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RESEARCH ARTICLE

Effects of Packaging Atmospheres on the Quality and Shelf Life of Beef Steaks [1]

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Summary

The effects of modified atmosphere packaging on the quality and shelf life of beef steaks were investigated. Beef steaks were packed under varying modified atmosphere conditions (A = 70% CO_2, 30% N_2; B = 80% CO_2, 20% O_2; C = 30% CO_2, 10% N_2, 60% O_2; D = 100% CO_2; E= 100% O_2) and vacuum. The lowest pH and TBA values were found in the samples packed with carbondioxide and vacuum. High level oxygen increased lipid oxidation during the storage. Oxygen in the pack also caused discoloration. Microbial inhibition was observed in case of presence of carbondioxide. The best quality was observed with 70% CO_2, 30% N_2 atmosphere. Shelf life of beef steaks packed under this atmosphere was 14 days.

Keywords: *Modified atmosphere packaging, Beef, Quality, Shelf life*

Sığır Etinin Kalitesi ve Raf Ömrü Üzerine Paketleme Atmosferlerinin Etkileri

Özet

Bu çalışmada sığır etinin kalitesi ve raf ömrü üzerine paketleme atmosferlerinin etkileri incelenmiştir. Sığır biftekleri değişik atmosfer koşulları (A = %70 CO_2, %30 N_2; B = %80 CO_2, %20 O_2; C = %30 CO_2, %10 N_2, %60 O_2; D = %100 CO_2; E = %100 O_2) ve vakum altında paketlenmiştir. En düşük pH ve TBA değerleri karbon dioksit ve vakumla paketlenmiş örneklerde bulunmuştur. Yüksek düzeylerde oksijen depolama sırasında lipid oksidasyonunu arttırmıştır. Paket içerisindeki oksijen renk bozulmalarına neden olmuştur. Karbondiokdit varlığında mikrobiyal inhibisyon gözlenmiştir. En iyi kalite %70 CO_2, %30 N_2 atmosferi ile elde edilmiştir. Bu atmosfer altında paketlenen sığır eti bifteklerinin raf ömrü 14 gün olmuştur.

Anahtar sözcükler: *Modifiye atmosferde paketleme, Sığır eti, Kalite, Raf ömrü*

INTRODUCTION

Natural and processed food s are exposed to serial changes according to the present medium conditions before consuming and because of this, loss of nutritive value and sensory characteristics are observed. In order to prevent or remove the spoilage, various food protection methods are used [1]. Modified atmosphere packaging (MAP) technology is one of the protection methods in which the surrounding atmosphere of the food is changed [2]. Basic process in MAP is to remove the air inside the package and put in a gas or gas combination instead, and then seal hermetically.

Beef is a highly perishable food commodity with a short shelf life. Microbial growth and metabolic activities are an important cause of spoilage in fresh meat. Such spoilage may occur as visible growth, textural changes or through the development of off-odours and off-flavours. Spoilage of meats leads to significant economic losses for the meat industry [3]. During production, processing, distribution and storage, food undergoes deterioration from chemical and microbial processes. After slaughter, there are many factors that determine meat quality,

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including temperature and time of ageing and packaging [4]. Shelf life and quality of fresh beef are strongly influenced by initial meat quality, package parameters and storage conditions [5]. Gas combination used in MAP is specific. The effectiveness of the gas on prevention of food spoilage should be known [2]. In the search of a consumable and desirable meat, researchers continue to find out the right gas combination such as using carbon monoxide to stabilize the meat color [6-7].

The objective of the present study was to investigate the effects of modified atmosphere packaging on the quality and shelf life of beef steaks using different gas combinations, in order to propose the most suitable MAP gas combination to maintain the preferred chemical, physical and microbiological beef steak quality.

MATERIALS and METHODS

Materials

The study was performed at a private meat processing factory. Meat and packaging materials were provided by this factory. Beef steaks taken from *Longissimus dorsi* muscle were used as research material. The meat was trimmed of external fat, cut into pieces of 1-1.5 cm, placed on plastic foam trays in pouches of polyamide (80 µm; oxygen transmission rate 20 cm^3/m^2 d bar at 20°C and water vapour permeability <1.5 g/m^2 d bar at 20°C)/polyethylene (250 µm; oxygen transmission rate 68 ml/m^2 d atm at 10°C, 23.08 ml/m^2 d atm at 25°C and water vapour permeability of 1.37 ml/m^2 at 37.8°C) of size 17.5 x 13.5 x 2.5 and packed under vacuum and modified atmospheres (A = 70% CO_2, 30% N_2; B = 80% CO_2, 20% O_2; C = 30% CO_2, 10% N_2, 60% O_2; D =100% CO_2; E = 100% O_2). The packages were gas flushed and sealed using MULTIVAC packaging machine (R230/719) in the factory. Each pouch contained approximately 300 g beef steaks (17.5 cm x 13.5 cm x 2.5 cm). Packed meat steaks were transferred to the laboratory belonging to university, stored at 4°C and analyzed on the 0, 7 and 14th day of storage. Experiment was repeated with samples packaged at two separate times. For chemical and microbiological analyses, meats were homogenized using a kitchen blender, the homogenates were pooled for each treatment. All assays were conducted on duplicate samples from the homogenate pool for each replication. Results were expressed as means of four duplicates.

