

International Journal of Food Microbiology 79 (2002) 35-45

INTERNATIONAL JOURNAL OF FOOD Microbiology

www.elsevier.com/locate/ijfoodmicro

Preservation of fresh meat with active and modified atmosphere packaging conditions

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Received 1 April 2002; received in revised form 23 April 2002; accepted 26 April 2002

Abstract

The sensory, microbiological and physicochemical attributes of fresh meat stored at 5 and 15 °C were affected by the combined effect of volatile compounds of oregano essential oil and modified atmosphere packaging conditions (40% CO₂/30% N₂/30% O₂, 100% CO₂, 80% CO₂/20% air, vacuum pack and air). It was found that the extension of shelf life of meat samples depended on the packaging conditions and augmented in the order: air < vacuum pack < 40% CO₂/30% N₂/30% O₂ < 80% CO₂/20% air < 100% CO₂. Longer shelf life was observed in samples supplemented with the volatile compounds of oregano essential oil and stored under the same packaging conditions mentioned above. The extension of shelf life may be due to the synergistic effect of volatile compounds of oregano essential oil and the modified atmosphere packaging used on the microbiological and physicochemical characteristics of meat. Indeed, both these hurdles can prolong and delay microbial growth or suppress the final counts of the spoilage microorganisms in comparison with the 'control' samples. The effect of essential oil volatile compounds was even more pronounced on the physicochemical changes of meat samples caused by microbial association. Oregano essential oil delayed glucose and lactate consumption, both indicators of meat spoilage aerobically as well as under 40% CO₂/30% N₂/30% O₂, and 100% CO₂. Finally, changes in other metabolites such as formic acid were also observed. © 2002 Elsevier Science B.V. All rights reserved.

Keywords: Meat; Active packaging; Spoilage; Quality index; Preservation; Natural products; Essential oil; Inhibition; Ecosystem

1. Introduction

It is well known that packaging makes food more convenient and gives the food greater safety assurance from microorganisms, biological and chemical changes so that the packaged foods may have a longer shelf life. As a result, packaging has become an indispensable element in the food manufacturing process. In order to meet the huge demand of the food industry, there has been a remarkable growth in the development of food packaging in the past decades. Among the packaging technologies developed by and for the food industry, modified atmosphere packaging has led the evolution of fresh and minimally processed food preservation, specially in meat and meat products for the past two decades. In such packaging, an initial atmosphere is generated by either permitting air to be enclosed or by injecting a

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desired initial gas mixture. This blend then changes as a result of multiple variables including: (i) permeation of oxygen, carbon dioxide, and water vapor through the package material; (ii) transmission of oxygen, carbon dioxide, and water vapor through the seal and defective structural areas; (iii) temperature of the package material which may lead to small changes in permeation; (iv) surface area of the package material; and (v) thickness of the package material (Tsigarida and Nychas, 2001). Such changes may influence/affect the contribution of different members of microbial association and as a consequence, an extension of shelf life can be achieved. Despite the extended shelf life of refrigerated products stored under vacuum pack/modified atmosphere packaging conditions, there is an increased concern about the growth/survival of microaerophilic psychrotrophic pathogens (Garcia de Fernando et al., 1995). Thus, additional hurdle(s) should be used to ensure the safety of such products. Smart, interactive and active packagings are terms that have been used to describe the innovative concept of package structures. It can be defined as a type of packaging that changes the condition of the packaging to extend shelf life or improve safety or sensory properties while maintaining the quality of the food. Since most food packaging systems consist of the packaging material, the food, and the headspace in the package, antimicrobial agents may either be incorporated into the packaging materials initially and migrate into the food through diffusion and partitioning, or be released through evaporation in the headspace. The latter can be achieved with essential oils that are volatile and are regarded as "natural" alternatives of chemical preservatives. In addition, their use in foods meets the current demands of consumers for mildly processed or natural products (Nychas, 1995). However, since their practical application is limited due to flavor considerations, as well as because their effectiveness is moderated due to interaction with food ingredients and structure, their application in active packaging can be of great importance (Juven et al., 1994; Skandamis and Nychas, 2000; Skandamis et al., 2000, 2002).

