EFFECT OF TEMPERATURE AND PACKAGING CONDITIONS ON MICROBIAL GROWTH IN LAMB

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Abstract – The aim of this study was to investigate the effect of temperature and type of packaging on lamb preservation. Legs from twenty lamb carcasses were removed and cut. Chops were vacuum skin and modified atmosphere packaged and refrigerated at -0,5 and 4 °C. Psychrotropic total viable counts, Enterobacteriaceae, Pseudomonas spp. and lactic acid bacteria were determined. Gas composition of modified atmosphere packs was also measured throughout the storage period. Packs stored at 4 °C showed higher counts than those were refrigerated at -0,5 °C for all the microorganisms determined. Vacuum skin packaged meat resulted in a better stability than those was packaged in modified atmosphere, showing significant lower counts for psychrotropic total viable counts and Pseudomonas spp. Samples stored at 4 °C registered an increase in the percentage of CO₂ and a simultaneous reduction in the percentage of O₂, probably due to microbial growth. In conclusion, refrigeration at -0,5 °C combined with vacuum skin packaging resulted in the highest inhibition of microbial growth.

Key Words – refrigeration, vacuum skin packaging, modified atmosphere packaging.

I. INTRODUCTION

Lamb is a high perishable product and its short shelf life is one of the factors that most concern meat industry [1]. Meat acceptance varies throughout the preservation period; a decrease could be possible for many reasons: development of brown colour, formation of off odours and flavours, texture deterioration and/or superficial slime formation. Microbial growth has a significant role in meat spoilage, so several technologies have been developed with the aim of avoiding it.

Nowadays, the trend is to sell packaged meat in refrigerated self-services display cases [2]. The effect of cool applied to meat is well known, as when temperature decreases microbial growth is reduced. The temperature applied for meat preservation is commonly 4 °C, however low

temperatures could extend lamb shelf life without quality decrease. Moreover, packaging has great advantages for dealers and markets because it is a mechanical barrier, reduces evaporative losses and has microbial and oxidative inhibitory effects if meat is surrounded with an adequate atmosphere. Modified atmosphere (MAP) and vacuum skin packaging (VSP) are the most used technologies for packaging meat to sell in self-services. In this conditions, Pseudomonas spp., lactic acid bacteria (LAB) and Brochotrix thermosphacta, which are the predominant gena, are responsible of microbial spoilage [3, 4]. With the aim of preserving lamb for an extended period, the temperature and type of package chosen should be optimized. Therefore, the main objective of this study was to assess the effect of temperature and type of packaging on lamb chops microbial growth during the preservation time.

II. MATERIALS AND METHODS

Twenty lamb carcasses were randomly chosen and chilled for 24 hours (-1,5 to 0,5 °C). Later, the two legs of each carcass were removed and cut into chops, which were vacuum skin (Multivac R570 CD) and modified atmosphere (40% $O_2/$ 30% CO₂/ 30% Ar) packaged (ULMA-SMART-500). Samples were divided into four batches and stored in darkness at 4 ± 0.5 °C or -0.5 ± 0.5 °C for 28 days. Gas composition was measured before opening the package with a portable gas analyser (Oxibaby). For microbiological examination, samples were aseptically collected from the chops swabbing an area of 10 cm² delimited with a sterile aluminium template (10 cm²). Each sample was homogenised in 0.1% peptone water (Biolife) and serial dilutions were done. 1 ml of the appropriate dilution was plated in the following manner: psychrotropic total viable counts (PTVC), on plate count agar (PCA) (Merck) at 10 °C for 96 h; Enterobacteriaceae on violet red bile dextrose agar (VRBD) (Merck) at 37 °C for 48 h; lactic acid bacteria on man, rogosa and sharpe (MRS) (Merck) for 96h at 37 °C in anaerobiosis and Pseudomonas spp. on Cephalothin-Sodium Fusidate-Cetrimide Agar (CFC) (Merck) at 20 °C for 24h. All microbial counts were expressed as base-10 logarithms of colony forming units per cm² of surface area (log CFU cm⁻²) All data were statistically analyzed by the general linear model (GLM) procedure of IBM SPSS version 19 (IBM SPSS, 2010). The model included temperature, type of packaging and refrigeration storage duration as main effects and also their interaction. Tukey post hoc test was used to assess differences between mean values when $P \leq 0.05$.

III. RESULTS AND DISCUSSION

Gas composition of modified atmosphere packages:

Figure 1. O_2 and CO_2 concentrations inside the modified atmosphere packages.



