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Shelf life and quality of dehydrated meat packed in edible coating under modified atmosphere

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Abstract

Effects of modified atmosphere packaging, with the addition of an active edible coating on quality and shelf-life of osmotically dehydrated pork meat was investigated. The pork was osmotically dehydrated, coated with starch based active edible coating with oregano essential oil and packed under atmospheric conditions and under modified atmosphere. The packaged meat was kept at 4°C for 2 months and was analyzed physico-chemically, microbiologically and sensorially. All physico-chemically parameters were improved and shelf-life was prolonged. Meat quality was promoted by using modified atmosphere packaging with active edible coating. Lipid oxidative changes were less pronounced in samples packed under modified atmosphere. The microbiological profile showed that the osmotic dehydration is hygienically safe, but samples packed under modified atmosphere showed higher level of microbial stability compared to samples packed in atmospheric conditions, although all groups remained stable during 60 days (which is a result of the active edible coating application).

Keywords

Packaging, edible coating, modified atmosphere packaging, osmotic dehydration, meat

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Introduction

Meat represents cellular, biochemically and structurally complex system, susceptible to many undesirable changes: microbial growth, lipid oxidation and sensorial change. This is directly related to consumer's acceptance (C. KENEDY & al. [1]). One of the main meat components is water (E. PUOLANNE & M. HALONEN [2]), placed in myofibrils, but it can also be found in the intracellular space between the myofibrils and sarcoplasm (J. BARAT & al. [3]). In order to decrease the water content in the meat, along with a_w value decrement, osmotic dehydration (OD) could be applied to fresh meat. This way, undesirable microbial and biochemical reactions retard, which implies prolonged shelf-life (S.J. SANTHURN & al. [4]). OD involves food sinking in a solution of high concentration (hypertonic= osmotic), which partially dehydrates food (H. MUJICAPAZ & al. [5]). The driving force for osmotic removal is the water concentration gradient. OD prolongs dehydrated product sustainability, reduces aroma loss and neutralizes unwanted textural changes and product shrinkage (K.B. PETROTOS & H.N. LAZARIDES [6]).

Oxygen promotes several types of deteriorative reactions in foods, including fat oxidation, browning reactions and pigment oxidation. Most of the common spoilage bacteria and fungi require O_2 for growth. Therefore, to increase shelf life of foods the pack atmosphere should contain a low concentration of residual oxygen (SANDHYA [7]). Refrigeration combined with modified atmosphere packaging (MAP) to reduce the microbial development and keep color attractive to consumers is one of the most widespread solutions (M. PEREIRA & M. MALFEITOFERREIRA [8]). MAP is a type of packaging that implies complete air removal, after which the vacuum is filled with gas or mixture of gases (CO_2 , O_2 and N_2). MAP can increase the shelf life of fresh meats by 50% to 400% compared to atmospheric packaging (D.N. RAO & N.M. SACHINDRA [9]). Used singly or in combination, these gases are commonly used to balance safe shelf-life extension with optimal sensorial properties of the food (SANDHYA [7]). In addition to CO_2 , by dissolving in the aqueous phase of meat, builds carbonic acid, which lowers the pH of meat and has a proven antimicrobial effect. CO_2 is very efficient for the preservation of red meat color, as it has 20 times higher affinity for binding with Mb, compared to O_2 's ability (M. BOECKMAN [10]). It was found that the extension of shelf life of meat samples depended on the packaging conditions and augmented in the order: air < vacuum < 40% CO_2 /30% N_2 /30% O_2 < 80% CO_2 /20%air < 100% CO_2 (P.N. SKANDAMIS & G.J.E. NYCHAS [11]).