The aim of the present study was to evaluate the efficacy of volatile compounds of oregano essential oil in combination with the use of modified atmosphere packaging conditions.

2. Materials and methods

2.1. Extraction of essential oil

Five hundred grams of dried oregano (*Origanum vulgare*) was purchased from a local retail spice market and placed in a 2-l flask and 1 l of distilled water was added. A continuous steam distillation extraction was performed for approximately 3 h and the oil was collected and stored at 4 °C (Skandamis et al., 2000; Tsigarida et al., 2000).

2.2. Preparation of samples

Fresh beef of normal pH (pH 5.4-5.6) was transported to the laboratory within 30 min of purchase around 09:00 h from a local supermarket with a butcher department and transported to the laboratory where it was held about 1 °C for 1-2 h. The meat was cut into pieces of 25 g, thickness 0.8 cm and placed into plastic containers. Treatment of samples with oregano essential oil was performed as follows: Whatman paper No. 6 was cut into square $(2 \times 2 \text{ cm})$ pieces and each piece was immersed into pure essential oil extract for 10 s. Then each piece was drained for 30 s on the surface of common paper and then it was placed within the container but not in contact with the meat. Samples stored aerobically were simply enclosed into permeable polyethylene bags. Samples, stored under modified atmospheres, were packed individually into plastic pouches, supplemented or not with the oregano essential oil as described above. Each pouch was 300 mm wide \times 200 mm long, with oxygen permeability of 1.7 cm 3 m $^{-2}$ 24 h $^{-1}$ at 23 °C and 75% RH. The pouches were evacuated and flushed three times before being filled (3 1). After filling, the pouches were double heatsealed. The whole packaging was carried out using a HencoVac Machine. A limited number of samples were freeze-stored to serve as controls during sensory evaluation of color and odor.

2.3. Experimental design

Two independent experiments were designed. At the first stage, a preliminary factorial experiment was designed to assess the effect of active packaging (i.e. effect of volatile compounds of oregano essential oil within the package) on the shelf life of fresh beef stored at different packaging atmosphere conditions and temperatures. Five packaging atmospheres (air, 40% $\rm CO_2/30\%$ $\rm O_2/30\%$ $\rm N_2$, 100% $\rm CO_2$, 80% $\rm CO_2/20\%$ air and vacuum pack) and four temperatures (0, 5, 10 and 15 °C) with or without oregano essential oil within the packs were studied. At this stage, only a sensory assessment of the samples was performed.

At the following stage, a second factorial design was pursued in order to examine the effect of oregano essential oil, packaging atmosphere and temperature on the microbiological and physicochemical changes of fresh beef. Packaging atmospheres and temperatures were mainly selected on the basis of commercial importance and temperature abuse of meat during the chill chain. This resulted in the study of three packaging atmospheres (air, 40% CO₂/30% O₂/30% N_2 and 100% CO_2) and two storage temperatures (5 and 15 °C) with or without oregano essential oil (procedure described above). Vacuum packaging was excluded since it showed no better results than 40% CO₂/30% O₂/30% N₂ in terms of its visual appearance. In these experiments, sensory and microbiological analyses of samples were carried out as well as the physicochemical analysis of endogenous glucose and organic acids during meat storage.

The above procedure was performed twice and duplicate samples for each treatment were taken.

