

Analysis of low-marbled Hanwoo cow meat aged with different dry-aging methods

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Objective: Different dry-aging methods [traditional dry-aging (TD), simplified dry-aging (SD), and SD in an aging bag (SDB)] were compared to investigate the possible use of SD and/or SDB in practical situations.

Methods: Sirloins from 48 Hanwoo cows were frozen (Control, 2 days postmortem) or dry-aged for 28 days using the different aging methods and analyzed for chemical composition, total aerobic bacterial count, shear force, inosine 5'-monophosphate (IMP) and free amino acid content, and sensory properties.

Results: The difference in chemical composition, total aerobic bacterial count, shear force, IMP, and total free amino acid content were negligible among the 3 dry-aged groups. The SD and SDB showed statistically similar tenderness, flavor, and overall acceptability relative to TD. However, SDB had a relatively higher saleable yield.

Conclusion: Both SD and SDB can successfully substitute for TD. However, SDB would be the best option for simplified dry-aging of low-marbled beef with a relatively high saleable yield.

Keywords: Low-marbling; Hanwoo Cow Meat; Dry-aging Methods; Sensory Property

INTRODUCTION

Consumers' concern on health has led increasing consumption of low-marbled beef [1,2]. However, it conflicts with consumer preferences as marbling has a positive impact on eating quality attributes of beef, including tenderness, flavor, and juiciness [3,4]. Therefore, various attempts have been made to improve the palatability of low-marbled beef, using mechanical (tender stretch, tender cut, and wrapping) and physicochemical (high hydrostatic pressure, ultrasound treatment, and marination and/or injection) methods [5]. However, except for marination and/or injection, most of the other methods are focused on enhancing the tenderness of beef.

Aging is another approach used to improve the tenderness, flavor, and juiciness (water holding capacity) of raw beef and has been recently applied to low-marbled products [6,7]. There are two types of aging methods: wet- and dry-aging. Wet-aging involves vacuum-packaging of the beef followed by its storage at refrigeration temperature during the aging period, whereas traditional dry-aging (TD) involves exposure of the beef to a controlled temperature, relative humidity (RH), and air flow [8]. The TD should be strictly controlled in specialized facilities and with specialized techniques; therefore, these requirements have been an obstacle to popularizing dry-aged beef in the market, in addition to its risks of saleable yield and/or microbial safety. Even though wet- and dry-aging processes can equally enhance the tenderness of beef [9], there has been increasing demand for TD owing to its better ability to maintain a highly concentrated beefy flavor [10-12]. Furthermore, aging bags that allow high water vapor permeability have been suggested to reduce the economic losses and/or microbial contamination that result from atmospheric exposure during

the aging period [13].

There has been an effort to apply TD without the requirements of special facilities and techniques, and as a result, a newly developed appliance (an ordinary refrigerator with a built-in temperature and humidity controller) was introduced as a simplified dry-aging (SD) method. However, no scientific data have been provided for the SD method, including data on microbial safety. Therefore, in this study, different dry-aging methods (TD, SD, and SD within an aging bag [SDB]) were compared to investigate the possible use of SD and/or SDB for improving the palatability of low-marbled beef.

MATERIALS AND METHODS

Raw materials and dry-aging

A total of 48 sirloins were obtained from approximately 48-month-old Hanwoo cows (quality grade 2). The samples were transferred in a cooler (4°C) to the Korea Institute for Animal Products Quality Evaluation (Sejong, Korea). The initial pH of all samples was measured (average 5.48 ± 0.12) prior to the dry-aging process. As a control, Hanwoo cow meat was assigned on 2 days postmortem, vacuum-packed, and immediately frozen to -70°C ($n = 24$). The other 24 sirloins ($n = 8$ for each treatment) were dry-aged for 28 days with the different aging methods: i) TD (temperature, $1 \pm 1^\circ\text{C}$; RH, approximately 85%; air, 5 ± 3 m/s in a specialized facility); ii) SD (temperature, $2 \pm 1^\circ\text{C}$; RH, approximately 75% in the newly developed appliance); and iii) SDB (the same condition as SD except for the aging bag [Drybagsteak LLC, Minneapolis, MN, USA]: water vapor permeability of 8,000 g [$15 \mu\text{m}^2/24$ h and O_2 permeability of 2.3 mL/ m^2/d at 38°C and 50% RH]). The SDB group was vacuum-packed (HFV-600L, Hankook Fujee Co., Ltd., Siheung, Korea) to ensure that the surface of the bag was in contact with the samples. After aging, the dried surfaces (crust) of all dry-aged samples were trimmed off, and the sirloins were then vacuum-packed and frozen to -70°C for further analyses.