Nowadays, the rapid development of functional food market has led to the emergence of active packaging (C.A. CAMPOS & al. [12]). Synthetic preservatives and antioxidants have been replaced by substances declared as

natural (spices, organic acids, polypeptides, essential oils) that have a wide range of biological and the pharmacological activity and can be applied as active packaging (J.H. HAN [13]). Essential oils are aromatic compounds derived from plant material with strong active antimicrobial and antioxidant activity (S. NANASOMBAT & P. WIMUTTIGOSOL [14]). However, the use of essential oils in the food industry could alter sensory properties of food (L. SANCHEZ-GONZALES & al. [15]), for an effective dose of oil may exceed the acceptable sensory levels (M. VIUDA-MARTOS & al. [16]). To avoid these problems, an alternative is essential oils implementation in the composition of edible films and coatings. Edible films and coatings, as carriers of essential oils, have the effect of encapsulation matrix, which is reflected in the necessary minimum doses of essential oils, limited volatility and controlled and gradual release of active components from the surface of the food (D. SALARBASHI & al. [17]). Several studies have shown that edible coatings help to prolong the shelf life and are important for perishable foods such as fish products (E. TORRIERI [18]), ready-to-cook poultry (V. GIATRAKOU & al. [19]), shrimps (M. MASTROMATTEO & al. [20]), sausages (M. MASTROMATTEO & al. [21]), beef stakes (A. LA STORIA & al. [22]).

In this paper, fresh pork was osmotically dehydrated, coated with a starch edible coating containing oregano essential oil and packed under atmospheric conditions and under modified atmosphere. The packaged meat was kept at 4°C for 2 months and was analyzed physico-chemically, microbiologically and sensorially. The aim of this research was to investigate contribution of applied active starch edible coating and modified atmosphere packaging on osmotic dehydrated meat quality and sustainability during 2 months of storage.

Materials and Methods

Experimental setup: Fresh pork (*Musculus brachii*) (48h post mortem) was purchased at the local butcher shop in Novi Sad, shortly prior to the experiment. Fresh pork was trimmed of external fat and connective tissues and manually cut into approximately (1x1x1) cm cubes. Samples were osmotically treated in sugar beet molasses solutions (80 kg•l⁻¹). The experiment was conducted under atmospheric pressure at 22°C for 5 hours. Solution to sample ratio was 5:1 (w/w) to avoid significant dilution of the medium by water removal. Every 15 min, the meat samples in osmotic solutions were stirred to provide better homogenization of the osmotic solution, considering the amount of diffused water from the samples. After the process of OD, the meat samples were lightly washed with distilled water, gently blotted with paper to remove excessive water from the surface.

Starch edible coating was prepared by casting aqueous modified maize starch solution (1.5 mg•ml⁻¹). The solution was heated at 90°C for 60 min in a water bath. A weight of glycerol equal to 40% of the original starch was added and the solution was kept hot with mechanical stirring for 10 min. Guar-xanthan modified mixture was added in a portion of 0.1% of the initial starch weight and oregano essential oil in a portion of 2%. The edible coating solution was homogenized using homogenizer at 166.67 Hz for 1 min. Right after osmotic treatment, meat samples were dipped into an edible coating solution for 1 min. After 2 h procedure was repeated. Samples were left to dry for 2 h more on the table surface and packed.

Atmosphere modification and package sealing were performed using a packaging machine CFS COMPAKT 420. Packaging molds were formed of two different foils: the first foil (transparent PVC//PE-EVOH-PE) was in the form of the tray and the second foil (PET//PE-EVOH-PE) was covered. One sample group received gas treatment: 30%CO₂+70%N₂ (labeled as OD+S+MAP), while the other sample group was packed under atmospheric conditions (labeled as OD+S+ATM). Packaged meat samples were stored at (4±0.5) °C and sampled after 15, 30, 45 and 60 days for analyses.

Physico-chemical analysis: Water content was determined by the ISO 1442:1997 standard [23] and water activity (*a_w*) was measured using a water activity measurement device TESTO 650 with an accuracy of ±0.001 at 25°C. pH measurements were determined by ISO 11289:1993 [24]. Mineral matter content was determined in accordance with ISO 6869:2000 standard [25].

