 [e]	19.73 ± 2.84 [b]	17.83 ± 1.32 [c]	13.07 ± 1.22 [e]	15.63 ± 1.39 [d]	19.88 ± 1.89 [b]	12.98 ± 1.12 [e]	20.78 ± 2.74 [a]

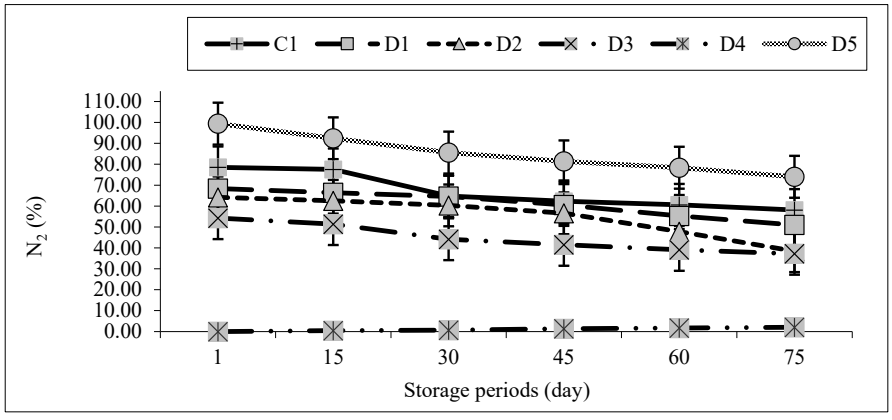
Method [1] RI, agrees with retention index literature; MS, compared with NIST 11 Mass Spectral Database; STD, agrees with mass spectral of standard chemicals; RT [2], retention time; RI [3], retention indices of unknown compounds on HP-5MS column calculated against the GC– MS retention time of *n*-alkanes (C_3–C_{25}); STD [4], agrees with mass spectral of standard chemicals; and ND [5], not detected. C1, control packed under aerobically atmosphere; C2, control packed under vacuum; D1, 10% CO_2/90% N_2; D2, 15% CO_2/85% N_2; D3, 25% CO_2/75% N_2; D4, 100% CO_2; and D5, 100% N_2; [a–f]: there is significant differences ($p < 0.05$) between any two means, within the same row, have the different superscript letters.

3.6. Gas Composition

Figure 2A,B shows the levels of both CO_2 and N_2 in the MAP-cheese samples during 75 days of storage. The CO_2 level showed significant changes ($p < 0.05$) in different packaging systems in the fresh period and throughout the storage period (Figure 2A). All cheese samples showed gradual increases ($p < 0.05$) in CO_2 co-occurring with parallel decreases in N_2 during refrigerated storage periods, except for the D4 treatment (100% CO_2), which showed a decrease. The increase in CO_2 could explain the clear decline in N_2 and O_2. In D4 treatment, CO_2 percentages decreased, reaching 91.82% after 75 days of storage, due to expected gas depletion from the initially high level of 100% CO_2. An increase in CO_2 percentage was obtained for D5 treatment (100% N_2), reaching 23.97% after 75 days of storage. However, CO_2 percentage in the under vacuum samples was not detected in the fresh period or throughout the storage periods. The N_2 levels showed significant changes ($p < 0.05$) in all packaging systems throughout the storage periods (Figure 2B). In C1, D1, D2, D3, and D5 treatments, a gradual decrease occurred in the N_2 levels during the storage period. In D4 treatment (100% CO_2), an increase in N_2 levels ($p < 0.05$) occurred during storage due to the clear decrease in CO_2 levels. However, N_2 in the under vacuum treatment was not detected at the starting day and throughout the storage periods.



(**a**)



(**b**)

Figure 2. CO_2 (**a**) and N_2 (**b**) levels of the Domiati cheese packaged under different modified atmosphere conditions; C1, control packaged under aerobically atmosphere; D1, 10% CO_2/90% N_2; D2, 15% CO_2/85% N_2; D3, 25% CO_2/75% N_2; D4, 100% CO_2; and D5, 100% N_2. Data are showed with standard errors.

