PS8.04 Effect of active packaging on the oxidative and microbial stability of beef aged for different times and stored in modified atmosphere 4.00

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Abstract— **Fresh beef steaks were treated with two different preparations: one group was packaged without oregano (0%), the second group was packaged with oregano (4%) active film. Each steak was placed into a polystyrene tray. All the trays were filled with a gas mixture of 80% O_2 + 20% CO_2. The samples were displayed at 2 ± 1 °C in a chilled cabinet, illuminated by a standard supermarket fluorescent, for 19 days the 2 days samples and 13 days the 7 or 14 days postmortem ageing time samples. Colour determination (CIE a*, MetMb %), lipid oxidation (TBARS), psychrotrophic bacterial counts (PCA), sensory red colour, discolouration and odour were determined. Results demonstrated that the use of oregano by active films resulted in an effective delay of oxidative deterioration and microbial developement of fresh meat at all stages of storage and *postmortem* ageing time. Shelf life was extended beyond that of control, according to evaluation of sensory attributes (P<0.05).**

Key Words: Beef, Postmortem ageing time, display, active packaging, oregano, antimicrobial activity, antioxidant activity.

I. INTRODUCTION

The colour of all meats was more desired when presented under a light source (Barbut, 2001). Therefore, at the store, consumers must mainly base their selection on visual appearance when choosing meat. Food retail market assign a relatively large display space to high value meat cuts (Eilert, 2005). Textural parameters also affect meat tenderness, which is appreciated by consumers after purchase. The meat market is very concerned about the influence of ageing on beef quality. However, not many studies have analysed the possible influence of *postmortem* ageing time on the shelf life of meat.

Fresh meat is a prosperous nutrient medium that provides a suitable environment for proliferation of meat spoilage microorganisms and common food-borne pathogens; therefore adequate preservation technologies must be applied in order to preserve its safety and quality. Traditional packaging concepts are limited in their ability to prolong the shelf-life of meat products.

To delay or minimize microbial and oxidative deterioration, effective antioxidants and antimicrobial compounds could be added in meat (Djenane, Sánchez-Escalante, Beltrán & Roncalés, 2003). Consumer demands high quality, convenient, innovative, regular and safe meat products with natural flavour and taste and an extended shelf-life. Oregano (*Origanum vulgare* L.) is well known for its antioxidative and antimicrobial activity. The combined use of natural compounds and modified atmosphere (MA) packaging for meat represents a realistic and attractive strategy to increase the shelf life of fresh meat (Giese, 1996). The interest in the application of naturally occuring antioxidants and antimicrobial has increased over recent years. In the last decade new packaging systems have contributed to extend the shelf-life of products (Nerín, Tovar, Djenane, Camo, Salafranca, Beltrán & Roncalés, 2006; Nerín, Tovar & Salafranca, 2008). Active Packaging is an innovative food packaging concept that has been introduced as a response to the continuous changes in current consumer demands and market trends. That combines advances in food technology, food safety, and packaging and material sciences in an effort to better meet consumer demands for fresh-like, safe products.

In this system the antioxidant and microbial substances would gradually migrate from the pack to the food through diffusion and partitioning or release through evaporation in the headspace during storage and distribution, thus being able to reduce the postprocessing contaminations in the surface of the food products (Han, 2005; Rodríguez, Batlle & Nerín, 2007). Besides this, the development of active packaging is currently attracting the attention of food technologists. One of the most promising fields is the incorporation of antimicrobials such as bacteriocins and plant extracts to the active packaging and their association to biodegradable packaging in order to reduce wastes and being environmental friendly.

In this research a new antioxidant active food packaging has been developed and evaluated. It consists of an innovative system in which natural

antioxidants from oregano have been immobilized in a polypropylene film. The influence of oregano in the polymer and the contact system between the active film and food have been studied on the display life of fresh beef packaged in modified atmospheres at different *postmortem* ageing times.

II. MATERIALS AND METHODS

II. 1. Preparation of samples. Beef carcasses were obtained from Mercazaragoza (Zaragoza, Spain) 48 h *postslaughter*. Three *Longissimus dorsi* portions were vacuum-packaged and aged at 4ºC for either 2, 7 or 14 days. At the end of each ageing period, steaks of about 30-50 g weight (1.5 cm thick and about 50 cm^2 surface) were aseptically cut, using sterile cutting boards and knives. They were randomly divided into two groups within each ageing group. The first group was packaged without extracts (control). The second group was packaged with an active film (4% of oregano).

