



The combined effects of superchilling and packaging on the shelf life of lamb



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ABSTRACT

The aim of this study was to evaluate the effect of superchilled storage at -1° on the shelf life of lamb slices packaged in an O_2 enriched (40% O_2 /30% CO_2 /30% Ar) or in an anaerobic atmosphere (vacuum skin packaging). Physicochemical, microbial and sensory analyses were performed. The effect of superchilled storage on lamb stability differed depending on the atmosphere surrounding the product. Superchilled ($-1^\circ C$) slices of lamb showed lower microbial counts than those refrigerated at $4^\circ C$ in both packaging conditions. Moreover, meat stored at $-1^\circ C$ had a higher colour stability under vacuum. Superchilled storage combined with an O_2 enriched atmosphere increased the rate of lipid oxidation, which reduced the shelf life reached by refrigerating at $4^\circ C$. Vacuum skin packaging strongly inhibited lipid oxidation independently of storage temperature. Thus, superchilled storage extended the shelf life at least twice compared to storage at $4^\circ C$ under anaerobic conditions while it was disadvantageous when an O_2 enriched atmosphere was used.

1. Introduction

Fresh lamb is a perishable product so it is commonly preserved by refrigeration combined with packaging either for storage or display (Berruga, Vergara, & Gallego, 2005; Vieira & Fernández, 2014). Refrigeration temperatures around $4^\circ C$ and modified atmosphere (MAP) or vacuum skin packaging (VSP) are usually used to extend the lifespan of lamb meat. Traditionally the gases used in MAP comprises O_2 , CO_2 and N_2 (Bellés, Alonso, Roncalés, & Beltrán, 2017a). Lamb is packaged in enriched O_2 atmospheres (70–80% O_2) to provide a desirable red colour to meat. However, the presence of O_2 could enhance lipid oxidation and ultimately myoglobin oxidation (O'Grady, Monahan, Burke, & Allen, 2000), so an atmosphere including a lower content of O_2 could be more suitable when an extended shelf life is required. Argon (Ar) is an inert, odourless and tasteless noble gas envisaged as a possible alternative to N_2 for MAP application (Heinrich, Zunabovic, Nehm, Bergmair, & Kneifel, 2016). Spencer and Humphreys (2002) suggested that Ar displaces O_2 more efficiently and offers a higher shelf life than N_2 based MAP, while Ruiz-Capillas and Jiménez-Colmenero (2010) and Herbert, Roissant, Khanna, and Kreyensmidt (2013) noted a better keeping of sensory properties in Ar based MAP. Despite N_2 is still more common, Ar based MAP is increasingly used for lamb retail cuts packaging in Spain since it could have some benefits on lamb preservation.

Regarding vacuum skin packaging, it is a relative new development which achieves a tight disposition of the film on meat surface, reducing

the formation of pockets and consequently, decreasing residual O_2 . Wrinkles, which are common in traditional vacuum packaging, are also avoided thus improving the visual appearance of packaged meat (Lagerstedt, Ahnström, & Lundström, 2011).

However, the shelf life reached by refrigerating at $4^\circ C$ hardly overpasses 21 days (Berruga et al., 2005; Lopacka, Pótorak, & Wierzbicka, 2016), which represents a limiting factor for industries interested in an extended life for distribution or storage. Freezing is commonly used for long-term meat preservation due to the extended shelf life of frozen meat resulting in a greater flexibility for distributors and retailers (Wheeler, Miller, Savell, & Cross, 1990). However, some studies have found that freezing could modify meat tenderness, juiciness and flavour (Kaale, Eikevik, Rustad, & Kolsaker, 2011; Vieira, Díaz, Martínez, & García-Cachán, 2009), and these variations have not gone unnoticed for consumers, who associate frozen meat with a lower quality product than chilled meat (Lagerstedt, Enfält, Johansson, & Lundström, 2008).

