

## IMPACT OF PACKAGING IN MODIFIED ATMOSPHERE OF SUSTAINABILITY OF MINCED MEAT PRODUCTS

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### Abstract

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This paperwork presents the results of examining the sustainability of burgers and kebabs packed in modified atmosphere with composition of gases 65% O<sub>2</sub> and 35% CO<sub>2</sub>. The examined samples were stored for 8 days in controlled conditions at a temperature of +4°C. The microbiological examinations included a total number of bacteria, presence of *Enterobacteriaceae*, *Escherichia coli*, *Salmonella*, lactic acid bacteria. The samples were microbiologically tested first, third, fifth and eighth day. The sensor features of kebabs and burgers were analyzed by means of descriptive test (colour and odour when fresh and consistency and taste after the backing). pH was checked every day, and the acidic and peroxide number were checked on the fourth and the eighth day. On the basis of the obtained results of the testing, we found that kebabs and burgers packed in modified atmosphere with 65% O<sub>2</sub> and 35% CO<sub>2</sub> were sustainable for six days.

*Key words:* kebabs, burgers, modified atmosphere

### Introduction

The industry for production of meat and meat products is a highly risky industry, and thus meat as a product of animal origin belongs to perishable food products.

Spoiling of meat may be defined as any change that makes meat and meat products unacceptable in a sensor aspect for the consumer (Gram et al., 2002; Petäjä-Kanninen and Puolanne, 2007). As a result of the reaction of microorganisms and their development and expansion in meat, unpleasant odour and taste is created whereby the intensity of sensory changes is related to the decomposing of nutrients in meat and the creation of unwanted volatile metabolites. (Jay et al., 2003; Doyle, 2007:) the unpleasant odour is due to the number of microorganisms above 7 log cfu/g, and the decomposition of free amino acids when the expansion of microorganisms reaches a value of 9 log cfu/g results in a strong odour in meat. Cooled meat in depth is sterile or contains a small number of bacteria (up to 100/g). They include anaerobic, facultative anaerobic and microaerophilic

bacteria species of genus: *Clostridium*, *Bacillus*, *Streptococcus*, *Lactobacillus*, *Enterobacter*. The bacteria *Enterobacter* and the lactic acid bacteria, as well as *Aeromonas*, *Moraxella*, *Acinetobacter*, *Flavobacterium* etc. (Labadie et al., 1999; Pin et al., 2002) contribute towards perishing of cooled meat. These bacteria species multiply fast in cooled meat and put aside the other microorganisms that can also multiply at low temperatures. Unpleasant odour of meat and meat products results from the presence of *Bronchothrix thermosphacta*. The chopped meat is placed on the market as minced or chopped shaped meat (kebabs, burgers). By chopping meat conditions are created for development of microorganisms. Surface of meat is increased, fascia and aponeuroses (muscle fibers connective tissue membrane) are torn apart and thus the biological barrier for entering of microorganisms deeply in the meat is interrupted. The damaged muscle fibers discharged liquid and this increases the humidity of minced meat. The initial microflora of meat is increased by secondary contamination during the preparation and adding of additives. As a result the minced meat biologically decomposes

The dominant microflora in minced meat is comprised by representatives of the *Pseudomonas* genus. They are followed by: *Lactobacillus*, *Enterobacter*, *Bronchothrix thermosphacta*, all most always present in minced meat. Rotten meat is primarily caused by the following bacteria: *Pseudomonas*, *Serratia*, *Enterobacter*, *Citrobacter*, *Yersinia*, *Klebsiella*. At higher temperatures, decay of this meat is caused by: *Bacillus*, *Aeromonas*, *Proteus*, *Serratia*, *Leuconostoc*, *Micrococcus*, *Streptococcus*, some species of *Clostridium*, *Escherichia coli*. As a result of the bacteria activity, greenish discolorations may appear on the surface or inside the chopped meat caused by peroxides of some lactobacilli, for example *Lactobacillus viridescens* or hydrogen sulfide of some species of the genus *Proteus*, *Citrobacter* etc. Even though microorganisms have significant role in meat rotting, still the final assessment on rotting is given after the sensor analysis (Nychas et al., 1998; Ellis and Goodachre, 2000).