2.4. Microbiological analysis

Samples (25 g) of meat were aseptically weighed, added to sterile quarter strength Ringer's solution (90 ml), and homogenized in a stomacher (Lab Blender 400, Seward Medical, London) for 60 s at room temperature. Decimal dilutions in quarter strength Ringer's solution were prepared and duplicate 1 or 0.1 ml samples of the appropriate dilutions were poured or spread on the following media: plate count agar (PCA; Merck, 1.05463, Darmstadt, Germany) for total viable count (TVC), incubated at 25 °C for 72 h; Brochothrix thermosphacta on STAA medium supplemented with streptomycin sulfate, thallous acetate and cycloheximide (actidione), this medium was made from basic ingredients in the laboratory, and incubated at 25 °C for 72 h; lactic acid bacteria on MRS (Merck, 1.10660), overlaid with the same medium and incubated at 25 °C for 96 h under anaerobic conditions; Pseudomonas spp. on cetrimide-fucidin-cephaloridine (CFC) agar (Oxoid, CM559 supplemented with selective supplement SR 103E, Basingstoke, UK) incubated at 25 °C for 48 h; yeasts on Rose Bengal Chloramphenicol Agar (Lab M, 36, supplemented with chloramphenicol supplement, X009, Bury, UK) incubated at 25 °C for 5 days; *Enterobacteriaceae* on Violet Red Bile Dextrose Agar (Merck, 1.10275) incubated at 37 °C for 24 h.

The data (growth counts) were transformed to log₁₀ values. The Baranyi model (Baranyi et al., 1993) was fitted to the logarithm of the viable cell concentration. For curve fitting, the in-house program DMFit (Institute of Food Research, Reading, UK) was used which was kindly provided by Dr. J. Baranyi.

2.5. Chemical analysis

The concentration of glucose in meat during storage was assayed as follows: beef (25 g) was reduced to a fine suspension with 100 ml cold water (3–5 °C) in an Omni mixer (Waring, New Hartford, UK). The suspension was shaken (orbital shaker, 100 r.p.m., 45 min) at 3 °C, centrifuged ($4000 \times g$, 5 min, 3 °C), filtered (Millipore 0.22 m) and the clear filtrate used to determine glucose applying the GOD-PERID kit (Boehringer, Mannheim).

2.6. HPLC analysis of organic acids

The profile of the organic acids (treated with Trifluoroacetic acid) of beef samples was analysed by high performance liquid chromatography (Spectra Physics P2000 two pump systems with UV/VIS detector using Low Inertia Scanning Technologysimilar to Photodiode array-and software, San Jose CA, USA) using a Rheodyne 7125 injector and a 300 × 7.8 mm Aminex HPX-87H 5 m (Bio-Rad Laboratories, Richmont, CA) as described by Koutsoumanis and Nychas (1999). The compounds were separated isocratically with 0.009 N H₂SO₄ in distilled water (flow rate 0.7 ml/min). Peak width was 12, peak threshold was 600 and 0.034 AUFS. The whole spectra (190-330 nm) of the chromatograms were analysed. The solvents were HPLC grade and for the identification of peaks, solutions of reference substances (citric, lactic, acetic, tartaric, malic, succinic, formic and propionic) were analysed using the same program and their retention times (RT) and

Packaging	Shelf life (days)													
	0 °C		5 °C		10 °C		15 °C							
	OEO –	OEO+	OEO –	OEO+	OEO –	OEO+	OEO –	OEO+						
Air	24ª	32	10	14	5	8	3	5						
40% CO ₂ /30% O ₂ /30% O ₂	42	55	18	22	14	18	5	7						
100% CO ₂	66	78	30	40	13	18	6	9						
80% CO ₂ /20% air	44	58	26	34	13	23	7	15						
Vacuum pack	57	65	20	27	14	22	3	5						

Table 1
Effect of volatile compounds of oregano essential oil (OEO) on the shelf life of meat stored under different conditions

spectra were compared. The contribution of each identified compound was expressed as the percentage (%) of its peak area to the total area of all peaks eluted in each chromatograph. The precision of the results was always better than $\pm 5\%$.

