Effects of Dry- and Wet-ageing on Flavor Compounds and Eating Quality of Low Fat Hanwoo Beef Muscles

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Abstract The present study aimed at investigating the effects of dry- and wet-aging methods on flavor compounds and sensory properties of low fat Hanwoo beef muscles. All the beef samples were obtained from 2-grade carcasses of Hanwoo cows. The beef samples used in the dry- and wet-aging methods were prepared in the forms of quarter beef (bone-in) and cuts (boneless), respectively. The dry-aging was carried out at 2℃–4℃ and humidity of 65%–85%, while the wet-aging was done at 1℃ for 0, 20, 40, and 60 d. At each aging time, three muscles: longissimus thoracis (LT), glutaeus medius (GM) and semimembranosus (SM) were taken from the corresponding quarters and cuts, and used for the flavor compounds and sensory analyses. Results showed that both aging methods significantly increased the concentrations of flavor compounds and total amount of all classes of the flavor compounds as the aging time increased (p<0.05). In the dry-aging method, the GM and SM muscles presented significantly higher total amounts of pyrazines and sulfur-containing compounds compared to the LT muscle (p<0.05). Both the aging methods improved the eating quality attributes, indicating by increased scores of sensorial attributes with increased aging time for all the muscles studied (p<0.05). However, compared to the wet-aging, the dry-aging method resulted in significantly higher scores of tenderness and flavor for the GM and SM muscles after 40 to 60 d. Thus, postmortem aging, especially the dry-aging method could be used to improve eating quality attributes (tenderness and flavor) of low fat beef muscles such as GM and SM.

Keywords dry-aging, wet-aging, flavor, eating quality, Hanwoo beef

Introduction

According to the statistical data, per capita consumption of beef in Korea is steadily increasing from 8.8 kg in 2010 to 11.3 kg in 2017 (MAFRA, 2018). In a recent survey by Hanwoo Meat Consumption-Distribution Monitoring (Hanwoo Board, 2016) showed that Korean consumers much prefer highly-marbled beef cuts rather than cuts with low fat content, and the consumption level largely varied among cuts for instance, loin (33.2%), brisket (27.6%), tenderloin (12.2%), and rib (6.5%). As a result, the domestic
beef producers and suppliers must face a lot challenges arising from fundamental imbalance between the consumption demand and production capacity for the highly-marbled beef cuts, as well as the redundancy of low fat cuts which account for a large proportion in each carcass. Therefore, it is necessary to find out suitable solutions to solve this problem for instance, by applying postmortem aging techniques to improve eating quality and reduce the variations in quality among beef cuts (Cho et al., 2016).

It is well recognized that postmortem aging produces beef that is naturally tender and flavorful (Huff-Lonergan and Lonergan, 2005; Kemp et al., 2010; Van Ba et al., 2017). In the meat industry, two the most commonly-applied aging methods which are wet-aging and dry-aging. The nature of wet-aging is placing beef primal and/or sub-primal cuts into plastic bags which are vacuum-sealed and stored in refrigerated temperature (Smith et al., 2008). For decades, wet-aging is the most predominantly used method in the beef industry, and its advantageous points are increased juiciness and tenderness due to retained meat’s moisture (Kim et al., 2019; Park et al., 2015; Van Ba et al., 2017) and flexibility of storage (Smith et al., 2008). While, the dry-aging is a traditional aging method, the nature of this method is hanging beef carcasses or quarter and primal cuts in a cold room without any protective packaging materials (Savell, 2008). Previous studies reported that during the dry-aging process, moisture is drawn out, which results in a beefier, more flavorful and tender beef (Berger et al., 2018; Li et al., 2014).

More to the point, the postmortem aging causes an increased muscle protein breakdown by endogenous proteases, resulting in formations of small peptides and free amino acids which are the important flavor precursors (Dashdorj et al., 2013; Koutsidis et al., 2008; Reina et al., 2014). During cooking, these precursors (e.g., amino acids) react with other components (e.g., reducing sugars) to produce a variety of volatile aroma compounds which subsequently contribute to the typical flavor characteristics of cooked beef (Calkins and Hodgen, 2007; Elmore et al., 2002; Mottram 1994; Van Ba et al., 2013).

