

Comparison of Beef Color Stability during Display of Two Muscles between Japanese Shorthorn Steers and Japanese Black Steers

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ABSTRACT : The beef color stability during display of two muscles, *m. longissimus thoracis* and *m. semitendinosus*, of Japanese Shorthorn steers (n=14) was compared with that of Japanese Black steers (n=14). The beef color of each carcass was evaluated according to the Japanese Grading Standards at 24 h post mortem. Steak samples from muscles were over-wrapped with PVC film and displayed under fluorescent lights at 4°C for 9 days. Metmyoglobin percentages of steak samples were determined at days 0, 3, 6 and 9. The overall grade of beef color of the carcasses of Japanese Shorthorn steers was significantly ($p<0.05$) lower than that of Japanese Black steers. The metmyoglobin percentages during the display of two muscles of Japanese Shorthorn steers were significantly ($p<0.05$) lower than those of Japanese Black steers. These results suggested that though beef color evaluation of the carcasses of Japanese Shorthorn steers was lower than that of Japanese Black steers, the beef color stability during the display of the muscle of Japanese Shorthorn steers was higher than that of Japanese Black steers. (*Asian-Aust. J. Anim. Sci.* 2004, Vol 17, No. 9 : 1303-1308)

Key Words : Beef, Color Stability, Metmyoglobin, Japanese Shorthorn Steers, Japanese Black Steers

INTRODUCTION

Japanese Shorthorn is one of four breeds of Wagyu (Japanese beef cattle). Japanese Shorthorn is bred mainly in the northern area of Iwate Prefecture in Japan. The evaluation of beef quality of Japanese Shorthorn is generally lower than that of Japanese Black, because the beef of Japanese Shorthorn does not become marbling beef which is generally preferred by Japanese consumers. Recently, however, the number of Japanese consumers who prefer less marbling, or lean beef, has been increasing. In evaluating the quality of lean meat, color, firmness and texture are the most important factors. Sanders et al. (1997) reported that 58% of Japanese survey participants (n=10,941) identified beef color as the most important factor in selecting beef products. The red color of fresh beef is due to oxymyoglobin. The discoloration of beef from red to brown during display results from the oxidation of oxymyoglobin to metmyoglobin (Faustman and Cassens, 1990). Several reports have addressed the beef color stability of the muscle of Japanese Black steers (Mitsumoto et al., 1995; Mitsumoto et al., 1998; Muramoto et al., 2003a; Muramoto et al., 2003b). However, there are few reports regarding the beef color stability of the muscle of Japanese Shorthorn steers (Ito and Kondo, 1983). The purpose of this study was to compare the beef color stability during display of the muscles of Japanese Shorthorn steers and Japanese Black steers.

MATERIALS AND METHODS

Animals and diets

Fourteen Japanese Shorthorn steers (10 months of age) and fourteen Japanese Black steers (10 months of age) were fed commercial formula feed and hay until slaughtered. Japanese Shorthorn steers and Japanese Black steers were slaughtered at 28.3 ± 0.7 and 29.3 ± 1.3 months of age, respectively.

Beef color evaluation of carcasses

After slaughter, the carcasses were kept in a 0°C refrigerator. For beef color evaluation, each carcass was graded at the 6-7th rib interface by official Japanese graders in accordance with Japanese Grading Standards (JMGA, 1989) at 24 h post mortem. The beef color was evaluated according to the Beef Color Standard (BCS) prepared as seven continuous standards from No. 1 (pale) to No. 7 (dark). The range of BCS Nos. 1 to 7 was graded as "Grade 2 (Below Average)" or upper grades. The range of BCS Nos. 1 to 6 was graded as "Grade 3 (Average)" or upper grades. The range of BCS Nos. 2 to 6 was graded as "Grade 4 (Good)" or upper grade. The range of BCS Nos. 3 to 5 was graded as "Grade 5 (Very Good)". Other beef color was graded as "Grade 1 (Inferior)". The beef brightness was evaluated by visual appraisal. The beef brightness of "Very Good", "Good", "Average", "Below Average" and "Inferior" was graded as "Grade 5", "Grade 4", "Grade 3", "Grade 2" and "Grade 1", respectively. The lowest grade of the two items was graded as the overall beef color grade of each carcass.