2.7. Sensory analysis

Sensory evaluation of meat samples was performed during storage according to Gill and Jeremiah (1991) by a sensory panel composed of four members (staff from the laboratory). The same trained persons were

used in each evaluation, and all were blinded to which product was being tested. The sensory evaluation was carried out in artificial light and the temperature of packed product was similar to ambient temperature. Special attention was given to the color and the presence of exudate in the pack prior to opening and the assessment of abnormal odors during the opening of the pack (Kotzekidou and Bloukas, 1996). Each attribute was scored on a three-point hedonic scale where: 1 = acceptable; 2 = marginal; and 3 = unacceptable. Assessment was designed to identify spoilage conditions exclusively. Odor characteristic of raw beef,

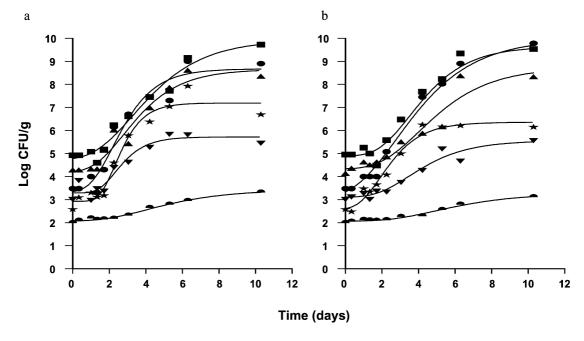


Fig. 1. Changes of microbial association of meat stored under aerobic conditions at 5 °C without (a) or with (b) the presence of volatile compounds from oregano essential oil [total viable counts, \blacksquare ; pseudomonads, \bullet ; *B. thermosphacta*, \blacktriangle ; LAB, \blacktriangledown ; *Enterobacteriaceae*, \bigstar ; yeasts, \blacktriangle].

^a Shelf life in days—as criterion of sensory analysis of sample.

as exemplified by special samples from frozen storage that were thawed prior to each sensory evaluation, was regarded as acceptable. Distinct putrid, sweet, sour or cheesy odors were regarded as indicative of spoilage and therefore unacceptable. Bright colors typical of fresh oxygenated meat were considered acceptable. A persistent dull appearance, or unusual color or appearance was considered unacceptable. The time in days before the taste panel considered the quality to be at the limit of acceptability (score=2-1) was defined as the sensory shelf life of samples, under the specific packaging conditions and oregano essential oil concentrations. The shelf life limit was defined as the point when 50% of the panelists rejected the sample.

3. Results

3.1. Sensory changes

Shelf life of beef samples at different packaging atmospheres and temperatures with or without treat-

ment with oregano essential oil are presented in Table 1. It is evident that in all cases, the volatile compounds of essential oil increased shelf life of meat compared to the control. The presence of essential oil contributed to the maintenance of visual appearance of meat for quite a long time (photographs not shown) with the exception of samples in 100% CO₂ where fresh color of meat disappeared immediately after packaging. Such effect was more pronounced at low temperatures. Sensory evaluation showed similar shelf lives for samples as those of the experiments of the first stage.

3.2. Microbiological changes

Aerobic storage of meat, at both temperatures, allowed total aerobic counts to reach high levels, with *Pseudomonas* spp. being the dominant microorganism, followed by *B. thermosphacta* and then lactic acid bacteria (Figs. 1a and 2a). Packaging under 40% CO₂/30% N₂/30% O₂ delayed and restricted growth of pseudomonads, whereas favored growth of *B*.

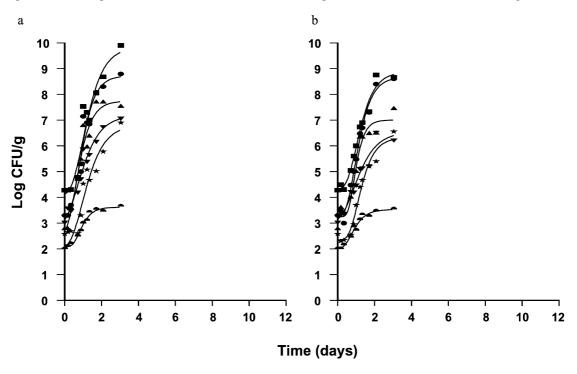


Fig. 2. Changes of microbial association of meat stored under aerobic conditions at 15 °C without (a) or with (b) the presence of volatile compounds from oregano essential oil [total viable counts, ■; pseudomonads, ●; B. thermosphacta, ▲; LAB, ▼; Enterobacteriaceae, ★; yeasts, ▲].

