

ARTICLE

Determination of Salable Shelf-life for Wrap-packaged Dry-aged Beef during Cold Storage

Hyun Jung Lee¹, Juhui Choe¹, Ji Won Yoon¹, Seonjin Kim¹, Hyemin Oh²,
Yohan Yoon², and Cheorun Jo^{1,3,*}

¹Department of Agricultural Biotechnology, Center for Food and Bioconvergence, and Research Institute of Agriculture and Life Science, Seoul National University, Seoul 08826, Korea

²Department of Food and Nutrition, Sookmyung Women's University, Seoul 04310, Korea

³Institute of Green Bio Science and Technology, Seoul National University, Pyeongchang 25354, Korea

 OPEN ACCESS

Received January 25, 2018

Revised January 31, 2018

Accepted January 31, 2018

*Corresponding author : Cheorun Jo
Department of Agricultural Biotechnology,
Center for Food and Bioconvergence,
Research Institute of Agriculture and Life
Science, Seoul National University, Seoul
08826, Korea
Tel: +82-2-880-4804
Fax: +82-2-873-2271
E-mail: cheorun@snu.ac.kr

Abstract We investigated microbial and quality changes in wrap-packaged dry-aged beef after completion of aging and subsequent storage in a refrigerator. After 28 days of dry aging (temperature, 4°C; RH, approximately 75%; air flow velocity, 2.5 m/s), sirloins were trimmed, wrap-packaged, and stored at 4°C for 7 days. Analyses of microbial growth, pH, volatile basic nitrogen (VBN), 2-thiobarbituric acid-reactive substance (TABRS), and instrumental color, myoglobin, and sensory evaluation were conducted on days 0, 3, 5, and 7. The results show that the number of total aerobic bacteria (TAB), yeast, and lactic acid bacteria increased with an increase in storage days, whereas no change in the growth of mold was observed during 7 days of storage. Based on the legal standard for TAB count, the estimated shelf-life of wrap-packaged dry-aged beef was predicted to be less than 12.2 days. However, the shelf-life should be less than 6.3 days, considering the result of sensory quality (odor, taste, and overall acceptance). No significant change in visible appearance was also observed during 7 days of storage. The results suggest that the present quality indicators for meat spoilage (pH, VBN, and TBARS) should be re-considered for dry-aged beef, as its characteristics are different from those of fresh and/or wet-aged beef.

Keywords dry-aged beef, wrap-packaging, shelf-life

Introduction

Dry aging, one of the aging methods [wet (vacuum) or dry aging], exposes raw beef to controlled conditions (temperature, RH, and air flow) to improve its tenderness, flavor, and juiciness (Lee et al., 2017). It is a traditional aging method used prior to the introduction of vacuum packaging. However, the low efficiency in processing and reduced salable yield resulted in decreased market value of dry-aged beef since 1980's (Dashdorj et al., 2016). In recent years, there has been an increasing demand

for dry-aged beef with a premium value owing to its characteristic concentrated flavor (beefy and roasted), which is absent in wet-aged beef (Oh et al., 2017).

Dry-aged beef is produced without packaging and is prone to microbial contamination and growth of mold/yeast on the crust during dry aging process (Lee et al., 2017). Hence, microbial safety has been issued for dry-aged beef and several researchers have studied the microbial properties of dry-aged beef. Li et al. (2013) observed a significant increase in the numbers of total aerobic bacteria [TAB; from 1.2 to 5.2 Log colony-forming unit (CFU)/g] and yeast (from 0.01 to 3.0 Log CFU/g) after 14 days of dry aging. In addition, an increase in microbial counts (TAB, coliform, and yeast) was observed in dry-aged beef after 8 or 19 days of aging (Li et al., 2014), consistent with a previous study (Lee et al., 2017). In addition, our previous study concluded that wrap packaging (aerobic condition) may increase the chances of microbial contamination following the completion of dry aging process (unpublished data). As dry-aged beef is usually wrap-packaged in market, microbial contamination during the dry aging process and distribution may raise safety concerns while sale and/or consumption. However, no standard methods and/or information are available on the shelf-life of wrap-packaged dry-aged beef.

Quality deterioration may occur with an increase in the microbial count during storage and is related to meat spoilage, which results in low consumer acceptability. Therefore, there are recommended standard methods to assess meat spoilage, such as the analysis of pH, volatile basic nitrogen (VBN), ammonia, Walkiewicz reaction, and trimethylamine level (Jang et al., 2014). However, no scientific information is available on the storage stability of dry-aged beef as compared with fresh or wet-aged beef, although there is a considerable difference in the process (drying and aging) and environment. Therefore, the objective of this study was to investigate microbial and quality changes in wrap-packaged dry-aged beef during 7 days of storage. The results of this study may help small scale producers, vendors, and consumers as well as authorities to understand the storage characteristics of wrap-packaged dry-aged beef.

Materials and Methods

Dry aging and packaging condition

A total of nine sirloins were obtained from nine beef carcasses (Holstein, quality grade 3) at three different slaughter days (3 sirloins/trial) and subjected to dry aging for 28 d (temperature, 4°C; RH, approximately 75%; air flow velocity, 2.5 m/s). After dry aging, the external crust was trimmed and the samples were cut (length×width×height, 12.7×7.6×2.54 cm³), wrap-packaged (aerobic condition), and stored at 4°C for 7 d. The samples were obtained at days 0, 3, 5, and 7 for further analyses.