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Table 1. Beef Color Standard numbers, grades of beef brightness and overall grades of beef color of carcasses of Japanese Shorthorn steers and Japanese Black steers

	BCS No. ¹		Beef brightness		Overall grade	
	MN	SE	MN	SE	MN	SE
Japanese Shorthorn steers	5.4 ^a	0.3	1.6 ^b	0.1	1.6 ^b	0.1
Japanese Black steers	3.1 ^b	0.2	3.7 ^a	0.2	3.7 ^a	0.2

^{a, b} Means within a column with a different superscript letter differ ($p < 0.05$). ¹ Beef color standard numbers (JMGA 1988).

Muscle samples

Two muscles, *m. longissimus thoracis* and *m. semitendinosus*, were identified and removed from the left side of each carcass. A part of each muscle was ground twice through a 3 mm plate of a laboratory meat grinder for analyses of crude fat and α -tocopherol concentrations. The ground meats were stored in a -40°C refrigerator until required. The remainder of each muscle was vacuum-packaged and stored for an additional 5 days at 4°C for beef color and metmyoglobin analyses.

Crude fat and α -tocopherol analyses

Crude fat concentration in each muscle was determined by the ether extract method according to AOAC (1984). α -Tocopherol concentration in each muscle was determined by the HPLC method described by Bennink and Ono (1982). The 0.5–1.0 g samples were placed in a 60 ml tube with 1 ml of 1% (W/V) sodium chloride and 10 ml of 3% pyrogallol-ethanol. The tubes were mixed and preincubated in a 70°C water bath for 5 minutes before 2 ml of 60% (W/V) KOH was added. The samples were saponified at 70°C for 30 minutes and were allowed to cool before 22 ml of 1% (W/V) sodium chloride and hexane:ethyl acetate (9:1) were added. The saponified mixture and hexane:ethyl acetate were shaken and then centrifuged to separate the aqueous and organic phases. An aliquot of the hexane phase which contained the α -tocopherol was removed and dried under a stream of nitrogen at 60°C . α -Tocopherol standards were carried through the same procedure as described for the muscle samples. α -Tocopherol was determined by high-performance liquid chromatography with fluorometric detection. Dried lipid from the hexane and injected onto a 0.46×25 cm silicagel column (PEGASIL Silica 60-5, Senshu Scientific Co., Ltd., Tokyo, Japan) and α -tocopherol were eluted with hexane:acetic acid:isopropanol (1,000:6:5) at a flow rate of 1.5 ml/min. Detection of the α -tocopherol in the eluant was performed using a spectrofluorometer (RF-10A XL, SHIMADZU CO, Kyoto, Japan) equipped with a flow cell. α -Tocopherol was detected by measuring extraction at 298 nm and emission at 325 nm. Standard curves for α -tocopherol were prepared by injecting known amounts of α -tocopherol and measuring resultant peak heights.

Beef color and metmyoglobin analyses

Muscle samples for beef color and metmyoglobin analyses were sliced into 1.5 cm-thick steaks, and three pieces of 3 cm square cores were cut from these. These samples were placed in a 100 ml disposable weighing boat and stored for 30 minutes at 4°C . After that, CIE (Commission Internationale de l'Eclairage) L^* values (lightness) and a^* values (redness) and reflectance (360–740 nm) of triplicate samples were measured using a spectrophotometer (CM-2500d, KONICA MINOLTA HOLDINGS INC., Tokyo, Japan). The measuring time was less than one second. Each sample was placed in the boat again, over-wrapped with oxygen-permeable PVC film, and displayed under cool white fluorescent lights (1,000–1,500 lux) at 4°C for 9 days. L^* and a^* values and reflectance of triplicate samples were obtained at display days 3, 6 and 9 using the spectrophotometer. Metmyoglobin percentages were obtained according to the method of Stewart et al. (1965).

Statistical analysis

Data were analyzed using the General Linear Model procedure of SAS (1985). Differences between breed treatment means were detected by the Student's *t* test.

