SHELF-LIFE EXTENSION BY CARNOSINE OF BEEF PACKAGED IN ATMOSPHERES VARYING IN OXYGEN AND CARBON DIOXIDE.

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INTRODUCTION

Oxidative processes, including lipid oxidation, are deleterious to meat quality, particularly when stored for long periods. Carnosine has been shown to inhibit lipid oxidation catalyzed by most oxidants. The antioxidant mechanism of carnosine has been suggested to be due to a combination of metal chelation and free radical scavenging (Kohen *et al.*, 1988; Decker *et al.*, 1992). An antioxidant effect of the dipeptide has been demonstrated in model systems, as well as in minced meat (Decker and Crum, 1991), beef homogenates (Lee and Hendricks, 1997) and chicken meat (O'Neill et al., 1997). Carnosine appears then to have an excellent potential as a natural antioxidant for its use in meat storage. Our aim was to apply the antioxidant effect of carnosine to shelf-life extension of beef packaged in a standard modified atmosphere. We focused also on the effect of decreasing oxygen and increasing carbon dioxide concentrations on carnosine action.

MATERIALS AND METHODS

Preparation of Samples. Three bovine *Longissimus lumborum* muscles were injected with carnosine solutions (25mM and 50mM) or distilled water. The injected volume was the 2% of muscle's weight. The muscles were stored at 1±1°C for 5 h before packaging beef steaks (2 cm thick). Gas mixtures consisted of: 70% O_2 + 20% CO_2 + 10% N_2 (v/v) (CMA), 60% O_2 + 40% CO_2 (v/v) and 40% O_2 + 60% CO_2 (v/v). For each of the three atmospheres, three samples were evaluated. **Lipid Oxidation.** Lipid oxidation was assessed by the 2-thiobarbituric acid method of Witte *et al.*, (1970). TBA values were expressed as mg TBARS/kg sample. **Microbial Analysis.** Microbial analysis were done by sprinkling an area of 10 cm^2 of meat surface. Using conventional dilution procedures (in 0.1% peptone water), counts of aerobic psychrotrophic flora were determined from plates bearing 20-200 colonies in PCA, incubated at 7°C for 10 days; and *Brochothrix thermosphacta* in STAA agar, with streptomycin sulfate (500mg/L), thallous acetate (50mg/L), and cycloheximide (50mg/L), incubated at 25°C for 3 days. **Colour Instrumental Measurement.** CIE L*, a*, b* parameters were measured at the surface of meat samples using a spectrophotometer (Minolta Chroma Meter CM-2002), 30 min after package opening. Each value of this parameters was the mean of 20-25 determinations. **Metmyoglobin Measurement.** Metmyoglobin percentage was estimated by measuring the surface reflectance at 525 and 572 nm according to Stewart *et al.* (1965), using Kubelka-Munk (K/S) ratios. The maximum value of the quotient between K/S_{572} and K/S_{525} was fixed as 0% MetMb, while 100% MetMb was obtained after oxidizing a sample in a 1% (w/v) solution of potassium ferricyanide. **Sensory Analysis.** Meat samples were evaluated by a trained panel of six members. Three open-discussion sessions were held in order to familiarize the individual with the attributes to study and the scale to use. Discolouration was scored using a 9-point scale; 9 denoted extremely high and 1 denoted extremely low.

RESULTS AND DISCUSSION

Colour Instrumental Measurement. Changes in a* values (Figure 1) demonstrated that redness of fresh beef steaks was little infuenced by atmosphere, while it was by the presence of carnosine. Steaks injected with carnosine, both 25mM and 50 mM, showed a* values significantly higher. A delay of 2-4 days in redness decay was evident in all cases.

Metmyoglobin Measurement. Figure 2 shows the changes in MetMb percentage. The amount of MetMb formed was higher in samples packaged with elevated concentrations of CO_2, which reached a critical acceptable level (30-40% of metmyoglobin) as soon as 11-14 days, whereas samples packaged in CMA reached this level after 16-18 days. Carnosine gave rise to a delay of 3-6 days for all atmospheres assayed in MetMb formation.

Sensory analysis. Results of Discolouration displayed in Figure 3 showed that no effect of the atmosphere was evident. Carnosine, on the contrary, caused a delay of about 4 days in discolouration appearance.