Methods

Surface color measurements were determined using a CR-400 chroma meter (Minolta Co., Osaka, Japan). A white calibration plate was used to calibrate the instrument. The L* value represents lightness, a* and b* values represent redness and yellowness, respectively. Four zones of each beef steaks were measured and the results were expressed as means of four measurements. Penetrometer (Humboldt) equipped with a total of 45 g load weight was used to determine the hardness of beef steaks. The needle of instrument was placed at the surface of beef steaks and the instrument was turned on for 10 s to produce a puncture. The depth of puncture was measured in mm with greater depth indicating lower hardness. Penetration measurements were done in 8 different areas of the meat surfaces. Data are means of eight measurements. The pH of beef steak was measured using pH-meter (WTW Inolab, Weilhem, Germany). The pH electrode was dipped into a mixture of homogenized sample and distilled water (1/1) [8]. The TBA distillation method was performed [9]. Homogenized 10 g sample was distilled after addition of 2.5 ml HCl + distilled water solution (1/2). A 5 ml of distilled solution was transferred into the stoppered test tube and 5 ml TBA solution (0.288 g TBA/100ml distilled water) was added, test tube was shaken and left in the water bath at 110°C for 35 min. The absorbance was determined by a spectrophotometer at 538 nm against a blank containing distilled water and TBA solution. The results were expressed as mg malonaldehyde/kg. For microbiological analysis, a 25 g sample was added to 225 ml of sterile 0.1% (w/v) saline peptone water (0.1% peptone and 0.85% NaCl) and homogenized in a stomacher (Stomacher 80, Seward Medical, London, UK) for 2 min at low speed at room temperature. Serial decimal dilutions were made and plated onto appropriate culture media. Aerobic mesophilic counts (APC) were determined using Plate Count Agar (PCA- Oxoid CM0463B) (35°C for 48 h under aerobic conditions). Lactic acid bacteria (LAB) were determined on APT agar (All Purpose Agar with Tween 80-Acumedia 7302) (25°C for 72 h under anaerobic conditions). The colonies from selected dilutions were enumerated as colony forming units. Data were analyzed by a two factor factorial arrangement in a completely randomized design. The two factors were the five packaging methods (A,B, C,D,E,V) and the three storage days (0, 7, 14). Analysis was conducted using the SAS software (Statistical Analysis System, Cary, NC, USA). When main effects or interactions were significant, Duncan's Multiple Range test was used.

RESULTS

There were no statistical differences in lightness (L* value) among the atmospheres at the end of the storage period *(Table 1)*. Redness (a* value) decreased in the samples packed with oxygen during the storage ($P<0.01$). While the redness of the samples packed under 80% CO_2, 20% O_2 atmosphere low, yellowness (b* value) reached the highest levels *(Table1)*, b* values shown little fluctuation during the storage period.

Hardness of meat packed under 70% CO_2/30% N_2 and 100% CO_2 atmospheres significantly decreased during the storage *(Table 2)*. There were no significant differences in texture values of the samples packed under 30% CO_2/10%

Table 1. *Effects of packaging and storage time on the color values (L,a,b) of beef steaks stored at 4°C* [1]

Tablo 1. *4°C'de depolanan sığır etinin renk değerleri üzerine paketleme ve depolama süresinin etkileri*

Color	Packaging [2]	Storage Days		
		0	7	14
L*	A	27.79±0.45 a,x	31.25±0.54 b,x	29.90±0.63 b,x
	B	27.79±0.45 a,x	34.41±1.6 b,y	32.90±0.98 b,x
	C	27.79±0.45 a,x	29.74±1.19 ab,x	31.07±1.06 b,x
	D	27.79±0.45 a,x	32.42±1.42 b,xy	33.47±1.14 b,x
	E	27.79±0.45 a,x	30.43±0.45 ab,y	32.25±2.12 b,x
	V	27.79±0.45 a,x	31.22±0.66 b,y	30.25±0.43 a,x
a*	A	19.7±0.53 a,x	19.26±2.06 a,x	17.87±0.72 a,x
	B	19.7±0.53 a,x	16.88±1.35 a,x	10.58±1.52 b,y
	C	19.7±0.53 a,x	16.60±0.95 b,x	14.24±0.78 b,z
	D	19.7±0.53 a,x	19.06±0.82 a,x	18.98±0.83 a,x
	E	19.7±0.53 a,x	16.97±0.79 b,x	17.42±0.59 b,x
	V	19.7±0.53 a,x	16.55±0.62 b,x	19.36±0.80 a,x
b*	A	6.18±0.19 a,x	5.97±0.93 a,x	5.64±0.24 a,x
	B	6.18±0.19 a,x	5.97±0.73 a,x	7.20±1.78 a,y
	C	6.18±0.19 a,x	4.48±0.78 a,x	5.45±0.56 a,z
	D	6.18±0.19 a,x	6.06±0.75 a,x	5.34±0.59 a,x
	E	6.18±0.19 a,x	4.47±0.45 b,x	5.14±0.59 ab,x
	V	6.18±0.19 a,x	5.92±0.32 a,x	5.56±0.43 a,x