Microbial growth analysis

Each sample (5 g) was blended in sterile saline (45 mL, 0.85%) for 2 min using a laboratory blender (BagMixer 400 P, Interscience, France). Each dilution (100 µL) was spread on plate count agar (Difco Laboratories, USA), yeast mold (YM) agar (Difco Laboratories), and de man, rogosa and sharpe agar (Difco Laboratories) for enumeration of TAB, mold/yeast, and lactic acid bacteria (LAB), respectively. The agar plates for TAB and LAB were incubated at 37°C for 48 h, whereas YM plates were incubated at 25°C for 120 h. After incubation, microbial counts were enumerated and expressed as Log CFU/g.

Spoilage indicators and lipid oxidation

Changes in pH, VBN, and lipid oxidation of the wrap-packaged dry-aged beef were determined on 0, 3, 5, and 7 days of storage. pH and VBN values were analyzed as spoilage indicators (Jang et al., 2014) and lipid oxidation [2-thiobarbituric

acid-reactive substance (TABRS)] was measured for quality deterioration. All analyses were conducted based on the methods from Lee et al. (2012).

Instrumental color and myoglobin analysis

Meat color of wrap-packaged dry-aged beef was measured using a colorimeter (CM-5, Minolta Co., Ltd., Japan; 8 mm diameter aperture and D65 illuminant) and expressed as CIE (L^* , a^* , and b^*) values following 30 min blooming time. Three measurements were averaged and used as one replication for each sample. Chroma ($[a^{*2} + b^{*2}]$) and hue-angle ($\tan^{-1} [b^*/a^*]$) were calculated from a^* and b^* values. Myoglobin (Mb) content and its composition [deoxymyoglobin (deoxyMb), oxymyoglobin (oxyMb), and metmyoglobin (metMb)] were analyzed following the methods of Krzywicki (1979).

Sensory evaluation

Sensory evaluation was conducted by a consumer panel (total 10 panelists) to observe the changes in sensory properties of wrap-packaged dry-aged beef during 7 days of storage. Each sample was cut into same size ($4 \times 2 \times 2.54 \text{ cm}^3$) and grilled until the core temperature reached 72°C . The cooked meat was maintained at 72°C before serving to the panel. A 9-point hedonic scale (1, extreme dislike; 9, extreme like) was used to score the appearance, odor, taste, and overall acceptability. Three trials were conducted and the average score from each trial was used as one replication.

Estimation for shelf-life

Each regression equation for all traits of wrap-packaged dry-aged beef tested was obtained using the data from different storage days (Table 1). In the equation, 'x' represents estimated shelf-life and 'y' represents quality limit. Quality limit was based on the legal standard from Ministry of Food and Drug Safety, Korea (MFDS, 2014) for TAB. The other quality limits were calculated using other equations (see supplementary materials) between the data and overall acceptance from sensory evaluation (>5 considered as acceptable).

Statistical analysis

A randomized incomplete block design was applied using the trial as the block. Samples with different storage days (0, 3, 5, and 7 days) were analyzed in the trial. In each trial, three measurements were averaged and used as one replication ($n = 3$). A general linear model was performed using SAS 9.3 (SAS Institute Inc., USA) and results were reported as mean values with standard error of the means. Significant differences among the mean values were determined on the basis of Tukey's multiple comparison test at a significance level of $p < 0.05$. Correlation coefficient (r^2) between microbial and quality changes in wrap-packaged dry-aged beef was also calculated.

Results and Discussion

Microbial growth

Although meat spoilage is associated with several inter-related factors (temperature, oxygen, enzymes, moisture, light, etc.), the most important factor is the type and number of microorganisms (Lambert et al., 1991). Therefore, the estimation of microbial count is performed as the legal standard method to evaluate meat spoilage ($< 7.0 \text{ Log CFU/g}$ for TAB; MFDS, 2014). In this study, the initial microbial count of wrap-packaged dry-aged beef was 4.26, 2.86, 2.60, and 1.78 Log CFU/g for TAB, mold, yeast, and

LAB, respectively (Fig. 1). The number of TAB, yeast, and LAB increased with an increase in storage days, whereas no change was observed in the mold count during 7 days of storage. Based on the legal standard value for TAB count (7 Log CFU/g meat), the shelf-life of wrap-packaged dry-aged beef could be estimated to be less than 12.2 days (Table 1). In addition, the number of yeast and LAB was 4.9 and 3.4 Log CFU/g, respectively, when the overall acceptance was less than 5 (Table 5).