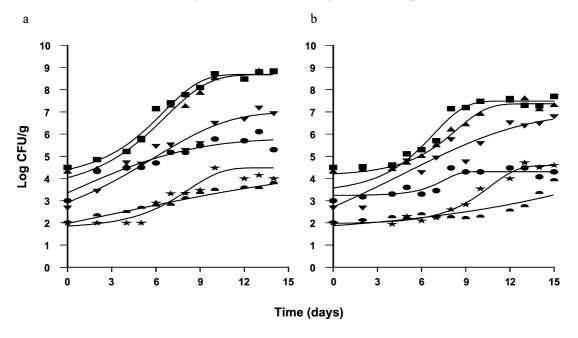


Fig. 3. Changes of microbial association of meat stored under 40% CO₂/30% O₂/30% N₂ MAP conditions without (a) or with (b) the presence of volatile compounds of oregano essential oil at 5 °C [total viable counts, \blacksquare ; pseudomonads, \bullet ; *B. thermosphacta*, \blacktriangle ; LAB, \blacktriangledown ; *Enterobacteriaceae*, \bigstar ; yeasts, \blacktriangle].

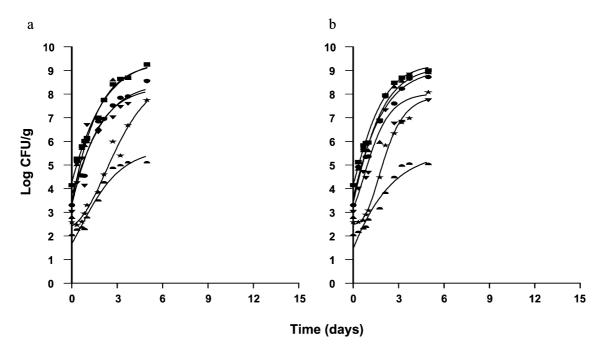


Fig. 4. Changes of microbial association of meat stored under 40% CO₂/30% O₂/30% N₂ MAP conditions without (a) or with (b) the presence of volatile compounds of oregano essential oil at 15 °C [total viable counts, \blacksquare ; pseudomonads, \bullet ; *B. thermosphacta*, \blacktriangle ; LAB, \blacktriangledown ; *Enterobacteriaceae*, \bigstar ; yeasts, \blacksquare].

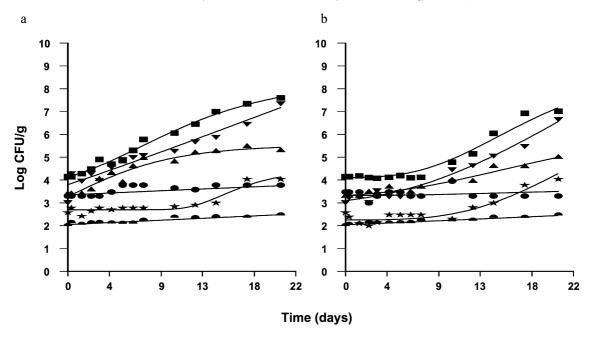


Fig. 5. Changes of microbial association of meat stored under 100% CO₂ conditions at 5 °C without (a) or with (b) the presence of volatile compounds from oregano essential oil [total viable counts, \blacksquare ; pseudomonads, \bullet ; *B. thermosphacta*, \blacktriangle ; LAB, \blacktriangledown ; *Enterobacteriaceae*, \bigstar ; yeasts, \blacktriangle].