Although the postmortem aging generally improves eating quality typically the flavor characteristics of beef as mentioned above, also the huge contributions of volatile aroma compounds to the cooked beef flavor development have been demonstrated (Elmore et al., 2002; Mottram 1998; Stetzer et al., 2008), there has been no published research detecting and identifying the volatile aroma compounds in dry-aged beef. Taken together, we hypothesized that type of aging methods (wet-aging and dry-aging) may cause different effects on the generations of flavor precursors (e.g., free peptides, amino acids and fatty acids) which subsequently affect the quality and quantities of volatile flavor compounds as well as flavor characteristics of the cooked beef products. Therefore, the objective of this study was to investigate the effects of wet- and dry-aging methods on the volatile flavor compounds and eating quality of low fat content Hanwoo beef muscles.

**Materials and Methods**

**Samples preparation and aging treatment**

In the present study, beef samples obtained from 2-grade carcasses of Hanwoo cows (34–60 months old) were used for the aging treatment. The cattle were slaughtered in a slaughterhouse in Chungbuk (Korea) under the commercial slaughtering process. After chilling for 24 h, hindquarters and bone-in loins were obtained from both carcass sides and used for the dry-aging. While, three cuts (boneless): Loin, top-round and rump obtained from the carcasses were used for the wet-aging. All the samples were then transported to the meat pilot plant of National Institute of Animal Science (NIAS, Jeonju, Korea) under cooling condition. For the dry-aging treatment, the samples were aged by direct exposure to the aging environment (2–4±0.5°C and 65%–85% humidity) without any package. The samples assigned to the wet-aging were individually vacuum-
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The aging was carried out under the controlled conditions (as shown in Table 1) in the aging rooms where the samples were kept on stainless steel gratings for 0, 20, 40, and 60 d.

At the end of each aging period, three muscles: *Longissimus thoracis* (LT), *semimembranosus* (SM) and *gluteus medius* (GM) (n=3 each) were taken from the corresponding bone-in loins and hindquarters (in the dry-aging) or from loin, top-round and rump (in the wet-aging), respectively. The muscles were trimmed off all visual fats and connective tissues, and then cut into sub-samples depending on analyses.

### Analysis of volatile flavor compounds

At each aging period, the dry-and wet-aged beef muscle samples were subjected to the volatile flavor compounds analysis. The sample preparation, extraction of flavor compounds and separation conditions used were same as those described in previous study (Van Ba et al., 2010). Particularly, about 1.0 g of cooked sample was taken and placed into a 20-mL headspace vial (Part No. 5188-2753, Agilent, USA) and 1.0 µL of internal standard (2-methyl-3-heptanone, 816mg /mL in methanol) was also added. The vial was then sealed with PTFE-faced silicone septum for extraction. The extraction of volatile flavor compounds was done using solid-phase micro-extraction (SPME). A SPME device containing carboxen–polydimethylsiloxane (75 µm) fiber (Supelco) was used for extraction of the compounds. All steps such as; extraction, absorption, desorption of the flavor compounds were done using a fully automated SPME sample preparation instrument (Model: AOC-5000 Plus) connected to Gas Chromatography (Model: 7890B GC, Agilent Technologies) with Mass Spectrophotometry (Model: 5977B MSD, Agilent Technologies). The extraction was carried out at 65°C and agitated at 250×rpm for 60 min. The GC/MS conditions set were same as those mentioned in the above cited literature. Identifications of volatile compounds were performed by comparing their mass spectra with those already present in the Wiley registry of mass spectral data (Agilent Technologies) and/or by comparing their retention times with those of authentic external standards. Approximated quantities of the volatile compounds were quantified by comparison of their peak areas with that of the internal standard obtained from the total ion chromatogram using a response factor of 1.

### Sensory evaluation

The sensory evaluation was carried out in a test room of Animal Product Utilization Division (NIAS), using the method as described by Cho et al. (2016) with minor modifications. The sensory evaluation procedure was approved by the Institutional Review Board of National Institute of Animal Science (No.11-1390744-000007-01). The panel consisted of 7 trained members (at 26 to 40 years old). Prior to use, the vacuum-packaged sub-sample blocks were removed from a freezer (–20°C) and thawed for 4 h in a 4°C cooler, and then were sliced into 4-mm thick slices using a meat slicer. For each the sample, 7 representative slices (50×75×4 mm) were finally made and chosen for sensory evaluation. Each session had 7 panelists; each