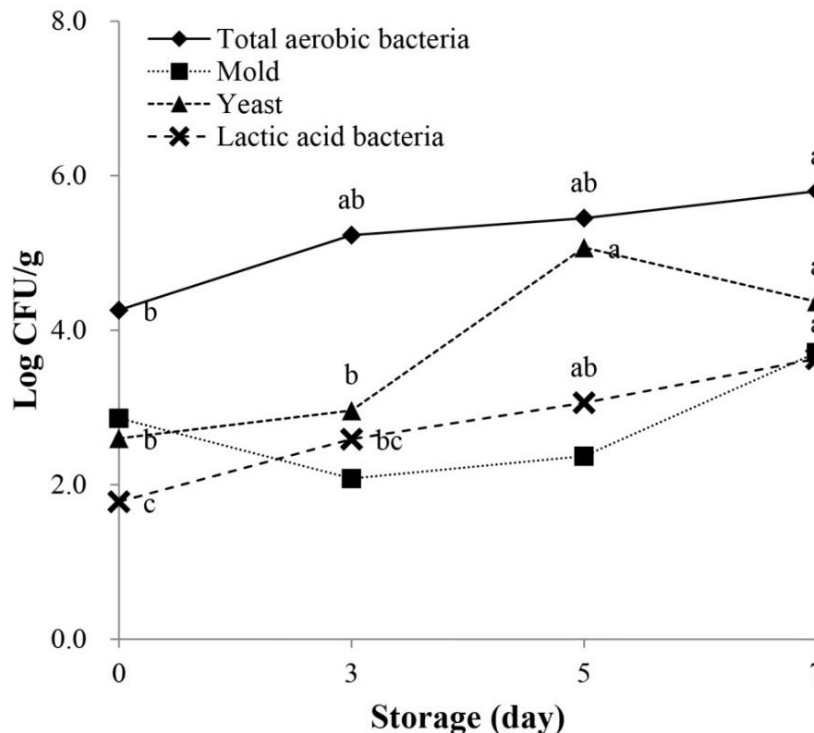


Fig. 1. The microbial growth (Log CFU/g) of wrap-packaged dry-aged beef during the storage period of 7 days (mean±SD). ^{a-c} Different letters within the same microorganism indicate a significant difference ($p < 0.05$).

Table 1. The estimated shelf-life of wrap-packaged dry-aged beef with quality standards

Traits	Quality limit ¹⁾	Shelf-life (day)	Regression equation	r ²
TAB (Log CFU/g)	7.0	<12.2	$y = 0.2155x + 4.3768$	0.951
Mold (Log CFU/g)	12.9	<102.6	$y = 0.1025x + 2.3680$	0.183
Yeast (Log CFU/g)	4.9	<7.5	$y = 0.3199x + 2.5479$	0.676
LAB (Log CFU/g)	3.4	<6.3	$y = 0.2619x + 1.7830$	0.999
pH	6.07	<84.3	$y = 0.0040x + 5.7327$	0.057
VBN (mg%)	89.31	<6.3	$y = 3.2654x + 68.8790$	0.894
TBARS (mg MDA/kg meat)	1.35	<16.4	$y = -0.0146x + 1.5896$	0.276
Appearance	>5	<15.8	$y = -0.0711x + 6.1216$	0.590
Odor	>5	<6.7	$y = -0.1026x + 5.6905$	0.785
Taste	>5	<7.4	$y = -0.1844x + 6.3716$	0.798
Overall acceptance	>5	<6.3	$y = -0.1519x + 5.9596$	0.852

¹⁾ Quality limit of TAB was the legal standard from Ministry of Food and Drug Safety (MFDS, 2014) and the other quality limits were calculated using other equations between the data and overall acceptance from sensory evaluation (>5 considered as acceptable). TAB, total aerobic bacteria; LAB, lactic acid bacteria; VBN, volatile basic nitrogen; TBARS, 2-thiobarbituric acid-reactive substance.

Spoilage indicators and lipid oxidation

In general, pH and VBN are the most reliable spoilage indicators for fresh meat among the recommended standard parameters (Jang et al., 2014). In Korea, pH and VBN values of meat are limited to 6.2 and 20-30 mg% (20 mg% for fresh meat and 30 mg% for wet-aged meat), respectively, and values higher than these may deem the meat as spoiled (Jang et al., 2014). During meat spoilage, pH and VBN values tend to increase, owing to the production of amines/ammonia by microorganisms (Flores et al., 1997; Pearson, 1968). In this study, however, pH may not serve as a proper spoilage indicator for dry-aged beef ($r^2 = 0.057$, Table 1) because we failed to observe any significant change in pH value during 7 days of storage and its correlation to microbial counts (Tables 2 and 3). The non-significant change in pH value may be associated with the lactic and acetic acids produced from LAB and mold/yeast that offset the increase in pH. Flores et al. (1997) reported the effect of acids produced by inoculated yeast and LAB on the pH of dry-cured sausages. VBN value of wrap-packaged dry-aged beef was high even at day 0 (66.14 mg%) and reached 89.49 mg% after 7 days of storage (Table 3). This value was 2.8-3.8 times higher than that reported for wet-aged beef sirloin aged for 14 wk (Jang et al., 2014). Our previous study showed that this observation may be attributed to the higher rate of proteolysis in dry-aged beef than wet-aged beef (data not shown), possibly due to the growth of mold/yeast on the surface of beef during the dry aging process. VBN content significantly increased at day 3 and the level was maintained until the end of storage days in the presence of TAB and LAB ($r^2 = 0.88$ and 0.84 , respectively; $p < 0.01$; Tables 2 and 3). VBN may serve as the spoilage indicator for dry-aged beef ($r^2 = 0.894$, Table 1); however, the present recommendation for meat spoilage (20-30 mg% for fresh or wet-aged beef) should be re-considered. Based on the results of sensory analysis in this study, VBN content below 89.31 mg% was acceptable for dry-aged beef.

Lipid oxidation, measured using TBARS value, is an important indicator of quality deterioration in meat (Ladikos and Lougovois, 1990). In this study, TBARS value of wrap-packaged dry-aged beef varied from 1.43 to 1.63 mg malondialdehyde/kg meat (Table 3), similar to that reported for dry-aged beef by DeGeer et al. (2009). No significant change in TBARS value was observed during 7 days of storage. This observation is in line with the results of our previous study, wherein no significant increase in TBARS value was observed regardless of temperature, packaging methods, and storage days (data not shown). TBARS value had no correlation with other quality attributes (Table 2) and may not be an appropriate indicator of the quality deterioration of dry-aged beef. In addition, lipid oxidation has been reported to have a positive impact on the flavor of the dry-aged meat (DeGeer et al., 2009).