$\% O_2 MAP 4 C$	-% CO ₂ MAP 4 °C
•••••• % O ₂ MAP -0, 5 °C	• '% CO ₂ MAP -0, 5 °C

As can be seen in figure 1, significant differences were found in the evolution of gas composition of modified atmosphere packages refrigerated at different temperatures. Packages stored at -0,5 °C did not show big changes to the initial atmosphere, whereas those were chilled at 4 °C showed severe variations both the O₂ and CO₂ percentages. A dramatic increase in the percentage of CO₂ and a simultaneous reduction in the O₂ started after 14 days of refrigeration and continued until the end of the experimental period. The phenomenon described could be due to microbial metabolism; aerobes bacteria growth results in a reduction in O_2 meanwhile CO_2 is produced [5].

Psychrotrophic total viable counts

Psychrotropic total viable counts obtained are presented in figure 2. Growth in meat refrigerated at -0, 5 °C was significantly lower than those was stored at 4 °C. Microbial growth remained stable from the beginning to the seventh day of storage in lamb maintained at lower temperature. Then, it was faster from the 7th to the 28th day of storage, when it reached 6 log CFU/ cm^2 . Fernandes *et al*. [6] also noted an initial stability in PTVC until 7th day of storage in vacuum and modified atmosphere packaged (75% O₂-25% CO₂) lamb loins stored at 1 °C. Meat preserved at 4 °C showed a different behavior. Microbial growth was observable since the initial day until the 21th day of storage. Significant differences were not found between the counts registered this day and at the final of storage. Regarding to packaging, differences in PTVC were significant between MAP and VSP meat refrigerated at 4 °C for all days. However, in lamb refrigerated at -0,5 °C differences only were significant after 28 days of preservation.

Only MAP lamb maintained at 4 °C reached 8 log CFU/ cm², in contrast, the others treatments didn't overcome this limit for microbial spoilage (7-8 log CFU/ cm² or g) reported by some authors [7, 5].

Figure 2. Psychrotropic total viable counts on lamb chops



Enterobacteriaceae counts

Figure 3. *Enterobacteriaceae* counts on lamb chops



Significant differences between *Enterobacteriaceae* counts of lamb refrigerated at different temperature were noted after 28 days of storage. However, differences were not registered between the counts of MAP and VSP meat stored at the same temperature, neither in -0,5 nor in 4 °C. Counts were dramatically higher in meat stored at 4 °C than in refrigerated at -0,5 °C, in which *Enterobacteriaceae* growth was strongly inhibited. These results are consistent with those were obtained by Sheridan *et al.* [8], who also

found significant lower counts for lamb shoulders refrigerated at 0 °C than those were stored at 5 °C, both in vacuum and in MAP (80% $O_2/20\%$ CO₂). In contrast, different authors cited lower *Enterobacteriaceae* growth in MAP than in vacuum packaged lamb due to the inhibitor effect of CO₂, however, the atmosphere used (80% $O_2/20\%$ CO₂ and 75% $O_2/25\%$ CO₂ vs 40% O_2 /30% CO₂ /30% Ar) and the type of vacuum (traditional vs skin vacuum packaging) were different.

As can be observed in figure 3, *Enterobacteriaceae* growth reached 6 log CFU/ cm² in VSP and MAP lamb stored at 4 °C. Similar counts were obtained by Sheridan *et al.* [8] in vacuum packaged lamb shoulders refrigerated at 5 °C. However, in this trial the counts obtained were lower in MAP than in vacuum (4, 8 vs 6, 1 log CFU/ cm²).

Lactic Acid Bacteria counts

Figure 4. Lactic acid bacteria counts on lamb chops



Lactic acid bacteria counts achieved 6 log UFC/cm² after 28 days post-packaging in meat stored at 4 °C both in MAP and in VSP lamb. Higher counts (7-8 log UFC/cm²) were registered by Berruga *et al.* [9] at the same time of storage. On the other hand, significant lower growth was noted in packs refrigerated at lower temperature. In this case, neither VSP nor MAP reached 4 log UFC/cm².

Several authors registered higher counts in vacuum than in modified atmosphere packaged meat [9, 10] by contrast, in this trial differences were not found.

Pseudomonas spp. counts

Pseudomonas spp. growth can be observed in figure 5. Significant differences were obtained between each one of the preservation conditions at the end of the trial. MAP lamb refrigerated at 4 °C, which achieved 7 log CFU/ cm², showed the highest counts. Regarding meat preserved at lower temperature, MAP also showed higher counts than VSP, which registered the lowest growth for *Pseudomonas spp.* (3, 1 log CFU/ cm²). Results agree with those obtained by Sheridan *et al.* [8] who also found higher growth in MAP than in vacuum packaged lamb at 5 and 0 °C.

Figure 5. Pseudomonas spp. counts on lamb chops



IV. CONCLUSION

According to the results, we conclude that lamb stored at -0, 5 °C had a dramatically better stability than those was refrigerated at 4 °C. On the other hand, vacuum skin packaging was more efficient than modified atmosphere packaging in reducing aerobes psychrotropic microorganisms and *Pseudomonas spp.* growth. However, other combinations of gases should to be tested to achieve a better stability of refrigerated lamb meat.

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