RESULTS AND DISCUSSION

The BCS numbers, grades of beef brightness and overall grades of beef color of the carcasses of Japanese Shorthorn steers and Japanese Black steers are shown in Table 1. The BCS number of carcass of Japanese Shorthorn steers was significantly ($p < 0.05$) higher than that of Japanese Black steers. The grade of beef color of the carcasses of Japanese Shorthorn steers and Japanese Black steers was Grade 4 and Grade 5, respectively. The grade of beef brightness of the carcasses of Japanese Shorthorn steers (Grade 1) was significantly ($p < 0.05$) lower than that of Japanese Black steers (Grade 3). The overall grade of beef color of the carcasses of Japanese Shorthorn steers (Grade 1) was significantly ($p < 0.05$) lower than that of Japanese Black steers (Grade 3). These results suggested that the difference in the overall evaluation of beef color of the carcass of Japanese Shorthorn steers and Japanese Black steers had been caused mainly by the beef brightness.

A comparison of CIE L^* values during the display of *m. longissimus thoracis* of Japanese Shorthorn steers and

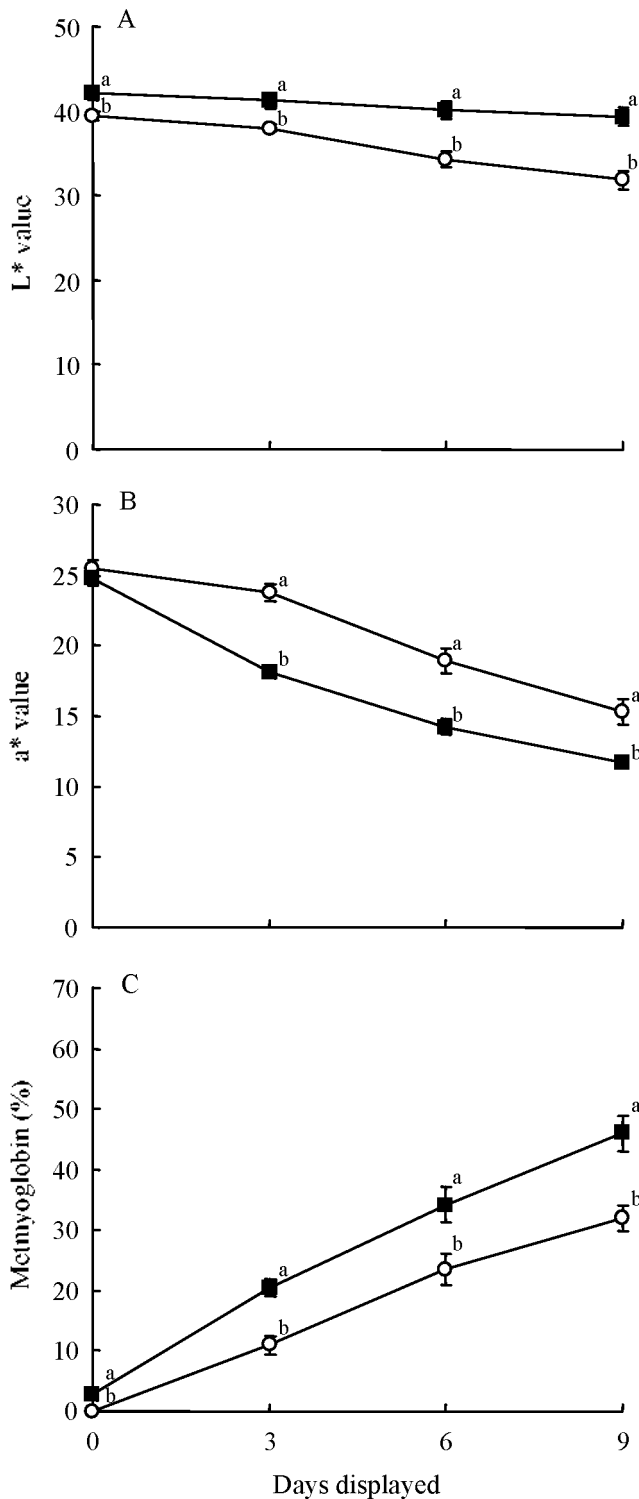


Figure 1. L* values (A), a* values (B) and metmyoglobin percentages (C) during display of *m. longissimus thoracis* of Japanese Shorthorn steers (○) and Japanese Black steers (■). Each muscle sample was over-wrapped with PVC film and displayed under fluorescent light at 4°C for 9 days. Standard error bars are indicated. ^{a, b} Values on the same display days with a different superscript letter differ ($p < 0.05$).