| Table 1. Aging conditions for Hanwoo cow beef muscles |
|-----------------------------|-----------------|----------------|
| Aging method               | Conditions       | Sample collection days (d) |
| Dry-aging                   | 2°C and 65% humidity (20 d) | 0, 20, 40, 60 |
|                             | 2°C and 75% humidity (20 d) |                      |
|                             | 4°C and 85% humidity (20 d) |                      |
| Wet-aging                   | 1°C             | 0, 20, 40, 60 |
panelist evaluated 7 samples, and two sessions per day were carried out. The slices were cooked on an open tin-coated grill for approximately 2 min and turned 30 s intervals. The cooking temperature was monitored using an infrared thermometer and was maintained at around 130°C–140°C. One set of grill was used where the grill was set to cook 7 slices of each sample. Immediately after cooking, the samples were placed on individual dishes and served to the panelists. The panelists then handled the cooked samples with an approved odorless plastic fork and tasted for flavor, juiciness, tenderness and overall acceptability using a 6-points scale as described by Meilgaard et al. (1999). In which, tenderness score ranged from 1 (very tough) to 6 (very tender), juiciness score ranged from 1 (very dry) to 6 (very juice), flavor and overall liking scores ranged from 1 (extremely dislike) to 6 (extremely like).

Statistical analysis

The results of this experiment were analyzed using the Statistic Analysis System (SAS) package (SAS Institute, Cary, NC, USA, 2010). Significance among the treatments was verified at the 5% level by Student-Newman-Keul multiple test. Analysis of the heat map was made using R-Studio (http://www.rstudio.com) version 1.1.453.

Results and Discussion

Effects of aging method on volatile flavor compounds

The volatile compounds formed in meat during cooking are the main components responsible for the development of cooked meat flavor characteristics (Mottram, 1998). To date, approximately thousands of volatile compounds with various flavor notes, have been detected and identified in cooked meat (Mottram, 1994; Aaslyng and Meinert, 2017). These compounds are usually derived from the flavor precursors such as, fatty acids, free amino acids, reducing sugars etc. present in the raw meat through the Maillard reactions and oxidation/degradation during cooking/heating process (Aaslyng and Meinert, 2017; Mottram, 1998; Van Ba et al., 2013). In the present investigation, the changes in the concentrations of the frequently-detected flavor compounds in cooked beef muscles by different aging methods and periods are presented in Figs. 1, 2, and 3. It was observed that most of volatile compounds increased in their concentrations with increased aging time for all the muscles (LT, GM, and SM) and aging methods. However, compared to the wet-aging, all the dry-aged muscles presented significantly higher concentrations of all the compounds on all days examined. Among 12 aldehydes identified, hexanal, heptanal, octanal, nonanal and 17-octadecanal were the most predominant compounds in the cooked LT (Fig. 1), GM (Fig. 2) and SM muscle (Fig. 3) throughout the aging periods.

Hexanal is known to be formed from the oxidation/degradation of C18:2n-6 while, heptanal, octanal and nonanal are derived from C18:1n-9 (Elmore et al., 2002; Van Ba et al., 2013). This result could be attributed to the increased C18:1n-9 and C18:2n-6 degradation levels resulting from lipolysis during aging, with higher rate in the dry-aged muscles compared to the wet-aged ones. Researchers have reported that aldehydes are the important lipid-derived flavor compounds which significantly contribute to meat flavor due to their low odor-detection threshold (Elmore et al., 1999). Particularly, hexanal contributes positively to meat flavor but it may also produce undesirable flavors at higher concentrations (Calkins and Hodgen, 2007). While, octanal and nonanal with their pleasant odor notes (e.g., fatty-sweet-green-oily) are important for the cooked-beef flavors (Rochat and Chaintreau, 2005; Specht and Baltes, 1994). Furthermore, the total amount of aldehydes (averaged from all the aging periods) in the cooked beef muscles are presented in Fig. 4A. All the dry-aged muscles (except for SM) had significantly (p<0.05) higher total amount of aldehydes compared to those aged by the wet-aging method. Under
the same dry-aging condition, the LT muscle exhibited significantly higher total amount of aldehydes, whereas under the wet-aging condition this muscle showed a lower total amount compared to the GM and SM muscles (p<0.05).

