# EFFECTS OF PACKAGING ATMOSPHERES ON SHELF-LIFE AND QUALITY CHARACTERISTICS OF HEAVILY MARBLED LONGISSIMUS STEAKS

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Abstract -This study investigated the effects of four typical modified atmosphere packaging (MAP) methods on shelf-life and quality characteristics of beef steaks with high marbling. The packaging methods used included 80%O<sub>2</sub>-MAP  $(80\% O_2 + 20\% CO_2)$ , 50%  $O_2$ -MAP  $(50\% O_2)$ O<sub>2</sub> + 30% CO<sub>2</sub> + 20% N<sub>2</sub>), carbon monoxide MAP  $(0.4\% \text{ CO} + 30\% \text{ CO}_2 + 69.6\% \text{ N}_2)$  and vacuum packaging (VP). Steaks were stored at 4 °C for 12 days. Shelf life and quality evaluations were based on color stability, oxidative stability and microbial counts. CO-MAP had the best preservation for beef steaks, due to more red color together with a lower oxidation and microbial loads. For steaks in both aerobic packaging, although microbial counts did not reach the recommended legal threshold, discoloration and oxidation were major factors limiting their shelf-life to 12 days or less. These results indicate anaerobic packaging extends shelf-life properties throughout storage.

#### Key Words –Modified atmosphere packaging, Preservation, Snowflake beef

#### I. INTRODUCTION

Highly-marbled fattened cattle, known as *snowflake* beef in China, are characterized by their ability to deposit a large amount of intramuscular marbling fat. *Snowflake* beef is widely preferred by consumers in East Asia, however, to our knowledge, few have investigated the microbial and color stability of *snowflake* beef during distribution and retail display; especially lacking its information on the effects of packaging systems.

The MAP with 80%  $O_2$  and 20%  $CO_2$  is the most popular MAP type for fresh red meat in the world because of the improvement in color stability, but it is also expected to increase the rate of meat oxidation [1]. The 50%  $O_2$ -MAP appears as one kind of commercial low oxygen MAP in recent years [2]. Lücke *et al.* (2014) indicate that reducing the  $O_2$  level in MAP to about 40-50% still sufficiently stabilizes meat

color while reducing meat oxidative processes [3]. One of the most recent meat packaging technologies introduced in U.S. is the CO-MAP with a low level of CO (0.4%), 20-30% CO<sub>2</sub>, and 69.6% N<sub>2</sub> [4]. CO binds strongly to the myoglobin to form carboxymyoglobin, which provides meat a bright cherry-red color for a long time, however, this stable red color may conceal spoilage and may put consumers at risk [5]. Vacuum packaging (VP) produces anaerobic conditions which inhibit both microbiological growth and oxidation of meat. But the purple color and visible purge loss of VP meat are often regarded as less attractive by consumers [6]. This work aimed to compare the effects of four different packaging conditions on the shelf-life and quality of *snowflake* beef and determine the most suitable packaging methods.

#### II. MATERIALS AND METHODS

A. Animals, sampling and experimental design The *M. longissimus lumborum* were extracted from four fattened steers (Bos Japanese Black Cattle× Bos Yanbian Yellow cattle, aged 32 to 37 months old, carcasses weighted 392 to 423 kg, the fat content was  $22.76 \pm 5.03$ , and then vacuum packaged, kept frozen at -20 °C until transported to the laboratory. Before packaging, samples were thawed at 4 °C

Before packaging, samples were thawed at 4 °C for 24 h. Muscles were cut into 2 cm thick steaks. A total of 39 steaks was obtained from both sides of each carcass and mixed, and then randomly packed in 80% O<sub>2</sub>-MAP (80% O<sub>2</sub> + 20% CO<sub>2</sub>), or 50% O<sub>2</sub>-MAP (50% O<sub>2</sub> + 30% CO<sub>2</sub> + 20% N<sub>2</sub>), or CO-MAP (0.4% CO + 30% CO<sub>2</sub> + 69.6% N<sub>2</sub>), or vacuum packaging. Three replicates were performed for each treatment at each storage time interval (4, 8, 12 d). Three steaks from each carcass were analysed for initial data at time zero.

B. Experimental methods

The color attribute, including lightness  $(L^*)$ , redness  $(a^*)$ , yellowness  $(b^*)$ , hue  $(h^*)$ , chroma  $(C^*)$  and the relative content of metmyoglobin (MetMb%), was measured using the method described by [1]. Lipid oxidation was measured using the 2-thiobarbituric acid method described by Siu & Draper (1978) [7]. Protein carbonyls were quantified following the DNPH assay according to the modification of Oliver *et al.* (1987) [8].

Ten gram samples were aseptically placed into a stomacher bag containing 90 ml sterile 0.1% peptone water. After homogenization, a 10-fold dilution series was carried out for microbiological analysis. For total viable counts, diluted samples were cultured in Plate Count Agar (PCA; LandBridge Co., Ltd., Beijing, China) and incubated at 37 °C for 48 h.