Chemical composition

Ground meat (200 g) was prepared in a sample cup (FOSS, Hillerød, Denmark) and its chemical composition (moisture, fat, protein, and collagen contents) was determined using the Food-Scan Lab meat analyzer (FOSS, Denmark) based on official methods of analysis of AOAC [14].

Microbial analyses

A sample of the meat (5 g) was blended in sterile saline (45 mL, 0.85%) for 2 min using a laboratory blender (BagMixer 400 P, Interscience, St. Nom la Bretèche, France). Each dilution (100 μL) was spread on plate count agar (Difco Laboratories, Detroit, MI, USA), YM agar (Difco Laboratories, USA), and eosin methylene blue agar (Difco Laboratories, USA) for enumeration of total aerobic bacteria, mold/yeast, and coliforms, respectively. The agar

plates for total aerobic bacteria and coliforms were incubated at 37°C for 48 h, whereas the YM plates were incubated at 25°C for 120 h. After incubation, the microbial counts were calculated and expressed as log colony-forming unit (CFU)/g.

Warner-Bratzler shear force

The meat sample was vacuum-packed (HFV-600L, Hankook Fujee Co., Ltd., Korea) and boiled in a water bath until a core temperature of 72°C was reached. The cooked samples were then cut parallel to the muscle fiber into cuboidal subsections (2.54 cm height) and placed under a Warner-Bratzler shear probe, perpendicularly to the muscle fiber. Shear force (N) was measured using a texture analyzer (TMS-Touch, Food Technology Co., Sterling, VA, USA) with a cell load of 0.1 N and a cross-head speed of 400 mm/min.

Inosine 5'-monophosphate and free amino acid contents

Inosine 5'-monophosphate (IMP) was extracted from the samples according to the method of Lee et al [15] with a few modifications. The extract was filtered through a membrane filter (0.2 μm ; Whatman PLC., Kent, UK) into a glass vial and injected into a high-performance liquid chromatography (HPLC; Ultimate 3000, Thermo Fisher Scientific Inc., Waltham, MA, USA) system. The analytical conditions were as follows: injection volume, 10 μL ; mobile phase, 20 mM potassium phosphate monobasic (pH 5.5); flow rate and time, 1.0 mL/min for 25 min; column, Synergi Hydro-RP (250×4.6 mm², 4 μm particles; Phenomenex Inc., Seoul, Korea) at 30°C ; and detector, UV/Vis detector at 254 nm. The peak area was calculated from a standard curve obtained using a standard IMP (Sigma-Aldrich, St. Louis, MO, USA).

For free amino acid content, the samples were prepared according to the method of Lee et al [15] and injected into a HPLC (S 1125, Sykam GmbH, Eresing, Germany) system with post-column derivatization (Pinnacle PCX derivatization instrument, Pickering Laboratories, Mountain View, CA, USA). The analytical conditions were as follows: mobile phase, buffers A, B, C, and D (Ajoo Scientific, Gunpo, Korea); column, 4.6×150 mm² (Sykam GmbH, Germany) at 25°C ; and detector, UV/V is detector at 540 nm. The standard (Sigma-Aldrich, USA) was used to generate a standard curve for calculation of the peak area.

Sensory evaluation

Sensory evaluation was conducted by a consumer panel (total 30 panelists) to observe changes in the low-marbled Hanwoo cow meat after dry-aging and differences among the different dry-aging methods (TD, SD, and SDB). Each sample was cut into a similar size ($50 \times 20 \times 6$ mm³, length \times width \times height), grilled until the core temperature reached to 72°C , and served to the panelists. A 7-point hedonic scale (1, extremely dislike; 7, extremely like) was used to score juiciness, tenderness, flavor, and overall acceptability before and after dry-aging.

Statistical analysis

A randomized incomplete block design was applied, using the trial as the block. The control ($n = 12$, 24 sirloins per 2 trials) and the 3 different dry-aging methods ($n = 4$ for each treatment, 8 sirloins per 2 trials) were assigned, and the model was analyzed with the fixed effect (aging method) and the random effect (carcass and side of the carcass). Calculations based on the general linear model were performed using SAS 9.3 (SAS Institute Inc., Cary, NC, USA) and the results were reported as mean values with standard error of the means. Significant differences among the mean values were determined on the basis of the Student-Newman-Keuls multiple comparison test at a level of $p < 0.05$.