TBARS (2-thiobarbituric acid reactive substances) test was performed according to the method of N.A. BOSTOGLU & al. [26], with modifications. Total volume of trichloroacetic acid (TCA) was added to the sample and extraction was performed in ultrasonic bath XUB 12 (Grant Instruments, Cambridge, UK). Spectrophotometer Jenway 6300 (Jenway, Felsted, United Kingdom) was used. TBARS values were expressed as milligrams of malondialdehyde per kilogram of the sample. TBARS test was performed on three samples from each batch in duplicate.

Microbiological analysis: Total Viable Counts (TVC) and Enterobacteriaceae were determined in accordance with appropriate ISO standards 4833:2003 [27] and 21528-2:2004 [28]. Total yeasts and molds count was enumerated according to ISO 21527-2:2008 [29]. *Escherichia coli* and *Clostridium* spp. were determined according to appropriate ISO standards: ISO 16649-2:2001 [30] and ISO 15213:2003 [31].

Color measurements: Color of each sample was measured immediately after slicing. The CIE L* (lightness), CIE a* (redness) and CIE b* (yellowness) color coordinates

were determined using MINOLTA Chroma Meter CR-400 (Minolta Co., Ltd., Osaka, Japan) using D-65 lighting, a 2° standard observer angle and an 8-mm aperture in the measuring head (B. ŠOJIĆ & al. [32]). The Chroma Meter was calibrated using a Minolta calibration plate (No. 11333090; $Y = 92.9$, $x = 0.3159$; $y = 0.3322$). Six replicates measures of surface color were performed on three samples from each batch.

Sensory evaluation: Dehydrated meat in molasses was assessed by a panel of six members and ISO standards were applied [33, 34]. Panelists identified descriptors and set up descriptive profiles according to ISO 4121:2003 [35]. The selected descriptors were rated using a 5-point intensity scale, where: 0-is not observed, and 5-strong in relation to the selected property.

Statistical analysis: Descriptive statistical analyses for calculating the means and the standard error were performed using MicroSoft Excel software. Regression analysis and the evaluation of analysis of variance (ANOVA) of obtained results were performed for comparison of means, and significant differences are calculated according to post-hoc Tukey's HSD test at 95% confidence limit, using StatSoftStatistica 10.0. Principal Component Analysis (PCA) and Cluster Analysis (CA) have been applied to classify and discriminate the different samples.

Results and Discussions

Physico-chemical analysis

Physicochemical data for fresh meat, after OD (beginning of the storage period) and after storage period are given in Table 1. Water content decreased from 75.127% to 49.703% after OD, followed by *a_w* reduction from 0.928 to 0.802, resulting with more stable meat products. After 60 days water content was slightly reduced so as *a_w* value, compared to the beginning. The obtained results are in agreement with V. FILIPOVIĆ & al. [36] and D. ŠUPUT & al. [37]. The post-hoc Tukey's HSD test was used for water content, *a_w* and pH comparison between samples. Statistically significant difference was found between all samples, before and after OD, $p < 0.05$ (Table 1).

Sugar beet molasses is an excellent medium for OD, because of its high dry matter content and specific nutritional composition with all mineral components in the dissolved state. Sugar beet molasses significantly enrich the content which is dehydrated in terms of vitamins and minerals, as shown in Table 2. The obtained results are in agreement with the D. ŠUPUT & al. [37] findings. The comparison between samples, concerning minerals content, were done using post-hoc Tukey's HSD test, and statistically significant difference was found in all samples, $p < 0.05$ (Table 2).