### C. Statistical analysis

Statistical analysis was carried out with the Statistical Analysis System (Version 9.0, SAS Inst., Inc., Cary, NC, USA). Experiments adopted a split-plot design. For the whole plot, cattle (n=4) served as blocks (replicates). Within the subplot, steaks from each cattle were randomly assigned to the combinations of 4 packaging treatments and 3 storage times (d 4, 8, 12). The MIXED procedure was used to estimate the effects on variation in samples of packaging treatments, storage time and their interaction as fixed factors and animal as random factor. Differences were considered significant at P < 0.05.

# III. RESULTS AND DISCUSSION

The evolution of L<sup>\*</sup>, a<sup>\*</sup>, b<sup>\*</sup>, C<sup>\*</sup> and h<sup>\*</sup> is shown in Table 1. Compared with other three packaging types, CO-MAP steaks showed the best color protection effect with higher L<sup>\*</sup>, a<sup>\*</sup>, b<sup>\*</sup>, C<sup>\*</sup> values than others during display. Although vacuum packaging steaks maintained lower L<sup>\*</sup>, a<sup>\*</sup>, their color was more stable than aerobic packaging steaks. The samples in 80% O<sub>2</sub>-MAP and 50% O<sub>2</sub>-MAP had a faster discoloration than anaerobic packaging. 50% O<sub>2</sub>-MAP samples showed the lowest color stability within 12 days and the L<sup>\*</sup>, a<sup>\*</sup> was significantly lower than 80%O<sub>2</sub>-MAP at day 8 (P<0.05). This might be caused by a particularly pronounced lipid oxidation in meat, which induced dissolved oxygen depletion and promoted the formation of metmyoglobin. Since Greene *et al*, (1971) reported that an above 40% MetMb caused meat rejection by consumers [9], the shelf-life of snowflake beef steaks in aerobic packaging should not exceed 12 d (Table 1).

Compared with two aerobic packaging methods used in this study, CO-MAP and vacuum packaging steaks remained lower TBA values and carbonyl content during storage (Table 2), which indicated anaerobic packaging could effectively help reduce the lipid and protein oxidation rate of snowflake beef steaks. 80%O<sub>2</sub>-MAP samples had the highest TBA values and carbonyl content during display and their values were significantly higher than 50%O<sub>2</sub>-MAP samples at day 12 (P < 0.05). Considering the threshold for rancidity in beef meat was 2.28 mg MDA/kg [10], the present study suggests that the shelf-life of snowflake beef in aerobic packaging should also not exceed 12 days due to the pronounced lipid oxidation.

The CO-MAP and vacuum packaging showed the best microbial inhibitory effect for snowflake beef steaks.  $50\%O_2$ -MAP samples had lower microbial loads compared with  $80\%O_2$ -MAP. Among these four packaging methods, microbial counts did not reach the recommended legal threshold during the 12-day storage period.

# IV. CONCLUSION

It's suggested that adopting anaerobic packaging for snowflake beef. As a realistic alternative for beef industry, CO-MAP had the best preservative effect. If using aerobic packaging for snowflake beef, discoloration and oxidation were two major factors limiting their shelf-life to 12 days or less.