RESULTS AND DISCUSSION

Chemical composition

Differences in chemical composition were found between the control and the dry-aged groups (Table 1). After 28 days of dry-aging, the moisture content in the 3 dry-aged groups was significantly lower than that in the control. This typical result from the dry-aging process is attributable to moisture evaporation [10,11,16], and is important for concentrating the flavor components in dry-aged beef [17]. As the RH can promote moisture evaporation during the dry-aging process [18], SD and SDB groups (RH, approximately 75%) were expected to have a lower moisture content than TD group (RH, approximately 85%). However, no significant difference in moisture content was detected among the 3 dry-aged groups. This was probably due to moisture evaporation in TD, promoted by air flow (5 ± 3 m/s), whereas the bag used in SDB did not interfere with moisture evaporation as it was highly water vapor permeable ($8,000 \text{ g}/15 \mu\text{m}^2/24 \text{ h}$). This phenomenon is noteworthy, given the possibility of applying SD and SDB instead of TD for aging beef at similar rate of dry-aging. The fat and protein contents showed an increasing trend during the 28 days of aging, but this was a consequence of the reduction in moisture content rather than actual changes in their content during the dry-aging process [19]. For similar reasons, the different dry-aged methods did not show differences in the fat and protein contents as their moisture contents were similar. The moisture content was negatively correlated with fat ($r = -0.9$) and with protein ($r = -0.3$). The different dry-aging methods did not cause

Table 1. Effects of different dry-aging methods on the chemical composition (%) of sirloin from low-marbled Hanwoo cow meat after 28 days

Items	Control ¹⁾	TD ¹⁾	SD ¹⁾	SDB ¹⁾	SEM ²⁾
Moisture	67.07 ^a	63.81 ^b	62.17 ^b	64.44 ^b	0.757
Fat	10.00 ^b	12.82 ^a	13.53 ^a	12.00 ^{ab}	0.793
Protein	21.10 ^b	21.73 ^{ab}	22.47 ^a	21.91 ^{ab}	0.270
Collagen	1.83	1.65	1.84	1.66	0.078

¹⁾ Control, 2 days postmortem and not aged; TD, traditional dry-aging; SD, simplified dry-aging; and SDB, simplified dry-aging in a highly water vapor-permeable bag.

²⁾ Standard error of the means ($n = 48$).

^{a,b} Different letters within the same row indicate a significant difference ($p < 0.05$).

any significant changes in the collagen content.

Microbial analyses

No mold/yeast and coliform contamination were found in the meats before and after dry-aging (Table 2). Total aerobic bacteria count in the 3 dry-aged groups was higher than that in the control. Among the dry-aged groups, SDB showed the highest number of bacteria, which was not expected as most of the previous studies have reported that the bag could inhibit microbial contamination [11,13]. This result might be attributable to the different amounts and depths of the crust and/or its formation rate as it has a role in preventing microbial penetration into the meat [20]. When we estimated the amount and depth of crust from the trimming loss among the dry-aged groups (data not shown), the trimming loss was the highest in SD group (26.31%), followed by TD (19.54%) and SDB (18.16%) groups. The results are in the same order as those of the microbial counts in Table 2. It would seem that crust formation during the early dry-aging process may be more effective in preventing microbial penetration than application of the aging bag. However, it can be a disadvantage from the economic aspect of the dry-aged beef. In this study, the saleable yield of SD group decreased approximately 13.67% relative to that of TD group, whereas SDB group maintained a similar level to TD group ($p < 0.05$, data not shown). Thus, SDB would be a better choice for the dry-aged beef that have a relatively higher saleable yield, because the difference of microbial counts among the dry-aged groups ($< 1 \text{ log CFU/g}$), albeit statistically significant, can be ignored in practical situations.

Table 2. Effects of different dry-aging methods on the microbial counts (log CFU/g) in sirloin from low-marbled Hanwoo cow meat after 28 days

Items	Control ¹⁾	TD ¹⁾	SD ¹⁾	SDB ¹⁾	SEM ²⁾
Total aerobic bacteria	5.60 ^c	6.58 ^{ab}	6.18 ^{bc}	7.01 ^a	0.212
Mold/yeast	nd ³⁾	nd	nd	nd	-
Coliforms	nd	nd	nd	nd	-

CFU, colony-forming unit.

¹⁾ Control, 2 days postmortem and not aged; TD, traditional dry-aging; SD, simplified dry-aging; and SDB, simplified dry-aging in a highly water vapor-permeable bag.