Japanese Black steers is shown in Figure 1A. The L* values

of *m. longissimus thoracis* of Japanese Shorthorn steers on display days 0, 3, 6 and 9 were significantly ($p < 0.05$) lower than those of Japanese Black steers. CIE a* values of *m. longissimus thoracis* of Japanese Shorthorn steers and Japanese Black steers during display are shown in Figure 1B. There was no difference ($p > 0.05$) between breeds regarding the a* value of *m. longissimus thoracis* at display day 0. However, the a* values of *m. longissimus thoracis* of Japanese Shorthorn steers on display days 3, 6 and 9 of display were significantly ($p < 0.05$) higher than those of Japanese Black steers. Comparison of L* values of *m. semitendinosus* of Japanese Shorthorn steers and Japanese Black steers during display are shown in Figure 2A. The L* value of *m. semitendinosus* of Japanese Shorthorn steers on display day 0 was significantly ($p < 0.05$) higher than that of Japanese Black steers. No differences ($p > 0.05$) were found in the L* values of *m. semitendinosus* on display days 3, 6 and 9 between breeds. The a* values of *m. semitendinosus* of Japanese Shorthorn steers and Japanese Black steers during display are shown in Figure 2B. No difference ($p > 0.05$) was found in the a* value of *m. semitendinosus* between breeds on display day 0. However, the a* values of *m. semitendinosus* of Japanese Shorthorn steers on display days 3, 6 and 9 were significantly ($p < 0.05$) higher than those of Japanese Black steers. These results suggested that the redness of *m. longissimus thoracis* and *m. semitendinosus* of Japanese Shorthorn steers during display were maintained compared to those of Japanese Black steers. However, the lightness of *m. longissimus thoracis* of Japanese Shorthorn steers during display was not maintained compared to that of Japanese Black steers. In Japan the beef color of each carcass is evaluated mainly in terms of the color of the *m. longissimus thoracis*. Therefore, the low lightness of *m. longissimus thoracis* of Japanese Shorthorn steers may be one of the reasons for that the color evaluations of the carcass and the beef of Japanese Shorthorn steers were lower than those of Japanese Black steers.

Metmyoglobin percentages during display of *m. longissimus thoracis* of Japanese Shorthorn steers and Japanese Black steers are shown in Figure 1C. The percentages of metmyoglobin of *m. longissimus thoracis* of Japanese Shorthorn steers on display days 0, 3, 6 and 9 were significantly ($p < 0.05$) lower than those of Japanese Black steers. Percentages of metmyoglobin of *m. semitendinosus* of Japanese Shorthorn steers and Japanese Black steers during display are shown in Figure 2C. The metmyoglobin percentages of *m. semitendinosus* of Japanese Shorthorn steers on display days 0, 3, 6 and 9 were significantly ($p < 0.05$) lower than those of Japanese Black steers. Green et al. (1971) reported that consumers would reject beef containing over 30-40% metmyoglobin. In this study, 30% metmyoglobin was chosen as a threshold