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Attributo	PM	Storage time (days)				CE.	P-value		
Attribute		0	4	8	12	SE	PM	Day	PM×Day
$L^*$	80%O <sub>2</sub>	43.69 <sup>A</sup>	45.83 <sup>aAB</sup>	46.95 <sup>aB</sup>	47.09 <sup>aB</sup>	0.90	< 0.001	< 0.001	0.083
	$50\%O_2$	43.69 <sup>A</sup>	44.93 <sup>aA</sup>	$47.44^{aB}$	47.41 <sup>aB</sup>	0.90			
	CO-MAP	43.69 <sup>A</sup>	$44.78^{abA}$	46.83 <sup>aB</sup>	$47.62^{aB}$	0.90			
	VP	43.69 <sup>AB</sup>	$41.42^{bA}$	$42.48^{bAB}$	44.11 <sup>bB</sup>	0.90			
$a^*$	$80\%O_{2}$	26.37 <sup>A</sup>	26.32 <sup>abA</sup>	21.85 <sup>aB</sup>	16.41 <sup>aC</sup>	0.43	< 0.001	< 0.001	< 0.001
	50%O <sub>2</sub>	26.37 <sup>A</sup>	$25.26^{aA}$	$17.70^{bB}$	$14.26^{aC}$	0.43			
	CO-MAP	26.37 <sup>A</sup>	$28.49^{bA}$	30.60 <sup>cAB</sup>	31.23 <sup>bB</sup>	0.43			
	VP	26.37 <sup>A</sup>	21.13 <sup>cB</sup>	20.37 <sup>aB</sup>	20.52 <sup>cB</sup>	0.43			
$b^*$	$80\%O_2$	$18.58^{AB}$	19.19 <sup>aA</sup>	17.33 <sup>aB</sup>	15.55 <sup>aC</sup>	0.30	< 0.001	< 0.001	< 0.001
	$50\%O_2$	18.58 <sup>A</sup>	18.32 <sup>aA</sup>	15.37 <sup>bB</sup>	14.69 <sup>aB</sup>	0.30			
	CO-MAP	$18.58^{AB}$	17.89 <sup>aA</sup>	19.73 <sup>cB</sup>	20.13 <sup>bB</sup>	0.30			
	VP	$18.58^{A}$	11.55 <sup>bB</sup>	11.61 <sup>dB</sup>	12.25 <sup>cB</sup>	0.30			
Chroma	$80\%O_{2}$	32.26 <sup>A</sup>	32.57 <sup>aA</sup>	$27.90^{aB}$	22.65 <sup>acC</sup>	0.49	< 0.001	< 0.001	< 0.001
	50%O <sub>2</sub>	32.26 <sup>A</sup>	31.20 <sup>aA</sup>	23.43 <sup>bB</sup>	$20.54^{aC}$	0.49			
	CO-MAP	32.26 <sup>A</sup>	33.64 <sup>aA</sup>	36.47 <sup>cB</sup>	37.16 <sup>bB</sup>	0.49			
	VP	32.26 <sup>A</sup>	24.08 <sup>bB</sup>	23.45 <sup>bB</sup>	23.90 <sup>cB</sup>	0.49			
Hue	$80\%O_2$	35.13 <sup>A</sup>	36.10 <sup>aA</sup>	$38.52^{aB}$	43.97 <sup>aC</sup>	0.57	< 0.001	< 0.001	< 0.001
	50%O <sub>2</sub>	35.13 <sup>A</sup>	35.97 <sup>aA</sup>	$41.04^{bB}$	46.60 <sup>bC</sup>	0.57			
	CO-MAP	35.13 <sup>A</sup>	32.15 <sup>bB</sup>	32.77 <sup>cB</sup>	32.83 <sup>cB</sup>	0.57			
	VP	35.13 <sup>A</sup>	28.63 <sup>cB</sup>	$29.70^{dBC}$	30.85 <sup>dC</sup>	0.57			
MetMb%	$80\%O_{2}$	4.67 <sup>A</sup>	$12.88^{aB}$	$27.22^{aC}$	$48.79^{\mathrm{aD}}$	0.58	< 0.001	< 0.001	< 0.001
	50%O <sub>2</sub>	4.67 <sup>A</sup>	13.95 <sup>aB</sup>	33.70 <sup>aC</sup>	57.29 <sup>bD</sup>	0.58			
	CO-MAP	4.67	12.39 <sup>a</sup>	11.14 <sup>b</sup>	11.26 <sup>c</sup>	0.58			
	VP	4.67	2.54 <sup>b</sup>	5.96 <sup>b</sup>	5.78 <sup>c</sup>	0.58			

 Table 1 Effects of packaging methods (PM) and storage time (days) on meat color stability of snowflake beef steaks at 4 °C.

Means with different superscript capital letters within the rows (effect of storage time) differ at P < 0.05. Means with different superscript lowercase letters within the columns (effect of packaging) differ at P < 0.05.

Table 2	
Evolution of oxidative parameters of <i>snowflake</i> beef steaks packaged under different MAP at 4 °C.	

Attribute	РМ	Storage time (days)			SE	P-value			
Attribute		0	4	8	12	36	PM	Day	PM×Day
TBARS	$80\%O_2$	0.35 <sup>A</sup>	1.33 <sup>aB</sup>	2.35 <sup>aC</sup>	3.14 <sup>aD</sup>	0.04	< 0.001	< 0.001	< 0.001
(mg MDA/kg)	50%O <sub>2</sub>	0.35 <sup>A</sup>	$1.18^{aB}$	2.23 <sup>aC</sup>	2.93 <sup>bD</sup>	0.04			
	CO-MAP	0.35	0.39 <sup>b</sup>	0.37 <sup>b</sup>	0.38 <sup>c</sup>	0.04			
	VP	0.35	0.36 <sup>b</sup>	0.37 <sup>b</sup>	0.37 <sup>c</sup>	0.04			
Carbonyls	$80\%O_2$	0.64 <sup>A</sup>	1.51 <sup>aB</sup>	$2.02^{aC}$	$3.57^{aD}$	0.07	< 0.001	< 0.001	< 0.001
(nmol/mgprot)	50%O <sub>2</sub>	0.64 <sup>A</sup>	$1.46^{aB}$	$1.57^{bB}$	2.86 <sup>bC</sup>	0.07			
	CO-MAP	0.64 <sup>A</sup>	$0.52^{bA}$	0.65 <sup>cA</sup>	0.97 <sup>cB</sup>	0.07			
	VP	0.64	0.56 <sup>b</sup>	0.55 <sup>c</sup>	0.65 <sup>d</sup>	0.07			

Means with different superscript capital letters within the rows (effect of storage time) differ at P < 0.05. Means with different superscript lowercase letters within the columns (effect of packaging) differ at P < 0.05.

Fig.1 Total viable count of snowflakes beef steaks packaged under different MAP conditions during storage at 4  $^{\circ}\mathrm{C}$ 