Each steak was placed into a polystyrene tray of size 15.5×21.5×2.5 cm. All the trays were filled with a gas mixture of 80% O_2 + 20% CO_2. Gas mixtures were kindly supplied by Abelló Linde S. A. (Barcelona, Spain). The samples were displayed at 2 ± 1 °C in a chilled cabinet, illuminated by a standard supermarket fluorescent (Mazdafluor Aviva TF/36 w; Philips, Eindhoven, Holland). All illuminated samples were exposed to lighting at 1000 lux at the surface with interruption simulating retail conditions at supermarket. Light intensity was measured using a luxometer Chauvin Arnoux 810 (Paris, France). The positions of the samples in the cabinet were rotated every 24 h to minimize light intensity differences and possible abuse temperature at the surface of meat.

Six samples were taken at each selected time (0, 5, 9, 13, 17, 19 and 21 days) for subsequent analysis. Two of them were used for microbial analysis, two were used for sensory analysis, and two were used for the quantification of TBARS.

II.2. Meat colour and metmyoglobin analysis. Meat colour was measured at the surface of lamb steaks using a reflectance spectrophotometer (Minolta CM-2002; Osaka, Japan), 30 min after pack opening, in order to allow colour stabilisation on air exposure. CIE L^* (lightness), a^* (redness) and b^* (yellowness) parameters were recorded (CIE, 1978). The average value for each steak was the mean of 10 determinations. The metmyoglobin (MetMb) percentage of the total myoglobin perceptable at the steak surface was estimated spectrophotometrically, according to Stewart, Zipser & Watts (1965), by measuring steak surface reflectance at 525 and 572 nm (Minolta CM-2002; Osaka, Japan). The maximum value of the ratios of $(K/S)_{572}$ to $(K/S)_{525}$ at the beginning of the experiment was fixed as 0% metmyoglobin; K and S were the absorption and the scattering coefficients, respectively, and K/S ratios were calculated from reflectivity (R_∞) values using the Kubelka–Munk equation. The value of 100% MetMb was obtained following the same procedure after oxidising a sample in a 1% (w/v) solution of potassium ferricyanid (Ledward, 1970). The average value for each steak was the mean of 10 determinations.

II.3. Lipid oxidation analysis (TBARS). Lipid oxidation was assessed in duplicate by the 2-thiobarbituric acid (TBA) method of Pfalzgraf, Frigg & Steinhart (1995), using 10 g of meat. TBARS values were calculated from a standard curve of malondialdehyde and expressed as mg malondialdehyde per kg meat.

II.4. Microbial sampling and analysis. For microbial analysis: Two sterile cotton swabs moistened in 0.1% peptone water were used for swabbing 10 cm^2 of meat surface, delimited by a sterile stainless steel template. Swabs were stirred thoroughly in 10 ml of 0.1% peptone water. Serial 5-fold dilutions were prepared by diluting 1 ml in 9 ml of 0.1% peptone water. Two duplicate plates were prepared from each dilution by pouring 1 ml in fluid plate count agar (PCA; Merck; Darmstadt, Germany); plates were incubated at 10 °C for 7 days (ICMSF, 1983). Counts of aerobic psychrotrophic flora were determined from plates bearing 30–300 colonies. Counts were expressed as the $\log_{10}$ of colony forming units (CFU) per cm^2.

II.5. Sensory evaluation. Meat samples were evaluated by a six-member expert panel, trained according to the method of Cross, Moen. & Stanfield (1978). Three open-discussion sessions were held to familiarise panellists with the attributes and the scale to be used. The attributes studied were: **''***Red colour***'',** **''***Discolouration***''** and **''***off odour***''**. All attributes were scored using a 5-point descriptive scale, according to Djenane, Sanchez-Escalante, Beltrán, & Roncalés (2001), using a paper scorecard. **''**Red colour**''**,1 denoted extremely high and 5 denoted extremely low. Scores for **''**discolouration**''** referred to percentage (%) of discoloured surface: 1 = none, 2 = 0–10%, 3 = 11–20%, 4 = 21–60%, and 5 = 61–100%. Scores for **''**off odour**''** referred to the intensity of odours associated to meat spoilage: 1 = excellent, not different from fresh meat; 2 = good, but slightly poorer than fresh meat; 3 = acceptable, but obviously poorer than fresh meat; 4 = hardly acceptable as fresh meat; and 5 = non acceptable.

II.6. Statistical analysis

The significance of differences among samples at each day of storage was determined by analysis of variance (ANOVA) using the least square difference method of

the General Linear Model procedure of SPSS (1995). Differences were considered significant at the P<0.05.