Regarding alcohols, 1-pentanol, 1-octen-3-ol and 1-octanol were the most frequently-found compounds in the cooked beef muscles in which the dry-aged samples generally had higher amounts compared to the wet-aged ones (Fig. 1–3). As a result, the total amount of alcohols was also significantly (p<0.05) higher in all the dry-aged muscles (except for MS) compared to the wet-aged ones (Fig. 4B). Similar to the results observed for the aldehydes, the LT muscle aged by the dry-aging method presented significantly higher total amount of alcohols, whereas it showed a lower amount compared to the other remaining muscles when aged by the wet-aging method (p<0.05). Alcohols partly contribute to the cooked meat flavors due to their low odor-detection threshold (Sabio et al., 1998). Amongst, 1-octanol, and 1-pentanol and 1-octen-3-ol are known to be formed from C18:1n-9 and C18:2n-6 fatty acids, respectively (Van Ba et al., 2013).
Pyrazines, the products of Maillard reaction between amino acids and reducing sugar, have been reported to significantly contribute to the roasted odor of cooked meat (Aaslyng and Meinert, 2017; Mottram, 1998). Five pyrazines including: pyrazine, methylpyrazine, 2,5-dimethylpyrazine, 2-ethylpyrazine and 2,5-dimethyl-3-ethylpyrazine were frequently detected in the cooked beef muscles aged by the dry-and wet-aging methods (Figs. 1–3). Compared to the lipids-derived compounds such as aldehydes, pyrazines were found at lower concentrations, which agrees with finding of Van Ba et al. (2017). We also found that the concentrations of all the pyrazines in all the muscles increased with increased aging time in both aging methods, however, the dry-aging resulted a greater amount compared to the wet-aging. Similarly, total amount of pyrazines was significantly higher in all the dry-aged muscles compared to those aged by the wet-aging method (Fig. 5A). In contrast to the results obtained for the lipids-derived flavor compounds, the total amount of pyrazines was significantly higher in the GM and SM muscles compared to the LT muscle when aged under the same dry-aging condition.

Nitrogen- and sulfur-containing heterocyclic compounds are known to be the products of Maillard reaction between amino
acids and reducing sugar, the compounds have been shown to contribute to the ‘meaty’ and ‘onion’ flavors in cooked meat products (Benet et al., 2015; Mottram, 1998; Thomas et al., 2014). In the present study, the most frequently-found sulfur-and nitrogen-containing compounds in the aged beef muscles were dimethyldisulfide and 2-acetylthiazole, respectively (Figs. 1–3). Regarding the total amounts of sulfur-and nitrogen-containing compounds, it was observed that the GM muscle exhibited significantly (p<0.05) higher amount compared to the LT and SM muscle in both aging methods (Fig. 5C). In general, the dry-aging method resulted in higher total amounts of sulfur-containing compounds for all the muscles, however, significant (p<0.05) difference only was found for the LT muscle. Type of aging method also affected the total amount of nitrogen-containing compounds in which the muscles aged by the dry-aging method presented significantly (p<0.05) higher amounts compared to those aged with wet-aging method (Fig. 5D).

Fig. 3. Heat map representing the color-coded concentrations of volatile flavor compounds of SM muscle aged by dry- (A) and wet- (B) aging methods at different aging periods (0, 20, 40, and 60 d). SM, M. semimembranosus.
Fig. 4. Total amount (averaged from 0, 20, 40, and 60 d aging) of flavor class: aldehydes (A), alcohols (B), hydrocarbons (C) and ketones (D). ST, M. Longissimus thoracis; MG, M. glutaeus medus; SM, M. semimembranosus.

Fig. 5. Total amount (averaged from 0, 20, 40, and 60 d aging) of flavor class: pyrazines (A), furans (B), sulfur-containing compounds (C) and nitrogen-containing compounds (D) during aging periods (0, 20, 40, and 60 d). ST, M. Longissimus thoracis; MG, M. glutaeus medus; SM, M. semimembranosus.
Overall, during the postmortem aging, a substantial change in the concentrations of flavor precursors (e.g., small peptides, free amino acids and free fatty acids) can be expected due to the proteolytic and lipolytic activities by endogenous enzymes, which results in an increased amount of Maillard reaction-derived products (e.g., pyrazines, sulfur-and nitrogen-containing compounds) and lipids-derived compounds (aldehydes) during cooking (Aaslyng and Meinert, 2017). Unfortunately, levels of these flavor precursors were not determined in the present study. The results indicating higher concentrations of flavor compounds in the dry-aged beef muscles compared to the wet-aged ones, although we cannot yet offer a satisfactory explanation for this results, could be attributed to: (i) the greater levels of flavor precursors and, (ii) the significant evaporation and moisture loss induced the meat becomes more concentrated after dry aging (Li et al., 2014).

**Effects of aging method on beef eating quality**

Eating quality attributes, specifically tenderness and flavor are the most important factors affecting the purchasing decision by consumers for beef (McCathy et al., 2017). To date, the substantial improvement in beef eating quality due to the postmortem aging treatment (e.g., wet-and dry-aging) has been well recognized and widely reported in literature, however, most of previously-published works only the longissimus dorsi or longissimus lumborum was used as the representative muscle sample (Berger et al., 2018; Kim et al., 2016; Li et al., 2014). Whereas, the other remaining commercial beef muscles have not extensively been studied. In the present investigation, the LT, GM and SM muscles obtained from low grade Hanwoo carcasses were used to elucidate whether their eating quality can be improved during postmortem aging. The effect of type of aging method on the eating quality attributes of all three muscles studied is presented in Table 2.