Instrumental color and myoglobin analysis

During the early storage, a significant discoloration was observed for wrap-packaged dry-aged beef, as evident from the decrease in CIE (L^* , a^* , and b^*), chroma, and hue-angle values at day 3. The values were maintained after 3 days of storage ($p < 0.05$, Table 4). Discoloration in meat is mainly attributed to the oxidation of Mb to metMb (Faustman et al., 2010). However, we found that metMb level of wrap-packaged dry-aged beef was significantly decreased at day 3 and increased thereafter, contrary to oxyMb level. DeoxyMb level was not changed until day 5 and significantly decreased. Therefore, in this study, discoloration can not be fully elucidated with Mb composition and may be more related to its content. CIE L^* and a^* values showed higher relative correlation with Mb content as compared with oxyMb or metMb levels ($p > 0.05$, data not shown). As Mb imparts meat color, the significant decrease in Mb content resulted in the discoloration of meat. However, these changes were visibly negligible before/after cooking, regardless of the storage days (Tables 3 and 4).

Table 2. The correlation coefficient (r^2) of quality attributes of wrap-packaged dry-aged beef during 7 days of storage

	TAB	Mold	Yeast	LAB	pH	VBN	TBARS	L*	a*	b*	Chroma	Hue-angle	Appearance	Odor	Taste	Overall acceptance
TAB	1	0.18	-0.63*	0.97***	0.10	0.88**	-0.11	-0.49	-0.45	-0.26	-0.39	0.24	-0.46	-0.60*	-0.66*	-0.79**
Mold		1	-0.54	0.40	0.20	0.06	-0.05	0.07	0.18	0.29	0.22	0.24	-0.12	0.01	-0.01	-0.20
Yeast			1	-0.73**	-0.30	-0.61*	0.11	0.17	0.28	0.05	0.19	-0.48	0.43	0.69*	0.69*	0.72**
LAB				1	0.14	0.84**	-0.12	-0.44	-0.41	-0.21	-0.34	0.27	-0.47	-0.56	-0.65*	-0.82**
pH					1	-0.15	0.07	0.25	0.06	0.17	0.10	0.24	0.07	-0.30	-0.12	0.02
VBN						1	-0.21	-0.70*	-0.66*	-0.51	-0.62*	0.14	-0.31	-0.65*	-0.78**	-0.87**
TBARS							1	-0.19	-0.12	-0.15	-0.13	-0.05	-0.13	-0.15	0.01	0.15
L*								1	0.85**	0.83**	0.87**	0.23	0.09	0.48	0.45	0.57
a*									1	0.91***	0.99***	0.18	0.24	0.55	0.55	0.72**
b*										1	0.96***	0.55	0.23	0.32	0.43	0.58*
Chroma											1	0.33	0.24	0.47	0.51	0.68*
Hue-angle												1	0.11	-0.46	-0.16	0.00
Appearance													1	0.17	0.22	0.59*
Odor														1	0.82**	0.58*
Taste															1	0.77*
Overall Acceptance																1

* $p<0.05$; ** $p<0.01$; *** $p<0.001$.

TAB, total aerobic bacteria; LAB, lactic acid bacteria; VBN, volatile basic nitrogen; TBARS, 2-thiobarbituric acid-reactive substance.

Table 3. The spoilage indicators and lipid oxidation of wrap-packaged dry-aged beef during 7 days of storage

Traits	Storage (day)				SEM ¹⁾
	0	3	5	7	
pH	5.75	5.69	5.81	5.74	0.075
VBN (mg%)	66.14 ^b	83.26 ^a	85.60 ^a	89.49 ^a	1.946
TBARS (mg MDA/kg meat)	1.57	1.52	1.62	1.43	0.168

¹⁾Standard error of the means (n=12).^{a,b}Different letters within the same row indicate a significant difference ($p<0.05$).

VBN, volatile basic nitrogen; TBARS, 2-thiobarbituric acid-reactive substance; MDA, malondialdehyde.

Sensory evaluation

The odor and taste of wrap-packaged dry-aged beef was significantly deteriorated at day 5, while the overall acceptance significantly decreased at day 3 as compared to day 0 (Table 5). These changes were significantly related to the growth of TAB, yeast, and LAB, but not mold (Table 2). TAB and LAB count had a negative effect on the odor ($p<0.05$, TAB only), taste ($p<0.05$), and overall acceptance ($p<0.01$), while yeast had a positive effect on these attributes. Yeast is reported to exhibit a positive impact on sensory qualities, as it promotes flavor development in meat via lipolysis and/or proteolysis (Toldra, 1998). Molds play a similar role; however, we failed to observe any increase in mold count during 7 days of storage, resulting in the absence of any significant effect on sensory qualities (Fig. 1 and Table 2). Wrap-packaged dry-aged beef was acceptable until 6.3

Table 4. The instrumental color and myoglobin content/composition of wrap-packaged dry-aged beef during 7 days of storage