Table 1. Water content (%), pH and water activity (a_w)

	Fresh pork meat	OD meat - beginning of storage	OD meat - end of storage	
			OD+S+ATM	OD+S+MAP
Water content (%)	75.127±0.145 ^d	49.703±0.069 ^c	48.726±0.35 ^b	48.553±0.042 ^a
pH	6.183±0.008 ^c	6.297±0.006 ^d	6.259±0.011 ^b	6.217±0.007 ^a
a_w	0.928±0.001 ^c	0.802±0.0005 ^b	0.801±0.001 ^b	0.798±0.0005 ^a

^{a-d} Different letters written in the superscript in the rows of the table indicate the statistically different means between the samples.

Table 2. Mineral content (mg·100 g⁻¹)

	Mineral content (mg·100 g ⁻¹)				
	Fe	Mg	Ca	K	Na
Sugar beet molasses	4.41±0.5 ^b	86.39±0.8 ^b	666.31±0.6 ^c	2399.76±55.3 ^c	1046.21±10.6 ^c
Fresh pork meat	2.23±0.1 ^a	28.42±0.3 ^a	36.94±1.5 ^a	365.24±13.2 ^a	63.74±1.6 ^a
OD meat – beginning of storage	4.43±0.1 ^b	124.26±1.1 ^c	45.02±3.6 ^b	1156.33±16.5 ^b	247.52±3.3 ^b
OD meat – end of storage (OD+S+ATM)	4.53±0.2 ^c	127.20±5.6 ^c	46.09±7.1 ^b	1183.72±7.1 ^b	253.38±10.9 ^b
OD meat – end of storage (OD+S+MAP)	4.56±0.3 ^c	125.20±3.1 ^c	47.69±5.0 ^b	1190.36±14.9 ^b	258.77±6.7 ^b

^{a-c} Different letters written in the superscript in the rows of the table indicate the statistically different means between the samples.

Lipid oxidative changes of examined meat samples packed under MAP and ATM, expressed by TBARS test (mg malondialdehyde·kg⁻¹), are shown in Figure 1.

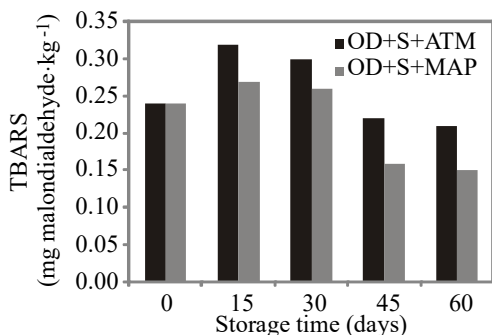


Figure 1. TBARS changes during storage period (OD – osmotic dehydration, S – starch active coating applied, ATM – packaging under atmospheric conditions, MAP – packaging under modified atmosphere)

At the beginning of the storage (0 day), TBARS value was low (0.24 mg MDA·kg⁻¹) which proves that OD was effective in preventing TBARS increase (D. ŠUPUT & al. [37]). During storage, increase in TBARS values was obtained for both sample groups as a result of lipid

oxidation (L. ZHANG & al. [38]). Maximum values were recorded after 30 storage days: 0.3 and 0.26 mg MDA·kg⁻¹ for samples packed under the atmosphere and modified atmosphere conditions, respectively. Still, the highest values were lower than those obtained without starch edible coating application (D. ŠUPUT & al. [37]). According to some authors, TBARS values higher than 0.5 mg MDA·kg⁻¹ (M.C. LANARI *et al.* [39]) lead to the emergence of rancidness in meat or according to others limit is 1 mg MDA·kg⁻¹ in meat products (B. ŠOJIĆ & al. [32]). After the 30th day, TBARS values decreased in both group samples. The reducing lipid oxidation in packed meat samples could be attributed to the presence of active phytochemicals in essential oil (A.D. GUPTA & al. [40]). Lower values were recorded in case of samples packed under modified atmosphere because of the CO₂ presence in the MAP composition (P. LÓPEZ-LORENCO & al. [41]) while, at the same time O₂ content is limited (D. ŠUPUT & al. [37], J.A. ORDONEZ & D.A. LEDWARD [42]). Studies showed that the initial reduction of O₂ in the atmosphere with a very low concentration and prevention of the entry of O₂ during storage by use of a gas-impermeable film does not lead to oxidation (R.D. NARASIMHA and N.M. SACHINDRA [43]). Another reason for low TBARS values is essential oil implementation compared to results previously obtained without using an active edible coating (D. ŠUPUT & al. [37]).