[1] *All values reflect mean±Standard error. Values in the same column bearing different letters (x, y, z, q, w) are significantly different (P<0.01). Values in the same row bearing different letters (a, b, c) are significantly different (P<0.01)*

[2] *A = 70% CO_2/30% N_2; B = 80% CO_2/20% O_2; C = 30% CO_2/10% N_2/60% O_2; D =100% CO_2; E = 100% O_2; V = Vacuum*

Table 2. *Effects of packaging and storage time on pH, TBA and texture values of beef steaks stored at 4°C* [1]

Tablo 2. *4°C'de depolanan sığır etinin pH, TBA ve tekstür değerleri üzerine paketleme ve depolama süresinin etkileri*

Parameter	Packaging [2]	Storage Days		
		0	7	14
pH	A	5.58±0.004 ab,x	5.61±0.009 a,x	5.57±0.009 b,x
	B	5.58±0.004 a,x	5.47±0.035 b,y	5.63±0.007 a,x
	C	5.58±0.004 a,x	5.76±0.05 b,z	5.53±0.03 a,x
	D	5.58±0.004 a,x	5.50±0.005 b,xy	5.13±0.007 c,y
	E	5.58±0.004 a,x	6.00±0.07 b,q	5.91±0.05 b,z
	V	5.58±0.004 a,x	4.90±0.0005 b,w	4.75±0.05 c,q
TBA (mg/kg)	A	0.22±0.04 a	1.37±0.06 b,x	0.26±0.08 a,x
	B	0.22±0.04 a	5.26±0.7 b,y	1.59±0.02 a,y
	C	0.22±0.04 a	1.33±0.48 a,x	3.34±0.27 b,z
	D	0.22±0.04 a	0.41±0.05 b,x	0.50±0.28 a,x
	E	0.22±0.04 a	2.98±0.04 b,z	1.79±0.03 c,y
	V	0.22±0.04 a	0.58±0.01 b,x	0.65±0.01 c,x
Texture	A	79.5±2.72 a,x	82.75±8.05 a,x	110.0±8.18 b
	B	79.5±2.72 a,x	84.75±6.53 a,x	86.25±3.58 a
	C	79.5±2.72 a,x	86.40±8.51 a,x	97.50±4.27 a
	D	79.5±2.72 a,x	100.75±3.18 b,x	86.50±1.8 a
	E	79.5±2.72 a,x	84.38±5.32 a,x	87.25±1.91 a
	V	79.5±2.72 a,x	62.87±4.40 b,y	98.87±2.78 c

[1] *All values reflect mean±standard error. Values in the same column bearing different letters (x, y, z, q, w)) are significantly different (P<0.01) Values in the same row bearing different letters (a, b, c) are significantly different (P<0.01)*

[2] *A=70% CO_2/ 30% N_2; B=80% CO_2/ 20% O_2; C=30% CO_2/ 10% N_2 /60% O_2; D=100% CO_2; E= 100% O_2; V= Vacuum*

N_2/60% O_2; and 100% O_2 atmospheres during the storage.

Lower pH values were found in the samples packed under 100% CO_2 atmosphere and vacuum during the storage (P<0.01) than the samples packed under other atmospheres *(Table2)*.

While TBA values in the samples packed under 80% CO_2, 20% O_2 and 100% O_2 atmospheres were high in the 7th day, decreased at the end of the storage (P<0.01). The lowest TBA values were found in the samples packed under 100% CO_2 and vacuum *(Table 2)*.

TMAB and LAB counts in all the samples packed under modified atmosphere and vacuum increased progressively during the storage (P<0.01) *(Table 3)*. The highest count was found in the samples packed under 100% O_2 atmosphere. Final total viable count of the samples packed under 100% O_2 atmosphere reached 8.75 log cfu/g.

accelerate the oxidation of myoglobin to metmyoglobin, which turns the color to brown; however, oxygen is one of the reasons of lipid oxidation, aerobic spoilage and unfavorable color.