Traits	Storage (day)				SEM ¹⁾
	0	3	5	7	
CIE L*	41.99 ^a	36.33 ^b	38.78 ^{ab}	37.22 ^{ab}	1.185
CIE a*	15.05 ^a	8.09 ^b	9.54 ^{ab}	10.59 ^{ab}	1.267
CIE b*	10.60 ^a	4.57 ^b	7.13 ^{ab}	8.19 ^{ab}	1.021
Chroma	18.43 ^a	9.33 ^b	12.02 ^{ab}	13.45 ^{ab}	1.548
Hue-angle	35.06 ^a	28.39 ^b	37.79 ^{ab}	37.71 ^{ab}	2.335
Mb (mM/L)	0.046 ^a	0.036 ^b	0.034 ^b	0.035 ^b	0.0009
DeoxyMb (%)	10.35 ^a	10.74 ^a	10.62 ^a	6.82 ^b	0.686
OxyMb (%)	62.80 ^b	71.73 ^a	59.91 ^b	48.84 ^c	1.834
MetMb (%)	26.85 ^b	17.53 ^c	29.48 ^b	44.34 ^a	1.370

¹⁾ Standard error of the means (n=12).

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

Mb, myoglobin; deoxyMb, deoxymyoglobin; oxyMb, oxymyoglobin; metMb, metmyoglobin.

Table 5. The sensory evaluation of wrap-packaged dry-aged beef during 7 days of storage¹⁾

Traits	Storage (day)				SEM ²⁾
	0	3	5	7	
Appearance	6.22	5.84	5.55	5.81	0.286
Odor	5.72 ^a	5.45 ^{ab}	4.95 ^b	5.11 ^{ab}	0.143
Taste	6.50 ^a	5.78 ^{ab}	5.08 ^b	5.36 ^b	0.231
Overall acceptance	6.11 ^a	5.28 ^b	5.11 ^b	5.06 ^b	0.130

¹⁾ 1, extreme dislike; 5, neither dislike nor like 9, extreme like.

²⁾ Standard error of the means (n=12).

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

days, as all sensory qualities (odor, taste, and overall acceptance) were higher than 5 (acceptable; Tables 1 and 5) and TAB count was lower than 7 Log CFU/g (Fig. 1). No significant change in visible appearance was also observed during 7 days of storage.

Conclusion

The results of this study show that the estimated shelf-life of wrap-packaged dry-aged beef was less than 6.3 days, during which it met the quality standards of both microbial count and sensory qualities. The present indicators for meat spoilage/quality deterioration such as pH, VBN, and TBARS values are unsuitable for dry-aged beef and may provide unreliable information to consumers. Therefore, further studies should be conducted to investigate appropriate indicators for the determination of quality standards of dry-aged beef.

Acknowledgments

This study was supported by “High Value-added Food Technology Development Program (Project No. 316048),” Korea

Institute of Planning and Evaluation for Technology in Food, Agriculture, Forestry and Fisheries.

References

- Dashdorj D, Tripathi VK, Cho S, Kim Y, Hwang I. 2016. Dry aging of beef; Review. *J Anim Sci Technol* 58:20.
- DeGeer SL, Hunt MC, Bratcher CL, Crozier-Dodson BA, Johnson DE, Stika JF. 2009. Effects of dry aging of bone-in and boneless strip loins using two aging processes for two aging times. *Meat Sci* 83:768-774.
- Faustman C, Sun Q, Mancini R, Suman SP. 2010. Myoglobin and lipid oxidation interactions: mechanistic bases and control. *Meat Sci* 86:86-94.
- Flores J, Marcus JR, Nieto P, Navarro JL, Lorenzo P. 1997. Effect of processing conditions on proteolysis and taste of dry-cured sausages. *Z Lebensm Unters Forsch A* 204:168-172.
- Jang A. 2014. Study of meat freshness (spoilage) standard evaluation. Ministry of Food and Drug Safety, Korea.
- Krzywicki K. 1979. Assessment of relative content of myoglobin, oxymyoglobin and metmyoglobin at the surface of beef. *Meat Sci* 79:1-10.
- Ladikos D, Lougovois V. 1990. Lipid oxidation in muscle foods: A review. *Food Chem* 35:295-314.
- Lambert AD, Smith JP, Dodds KL. 1991. Shelf life extension and microbiological safety of fresh meat - A review. *Food Microbiol* 8:267-297.
- Lee HJ, Choe J, Kim KT, Oh J, Lee DG, Kwon KM, Choi YI, Jo C. 2017. Analysis of low-marbled Hanwoo cow meat aged with different dry-aging methods. *Asian Australas J Anim Sci* 30:1733-1738.
- Lee HJ, Song HP, Jung H, Choe W, Ham JS, Lee JH, Jo C. 2012. Effect of atmospheric pressure plasma jet on inactivation of *Listeria monocytogenes*, quality, and genotoxicity of cooked egg white and yolk. *Korean J Food Sci An* 32:561-570.
- Li X, Babol J, Bredie WLP, Nielsen B, Tománková J, Lundström K. 2014. A comparative study of beef quality after ageing *longissimus* muscle using a dry ageing bag, traditional dry ageing or vacuum package ageing. *Meat Sci* 97:433-442.
- Li X, Babol J, Wallby A, Lundström K. 2013. Meat quality, microbiological status and consumer preference of beef *gluteus medius* aged in a dry ageing bag or vacuum. *Meat Sci* 95:229-234.
- MFDS (Ministry of Food and Drug Safety). 2014. Examination of microorganisms in food. Korea.
- Oh J, Lee HJ, Kim HC, Kim HJ, Yun YG, Kim KT, Choi YI, Jo C. 2017. The effects of dry or wet aging on the quality of the *longissimus* muscle from 4-year-old Hanwoo cows and 28-month-old Hanwoo steers. *Anim Prod Sci*. doi: 10.1071/AN17104.
- Pearson D. 1968. Assessment of meat freshness in quality control employing chemical techniques: A review. *J Sci Food Agric* 19:357-363.
- Toldra F. 1998. Proteolysis and lipolysis in flavour development of dry-cured meat products. *Meat Sci* 49:S101-S110.