The O_2 levels in the packages of cheese during 75 days of storage are recorded in Figure 3. The O_2 levels in the cheese decreased ($p < 0.05$) in all cheese groups throughout the storage period, except for C2, which did not indicate any presence of O_2 during the whole period of storage at 5 °C. Oxygen is the second principal component of the ambient air, whose initial value in the atmospheric air is around 21%. A significant decrease ($p < 0.05$) in O_2 level occurred in C1 treatment during cold storage. The O_2 levels of C1 treatment diminished from 19.98 to 0.00% during cold storage, probably due to microorganisms' consumption of O_2 inside the package.

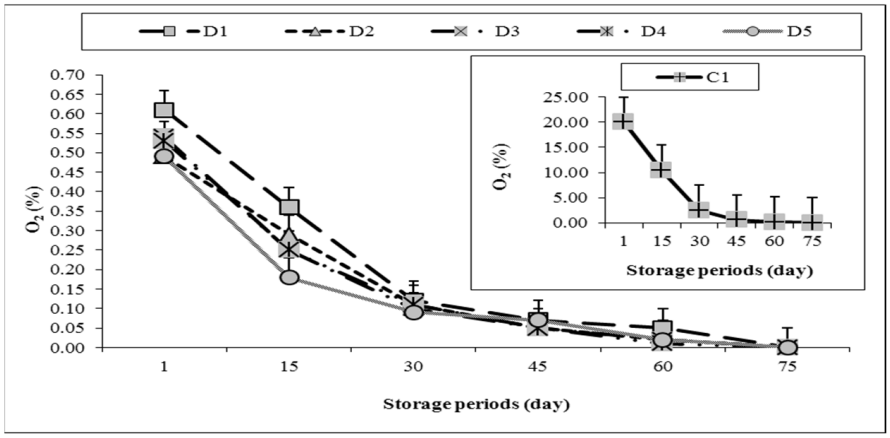


Figure 3. Levels of O_2 in the headspace of the Domiati cheese packaged under different modified atmospheres; C1, control packaged under aerobically atmosphere; D1, 10% CO_2/90% N_2; D2, 15% CO_2/85% N_2; D3, 25% CO_2/75% N_2; D4, 100% CO_2; and D5, 100% N_2. Data are presented as the means plus standard errors.

Table 5. Volatile ester and hydrocarbon compounds identified in cheese packaged under modified atmospheres (means ± SE).

Volatile Compounds	Method [1]	RT [2] (min)	RI [3]	Chemical Formula	One Day	Cheese Treatments after 75 Days						
						C1	C2	D1	D2	D3	D4	D5
Ester compounds (mg 100 g^{-1})												
Formic acid ethenyl ester	MS, STD	1.55	STD [4]	$C_3H_4O_2$	3.25 ± 0.09 ^{g}	11.78 ± 1.28 ^{a}	10.20 ± 1.09 ^{b}	7.45 ± 0.56 ^{d}	7.40 ± 0.78 ^{d}	6.01 ± 0.45 ^{e}	5.00 ± 0.20 ^{f}	9.10 ± 0.45 ^{c}
Acetic acid ethenyl ester	MS, STD	1.99	STD	$C_4H_6O_2$	4.56 ± 0.11 ^{g}	15.32 ± 1.12 ^{a}	14.03 ± 0.98 ^{b}	10.27 ± 1.20 ^{d}	10.28 ± 1.14 ^{d}	9.54 ± 0.74 ^{e}	8.02 ± 1.02 ^{f}	12.49 ± 1.42 ^{c}
Ethyl hexanoate	MS, STD	2.89	STD	$C_6H_{10}O_2$	4.21 ± 0.21 ^{g}	17.82 ± 1.32 ^{a}	16.11 ± 1.11 ^{b}	13.78 ± 1.41 ^{d}	13.72 ± 0.89 ^{d}	11.32 ± 0.97 ^{e}	9.78 ± 1.11 ^{f}	15.11 ± 1.35 ^{c}
δ-Nonalactone	MS, RI	29.11	1489.59	$C_9H_{16}O_2$	4.56 ± 0.31 ^{g}	19.25 ± 1.45 ^{a}	17.24 ± 1.56 ^{b}	14.71 ± 1.08 ^{d}	14.65 ± 0.79 ^{d}	11.12 ± 1.13 ^{e}	10.02 ± 1.33 ^{f}	16.21 ± 1.36 ^{c}
Ethyl decanoate	MS, RI	38.21	1491.23	$C_{10}H_{18}O_2$	5.21 ± 0.33 ^{g}	21.24 ± 2.31 ^{a}	20.01 ± 2.12 ^{b}	16.35 ± 1.23 ^{d}	16.32 ± 1.08 ^{d}	13.67 ± 1.23 ^{e}	11.21 ± 1.40 ^{f}	18.42 ± 1.78 ^{c}
Hydrocarbon compounds (mg 100 g^{-1})												
Heptane	MS, RI	3.32	687.88	$C_7H_{15}Cl$	ND [5]	6.54 ± 0.23 ^{a}	6.45 ± 0.22 ^{a}	6.50 ± 0.36 ^{a}	6.57 ± 0.12 ^{a}	6.49 ± 0.33 ^{a}	6.50 ± 0.23 ^{a}	6.52 ± 0.21 ^{a}

