COMPARISON OF FREE AMINO ACID CONTENT OF KOREAN HANWOO (*BOS TAURUS COREANAE*) LOIN BEEF WITH DIFFERENT QUALITY GRADES

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Abstract – This study was conducted to compare the free amino acid (FAA) content of Korean Hanwoo (*Bos taurus coreanae*) loin beef (*M. longissimus dorsi*, LD) with different quality grades (1, 1+, and 1++). The LD sample was aged at 2ºC for 0, 7, or 14 d and then utilized for the analysis of FAA content. At 14 d of aging, the beef of QG 1 had significantly ($P < 0.05$) higher histidine, methionine, glutamine, ornithine, and total FAA contents than that of QG 1+ or 1++. During aging, the contents of some FAAs significantly ($P < 0.05$) increased in all beef of three QGs. Total FAA content showed a tendency to increase in the beef of both QG 1 and 1+. These findings suggest that QG could affect the abundance of FAAs in Hanwoo beef.

Key Words – free amino acid, quality grade, aging, Hanwoo beef.

I. INTRODUCTION

In Korea, the quality grade (QG) of beef carcass is the most important parameter that determines the commercial value of beef. This is because the price of the Korean beef is dependent on its QG: the market price of the most highly marbled Hanwoo (Korean native brown cattle, *Bos taurus coreanae*) loin beef (QG 1++) averages US $10.2 per 100 g, whereas that of QGs 1 and 3 averages US $5.7 and $3.4 per 100 g, respectively (1).

High QG greatly influences the quality of beef with high intramuscular fat (IMF) content (2, 3, 4). Lee et al. (2) reported that Hanwoo striploin of QG 1++ had lower shear force value and higher L*, a*, and b* values and sensory scores compared to that of both QG 1+ and 1. Moreover, an increase in monounsaturated fatty acids (particularly, oleic acid (C18:1n9)) and a decrease in polyunsaturated fatty acids proportions with high QG were observed in another study on Hanwoo beef (3). Thompson (4) observed high IMF content had a positive effect on flavor and juiciness scores with consumer taste panels under constant tenderness, i.e., shear force value. However, there is still little information on the effect of QG on flavor-related compounds, such as sugars, nucleotides, peptides, and free amino acids (FAAs).

Therefore, this study was conducted to compare the FAA content of Korean Hanwoo loin beef with different quality grades.

II. MATERIALS AND METHODS

A. Reagents and chemicals

Using a Milli-Q Water Purifier (Millipore SAS, Molsheim, Alsace, France), deionized water was prepared before experiments. AccQ-Tag Ultra Derivatization Kit (186003836), Eluent A Concentrate (186003838), and Eluent B Concentrate (186003839) were purchased from Waters Corporation (Milford, Massachusetts, USA). The other reagents were purchased from Sigma-Aldrich Co. LLC. (St. Louis, Missouri, USA).

B. Sample preparation and design

Fresh loins (2 d post-slaughter) from the left sides of 15 Korean Hanwoo (*Bos taurus coreanae*) steer carcasses were purchased from a local meat plant. Beef loins were allocated to three different quality grade groups (n = 5 / group): 1, 1+, and 1++. After trimming, *M. longissimus dorsi* (LD) was vacuum-packaged and then aged at 2ºC for 0, 7, or 14 d.

C. Extraction of free amino acids

FAAs were extracted from beef sample with the process reported by Tikk et al. (5). Sample was mixed with 0.6 M perchloric acid at 2,600 g for 1 min using an Ultra-Turrax (T25 Basic, Ika Werke GmbH & Co., KG, Staufen, Baden-Wüttenberg, Germany). The homogenate was centrifuged at 2°C and 5,000 g (Avanti J-E Centrifuge, Beckman Coulter, Inc., Fullerton, CA, USA) for 10 min before filtering with a Whatman filter paper No. 1. After neutralizing with both 0.8 M KOH and 0.2 M K_2CO_3, the filtrate was centrifuged, and the supernatant was filtered with a 0.20 μm syringe filter. The filtrate contained 125 μM (final concentration) norvaline as an internal standard.

D. Derivatization of free amino acids

The derivatization of FAAs was performed as described by Waters (6). Two-hundred microliters of filtrate was transferred into a vial, and then 140 μl of borate buffer was added, followed by 40 μl of AQC reagent. Then, the mixture was heated in a 55°C water bath for 10 min.