Microbiological analysis

The results of the microbial counts are reported in Table 3. TVC values lower than 7 log CFU·g⁻¹ indicate good fresh meat quality (ICMSF [44]) TVC and Enterobacteriaceae count decreased significantly after OD treatment when comparing values of fresh meat and values at 0th day. This indicates that OD treatment is hygienically safe, which is in agreement of previous findings (D. ŠUPUT & al. [37]).

samples were below the limit value for the growth of most bacteria, which is a clear indicator that OD has a positive impact on the microbiological profile of osmotically dehydrated meat (ŠUPUT & al. [37]). On the other hand, oregano oil, applied as active component of edible coating, preserved microbial profile stable during the whole storage period in terms of bacterial strains. Still, yeast and moulds appeared in the second part of storage time, most probably because the a_w value of packed meat was adequate for their

Table 3. Microbiological profile during the storage period (log CFU·g⁻¹)

Microbiological profile (log CFU·g ⁻¹)					
Fresh meat					
Storage period	Total viable counts	<i>Enterobacteriaceae</i>	<i>E. coli</i>	<i>Clostridium spp.</i>	Yeasts and moulds
0	6.2	2.59	<1	<1	<1
OD+S+ATM					
0	3.18	2	<1	<1	<1
15	3.78	1.85	<1	<1	2.95
30	3.87	1.7	<1	<1	3.53
45	4.02	<1	<1	<1	3.78
60	4.11	<1	<1	<1	4.38
OD+S+MAP					
0	3.18	2	<1	<1	<1
15	3.34	1	<1	<1	<1
30	3.7	<1	<1	<1	3.43
45	3.86	<1	<1	<1	3.69
60	4.07	<1	<1	<1	3.93

„<1” – less than 1 means that microorganisms were not detected

During the storage, in both sample groups, there was an increase in the TVC, with the observation that values for samples packed under ATM were higher at all control points in comparison with samples packed under MAP. MAP had a significant effect on TVC because of low O₂ concentration presented in packaging (SANDHYA [7]). The correlation coefficient between samples packed in MAP and ATM for TVC was 0.988, statistically significant at p<0.01 level. All TVC values were lower compared to results obtained by D. ŠUPUT & al. [37] when starch edible coating wasn't applied in the same packaging system. Initial pH was lowered so that's why Enterobacteriaceae count decreased during storage period. The correlation coefficient between samples packed in MAP and ATM for Enterobacteriaceae was 0.991, statistically significant at p<0.01 level.

In both groups of OD packed meat *E. coli* and *Clostridium spp.* were not detected, so as in fresh pork. After the OD process, the a_w value of dehydrated meat

development. Another reason is the presence of starch edible coating, which is polysaccharide. Yeast and moulds value increased along with storage period.

Color measurements

Among all sensory attributes of meat, color is considered one of the most important physical traits because once color is deemed unacceptable, all other sensory attributes lose their significance to consumers (R.A. MANCINI & M.C. HUNT [45]). The color parameters, lightness (CIE L* value), redness (CIE a* value) and yellowness (CIE b* value) of the examined sausages are shown in Fig 2.