Supplementary Materials

1. Total aerobic bacteria (TAB): Estimated shelf-life 12.2 days

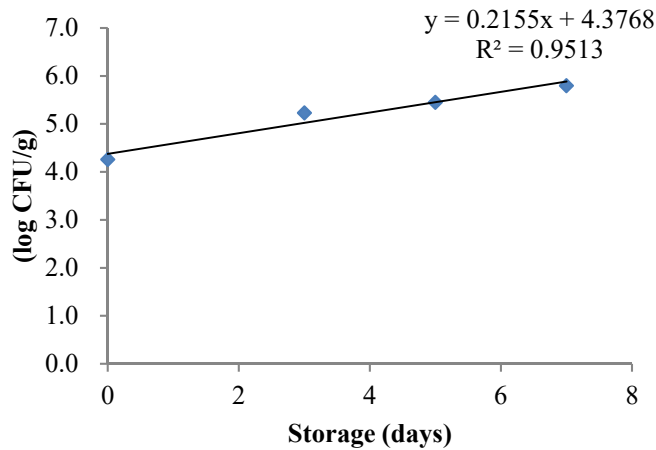


Fig. 1. Regression equation for estimated shelf-life (days) of wrap-packaged dry-aged beef based on total aerobic bacteria count (log CFU/g).

How calculated:

- 1) $y = 7$ (legal standard from Ministry of Food and Drug Safety).
- 2) substitute 7 (y) to the formula ($y = 0.2155x + 4.3768$) and calculate estimated shelf-life (x).

2. Mold: Estimated shelf-life 102.6 days

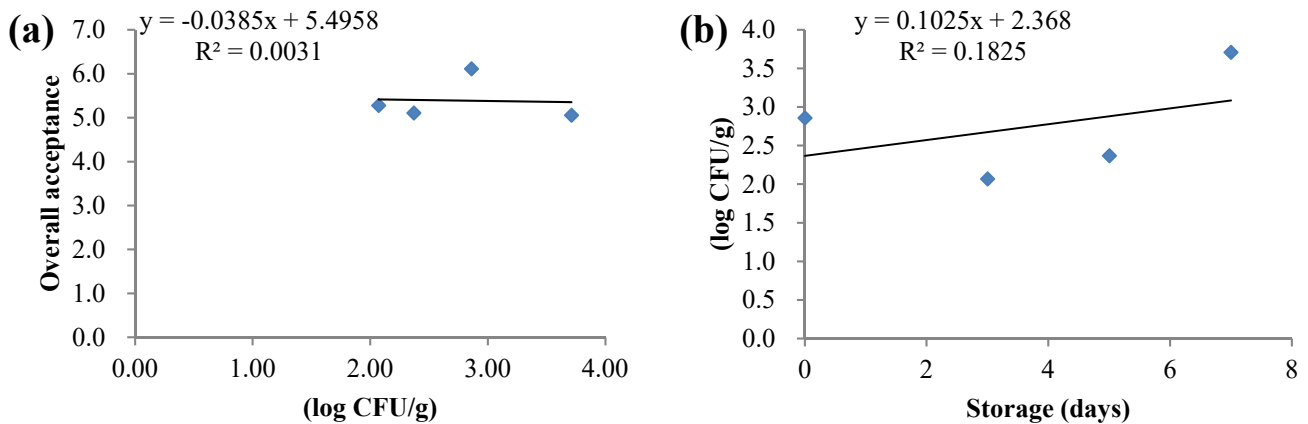


Fig. 2. Regression equation for (a) quality limit (using overall acceptance, 5 considered as acceptable) and (b) estimated shelf-life (days) of wrap-packaged dry-aged beef based on mold count (log CFU/g).

How calculated:

- 1) $y = 12.9$ (see Fig. 2a).
- 2) substitute 12.9 (y) to the formula ($y = 0.1025x + 2.368$) and calculate estimated shelf-life (x).