E. Ultra-performance liquid chromatography analysis

According to the Waters (6) method, FAA derivatives were analyzed using an ultra-performance liquid chromatography (UPLC; Waters Corporation, USA) equipped with an Ultra C18 column (100 mm length × 2.1 mm i.d. × 1.7 μm particle size, Waters Corporation, USA). Two microliters of derivatized sample was injected and chromatographically separated in a 55°C column at a flow rate of 0.7 ml/min using a gradient elution of (A) 5% Eluent A Concentrate and (B) 100% Eluent B Concentrate as follows: 0-0.54 min, 99.9% A/0.1% B; 0.54-5.74 min, 99.9-90.0% A/0.1-10.0% B; 5.74-7.74 min, 90.9-78.8% A/10.0-21.2% B; 7.74-8.04 min, 78.8-40.4% A/21.2-59.6% B; 8.04-8.64 min, 40.4% A/59.6% B; 8.64-8.73 min, 40.4-99.9% A/59.6-0.1% B; 8.73-11.5 min, 99.9% A/0.1% B. Ultraviolet detection was run at 260 nm. The area of the resulted chromatographic peak was quantified as mg each FAA per 100 g meat.

F. Statistical analysis

All data were statistically analyzed using the SPSS (7) program. As fixed factors, the effects of aging, quality grade, and their interaction (aging × quality grade) on FAA content were estimated by two-way analysis of variance using the general linear model. Significant differences among mean values were compared at $P < 0.05$ using the Duncan's multiple range tests.

III. RESULTS AND DISCUSSION

The comparison results of 21 each FAA content of loin beef (*M. longissimus dorsi*) from Korean Hanwoo with three different QGs during aging at 2ºC for 14 d was presented in Table 1. The QG had a remarkable impact on the contents of some FAAs. At 0 d of aging, aspartate content was significantly ($P < 0.05$) higher in the loin beef of QG 1 compared to that of QG 1+. At 7 d of aging, no significant differences observed for any FAAs among loin beef of QG 1, 1+, and 1++. At 14 d of aging, loin beef of QG 1 had significantly ($P < 0.05$) higher histidine and methionine contents than that of both QG 1+ and 1++. In addition, glutamine, ornithine, and total FAA contents (Figure 1) appeared to be higher ($P < 0.05$) in the loin beef of QG 1 than in that of QG 1++. Our results are similar to previous findings of Koutsidis et al. (8) who found that low-marbled beef (from silage-fed cattle) had higher histidine, asparagine, serine, glycine, threonine, methionine, tyrosine, phenylalanine, isoleucine, valine, leucine, and proline contents compared to high-marbled beef (from concentrate-fed cattle). In addition, Cho et al. (9) similarly reported higher contents of arginine, glycine, methionine, and leucine in Hanwoo loin of QG 1 compared to that of QG 1+.

Aging time showed a strong effect on FAA content of Hanwoo loin beef of three different QGs. Regardless of QG, serine, cystine, tyrosine, methionine, valine, isoleucine, and leucine contents significantly ($P < 0.05$) increased in Hanwoo loin beef for 14 d of aging. On the other hand, glutamine content significantly ($P < 0.05$) decreased during aging. Total FAA content did not exhibit significant differences among Hanwoo loin beef of QG 1, 1+, and 1++ but tended to increase in that of both QG 1 and 1+. These results are in agreement with a study of Koutsidis et al. (10) who observed that *post-mortem* conditioning at 4°C for 21 d increased the contents of most of FAAs. Moreover, in their study, glutamine content showed a tendency to decrease after 7 d of

conditioning. Locker (11) also found that glutamine content did not increase in aged beef.

Table 1. Comparison of each free amino acid content (mg / 100 g meat) of loin beef (*M. longissimus dorsi*) from Korean Hanwoo (*Bos taurus coreanae*) with different quality grades during aging at 2℃