According to obtained results, OD treatment changed color values: lightness (CIE L* value) decreased, while redness (CIE a* value) and yellowness (CIE b* value) increased which is in agreement with previous findings (D. ŠUPUT & al. [37]). It can be observed that color results for samples packaged in MAP do not change during storage period, according to ANOVA, statistically significant at

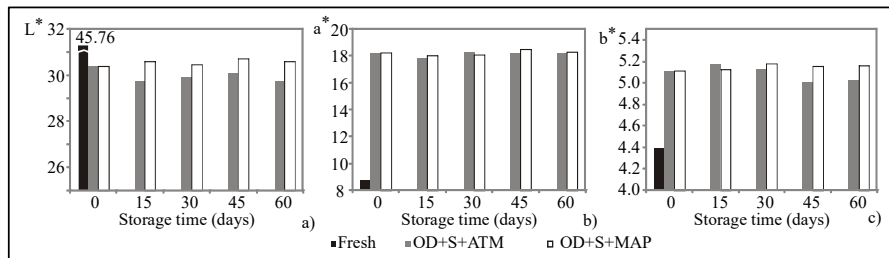


Figure 2. Changes in CIE Lab color coordinates during storage period (OD – osmotic dehydration, S – starch active coating applied, ATM – packaging under atmospheric conditions, MAP – packaging under modified atmosphere)

$p < 0.05$ level. The reason for this is MAP application since CO_2 is very efficient for the preservation of red meat color, as it has 20 times higher affinity for binding with Mb, compared to O_2 's ability (M. BOECKMAN [10]). O_2 promotes several types of deteriorative reactions in foods, including browning reactions and pigment oxidation. Therefore, meat color improvement depends on the atmosphere that should contain low concentration of residual O_2 (SANDHYA [7]) in order to avoid color defects (N. PENNEY & R.G. BELL [46]).

compared to samples packed under the same conditions, but without the active edible coating (D. ŠUPUT & al. [37]). Samples packed under MAP were better scored regarding all texture parameters compared to ATM samples (3.67 compared to 3.25). MAP and active edible coating positively influenced meat chewable property and elasticity (D. ŠUPUT & al. [37]) but also softness and juiciness.

Statistical analysis

A dendrogram for observed samples (based on the differences in physico-chemical, color, microbiological

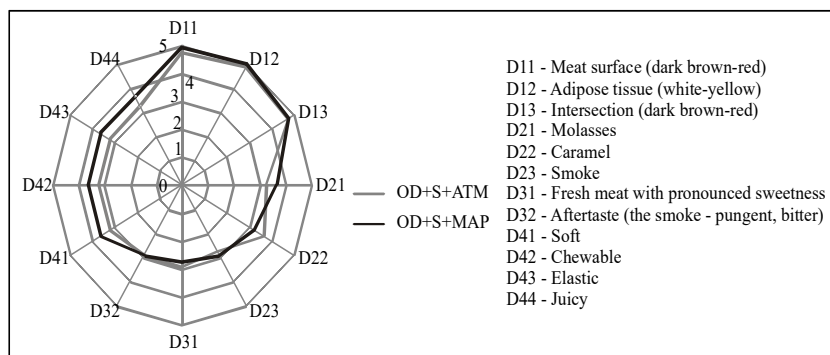


Figure 3. Sensory assessment (OD – osmotic dehydration, S – starch active coating applied, ATM – packaging under atmospheric conditions, MAP – packaging under modified atmosphere)

Sensory evaluation

Results related to sensory assessment are presented in Figure 3. Regarding color, samples packed under MAP obtained better scores, particularly in terms of meat surface and adipose tissue color. In terms of flavor, caramel and smoke flavor dominated in samples packed under MAP, while smoke flavor dominated in samples packed under ATM. The results of the taste evaluation were evaluated as average for both groups of samples. All marks were lower

and sensory characteristics) accusing complete linkage as an amalgamation rule and the City-block (Manhattan) distance as a measure of the proximity between samples is shown in Figure 4a. The dendrogram based on observed data showed the proper distinction between investigated samples (Fresh meat, OD meat, OD+S+ATM and OD+S+MAP), due to their different technological parameters preparation patterns. Fresh meat is the most distinct sample compared to other samples, being technologically untreated (amalgamation

distance is more than 1100 to other samples). There are also some minor differences between OD meat and OD+S+ATM and OD+S+MAP (amalgamation distance is almost 100, according to Fig. 4a).