Method [1] RI, agrees with retention index literature; MS, compared with NIST 11 Mass Spectral Database; STD, agrees with mass spectral of standard chemicals; RT [2], The retention time; RI [3], The retention indices of unknown compounds on HP-5MS column calculated against the GC– MS retention time of *n*-alkanes (C_3–C_{25}); STD [4], agrees with mass spectral of standard chemicals; and ND [5], not detected. C1, packed under aerobically atmosphere; C2, packed under vacuum; D1, 10% CO_2/90% N_2; D2, 15% CO_2/85% N_2; D3, 25% CO_2/75% N_2; D4, 100% CO_2; and D5, 100% N_2; $^{a–f}$: there is a significant difference ($p < 0.05$) between any two means, within the same row, have the different superscript letters.

4. Discussion

The modified atmosphere packaging (MAP) may be a suitable replacement for cheese preservation based on reducing level of O_2. MAP modifies the atmosphere surrounding the cheese, retarding microorganisms' growth without affecting the cheeses. The present study's goal was to investigate the pH, volatile compounds, and total free amino acids development of the Domiati cheese when packaged under vacuum and modified atmosphere during storage at 5 °C for different periods. Domiati cheese's microbiological, volatile compounds, and total free amino acids properties are critical quality characteristics because they play an essential role in consumers' acceptance of this type. Replacing N_2 by different proportions of CO_2 resulted in lower pH values after 75 days storage. These slight reductions in pH value may be due to the dissolution of CO_2 in water forming H_2CO_3. The decrease in pH level in the control samples (C1) may have resulted from the conversion of lactose to lactic acid by lactic acid bacteria or to the dissolution of CO_2 forming carbonic. Atallah et al. [13] reported a gradual pH, decline toward the end of the storage period in all MAP-preserved Domiati cheeses. Felfoul et al. [20] also observed the highest decrease of pH in MAP of the cheeses (Atms 40% CO_2/60% N_2, 60% CO_2/40% N_2 and 100% CO_2), while the lowest decrease recorded in treatments with air and 100% N_2, respectively. The CO_2 caused a decrease in pH because the formation of carbonic acid during storage. The decrease of pH, caused by carbon dioxide gas, was referred to by Farber [21]. Maintaining the pH at these intermediate levels in the combined gas packages may be beneficial for the quality of the cheese product.

The combined gas packaging systems (D1–D3) kept the level of total FAAs at intermediate levels ranging between 250 and 271 μM leucine equivalent, which is lower than the atmosphere air control (C1) treatment recoding 356 μM leucine equivalent and higher than the vacuum control treatment (C2) and the single gas packaging system (D4 and D5 treatments), in the range 204–222 μM leucine equivalent. In conclusion, the different noticed changes in the level of total FAA may be related to some factors, including high moisture percent and low pH levels caused higher residual chymosin activity, the major component of rennet. Small peptides and FAA are usually released under the action of chymosin, non-starter bacterial enzymes, and starter bacteria enzymes [22] or from any contributing microorganisms. The influence of the packaging conditions on the released FAA and small peptides may affect the sensorial quality of the developed and preserved cheese. Keeping the level of total FAA at intermediate levels may favor keeping the cheese quality while still counteracting the microbial contamination and degradation [22].