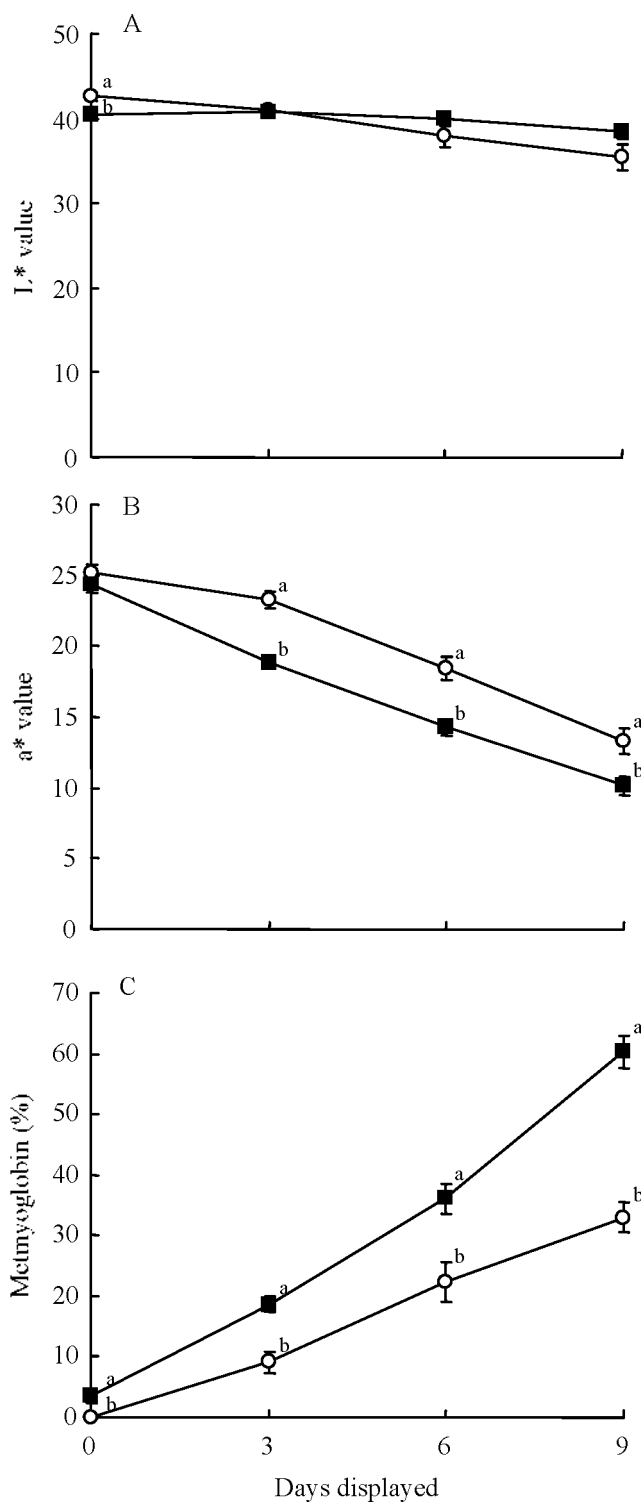


Figure 2. L* values (A), a* values (B) and metmyoglobin percentages (C) during display of *m. semitendinosus* of Japanese Shorthorn steers (○) and Japanese Black steers (■). Each muscle sample was over-wrapped with PVC film and displayed under fluorescent light at 4°C for 9 days. Standard error bars are indicated. ^{a, b} Values on the same display days with a different superscript letter differ ($p < 0.05$).

value. The metmyoglobin percentages of both *m.*

Table 2. Crude fat concentrations in two muscles of Japanese Shorthorn steers and Japanese Black steers (%)

	<i>M. longissimus thoracis</i>		<i>M. semitendinosus</i>	
	MN	SE	MN	SE
Japanese Shorthorn steers	3.3 ^b	0.4	2.2 ^b	0.2
Japanese Black steers	19.5 ^a	1.1	7.3 ^a	0.6

^{a, b} Means within a column with a different superscript letter differ ($p < 0.05$).

longissimus thoracis and *m. semitendinosus* of Japanese Black steers were beyond the threshold value on display day 6. On the other hand, the metmyoglobin percentages of both *m. longissimus thoracis* and *m. semitendinosus* of Japanese Shorthorn steers were beyond the threshold value on display day 9. The acceptable color shelf-lives of both *m. longissimus thoracis* and *m. semitendinosus* of Japanese Shorthorn steers were at least 3 days longer than those of Japanese Black steers, based on a threshold value of 30% metmyoglobin. These results suggested that beef color stability of both *m. longissimus thoracis* and *m. semitendinosus* of Japanese Shorthorn steers during display were higher than those of Japanese Black steers. It was expected that difference in myoglobin concentration in the muscles of Japanese Shorthorn steers and Japanese Black steers was one of factors causing the difference in beef color stability. However, Ito and Kondo (1983) reported that there was no difference in the myoglobin concentration in the rib muscles of Japanese Shorthorn steers and Japanese Black steers. Therefore, it was considered that the difference in the beef color stability of the muscles of Japanese Shorthorn steers and Japanese Black steers had been caused by other factors.