3. Yeast: Estimated shelf-life 7.5 days

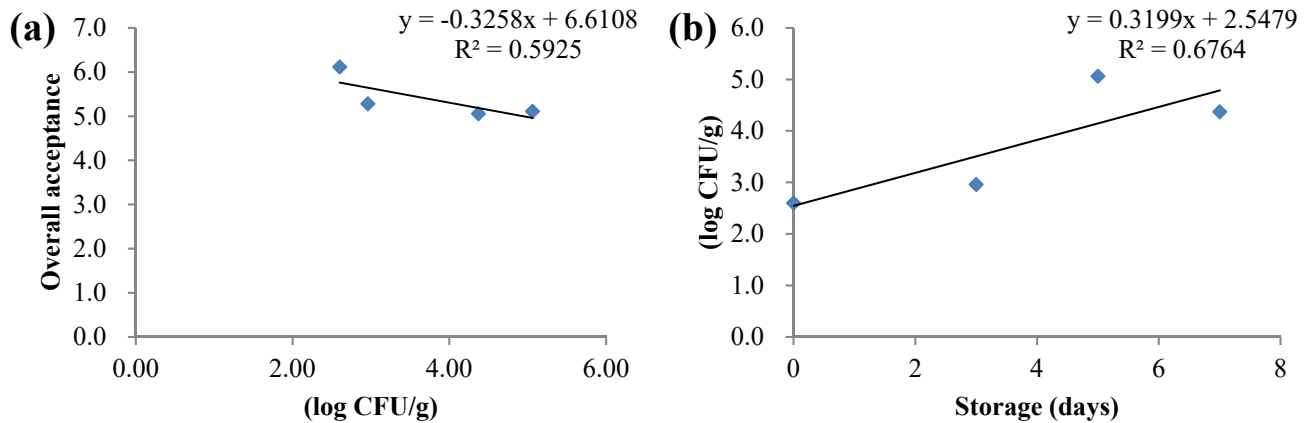


Fig. 3. Regression equation for (a) quality limit (using overall acceptance, 5 considered as acceptable) and (b) estimated shelf-life (days) of wrap-packaged dry-aged beef based on yeast count (log CFU/g).

How calculated:

- 1) $y = 4.9$ (see Fig. 3a).
- 2) substitute 4.9 (y) to the formula ($y = 0.3199x + 2.5479$) and calculate estimated shelf-life (x).

4. Lactic acid bacteria (LAB): Estimated shelf-life 6.3 days

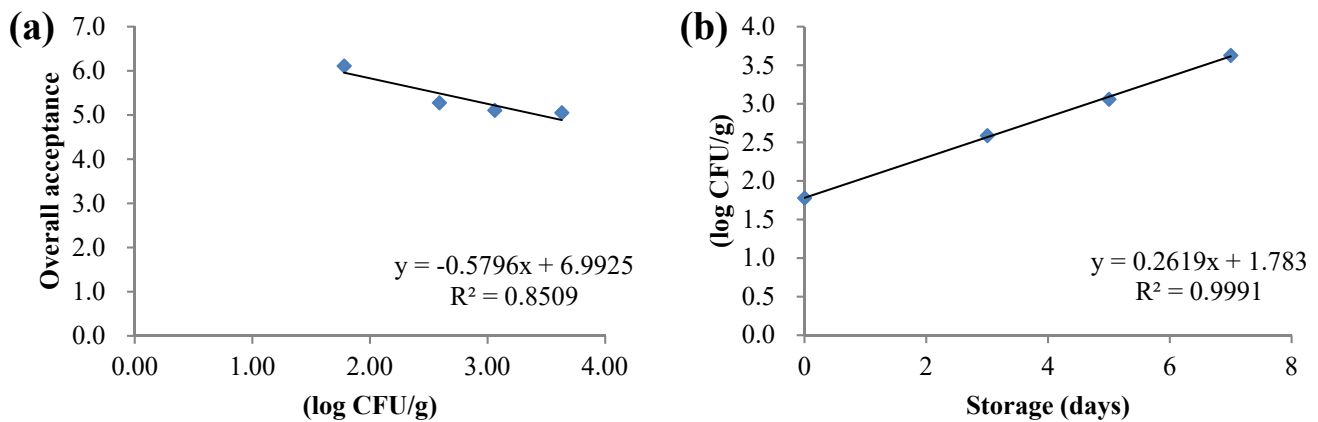


Fig. 4. Regression equation for (a) quality limit (using overall acceptance, 5 considered as acceptable) and (b) estimated shelf-life (days) of wrap-packaged dry-aged beef based on lactic acid bacteria count (log CFU/g).

How calculated:

- 1) $y = 3.4$ (see Fig. 4a).
- 2) substitute 3.4 (y) to the formula ($y = 0.2619x + 1.7830$) and calculate estimated shelf-life (x).

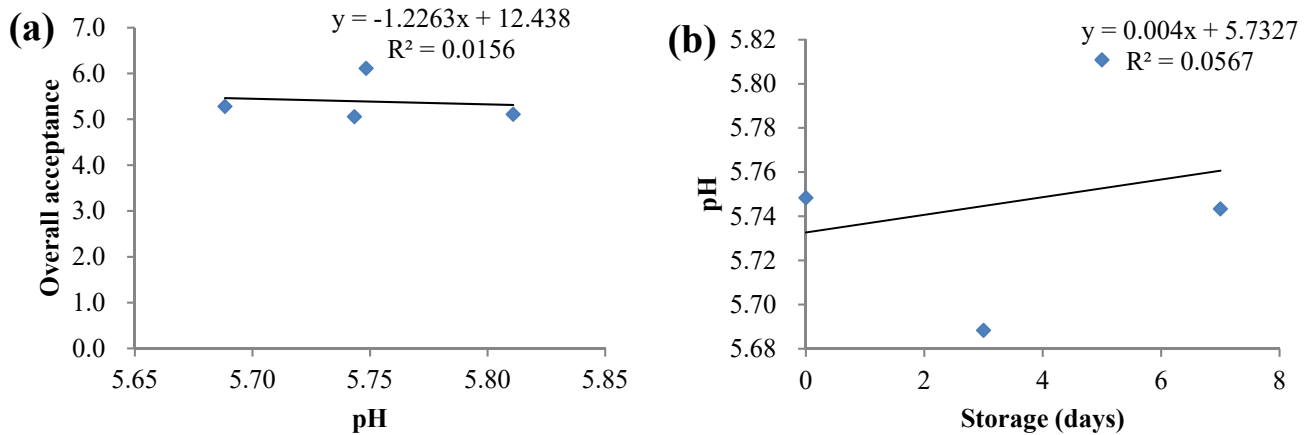
5. pH: Estimated shelf-life 84.3 days

Fig. 5. Regression equation for (a) quality limit (using overall acceptance, 5 considered as acceptable) and (b) estimated shelf-life (days) of wrap-packaged dry-aged beef based on pH.