While presence of oxygen in the pack maintained texture, in case of carbondioxide caused decrease in hardness.

Increase in acidity due to lactic acid bacteria growth and conversion of CO_2 into carbonic acid during storage is the probable reason why the pH value of the samples packed with CO_2 is low. Similar results were reported in the previous researches [14,15].

Thiobarbituric acid reactive substances (TBARS) were used as an index of lipid peroxidation. These substances accumulate during the first stage of oxidation and then decrease as the improvement due to degradation.

Table 3. *Effects of packaging and storage time on total viable count (TVC) and lactic acid bacteria (LAB) count (log cfu/g) of beef steaks stored at 4°C* [1]

Tablo 3. *4°C'de depolanan sığır etinin toplam canlı sayısı ve laktik asit bakteri sayısı üzerine paketleme ve depolama süresinin etkileri*

Parameter	Packaging [2]	Storage Days		
		0	7	14
TVC	A	5.13±0.03 a,x	6.22±0.002 b,x	7.44±0.05 c,x
	B	5.13±0.03 a,x	7.92±0 b,y	7.65±0.01 c,y
	C	5.13±0.03 a,x	7.94±0.01 b,y	8.05±0.01 c,z
	D	5.13±0.03 a,x	6.95±0.04 b,z	7.71±0.08 c,y
	E	5.13±0.03 a,x	7.96±0.07 b,y	8.74±0.07 c,q
	V	5.13±0.03 a,x	7.91±0.005 b,y	8.13±0.06 c,z
LAB	A	3.53±0.04 a,x	5.75±0.05 b,x	6.86±0.02 c,x
	B	3.53±0.04 a,x	6.91±0.04 b,y	6.94±0.005 b,x
	C	3.53±0.04 a,x	6.88±0.004 b,y	6.76±0.03 c,y
	D	3.53±0.04 a,x	6.62±0.05 b,z	7.72±0.03 c,z
	E	3.53±0.04 a,x	6.98±0 b,y	7.97±0.009 c,q
	V	3.53±0.04 a,x	6.78±0.003 b,q	8.10±0.03 c,w

[1] All values reflect mean±standard error. Values in the same column bearing different letters (x, y, z, q, w) are significantly different (P<0.01) Values in the same row bearing different letters (a, b, c) are significantly different (P<0.01)
[2] A=70% CO_2/ 30% N_2; B=80% CO_2/ 20% O_2; C=30% CO_2/ 10% N_2 /60% O_2; D=100% CO_2; E= 100% O_2; V= Vacuum

DISCUSSION

Color attributes and lightness have vital importance for the consumer. The color of meat depends on surface myoglobin concentration, its chemical state and structure of the muscle surface [10]. The proportion of the different states of myoglobin at the meat surface changes with storage and the atmosphere around the meat. The decrease in redness may result from the gradual formation of metmyoglobin on the meat surface. Presence of oxygen in the medium cause color disorders due to metmyglobin formation [11,12]. Good meat color was found between 55 and 80% oxygen [13]. High oxygen concentrations enhance bright-red color to fresh meat, but low concentrations

Other researchers reported that high levels of oxygen concentrations bring about higher lipid oxidation [12-16] and also, the process of myoglobin oxidation is a catalyst of lipid oxidation [17].

Oxygen plays a major role in determining the types of microorganisms present on stored beef. In general, a minimum of 20% CO_2 is needed for significant effects on microbiological growth [18]. In our research, it is obvious that total viable count increases in the presence of oxygen, whereas a decrease with the CO_2 level and this is supported by other researchers [19-21]. It is stated that spoilage occurs when total aerobic count reach 10^7 cfu/g [14]. Vacuum-packed samples had higher TVC than those packed under modified atmospheres except 100% O_2 atmosphere. Higher

levels of microbial load in the samples packed under vacuum than modified atmosphere containing carbon-dioxide were determined in another study [12]. In a previous study [22] it was pointed out that aerobic storage provoked TMAB to reach high levels. The lowest LAB count was found for samples packed under 70% CO_2/30% N_2 atmosphere at the end of storage.

In conclusion modified atmosphere packaging is an effective method to maintain color and lipid oxidation and inhibit microbial growth. High-level oxygen increased lipid oxidation during the storage. Oxygen in the pack also caused discoloration. Microbial inhibition was observed in case of presence of carbondioxide. High concentration (100%) of carbondioxide caused pack collapse because of the solubility of carbondioxide on the meat. Although the best quality was observed with A (70% CO_2, 30% N_2) and D (100% CO_2) atmospheres, A (70% CO_2, 30% N_2) was determined as the most appropriate atmosphere due to collapse of package with carbondioxide of 100%. Shelf life of beef steaks packed under A atmosphere was 14 days.

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