In this context, superchilling (also called deep chilling) emerges as a possible solution. It consists of meat storage at temperatures comprised between freezing and chilling, usually between -0.5 and $-2.8^\circ C$ (Beaufort, Cardinal, Le-Bail, & Midelet-Bourdin, 2009; Kaale et al., 2011). At superchilling temperatures microbial growth is strongly inhibited, extending the shelf life reached with conventional chilling by 1.4–4 times (Magnussen, Haugland, Torstveit Hemmingsen, Johansen, & Nordtvedt, 2008). Superchilling has been extensively used for seafood with promising results (Kaale & Eikevik, 2014) but the studies in meat preservation are scarce. Lan, Shang, Song, and Dong

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(2016) observed an extension of rabbit shelf life in 3–5.5 times by superchilled storage, which was also noted by Zhang, Wang, Li, Wu, and Xu (2016) for broiler meat. Nevertheless, severe myofibrils damages were detected, which may have been due to the formation and growth of ice crystals (Lan et al., 2016). These alterations could favour pro-oxidants release (Benjakul & Bauer, 2001) and consequently, lipid oxidation and colour deterioration. Superchilling can extend the shelf life of lamb but its effect on meat quality is a matter of concern. Moreover, there is also little knowledge about the effect of superchilling combined with packaging on meat shelf life and, as far as we know, there is not literature published concerning lamb.

Therefore, the aim of this study was to assess the effect of superchilled storage on physicochemical, microbiological and sensory quality of fresh lamb leg slices packaged in an O₂ enriched atmosphere (MAP) and in absence of O₂ (VSP). At the same time, differences in quality throughout storage were used to determine the shelf life reached by superchilling combined with both MAP and VSP.

2. Materials and methods

The lambs used for this trial were cared in accordance with the guidelines from the Spanish Ministry of Agriculture (Boletín Oficial del Estado (BOE), 2007).

2.1. Slaughtering, packaging, refrigerated storage and sampling

Twenty lamb carcasses were randomly chosen among commercial lambs of the Rasa Aragonesa, a medium wool breed that is reared for meat purpose in Spain. Animals were reared under intensive husbandry conditions with natural suckling until 40 days of age and fodder with concentrate and cereal straw ad libitum until they reached a body weight between 20 and 25 kg. The animals, aged about three months, were slaughtered in a commercial slaughterhouse (Mercazaragoza) following standard protocols. Within 15 min of dressing, carcasses were transported to the facilities of Casa de Ganaderos and Franco y Navarro S.A. and chilled for 24 h (-0.5 ± 0.5 °C, 90% RH, 1–2 m/s). The two legs of each carcass were removed and allocated among two packaging systems (MAP and VSP) and two storage temperatures (-1 °C and 4 °C), being the 10 legs of each group from different animals ($n = 10$). Legs were cut into 20-mm-thick slices and transported under refrigeration (4 °C) to the food technology pilot plant of the Faculty of Veterinary Science (University of Zaragoza). After that, slices were either vacuum skin (Multivac R570 CD, MultivacSepp Hagenmüller GmbH & Co. KG, Wolfertschwenden, Germany) or modified atmosphere packaged (40% O₂ + 30% CO₂ + 30% Ar) (ULMA-SMART-500, Ulma S. Coop., Guipúzcoa, Spain) with a product to gas ratio of 1:3 (2 slices per tray). The top film of the vacuum skin packages (VSP) (Cryovac® VST 0250 SKIN TOP WEB, Sealed Air S.L., Abrera, Spain) was 100 µm thick; its O₂ permeability rate at 23 °C was 1.5 cm³/m²/24 h/0% relative humidity (R.H.) and the water vapour transmission rate at 38 °C was 6 g/m²/24 h/90% R.H. The bottom film (Cryovac® EGA 008, Sealed Air S.L., Abrera, Spain) was 200 µm thick and O₂ permeability rate at 23 °C was 21 cm³/m²/24 h/atm/0% R.H. For modified atmosphere packaging, polystyrene trays (Linpac packaging S.A.U., Pravia, Spain) were used and sealing was done with a polyethylene and polyamide laminate film. The film was 30 µm thick, its O₂ permeability rate at 23 °C was 15 cm³/m²/24 h/0% R.H. and the water vapour transmission rate at 23 °C was 7 g/m²/24 h/85% R.H., (Linpac Packaging S.A.U., Pravia, Spain). After packaging, samples were refrigerated at conventional (4 ± 0.5 °C) or superchilling (-1 ± 0.5 °C) temperatures according to the assigned batch.