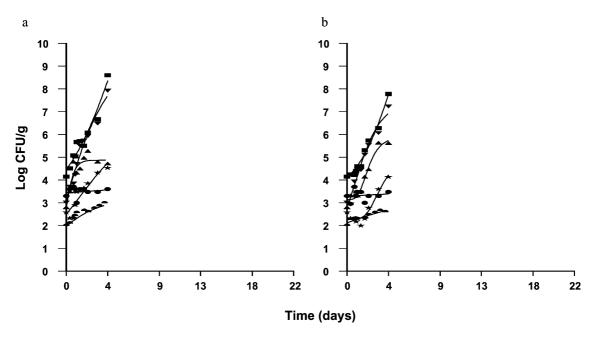


Fig. 6. Changes of microbial association of meat stored under 100% CO₂ conditions at 15 °C without (a) or with (b) the presence of volatile compounds from oregano essential oil [total viable counts, \blacksquare ; pseudomonads, \bullet ; *B. thermosphacta*, \blacktriangle ; LAB, \blacktriangledown ; *Enterobacteriaceae*, \bigstar ; yeasts, \blacktriangle].

thermosphacta and lactic acid bacteria (Figs. 3a and 4a). Lactic acid bacteria, which had the lowest initial population compared to the other members of natural microbial association of beef, indicated rapid growth and managed to reach the same levels as the other two microbial groups by the end of the storage period (Figs. 3 and 4), while in 100% CO₂ packaging, prevailed over the other members of microbial association figs. 5a and 6a. Enterobacteriaceae and yeasts contributed with the lowest percentage to the microbial association for the whole storage period at all packaging atmospheres and temperatures (Figs. 1a and 6a). The growth profile of microbial association was identical for the two storage temperatures. However, it should be noted that at 15 °C, the aforementioned changes occurred faster than at 5 °C.

The gradual release of volatile compounds of essential oil within the packaging affected the microbial association of meat packaged in modified atmospheres at 5 °C (Figs. 3b, 4b, 5b and 6b). In contrast, no significant changes compared to control samples were observed in aerobic conditions at both temperatures (Figs. 1b and 2b). Regarding the microbiological changes, the effectiveness of oregano essential oil in modified atmosphere packaging is evident since a difference on the population level of spoilage microorganisms of treated samples compared to the control samples, by $1-2 \log_{10}$ cfu/g, is consistently observed. Specifically, the inhibitory potential of essential oil followed the increased order: air < vacuum pack < 100% CO₂ < 40% CO₂/30% N₂/30% O₂ (Table 1).

3.3. Physicochemical changes

The inhibitory effect of essential oil at 5 °C was more pronounced on the physicochemical changes of meat during storage under modified atmospheres, i.e. on the rate of endogenous glucose consumption and the profile of changes in the organic acids (Fig. 7, Table 2). In particular, glucose and lactic acid were assimilated successively during aerobic storage and under 40% CO₂/30% N₂/30% O₂ packaging atmosphere regardless of the presence of oregano essential oil (Table 2). In samples stored under 100% CO₂, glucose consumption occurred in slower rate, whereas lactic acid increased slightly towards the end of storage (Table 2).

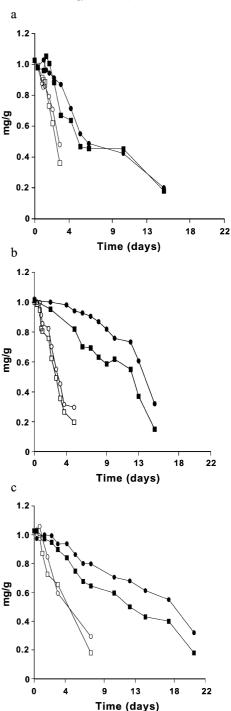


Fig. 7. Changes in D-glucose in meat stored under (a) aerobic conditions (b) 40% CO₂/30% O₂/30% N₂ at 5 °C (O, \square) and 15 °C (\bullet , \blacksquare), with (\bigcirc , \bullet) or without (\square , \blacksquare) the presence of volatile compounds derived from oregano essential oil.