Microbial Analysis. The mean $\log_{10}$ cfu cm^{-2} of total psychrotrophic aerobes (PCA) and *Brochothrix thermosphacta* (STAA) are shown in Figures 4 and 5. Psycrotrophic flora counts were higher in samples stored in CMA than in other atmospheres. A difference of 1-1.5 logarithmic cycles can be observed between the third and fourth week of storage period. Values higher than 10^7 cfu/cm^2 were only reached after 28 days of storage in the CMA packaged samples. Results agreed with Luño et al. (1998). No appreciable effect of carnosine was evident. *B. th.* grew quite rapidly to maximum levels near to 10^7 cfu/cm^2. A strong delay of *B. th.* growth was observed by effect of decreasing oxygen and increasing carbon dioxide. No effect of carnosine was observed.

Lipid Oxidation. Figure 6 shows the changes in TBA value. For all atmospheres, TBA values were lower in samples containing carnosine, which caused a delay of 4-6 days in TBARS formation. Therefore, carnosine was able to inhibit effectively lipid oxidation of beef steaks. The results are in good agreement with the findings of Lee and Hendricks (1997) and O'Neill et al. (1997).

CONCLUSION

Carnosine was effective in inhibiting oxidation, as revealed by TBA value. Shelf-life of beef packaged in modified atmosphere was extended by 4-6 days, according to results of a*, MetMb formation and discolouration, although microbial counts were not affected. Decreasing oxygen and increasing carbon dioxide inhibited microbial growth, though it did not result in shelf-life extension.

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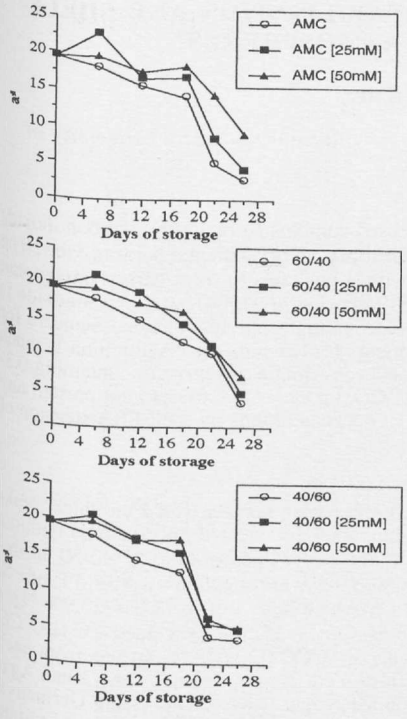

Fig. 1.- Values of a* for (*)

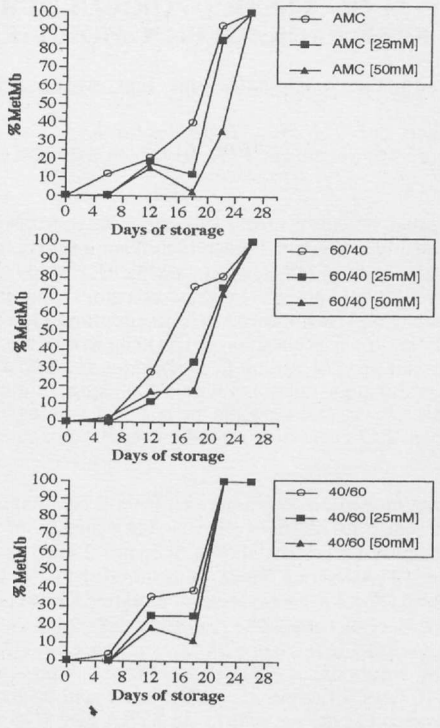

Fig. 2.- Metmyoglobin percentage of (*)

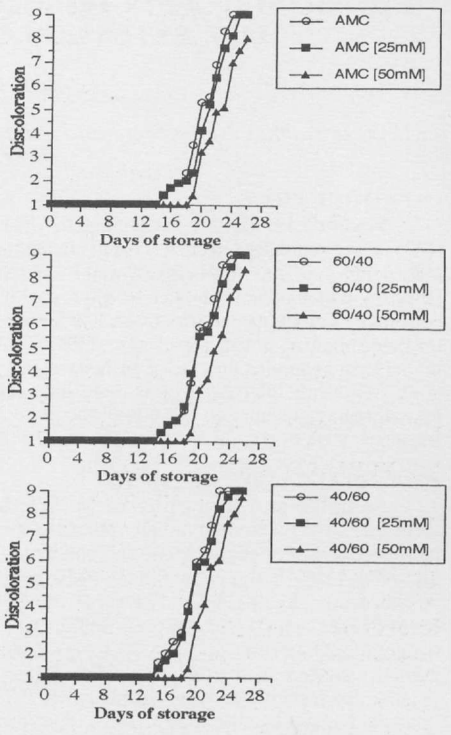

Fig. 3.- Sensory evaluation of discolouration of (*)

(*) Beef steaks with added carnosine (none: ○, 25 mM: ■, 50mM: ▲), packaged in atmospheres containing either 70% O2 + 20% CO2 + 10% N2 (CMA)(**A**), 60% O2 + 40% CO2 (**B**) or 40% O2 + 60% CO2 (**C**).