²⁾ Standard error of the means ($n = 48$).

³⁾ Not detected.

^{a,c} Different letters within the same row indicate a significant difference ($p < 0.05$).

Warner-Bratzler shear force

The shear force was significantly decreased (approximately 47%) in the dry-aged groups compared with the control (Figure 1). This improvement is due to the activities of calpains, cathepsins, proteasome, and caspases during the early postmortem period [21, 22]. Meanwhile, no difference in shear force was observed among the TD, SD, and SDB groups. This seems reasonable as the differences among the dry-aging groups involved only RH, air flow, and the application of the aging bag, showing that all 3 methods have similar abilities to improve the tenderness of low-marbled beef.

Inosine 5'-monophosphate and free amino acid contents

IMP and free amino acids are the degradation products of ATP and protein, respectively, after slaughter [23]. The free amino acid components are influential in the flavor formation of meat and meat products, as together with reducing sugars they have a specific role in the Maillard reaction. Among these degradation products, IMP and Glu have a positive and even synergistic impact on umami taste [15,24]. In this study, the IMP content had declined by 40% to 47% during the aging period, and no significant differences were found among the different aging methods (Table 3). IMP degradation to inosine and hypoxanthine is a constant but fast reaction at the early postmortem period, and because dry-aging takes a long time to change the meat characteristics, the increases and/or concentrated levels of free amino acids would be more important than those of nucleotides. Iida et al [25] calculated the umami intensity of beef during the dry-aging process and reported a significant role of Glu at the late aging period, whereas IMP was effective only during the early aging stage. A different pattern was found between the control and dry-aged groups for various free amino acids. Whereas there were no significant changes in Glu, the contents of Ala, Arg, Cys-cys, Gly, His, Ile, Leu, Phe, Ser, Thr, Val, and total amino acids were increased during the aging period and were significantly higher

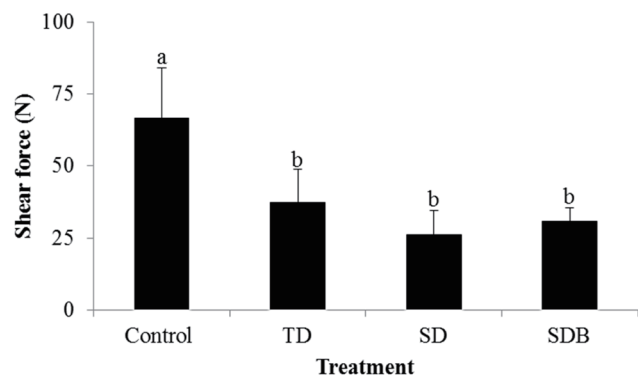


Figure 1. Effects of different dry-aging methods on the shear force (N) of sirloin from low-marbled Hanwoo cow meat after 28 days. Control, 2 days postmortem and not aged; TD, traditional dry-aging; SD, simplified dry-aging; and SDB, simplified dry-aging in a highly water vapor-permeable bag. ^{a,b} Different letters indicate they were significantly different ($p < 0.05$).

Table 3. Effects of different dry-aging methods on the inosine 5'-monophosphate (IMP) and free amino acid contents (mg/100 g) of sirloin from low-marbled Hanwoo cow meat after 28 days

Items	Control ¹⁾	TD ¹⁾	SD ¹⁾	SDB ¹⁾	SEM ²⁾
IMP	153.29 ^a	67.67 ^b	61.83 ^b	72.49 ^b	5.831
Ala	26.82 ^c	34.82 ^b	40.66 ^a	36.34 ^b	1.396
Arg	3.41	5.05	3.89	4.66	0.459
Asn	2.75	3.65	4.64	5.43	0.919
Asp	3.61	2.74	3.16	3.18	0.261
Car	1.89	1.39	1.52	1.54	0.523
Cys-Cys	4.16 ^b	7.41 ^a	8.47 ^a	8.64 ^a	0.727
Glu	15.77	18.25	16.85	14.65	2.324
Gly	10.66 ^b	15.85 ^a	18.33 ^a	14.12 ^{ab}	1.243
His	2.35 ^b	3.84 ^a	4.89 ^a	4.22 ^a	0.302
Ile	3.04 ^c	5.68 ^{ab}	6.44 ^a	4.75 ^b	0.429
Leu	7.68 ^b	16.18 ^a	18.62 ^a	16.40 ^a	1.320
Lys	17.97 ^a	6.13 ^b	6.59 ^b	14.60 ^a	1.576
Phe	3.96 ^b	8.13 ^a	9.32 ^a	8.57 ^a	0.604
Pro	2.07	2.01	2.50	1.60	0.277
Ser	5.48 ^b	10.90 ^a	12.79 ^a	10.32 ^a	0.883
Tau	7.32	6.76	6.81	7.93	0.888
Thr	3.32 ^b	6.54 ^a	7.87 ^a	6.22 ^a	0.581
Trp	7.34	6.92	6.88	9.23	0.991
Tyr	4.32 ^b	7.60 ^a	5.41 ^{ab}	3.08 ^b	0.797
Val	3.79 ^c	8.61 ^b	11.71 ^a	7.42 ^b	0.784
Total	137.73 ^b	178.46 ^a	197.33 ^a	182.84 ^a	8.446