Specific analyses were performed at 0 (approximately 24 h post mortem), 7, 14, 21 and 28 days post packaging. Samples destined to sensory analyses were vacuum packaged in each sampling day (-900 mbar of pressure) in polyethylene-polyamide bags with ethyl vinyl acetate sealant layer (30 × 25 cm, 90 µm thickness, water vapour

transmission rate at 23 °C of 2.8 g/m²/24 h/85% RH, an O₂ transmission rate at 23 ± 1 °C of 50 cm³/m²/24 h/75% RH; Eurobag & Film S.L., Spain) using a Tecnotrip EV-13-L-CD-SC machine (Tecnotrip S.A., Spain) and frozen stored at -20 °C until the evaluation was performed. All the analyses were performed on the *Semimembranosus* muscle.

2.2. Lipid oxidation

Lipid oxidation was determined as Thiobarbituric Acid Reactive Substances (TBARS) following the method described by Alonso et al. (2015).

2.3. Instrumental colour

A Minolta CM-2002 (Osaka, Japan) spectrophotometer was used to measure colour at the surface of a 20-mm-thick slice after opening the trays and exposing the samples to air for 2 h at 4 °C. The parameters registered were *L*^{*} (lightness), *a*^{*} (redness) and *b*^{*} (yellowness). A D65 illuminant was used at an observation angle of 10° and with an aperture of 30 mm. The instrument was calibrated using a white and black standard.

2.4. Microbial analyses

Samples were aseptically collected from the slices swabbing an area of 10 cm² delimited with a sterile aluminium template (10 cm²). Each sample was homogenized in 0.1% peptone water (Bioline) and serial dilutions were done.

For psychrotrophic total viable counts (PTVC), *Enterobacteriaceae* and lactic acid bacteria (LAB) determination, one milliliter of the correct dilution was inoculated in a Petri Plate and after that approximately 15 ml of the appropriate agar were added. Psychrotrophic total viable counts (PTVC) were investigated using plate count agar (PCA) (Merck) after incubation at 10 °C for 96 h. For *Enterobacteriaceae* counts violet red bile dextrose agar (VRBD) was used, covering plates after solidifying with 3–4 ml of VRBD (double layer) (Merck). Then, they were incubated at 37 °C for 48 h. Plates for LAB investigation were covered with man rogosa and sharpe agar (MRS) (Merck) and they were placed in an anaerobic jar with an anaerobic atmosphere generator kit (Anaerocult A) (Merck) together with an anaerobic conditions indicator strip (GazPack™). Incubation was done for 96 h at 37 °C. Regarding *Pseudomonas* spp. determination, 0.1 ml of the property dilution was inoculated on the surface of Cephalothin-Sodium Fusidate-Cetrimide Agar (CFC) (Merck) and the inoculum was spread using a sterile plastic handle. Plates were counted after incubation at 20 °C for 24 h.

All microbial counts were expressed as base-10 logarithm of colony forming units per cm² of surface area (log CFU/cm²).

2.5. Sensory analyses

Samples were thawed in tap water for 4 h and after breaking vacuum were wrapped in aluminium foil and cooked at 200 °C in a double-plate grill (Sammic GRS-5, Guipúzcoa, Spain) until an internal temperature of 72 °C was reached, which was monitored by an internal thermocouple Jenway 200 (Jenway Scientific, Ston, United Kingdom). After cooking, the muscle *Semimembranosus* was cut in portions (2 cm × 2 cm × 2 cm) wrapped individually in aluminium foil and assigned a single random three digit code. Samples were placed at 60 °C in a warming cabinet until they were tasted (≤ 10 min after being cooked).

A 9-member trained panel (ISO 8586–1, 1992) was used to evaluate the samples. It had been trained in sensory assessment (ISO 8586–1, 1992) and was familiar with sensory assessment of meat. Special training was undertaken in order to recognise attributes before beginning the process of evaluating the samples. Evaluations were done using a quantitative structured scale based on descriptors punctuated from 0

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