For the purpose of continuing the sustainability and preserving the quality of fresh meat, MAP packaging is used and it is proved to be very effective (Robertson, 1993; Lambropoulou et al., 1996; Sakota et al., 2002; Robertson, 2006; Koutsoumanis et al., 2008; Yilmaz and Demirci, 2010; Ozlem et al., 2011). MAP may be defined as a packaging in which air is pressed outside the packaging and replaced by a simple gas or a mixture of gases depending of the type of meat being packed (Kuzelov et al., 2012). In the modified atmosphere technology of packaging the following gases are used mostly: oxygen, carbon dioxide and nitrogen, used in different combinations and each of them performs a certain function (Xiong, 1999; Sorheim et al., 1997; Yam et al., 2005). Carbon dioxide is a main antimicrobial factor especially towards gram negative bacteria as *Pseudomonas species* that cause the unpleasant odors in meat. It damages the cell membrane by interacting with the lipids on the cell membrane whereby the ability of the membrane to receive ions is changed. The efficacy of this gas depends on its original and final concentration in packaging, the temperature of storing and the initial microflora. The big concentrations of CO<sub>2</sub> in gas mixtures lead to a greater number of *lactobacillus* and a smaller number of *Bronchothrix thermosphacta* (Sinell, 1988; Brody, 1989; Kleimer, 2004; Boeckman, 2006).

One of the main functions of oxygen is to maintain red pigment, myoglobin in oxymoglobin condition which is responsible for the light red colour related to freshness. The colour of meat is the most important sensor feature of fresh meat in retail. The use of greater amounts of oxygen in a packed product influences the maintaining of red colour, but can at the same time lead to increasing the oxidative reactions and increasing the rancidity of fats in meat which causes unpleasant smell and taste (Martinez et al., 2006).

Nitrogen prevents the rancidity of fats and inhibits the growth of aerobic microorganisms (Blakistone, 1998). Burgers and kebabs are very frequent in the retail network as part of products prepared by chopped and shaped meat in the Republic of Macedonia. These products are not thermally treated and their expiry date, according to the rulebook on quality of meat and meat products of the Republic of Macedonia, is 48 h if these products are kept at a temperature of +4°C packed.

The objective of this paperwork is to determine the sustainability of burgers and kebabs packed in modified atmosphere with a combination of mixture of gases comprised of 65% O<sub>2</sub> and 35% CO<sub>2</sub>.

## Materials and Methods

Burgers and Kebabs are produced of pork beef meat and bacon chopped at Wolf machine up to granulation of particles of around 3 mm by adding water, spices and mix for kebabs and burgers comprised of potassium polyphosphate, NaCl and ascorbic acid produced by Vanhes - Germany. Then burgers and kebabs are shaped and packed in a modified atmosphere (burgers – two per package, kebabs – ten pieces per package). Vacuum packaging machine is used for packaging (MULTI-VAC R 245), and vessels as a material for packaging Cryovac LidSus (Sealed Air USA) and upper foil F-Type, Lid HB-S (supplied by Din- Best Skopje) with dimensions 100 mm 180 mm 35 mm (width, length, height). The packaging is filled with a mixture of gases 65% O<sub>2</sub> and 35% CO<sub>2</sub>. The amount of mixture of gases amounted 100 - 200 ml for 100 g of product. After the packing in modified atmosphere, the burgers and kebabs were stored in a refrigerating chamber at a temperature of +4°C for eight days.

The samples of burgers and kebabs were microbiologically analyzed on the 1, 3, 5 and 8 day by means of the following methods:

Total number of bacteria - ISO 4833/2003, *Staphylococcus* ISO 6888-1/1999, Enterobacteria ISO 21528-1/2004. The number of *E. coli* is determined by the method ISO16649-2/2008. The presence of *Salmonella* is determined by ISO 6579/2008, the number of lactic acid bacteria is determined by ISO 15214/1998. The number of coagulase positive staphylococci is determined by ISO 688-2/2009. The number of *Bronchothrix thermosphacta* is determined by ISO 13722/1999. As regards chemical examinations, the acidic and peroxide numbers were checked on the fourth and the eight day of the experiment in accordance with ISO 660/2000 and ISO 3960/2001.

By means of the 9-degree scale, VNIMP Moscow defines the following sensor features colour and smell before trial,

backing and smell, consistency and taste after the trial of backing 1, 4, 6, and 8 day of the experiment. pH of burgers and kebabs is checked every day by means of pH meter. The results of the tests are processed using mathematical statistical program Microsoft Excel 2007.

## Results and Discussion

The results of the microbiological examination are presented in Tables 1 and 2. In the samples of the checked burgers and kebabs there was no presence of pathogen bacteria i.e. coagulase for positive staphylococci during the period of examination.

During the storage of packed burgers and kebabs at the temperature of + 4°C in a duration of 8 days, the total number of mesophilic bacteria showed a linear tendency of growth of 0.58 log/cfu/g per day whereby at the 8<sup>th</sup> day of storage the growth outreached the limit of acceptability of 7 log/cfu/g per day (ICMF, 1986). A gradual growth was detected as regards *Bronchothrix thermosphacta*, lactic acid bacteria and enterobacteria up to the 8<sup>th</sup> day of storage. The total number

of anaerobic psychrophilic bacteria during the examined period was 0.72 log/cfu/g at most for burgers and 0.68 log/cfu/g for kebabs.