Table 2 Changes in the chromatographic areas under, lactic, and formic acid peaks and unknown peaks with RT of 5.81 min (A), 7.55 min (B), 9.22 min (C), 10.9 min (D), 14.3 min (E) and 18.0 min (F) during storage of meat under different packaging atmospheres, without or with the presence of volatile compounds derived from the oregano essential oil, at 5 °C

Packaging atmosphere	Area	under p	peak (1	Area under peak $(10^3)^a$ at 210 nm														
	Without volatiles									With volatiles (h)								
	(h)																	
	0	48	73	120	170	247	290	336	518	0	48	73	120	170	247	290	336	518
Aerobic																		
A	31.4	20.3	10.8	nd^b	_	nd	_	_	_	31.4	23.2	28	16	_	4.2	_	_	_
В	nd	nd	2.9	6.1	_	nd	_	_		nd	1.2	1.3	5.8	_	3.1	_	_	_
C	5.5	3.2	nd	nd	_	nd	_	_	_	5.5	5.4	5.9	2.8	_	nd	_	_	_
Lactic acid	34.2	36	28.4	17	-	7	_	-	-	34.2	35	38.2	34	_	26	-	-	_
D	nd	nd	nd	nd	_	nd	_	-	_	nd	nd	nd	nd	_		_	-	_
Formic acid	16.7	29	3.2	2.9	-	1.1	_	-	-	16.7	17	28.6	26	_	8.6	-	-	_
E	10.9	5.9	3.1	nd	-	nd	_	-	-	10.9	6.9	7.5	5.4	_	5.3	-	-	_
F	233	120	97	113	-	255	-	-	-	233	150	265	240	-	342	-	_	-
40% CO ₂ :30% O ₂ :30%	6 N ₂																	
A	31.4	19	_	2	2	_	2	2	_	31.4	16	_	12	5	_	3	2	_
В	nd	nd	_	nd	1	_	2.5	2.5	_	nd	1.2	_	1.3	5.8	_	3.1	3.4	_
C	5.5	5.5	_	3.2	1.3	_	nd	nd	_	5.5	6.4	_	5	4.9	_	4.9	5	_
Lactic acid	34.2	30	_	30	29	_	26	20	_	34.2	37	_	42	39	_	43	45	_
D	nd	nd	_	nd	nd	_	nd	nd	_	nd	nd	_	2.3	6.5	_	5.1	5.1	_
Formic acid	16.7	23	_	11	14	_	13	4	_	16.7	11	_	3	nd	_	nd	nd	_
E	10.9	12	_	13	20	_	20	22	_	10.9	nd	_	nd	nd	_	nd	nd	_
F	233	192	_	197	181	_	191	208	-	233	236	-	235	230	-	300	350	-
100% CO ₂																		
A	31.4	17	11	12	_	_	nd	_	nd	31.4	21	15	8	_	_	6	_	6
В	nd	nd	nd	nd	_	_	nd	_	nd	nd	nd	nd	nd	_	_	nd	_	nd
C	5.5	4.5	5.1	5.2	_	_	1	_	nd	5.5	5	5.2	5.3	_	_	5	_	5
Lactic acid	34.2	36	30	29	_	_	32	_	25	34.2	37	40	39	_	_	40	_	46
D	nd	nd	nd	1.4	_	_	2	_	2.4	nd	nd	9	15	_	_	16	_	24
Formic acid	16.7	23	25	22	_	_	28	_	32	16.7	10	9	8	_	_	9	_	9
E	10.9	10	13	11	_	_	11	_	13	10.9	11	11	11	_	_	32	_	45
F	233	331	377	368	_	_	291	_	53	233	213	345	280	_	_	340	_	100

^{-,} not analysed.