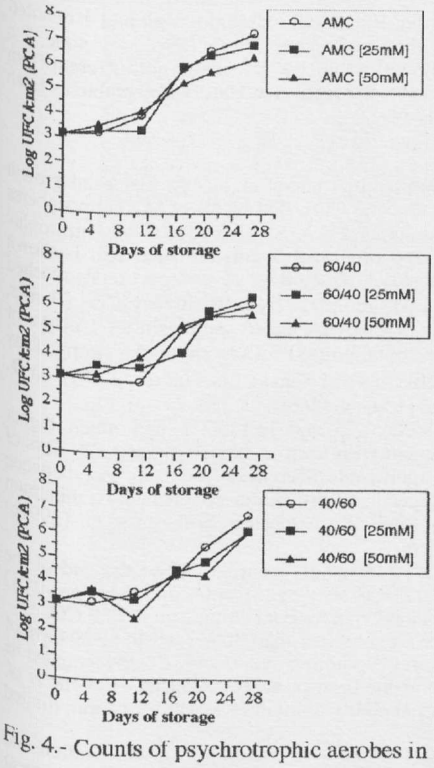

Fig. 4.- Counts of psychrotrophic aerobes in (*)

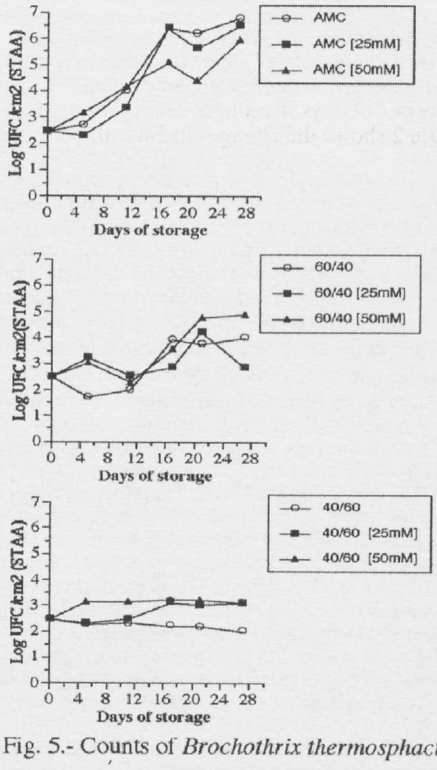

Fig. 5.- Counts of *Brochothrix thermosphacta* in (*)

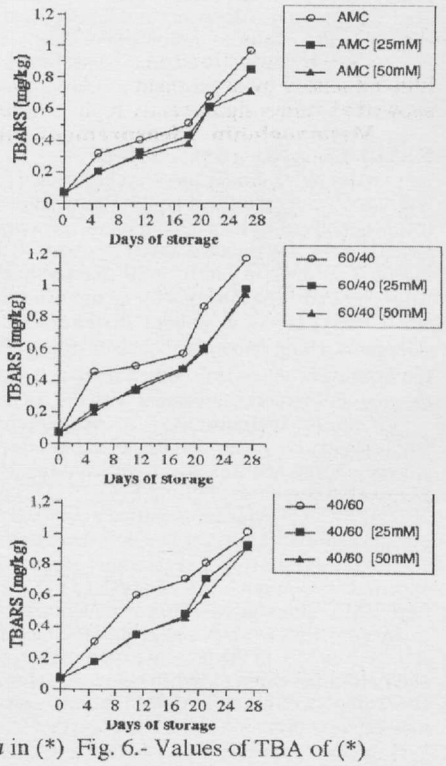

Fig. 6.- Values of TBA of (*)

(*) Beef steaks with added carnosine (none: ○, 25 mM: ■, 50mM: ▲), packaged in atmospheres containing either 70% O2 + 20% CO2 + 10% N2 (CMA)(**A**), 60% O2 + 40% CO2 (**B**) or 40% O2 + 60% CO2 (**C**).