Acid compounds' levels in all treatments showed significant changes with time and according to treatment. The level of each acid compound increased with the storage period, and the increases were particularly clear in pentanoic acid, hexanoic acid, heptanoic acid, benzoic acid, and *n*-decanoic acid. The highest level of acid compounds ($p < 0.05$) was detected in C1 (air atmosphere) treatment after 75 days of storage, while the lowest ones were associated with D5 (100% N_2) samples. These increases in the volatile acid compounds can be derived from the conversion of amino acids and carbohydrates. The initial stage in cheese ripening is the metabolic conversion of lactose to lactate by the microorganisms [23]. However, volatile acids may be observed in cheese as products of enzyme-mediated hydrolysis of protein and fat due to metabolic action of the growing microorganisms [21]. These volatile acid compounds may contribute to the good odor and flavor of cheese (especially acetic acid, butanoic acid, hexanoic acid, heptanoic acid, and *n*-decanoic acid) [24]. Colchin et al. [24] showed similar findings with respect to acid compounds observed in cheese stored under MAP. Juric et al. [25] did not detect any acids or esters during storage in sliced cheese under a CO_2 atmosphere either in the presence of light or in the dark. The high levels of acetic acid are responsible for the most taste and odor of dairy products, given the "acidic, pungent, vinegary" taste [26]. Moreover, hexanoic acid is a major source of odor, taste, and functionality in cheese, imparting a "flowery, pungent" taste [27]. The levels of heptanoic acid were detected in all treatments, and the highest heptanoic acid content after 75 days was recorded in C1 treatment. They are responsible for

the flavor and aroma qualities of cheese and other dairy products [28]. Moreover, Barakat et al. [29] found that volatile acid compounds were isolated in significant levels and given to sensory attributes of produced yogurt.

The air atmosphere control showed the highest levels of all the aldehyde compounds than MAP samples after 75 days of storage. Hexanal, heptanal, and benzaldehyde were the major aldehyde compounds found, accounting for 80% of the total aldehyde's compounds. Benzaldehyde was the only cyclic compound detected in all treatments. Benzaldehyde is responsible for the flavor and aroma to cheese and the other dairy products [30,31]. Heptanal was significantly ($p < 0.05$) affected by MAP techniques. It is an important volatile carbonyl compound that contributes to the "sweet, green" taste in cheese and yogurt [32]. Levels of heptanal can increase the taste and odor scores of cheeses and fermented milk products [33]. Hexanal and heptanal are the major aldehyde compounds in Camembert cheese [34]. Aldehyde compounds may be derived via two pathways: the oxidation of amino acids and the unsaturated fatty acids degradation [35]. It can be observed that neither of the two pathways is favored for a N_2 atmosphere packaging. Therefore, it is very probable that most aldehyde compounds originated from oxidation of lipid [35]. This fact is supported by aldehyde levels in air packaged samples (C1). The determination of higher levels of aldehyde compounds in samples exposed to C1, compared with the other treatments, implies that oxidation of lipid contributed to the aldehyde's formation [24,35]. Colchin et al. [24] showed similar findings with respect to aldehyde compounds observed in cheese stored under MAP.

The sample packaged under 100% CO_2 (D4) recorded the significantly ($p < 0.05$) lowest levels of the detected volatile ketones after 75 days. Compared to the normal control (C1), the levels of D5 were significantly higher, while those of D4 were significantly lower ($p < 0.05$). The samples of the combined treatments (CO_2/N_2) showed values higher than D4 but lower than D5. The sample (D3, 25% CO_2/75% N_2) was the best combined sample, showing significantly ($p < 0.05$) higher levels than the other combined samples (D1 and D2) or the 100% CO_2 sample (D4), but it remained in the same significant level of normal control (C1). The distinction of the 100% N_2 packaged samples in assuring higher levels of the volatile ketones may be due to compatibility between the hydrophobic nature of both the nitrogen and the volatile ketone compounds. Similar data were detected by Colchin et al. [24], who showed the presence of 2-heptanone, 2-pentanone, and 2-nonanone in shredded Cheddar cheese. These ketones are produced from pyruvate, which derives from citrate, protein, and lactose metabolisms [36] and contributes to the "creamy, buttery" taste and odor in cheese and fermented milks [37]. These components have been detected previously for dairy products [38,39].