Jayasingh et al. (2002) studied minced beef meat packed in modified atmosphere with a high percentage of oxygen. They found that the colour and total number of microorganisms are equal in treated and control groups. Ercolini et al. (2006) monitored the growth of *Pseudomonas* species, Enterobacteriaceae *Bronchothrix thermosphacta* for fresh beef meat and found a low growth of these microorganisms at samples packed in modified atmosphere with a combination of gases O<sub>2</sub> 60% CO<sub>2</sub> 40% as regards samples stored at air. Ozlem et al. (2011) found that chopped beef meat packed in modified atmosphere with a combination of gases 60% O<sub>2</sub> и 40% CO<sub>2</sub> favours growth of lactobacilli and enterobacteria, and inhibits the growth of *Pseudomonas species*, *Bronchothrix thermosphacta*. The results of the sensor assessment of burgers and kebabs are presented in Table 3.

The table shows that the colour and odour of burgers at the beginning of the examination are assessed as acceptable (9.52 +/- 0.25, 9.55 +/- 0.028, 9.55 +/- 0.28, 9.48+/-0.28). On the

**Table 1**  
Change of the number of microorganisms in burgers

Bacteria	Testing day			
	1	3	5	8
TVC	5.22 log/cfu/g	6.38 log/cfu/g	6.96 log/cfu/g	7.54 log/cfu/g
TVCP	0.25 log/cfu/g	0.32 log/cfu/g	0.47 log/cfu/g	0.72 log/cfu/g
EB	2.2 log/cfu/g	3.7 log/cfu/g	4.8 log/cfu/g	6.2 log/cfu/g
BCH	1.2 log/cfu/g	3.5 log/cfu/g	4.2 log/ cfu/g	5.5 log/cfu/g
LAB	1.12 log/cfu/g	2.1 log/cfu/g	3.7 log/cfu/g	5.2 log/cfu/g

Legends:

TVC Total count of aerobic  
TVCP Total count of aerobic  
EB Bacteria of the Enterobacteriaceae family  
BCH *Bronchothrix thermosphacta*  
LAB Lactic acid bacteria

**Table 2**  
Change of the number of microorganisms in kebabs

Bacteria	Testing day			
	1	3	5	8
TVC	5.20 log/cfu/g	6.35 log/cfu/g	6.90 log/cfu/g	7.48 log/cfu/g
TVCP	0.20 log/cfu/g	0.325 log/cfu/g	0.42 log/cfu/g	0.68 log/cfu/g
EB	2.0 log/cfu/g	3.2 log/cfu/g	4.5 log/cfu/g	5.2 log/cfu/g
BCH	1.5 log/cfu/g	3.5 log/cfu/g	4.5 log/cfu/g	5.2 log/cfu/g
LAB	1.15 log/cfu/g	2.10 log/cfu/g	3.50 log/cfu/g	5.20 log/cfu/g

Legends:

TVC Total count  
TVCP Total count of aerobic  
EB Bacteria of the enterobacteriaceae family  
BCH *Bronchothrix thermosphacta*  
LAB Lactic acid bacteria

**Table 3**  
Sensor assessment of burgers and kebabs before baking trial

Sensor features	Burgers				Kebabs			
	1 day	4 day	6 day	8 day	1 day	4 day	6 day	8 day
Colour	9.52+/- 0.25	9.48+/- 0.28	9.40+/-0.28	8.52+/-0.55	9.55+/-0.28	9.45+/- 0.20	9.38+/- 0.22	8.82+/-0.55
Odour	9.55+/-0.028	9.25+/-0.42	8.95+/-0.45	7.82+/-0.48	9.48+/-0.28	9.12+/-0.42	8.80+/-0.40	7.78+/-0.50

last day of examining the odour of burgers and kebabs, they received a low average grade (7.82 $\pm$  0.48, 7.78  $\pm$  0.50, 9.70 $\pm$ 0.28, 9.70 $\pm$  - 0.22) compared to the previous days of examining, whereas the colour was assessed with a higher grade (8.52  $\pm$  0.55, 8.82  $\pm$  0.55).

The results of the examination of smell, consistency and taste of burgers and kebabs after the baking trial are presented in the following Table 4.