Regarding tenderness, both aging methods produced a beneficial effect on tenderness improvement, indicating by increased scores for all the muscles as prolonging the aging time. However, the wet-aging method apparently resulted in a faster tenderization rate compared to the dry-aging, indicating by significantly (p<0.05) higher scores for the muscles (e.g., GM and SM) just after 20 d. While, the dry-aging method improved the tenderness after up to 40 to 60 d aging. This result could be related to the differences in the sample size/form and aging conditions. The effect of type of aging method only was observed for the GM muscle particularly, this muscle showed significantly (p<0.05) higher scores in the dry-aging method than in the wet-aging method after 40 and 60 d. Similarly, Kim et al. (2016) also reported no differences in tenderness scores between dry-aging and wet-aging for beef longissimus lumborum muscle.

For juiciness, both the aging methods led to significant increases in the scores for all the muscles studied. This finding agrees with that of Li et al. (2014) and Berger et al. (2018), who showed that both dry-and wet-aging methods improved juiciness of beef longissimus muscle as aging time increased.

Previous studies have reported that the improvement in flavor characteristic is one of the most typically advantageous points of the dry-aging method (Corbin et al., 2015; Kim et al., 2016; Savell, 2008). Our result showed that both the aging methods considerably affected the flavor of all the muscles studied (except LT muscle). Particularly, the GM and SM muscles aged with the dry-aging method had significantly higher flavor scores compared to those aged with the wet-aging method after 20 or 40 and 60 d (p<0.05), which agrees will with finding of Kim et al. (2016). These obtained results could be attributed to the significantly higher concentrations of flavor compounds (e.g., aldehydes, pyrazines, nitrogen-and sulfur-containing compounds etc.) in these dry-aged muscles (Fig. 4 and 5). Additionally, because of a greater moisture loss caused by evaporation during the dry-aging time (Li et al., 2014), the dry-aged muscles become more concentrated and flavorful. In contrast to the tenderness and juiciness results, the dry-aging method significantly increased the flavor scores of GM and SM muscles just after 20 to 40 d aging, whereas it took up to 60 d in the wet-aging method. Data similar to ours study for
flavor were reported by Savell (2008), who showed that dry-aging of beef for up to 30 d increases the flavor score (Li et al., 2014). Finally, both the aging methods significantly (p<0.05) increased the overall acceptability scores for all the muscles after 20 to 40 d aging; however, no differences were found for the scores between the dry- and wet-aged muscles (p>0.05).

**Conclusion**

The present study, for the first time detected and identified a variety of volatile flavor compounds in various beef muscles aged under dry-aging condition and compared with those aged with wet-aging method. The data from the present study...
indicated that both aging methods resulted in increases in concentrations of most of flavor compounds with increased aging time for all the beef muscles. However, compared to the wet-aging the dry-aging method resulted in significantly higher concentrations of the flavor compounds as well as total amounts of all classes of flavor compounds in the muscles. Furthermore, under the same dry-aging condition, the total amounts of pyrazines and sulfur-containing compounds associated with pleasant flavor notes were significantly higher in the GM and SM muscles compared to the LT muscle. Aging for 40 to 60 d generally improved the eating quality attributes of all the muscles, especially the dry-aging method resulted in the tenderer and more flavorful GM and SM muscles. Thus, from the results obtained in the present study, it may be concluded that postmortem aging especially the dry-aging method could be used to improve eating quality attributes of beef muscles (e.g., GM and SM) from the low quality grade carcasses. Further study in characterizing more descriptive flavor and odor attributes of the aged beef muscles by using a larger number of sensory panel, and detecting the changes in flavor precursors is necessary.

**Conflict of Interest**

The authors declare no potential conflict of interest.

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**Author Contributions**

Conceptualization: Ha Y, Cho S. Data curation: Ha Y. Formal analysis: Ha Y. Methodology: Ha Y, Van Ba H, Ryu S, Kim Y. Software: Kim Y. Validation: Hwang I, Cho S. Investigation: Kang S, Kim Y. Writing - original draft: Ha Y. Writing - review & editing: Ha Y, Hwang I, Van Ba H, Ryu S, Kim Y, Kang S, Kim J, Kim Y, Cho S.

**Ethics Approval**

This article does not require IRB/IACUC approval because there are no human and animal participants.

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