The presence of oregano essential oil in all packaging conditions at 5° C delayed significantly the occurrence of the aforementioned changes due to the metabolic activity of the microbial association and/or even alters the profile of these changes compared to the control samples. For instance, in the case of 40% $CO_2/30\%$ $N_2/30\%$ O_2 packaging, lactic acid showed a significant increase at the end of storage rather than the reduction observed in the control samples (Table 2), whereas under 100% CO_2 atmosphere, the increase

in lactic acid was greater in packaged samples containing oregano essential oil than in those without essential oil (Table 2). On the other hand, at 15 °C, there were no significant changes in the rate of glucose consumption and the profile of changes in lactic acid (results not shown); however, changes in the HPLC profile of (wherever observed) were greater than those of glucose (results not shown). Remarkable changes were also evident in the profile of formic acid between treated and control samples at both storage

 ^a Each sample is the mean of two samples taken from different experiments (coefficient of variation of the mean of samples taken from different experiments, <5%). Each sample was analysed in duplicate (coefficient of variation of samples from the same experiment, <0.65%).
 ^b nd. not detected.

temperatures (Table 2); however, even in this occasion changes were more significant at 5 °C. It was also noteworthy that changes in formic acid as well as in the unknown acid (unknown peak RT=10.95) in relation to the profile of lactic acid were in reverse order between control and samples treated with essential oil. For example, in 40% CO₂/30% N₂/30% O₂ packaging at 5 °C, there was a reduction in lactic acid and increase in formic acid in the control, whereas treated samples exhibited the opposite trend (Table 2).

4. Discussion

The results showed that volatile compounds of oregano essential oil are capable of affecting both growth and metabolic activity of microbial association of meat stored at modified atmospheres; however, such inhibition is not as strong as that due to the contact of pure essential oil with microorganisms when this is added directly on the surface of meat (Skandamis and Nychas, 2001; Tsigarida et al., 2000). For this reason, in the present study, the extension of shelf life of meat treated with essential oil at all packaging atmospheres cannot be explained solely by the microbiological results. Indeed, differences of lower magnitude than that of previous studies (Skandamis and Nychas, 2001; Tsigarida et al., 2000) on growth of microbial association at both storage temperatures were evident between treated and control samples (Figs. 1-6). On the contrary, the inhibition that was illustrated through the physicochemical changes, i.e. glucose consumption and changes in organic acids are of greater impact since glucose and organic acids are regarded as potential spoilage indicators (Dainty, 1996; Nychas et al., 1998).

This study, in combination with current knowledge on the potential limited use of oregano essential oil as preservative in foods, leads to the general conclusion that the volatile compounds of oregano essential oil can expand its application to extend the shelf life of meat by (1) delaying of growth of specific spoilage organisms, (2) inhibiting or restricting their metabolic activity that cause spoilage through the production of spoilage microbial metabolites and (3) by minimizing the flavor consideration. For instance, in 40% CO₂/ 30% N₂/30% O₂ and 100% CO₂ atmosphere, the dominant spoilage microorganisms were found to be

B. thermosphacta and lactic acid bacteria, respectively (Figs. 3-5), which proliferated similarly at treated and control samples. In samples with 100% CO₂, the growth of lactic acid bacteria may be also stimulated by the pH decrease caused by CO₂ It needs to be noted that the profile of lactic acid was different in samples packed in map conditions and with oregano essential oil compared to the control (Table 2). The latter may also be associated with changes in the metabolic activity of one or more members of the microbial association of meat (Kakouri and Nychas, 1994), such as those in the homofermentative or heterofermentative pathway of glucose metabolism by lactic acid bacteria (Nychas et al., 1998; Tsigarida and Nychas, 2001). Additionally, the rapid decrease of formic acid, the production of the unknown D peak (RT = 10.9), in samples with lower microbial population (i.e. samples with the presence of volatile compounds) in comparison with control samples (stored in map conditions) could also account for the effect of volatile compounds on the metabolic activity of dominant microbial flora of meat (Table 2).

Since the sensory evaluation of samples did not reveal any effect (i.e. flavor consideration), the above results increase the need to further investigate the application of oregano essential oil to different foods considering the specific microbial association of each food and to study in detail the physicochemical changes that control spoilage. Under this scope, essential oils should be examined for their potential ability/capacity to delay spoilage.

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