On the first and the fourth day after the baking trial of the sampled, the average grades on odour were (9.72  $\pm$  0.25, 9.72  $\pm$  0.22), odour is assessed as acceptable. During the examining, the average grade on odour for both sampled products decreased, thus on the last day of the examining the average grade for burgers was 7.82  $\pm$  0.48, and for kebabs 7.80  $\pm$  0.20. After the baking trial, on the 1<sup>st</sup>, 4<sup>th</sup> and the 6<sup>th</sup> day, the consistency of burgers and kebabs is assessed with high grades (9.82  $\pm$  0.22, 9.52  $\pm$  0.25, 9.35  $\pm$  0.20 for burgers, and 9.85  $\pm$  0.25, 9.55  $\pm$  0.28, 9.38  $\pm$  0.28 for kebabs). The eighth day of examining the consistency of both products received lower grades (9.10  $\pm$  0.22 and 9.12  $\pm$  0.32).

The taste of burgers and kebabs is assessed as very acceptable on the first and the fourth day of examining (10.0  $\pm$  0.0, 9.82  $\pm$  0.25, 10.0  $\pm$  0.0, 9.58  $\pm$  0.22), whereas on the eight day of the examining, the grades were lower for both sampled products (8.98  $\pm$  0.12, 8.85  $\pm$  0.48).

The average pH value of burgers during the examination was 5.52 $\pm$  0.010 with a variation coefficient 1.47 and for kebabs 5.58  $\pm$  0.07 with a variation coefficient 1.42. In contrast to the results that we obtained as regards the pH value Yilmaz and Demirci (2010) sampled kebabs packed in modified atmosphere with a mixture of gases comprised of 65% N<sub>2</sub> and 35% CO<sub>2</sub> found a decrease of pH value during the whole period of storing.

The results of examining the peroxide number and the acidic degree of burgers and kebabs are presented in Table 5. The value of the peroxide number points out that there were no oxidative changes of fats, and the values of the acidic degree on the eight day of examination point out that there were no hydrolytic changes of fats at the sampled burgers and kebabs as regards the fourth day of examining.

Ozlem et al. (2011) found that by packing beef minced meat in a modified atmosphere with a combination of gases

**Table 5**  
Results of the examining of the acidic and peroxide number of sampled burgers and kebabs

Products	Days of examining	Peroxide number	Acidic degree
Burgers	4	0.00 mmol/kg	3.60 mgKOH /g
	8	0.00 mmol/kg	3.72 mg KOH/g
Kebaps	4	0.00 mmol/kg	3.62 mgKOH/g
	8	0.00 mmol/kg	3.75 mgKOH /g

50% O<sub>2</sub>, 30% CO<sub>2</sub> and 20% N<sub>2</sub>, a lowest degree of oxidation of lipids is achieved.

## Conclusion

During the examined period, the total number of anaerobic psychrophilic bacteria at sampled burgers was 0.72 log/cfu/g at burgers and 0.68 log/cfu/g at kebabs. The last growth during the 8 day storage was noticed of some species of the genus *Enterobacter*, lactic acid bacteria and *Bronchothrix thermosphacta*. On the basis of the results of the peroxide number and the acidic degree, it can be concluded that there were no hydrolytic and oxidative changes in the sampled burgers and kebabs during the storage. Changes were not noticed neither with the sensor assessment as regards the rancidity and the burgers and kebabs were acceptable in view of colour, consistency, smell and taste during the storage.

On the basis of the obtained results, and especially on the basis of the recommended total number of anaerobic psychrophilic bacteria that must not be higher than 7.0 log/cfu/g and on the basis of the sensor features, burgers and kebabs packed in modified atmosphere with 65% O<sub>2</sub> and 35% CO<sub>2</sub> were sustainable for 6 days.

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**Table 4**  
Sensor assessment of burgers and kebabs after the baking trial

Sensor features	Burgers				Kebaps			
	1 day	4 day	6 day	8 day	1 day	4 day	6 day	8 day
Consistency	9.82 $\pm$ 0.22	9.52 $\pm$ 0.25	9.35 $\pm$ 0.20	9.10 $\pm$ 0.22	9.85 $\pm$ 0.25	9.55 $\pm$ 0.28	9.38 $\pm$ 0.28	9.12 $\pm$ 0.32
Odour	9.70 $\pm$ 0.28	9.70 $\pm$ 0.22	8.42 $\pm$ 0.32	7.82 $\pm$ 0.48	9.72 $\pm$ 0.25	9.72 $\pm$ 0.22	8.58 $\pm$ 0.28	7.80 $\pm$ 0.20
Taste	10.0 $\pm$ 0.0	9.82 $\pm$ 0.25	9.52 $\pm$ 0.48	8.98 $\pm$ 0.12	10.0 $\pm$ 0.0	9.58 $\pm$ 0.22	9.22 $\pm$ 0.20	8.85 $\pm$ 0.48



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