¹⁾ Control, 2 days postmortem and not aged; TD, traditional dry-aging; SD, simplified dry-aging; and SDB, simplified dry-aging in a highly water vapor-permeable bag.

²⁾ Standard error of the means ($n = 48$).

^{a,c} Different letters within the same row indicate a significant difference ($p < 0.05$).

than those of the control ($p < 0.05$). The amount of free amino acids responsible for bitter taste (bitter-FAAs: Arg, His, Ile, Leu, and Phe) achieved 2.0-fold increases on average, whereas the free amino acids responsible for sweet taste (sweet-FAAs: Ala, Gly, Ser, and Thr) increased 1.5-fold on average after dry-aging. Even though the bitter-FAAs showed a higher fold increase than did sweet-FAAs, the overall changes in free amino acid content would have a positive impact on flavor formation as the absolute amounts were approximately 2-times higher in sweet-FAAs after dry-aging. On the other hand, the differences in RH, air flow, and the application of the aging bag did not cause significant changes in both IMP and most of the free amino acid content, possibly because the final moisture content was similar among the three dry-aged groups (Table 1). Kim et al [17] reported that moisture evaporation is a cause of the concentrated flavor components during the dry-aging process.

Sensory evaluation

In the present study, improvement in the juiciness of low-marbled beef was expected, as Cambell et al [26] had elucidated that the fat content would concentrate with dry-aging through moisture evaporation. However, the juiciness was not significantly different among the different dry-aging groups (Figure 2), probably because their differences in moisture (62.17% to 67.01%) and

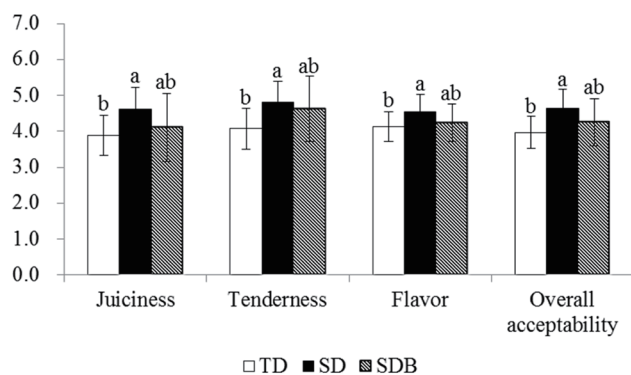


Figure 2. Effects of different dry-aging methods on the sensory properties of sirloin from low-marbled Hanwoo cow meat after 28 days. TD, traditional dry-aging; SD, simplified dry-aging; and SDB, simplified dry-aging in a highly water vapor-permeable bag. ^{a,b} Different letters indicate they were significantly different ($p < 0.05$).

fat (12.00% to 13.53%) contents were insignificant (Table 1). The palatability (juiciness, tenderness, flavor, and overall acceptability) of low-marbled beef was higher in SD, followed by SDB and TD and SD group was statistically similar to the results from SDB group (Figure 2). This is probably because TD resulted in the highest shear force and SD resulted in the highest total free amino acid content ($p > 0.05$, Figure 1, Table 3). The present results suggest that SD and SDB can be applied instead of TD to improve the palatability of low-marbled beef.

CONCLUSION

Both simplified dry aging methods (SD and SDB) resulted in the desirable overall acceptability among the dry-aged groups, thus can successfully substitute for TD. However, SDB could maximize the saleable yield without compromising significant hygienic problems, such as microbial growth. Therefore, SDB would be the best option for simplified dry-aging of low-marbled beef with a relatively high saleable yield.

CONFLICT OF INTEREST

We certify that there is no conflict of interest with any financial organization regarding the material discussed in the manuscript.

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