Items	Aging time (d)	Quality grade		
		1	1+	1++
HIS	0	1.31	1.25	1.17
	7	1.22	1.36	1.04
	14	1.75^{a}	1.17^{b}	1.02^{b}
ASN	0	0.20	0.17	0.15
	7	0.19	0.47	0.19
	14	0.23	0.23	0.29
SER	0	3.90^{B}	2.57^{C}	3.61^{B}
	7	4.84^{AB}	4.37^{B}	5.25^{AB}
	14	6.79^{A}	6.03^{A}	6.17^{A}
GLN	0	45.07^{A}	44.68^{A}	37.23^{A}
	7	37.76^{B}	36.91^{B}	28.79^{AB}
	14	34.49^{aB}	29.15^{abC}	24.85^{bB}
ARG	0	18.22	16.40^{B}	17.33
	7	19.35	17.75^{B}	18.42
	14	22.95	21.15^{A}	18.65
GLY	0	5.30	4.71	5.30^{A}
	7	4.39	3.59	3.88^{AB}
	14	3.92	3.98	3.39^{B}
ASP	0	0.79^{a}	0.62^{bB}	0.68^{ab}
	7	0.87	0.83^{A}	0.69
	14	1.00	0.88^{A}	0.72
GLU	0	11.88	11.21	12.68
	7	12.77	11.39	12.43
	14	15.86	14.25	11.73
THR	0	2.12^{B}	1.93^{C}	2.11
	7	2.34^{AB}	2.23^{B}	2.38
	14	2.71^{A}	2.54^{A}	2.33
ALA	0	15.91^{B}	15.02	15.74
	7	16.82^{B}	15.79	15.54
	14	19.65^{A}	17.86	15.69
PRO	0	3.64	3.34^{B}	3.71
	7	4.00	3.69^{B}	4.02
	14	4.66	4.32^{A}	3.76
ORN	0	1.49	1.42	1.44
	7	1.49	1.43	1.44
	14	1.58^{a}	1.48^{ab}	1.42^{b}
CYS	0	0.21^{C}	0.17^{C}	0.21^{B}
	7	0.41^{B}	0.42^{B}	0.49^{A}
	14	0.64^{A}	0.54^{A}	0.47^{A}
LYS	0	7.33^{B}	6.42^{B}	7.76
	7	8.77^{B}	7.85^{B}	8.98
	14	11.48^{A}	10.20^{A}	8.42
TYR	0	4.12^{B}	3.75^{C}	4.03^{B}
	7	4.59^{AB}	4.45^{B}	4.74^{A}
	14	4.93^{A}	4.98^{A}	4.73^{A}
MET	0	2.39^{B}	2.37^{B}	2.36^{B}
	7	2.57^{B}	2.66^{A}	2.68^{A}
	14	3.01^{aA}	2.69^{bA}	2.65^{bA}
VAL	0	3.69^{B}	2.81^{C}	3.51^{B}
	7	4.99^{AB}	4.24^{B}	5.13^{A}
	14	6.86^{A}	5.76^{A}	5.09^{A}
ILE	0	1.97^{B}	1.69^{C}	1.91^{B}
	7	2.32^{B}	2.19^{B}	2.54^{A}
	14	2.91^{A}	2.70^{A}	2.50^{A}
LEU	0	7.06^{B}	5.41^{C}	6.71^{B}
	7	8.77^{B}	8.11^{B}	9.56^{A}
	14	11.53^{A}	10.30^{A}	9.41^{A}
PHE	0	4.02^{B}	2.92^{C}	3.88^{B}
	7	5.06^{B}	4.75^{B}	5.73^{A}
	14	7.07^{A}	6.25^{A}	5.70^{A}
TRP	0	0.41^{B}	0.40	0.45
	7	0.48^{AB}	0.49	0.47
	14	0.55^{aA}	0.46^{ab}	0.45^{b}

$^{a-c}$Means in the same row with different superscripts differ significantly ($P < 0.05$).

$^{A-C}$Means in the same column with different superscripts differ significantly ($P < 0.05$).

[1]HIS: histidine; ASN: asparagine; SER: serine; GLN: glutamine; ARG: arginine; GLY: glycine; ASP: aspartate; GLU: glutamate; THR: threonine; ALA: alanine; PRO: proline; ORN: ornithine; CYS: cystine; LYS: lysine; TYR: tyrosine; MET: methionine; VAL: valine; ILE: isoleucine; LEU: leucine; PHE: phenylalanine; TRP: tryptophan.

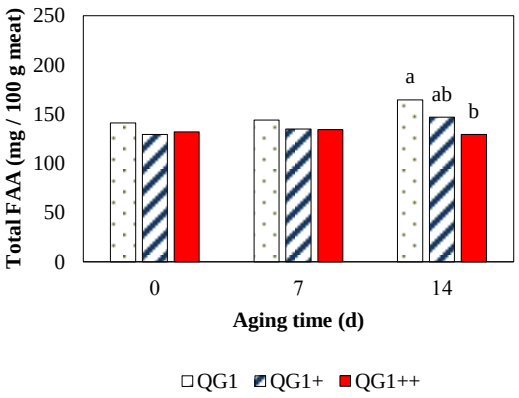

Figure 1. Comparison of total free amino acid (FAA) content (mg / 100 g meat) of loin beef (*M. longissimus dorsi*) from Korean Hanwoo (*Bos taurus coreanae*) with different quality grades (QGs) during aging at 2℃. These values are means. $^{a-b}$Different letters indicate significant differences among different quality grades within the same aging time ($P < 0.05$).

IV. CONCLUSION

This study compared the FAA content of Korean Hanwoo loin beef with three different QG during aging at 2℃ for 14 d. The beef of low QG had higher contents of some FAAs and total FAA compared to that of high QG. In all beef of three QG, the contents of some FAAs commonly increased during aging, while glutamine content decreased.

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