III. RESULTS AND DISCUSSION

Figures (1,2,3, a, b, c and d, respectively) show the results of CIE a*, Metmyoglobin %, TBARS and PCA counts throughout the storage of beef steaks packaged without oregano extracts and those packaged with an active film 4% of oregano. Results demonstrated that the active packaging with oregano extract effectively improved shelf life of fresh beef steaks, led to significant differences in CIE a* values (P<0.05) with the two groups from day 9 of storage onwards. Results demonstrated that the active film packaging effectively delayed metmyoglobin formation. This effect was evident from day 9 of storage, showing significant (P<0.05) differences with control samples. Samples packed with an active film showed a highly significant (P<0.05) inhibitory effect on the formation of TBARS. Differences were significant amongst samples of storage onwards (P<0.05) from day 9 and 5 respectively, for 48 h samples and 7, 15 days *postmortem* ageing time. Microbial counts of all samples gradually increased along storage, and reached final values of 6–8 $\log_{10}$ cfu $cm^{-2.}$ Results demonstrated that the active packaging with oregano extract effectively delayed counts of aerobic psychrotrophic flora. Samples treated with oregano active film exhibited lower counts during the whole period of storage, and the difference were significant (P<0.05) from day 5 of storage onwards.

Results of sensory analysis of meat samples group, including evaluation of red colour, discolouration and fresh meat odour, are summarised in **Tables 1,2,3.** These results clearly demonstrated that beef steaks packaged with active films were given lower scores than control samples. Beef loins in 4% oregano active packaging film obtained the highest sensory scores. This intensity of the protective ability on meat quality of the antioxidant treatments, as measured by sensory evaluation, consistently agreed with their effectiveness in preventing myoglobin oxidation, microbial development and lipid oxidation.

Tabel 1

Parameter	Samples (*0 day Postmortem ageing*)	Days of storage 0	5	9	13	17	21
Red colour	80/20 (O_2/CO_2)	1	1	1	3	3	5
	Oregano 4%	1	1	1	2	3	5
Discolouration	80/20 (O_2/CO_2)	1	1	1	3	4	5
	Oregano 4%	1	1	1	1	3	5
Off odour	80/20 (O_2/CO_2)	1	1	1	3	4	5
	Oregano 4%	1	1	1	2	3	4

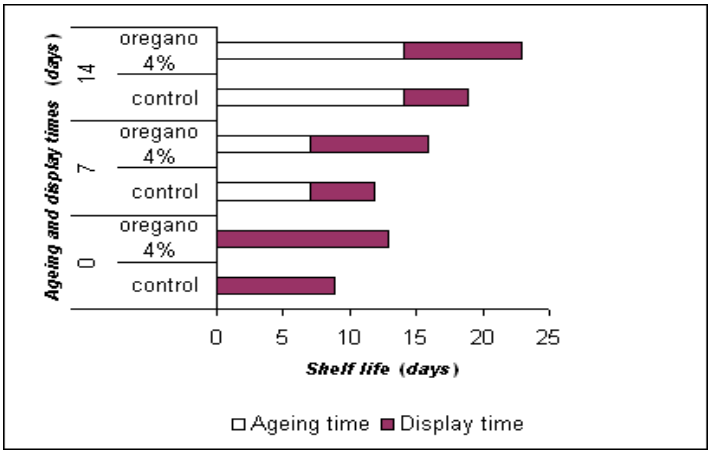
Table 2. Sensory analysis of meat samples (7 days *postmortem* ageing)

Parameter	Samples (*7 days Postmortem ageing*)	Days of storage 0	5	9	13
Red colour	80/20 (O_2/CO_2)	1	1	2	5
	Oregano 4%	1	1	2	5

Table 3. Sensory analysis of meat samples (14 days *postmortem* ageing)

Parameter	Samples (*14 days Postmortem ageing*)	Days of storage			
		0	5	9	13
Red colour	80/20 (O_2/CO_2)	1	1	2	3
	Oregano 4%	1	1	2	2

Fig 4. Effect of active packaging on the Shelf life extension of beef stored in modified atmospheres at different ageing time