Oxymyoglobin and cell membrane phospholipid oxidations are closely interrelated in meat and both are responsible for quality loss as well as shelf-life reduction (Kanner and Harel 1985; Schaefer et al., 1995). This is supported by observations that products of both myoglobin oxidation and lipid oxidation increase during storage, and that the addition of antioxidants can result in a reduction of both of these deteriorative processes (Green 1969; Faustman et al., 1989). Sasaki et al. (2001) reported that lipid oxidation in longissimus muscle during storage was negatively correlated with fat content. However, Muramoto et al. (2003b) reported that in spite of an increase in the crude fat concentration in *m. longissimus thoracis* of Japanese Black steers, the beef color shelf-life during display shortened with slaughter age. Table 2 shows the crude fat concentrations in *m. longissimus thoracis* and *m. semitendinosus* of Japanese Shorthorn steers and Japanese Black steers. In this study, crude fat concentrations in both *m. longissimus thoracis* and *m. semitendinosus* of Japanese Shorthorn steers were significantly ($p < 0.05$) lower than those of Japanese Black steers. This result indicated that

Table 3. α -Tocopherol concentrations in two muscles of Japanese Shorthorn steers and Japanese Black steers ($\mu\text{g/g}$ meat)

	<i>M. longissimus thoracis</i>		<i>M. semitendinosus</i>	
	MN	SE	MN	SE
Japanese Shorthorn steers	2.6	0.2	2.9 ^a	0.2
Japanese Black steers	2.3	0.2	1.9 ^b	0.1

^{a, b} Means within a column with a different superscript letter differ ($p < 0.05$).

though the crude fat concentration in the muscle of Japanese Shorthorn steers was lower than that of Japanese Black steers, the beef color stability during the display of the muscle of Japanese Shorthorn steers was higher than that of Japanese Black steers.

Vitamin E is absorbed by animals and incorporated into cellular membranes where it performs its antioxidant function. Dietary vitamin E supplementation of steers causes accumulation of α -tocopherol in muscle tissue, which delays oxymyoglobin oxidation and prolongs the color stability of beef (Chan et al., 1995; Liu et al., 1996). Table 3 shows the α -tocopherol concentrations in *m. longissimus thoracis* and *m. semitendinosus* of Japanese Shorthorn steers and Japanese Black steers. There was no difference ($p > 0.05$) in the α -tocopherol concentration in *m. longissimus thoracis* between breeds. However, the α -tocopherol concentration in *m. semitendinosus* of Japanese Shorthorn steers was significantly ($p < 0.05$) higher than that of Japanese Black steers. Arnold et al. (1993) found that α -tocopherol level of 3.3 $\mu\text{g/g}$ is sufficient to extend color stability in the muscle of steers. Mitsumoto et al. (1991) observed that α -tocopherol concentration over 3.5 $\mu\text{g/g}$ appeared to retard metmyoglobin formation in the muscle of steers. In this study, the α -tocopherol concentrations in *m. longissimus thoracis* and *m. semitendinosus* of Japanese Shorthorn steers were 2.6 and 2.9 $\mu\text{g/g}$, respectively. Therefore, it could be considered that the α -tocopherol concentrations in both *m. longissimus thoracis* and *m. semitendinosus* did not affect the differences in the beef color stability during display of the muscles between breeds.

CONCLUSION

Though beef color evaluation of the carcass of Japanese Shorthorn steers was lower than that of Japanese Black steers, the beef color stability during the display of muscle of Japanese Shorthorn steers was higher than that of Japanese Black steers. It was considered that the difference in the beef color stability of the muscles of these breeds is caused by other factors besides concentrations of crude fat and α -tocopherol and probably the myoglobin content in muscle.

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