Very low levels of volatile esters were detected on day zero and increased after 75 days of storage. The normal control (C1) treatment exposed to the atmospheric air showed the highest significant increases from zero-day levels. Dixon and Kell [40] observed that CO_2 gas, in addition to protecting spoilage microorganisms, decreases the level of several flavor components. Many studies have detected that CO_2 gas has an inhibitory effect on the growth of microorganisms that are basic for the development of flavor in cheeses [41]. Similarly, low levels of volatile esters have been recorded by Colchin et al. [24] in shredded Cheddar cheese. The low levels of volatile ester compounds are responsible for the "floral, fruity" taste and odor of cheese [42]. Hydrocarbon compounds increased in all treatments for up to 75 days of storage. These compounds were not significantly different between all treatments after 75 days. Among these components, heptane was the most prominent in all treatments after 75 days of storage. These compounds have also been detected because of microorganisms' action contributing positively to flavor compounds when present at low levels [40–43].

In D4 treatment, CO_2 percentages decreased after 75 days of storage due to expected gas depletion from the initially high level of 100% CO_2. An increase in CO_2 percentage was obtained for D5 (100% N_2) after 75 days of storage. These results agree with those of Felfoul et al. [20], who observed an increase in CO_2, together with a decrease in N_2, in

cheese with an atmosphere of 40% CO_2/60% N_2 and 60% CO_2/40% N_2 [44]. They also noted that the sensitivity of gas composition evolution in the package during storage was lower at higher CO_2 and N_2; thus, the variations may not be reliable. In C1, D1, D2, D3, and D5 treatments, a gradual decrease occurred in N_2 levels during the storage period. In D4 treatment (100% CO_2), an increase in N_2 levels ($p < 0.05$) occurred during storage due to the apparent decrease in CO_2 levels. However, N_2 in the under-vacuum treatment was not detected at the starting day and throughout the storage periods. These results are in accord with those for Burrata, and fresh cheese packaged under MAP [20,45]. As far as N_2 is concerned, its level inside the package was affected by N_2 penetration into the package when its concentration became < 78%, with a balanced relationship between N_2 and CO_2 percentages [44]. A significant decrease ($p < 0.05$) in the O_2 level occurred in C1 treatment during cold storage, due to microorganisms' consumption of O_2 inside the package [46]. The under-vacuum samples did not indicate any presence of O_2 during the whole period. The other groups also showed decreased levels of O_2 during the 75 days of storage at 5 °C, due to an initially limited residual air left between the cheeses that was adsorbed by the polystyrene tray and the following microbial activity consuming it. These data agree with those reported for sliced Mozzarella and fresh cheeses [20,46]. The different treatment may assure limited bacterial growth and contamination due to this limited oxygen levels. It is well-known that the overall quality of a given cheese product depends on several quality sub-indices. The shelf-life of the packaged Domiati cheese may be taken as the time at which one of these sub-indexes reaches its threshold. The two most critical sub-indexes for cheese samples are the microbial properties, which determine the shelf life. The increase in the microbial load properties was responsible for the short shelf life of Domiati cheese samples in the control treatment (C1). The use of MAP increased Domiati cheese samples' shelf-life.

5. Conclusions

The present study shows the impact of packaging cheese under different conditions, atmospheric air, vacuum, and modified atmospheres on the volatile components of the headspace and gas composition. Five different modified atmosphere conditions were studied. Control samples were packaged in the air (C1) and under vacuum (C2). Proteolysis during cheese storage was lower in MAP cheeses than in the air-atmosphere control cheeses (C1), which exhibited the highest FAA levels after 75 days storage. Alternatively, the lowest level of FAAs was detected in D4 (100% CO_2) treatment after 75 days of storage. Volatile organic acids (especially acetic acid, butanoic acid, pentanal, hexanal, benzaldehyde, 2-heptanone, acetoin, 2,3-butanedione, ethyl hexanoate, and heptane), which are important for cheese's good volatile compounds, were detected in the different MAP-kept cheeses. The CO_2, N_2, and O_2 percentages showed significant changes in all cheese varieties, both in the fresh period and throughout the storage period. However, further study is necessary to further determine the optimal CO_2 levels to package the Domiati cheese and correlating the treatment conditions with the compounds responsible for the specific taste of Domiati cheese, particularly the issued peptides and specific amino acids and determining the keeping action of these compounds.

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Conflicts of Interest: The authors declare no conflict of interest.

Abbreviations

MAP	modified atmosphere packaging
FAAs	free amino acids
IMCU	international milk clotting units
PA/PE	polyamide/polyethylene
SPME	solid-phase microextraction
GC	gas chromatography
GC–MS	gas chromatography–mass spectrometry
log10 CFU g−1	colony-forming units per gram
RIs	retention indices
RT	retention time
Ris	retention indices
RT	retention time

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