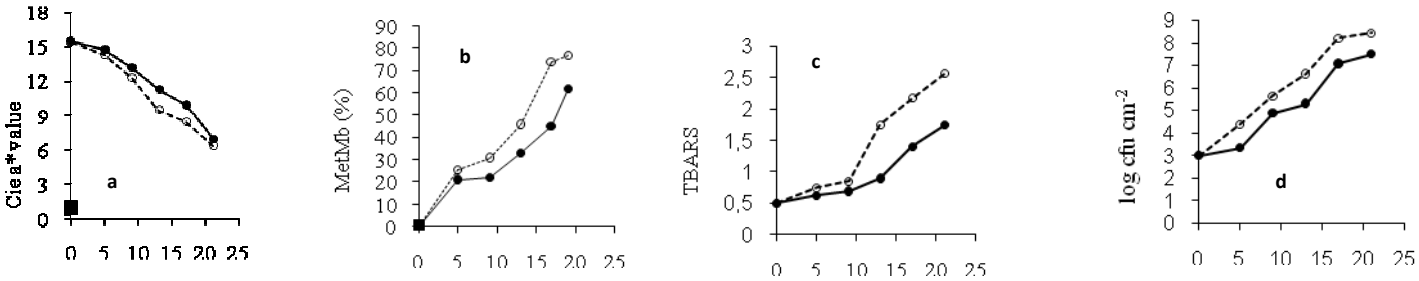
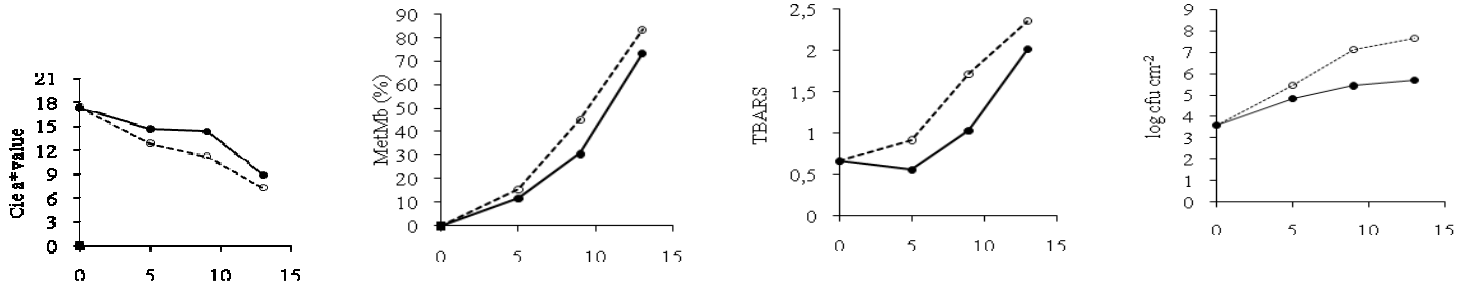


Fig. 1. Days of storage (samples 0 day *postmortem* ageing)



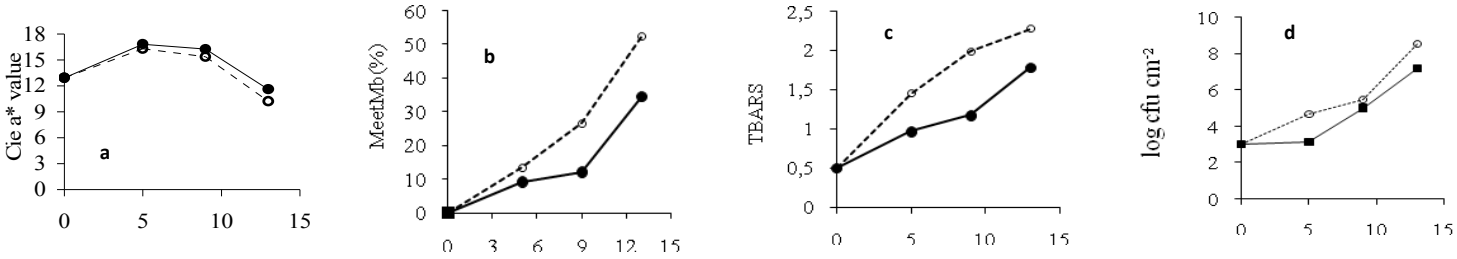


Fig. 3. Days of storage (samples 14 days *postmortem* ageing)

IV. CONCLUSION

Packaging with an oregano active film resulted in enhanced oxidative stability and effectively delayed count of aerobic psychrotrophic flora of fresh beef steaks obtained for beef *longissimus dorsi* at different ageing time packaged in modified atmosphere displayed at 2 ± 1 °C in a chilled cabinet, illuminated by a standard supermarket fluorescent. Lipids were protected against oxidation during storage, resulting in a better colour and odour stability than the control, extending the shelf life for at least 3 days.

Oregano active film retarded colour loss. Oxymyoglobin and lipids were protected against oxidation during storage, resulting in a better colour and odour stability than the control.

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