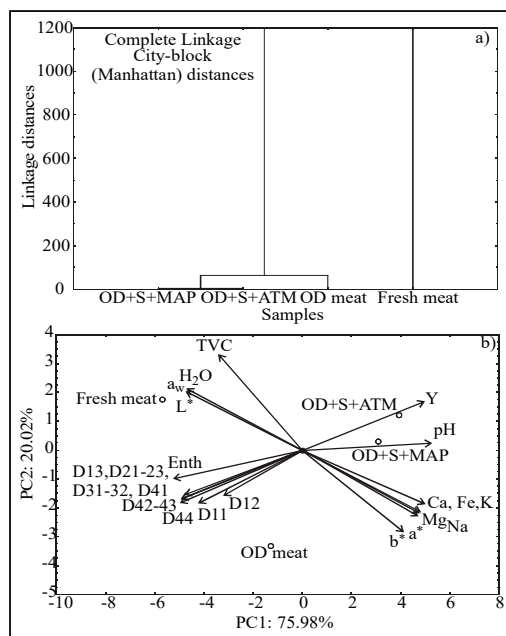


Figure 4. Cluster analysis (a) and PCA scatter biplot (b) of analyzed physico-chemical, color, microbiological and sensory characteristics of treated meat samples (OD – osmotic dehydration, S – starch active coating applied, ATM – packaging under atmospheric conditions, MAP – packaging under modified atmosphere)

The PCA of the present data explained that the first two principal components accounted for 96.0% of the total variance (76.0% and 20.0%, respectively) in the 26 variables, Figure 4b. The experimental data was transformed by autoscaling procedure, i.e. all values are subtracted to mean values and divided by standard deviations. Three Eigenvalues larger than 1 were obtained, 19.75, 5.20 and 1.04 for PCs. Considering PCA performed on the data, all variables exhibited almost the same contribution to the first principal component calculation, according to correlation. The positive contribution on the second principal component calculation was observed for TVC (which contributed 11.4% of the total variance), while negative scores on second principal component calculation was observed for: a* (5.1%) and b* (7.9%). The points shown in the PCA graphics, which are geometrically close to each other, indicate the similarity of patterns that represent these points. The orientation of the vector describing the variable in factor space indicates an

increasing trend of these variables, and the length of the vector is proportional to the square of the correlation values between the fitting value for the variable and the variable itself. The angles between corresponding variables indicate the degree of their correlations (small angles corresponding to high correlations).

Conclusions

Osmotic dehydration proved to be an effective preservation method for meat shelf-life prolongation along with keeping the product quality. Physico-chemical parameters were improved (water content decreased, a_w value decreased and pH increased) which corresponds to stable microbiological profile. However, optimal results were obtained for samples packed in modified atmosphere compared to samples packed in atmospheric conditions. Samples remained microbiologically stable during the whole storage period because the active edible coating was applied as additional protection. Also, the modified atmosphere application implies better results regarding lipid oxidation. Osmotic dehydration changed color values: lightness decreased, while redness and yellowness increased. Color results for samples packaged in modified atmosphere do not change during storage period since modified atmosphere packaging is very efficient for red meat color preservation. Osmotic dehydration provides shelf-life extension, but combination with modified atmosphere packaging is more effective in terms of product quality and sustainability. Application of active edible coating with oregano essential oil affected some sensorial properties. All three methods of preservation, osmotic dehydration, modified atmosphere packaging and active edible coating application, proved to be effective for meat quality and shelf-life improvement.

Conflict of interest disclosure

There are no known conflicts of interest in the publication of this article. The manuscript was read and approved by all authors.

Compliance with ethical standards

Any aspect of the work covered in this manuscript has been conducted with the ethical approval of all relevant bodies and that such approvals are acknowledged within the manuscript.

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