How calculated:

1) $y = 6.07$ (see Fig. 5a).

2) substitute 6.07 (y) to the formula ($y = 0.0040x + 5.7327$) and calculate estimated shelf-life (x).

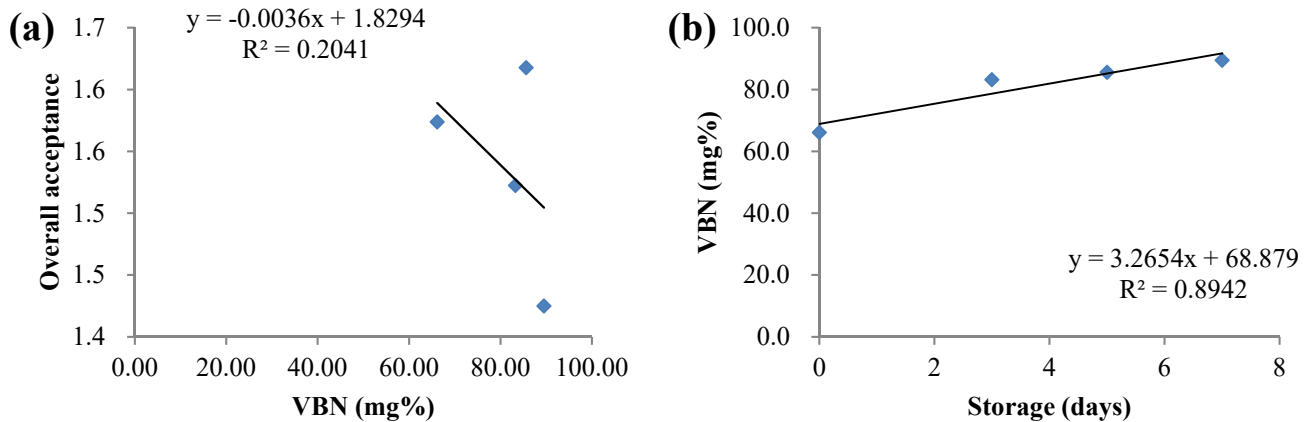
6. Volatile basic nitrogen (VBN): Estimated shelf-life 6.3 days

Fig. 6. Regression equation for (a) quality limit (using overall acceptance, 5 considered as acceptable) and (b) estimated shelf-life (days) of wrap-packaged dry-aged beef based on volatile basic nitrogen (VBN) content (mg%).

How calculated:

1) $y = 89.31$ (see Fig. 6a).

2) substitute 89.31 (y) to the formula ($y = 3.2654x + 68.8790$) and calculate estimated shelf-life (x).

7. 2-thiobarbituric acid-reactive substance (TBARS): Estimated shelf-life 16.4 days

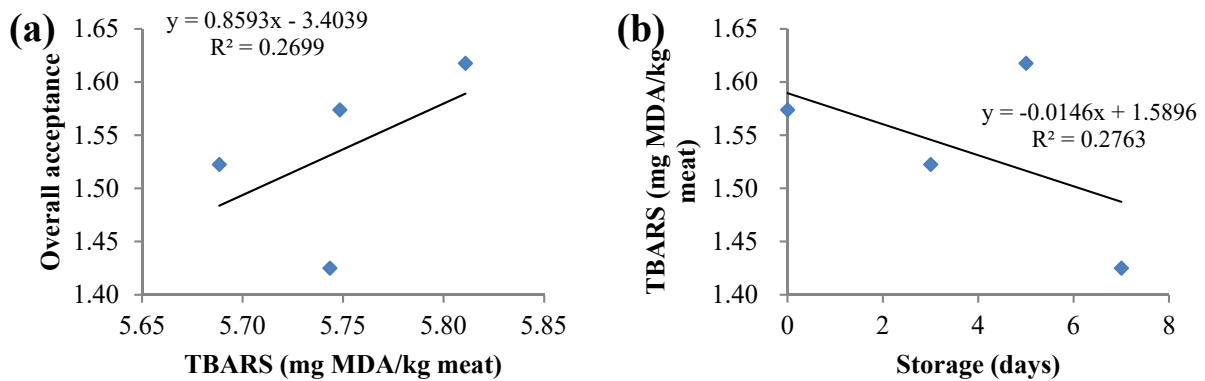


Fig. 7. Regression equation for (a) quality limit (using overall acceptance, 5 considered as acceptable) and (b) estimated shelf-life (days) of wrap-packaged dry-aged beef based on 2-thiobarbituric acid-reactive substance (TBARS) value (mg malondialdehyde/kg meat).

How calculated:

- 1) $y = 1.35$ (see Fig. 7a).
- 2) substitute 1.35 (y) to the formula ($y = -0.0146x + 1.5896$) and calculate estimated shelf-life (x).

8. Sensory evaluation: Estimated shelf-life 15.8 (appearance), 6.7 (odor), 7.4 (taste), and 6.3 days (overall acceptance)

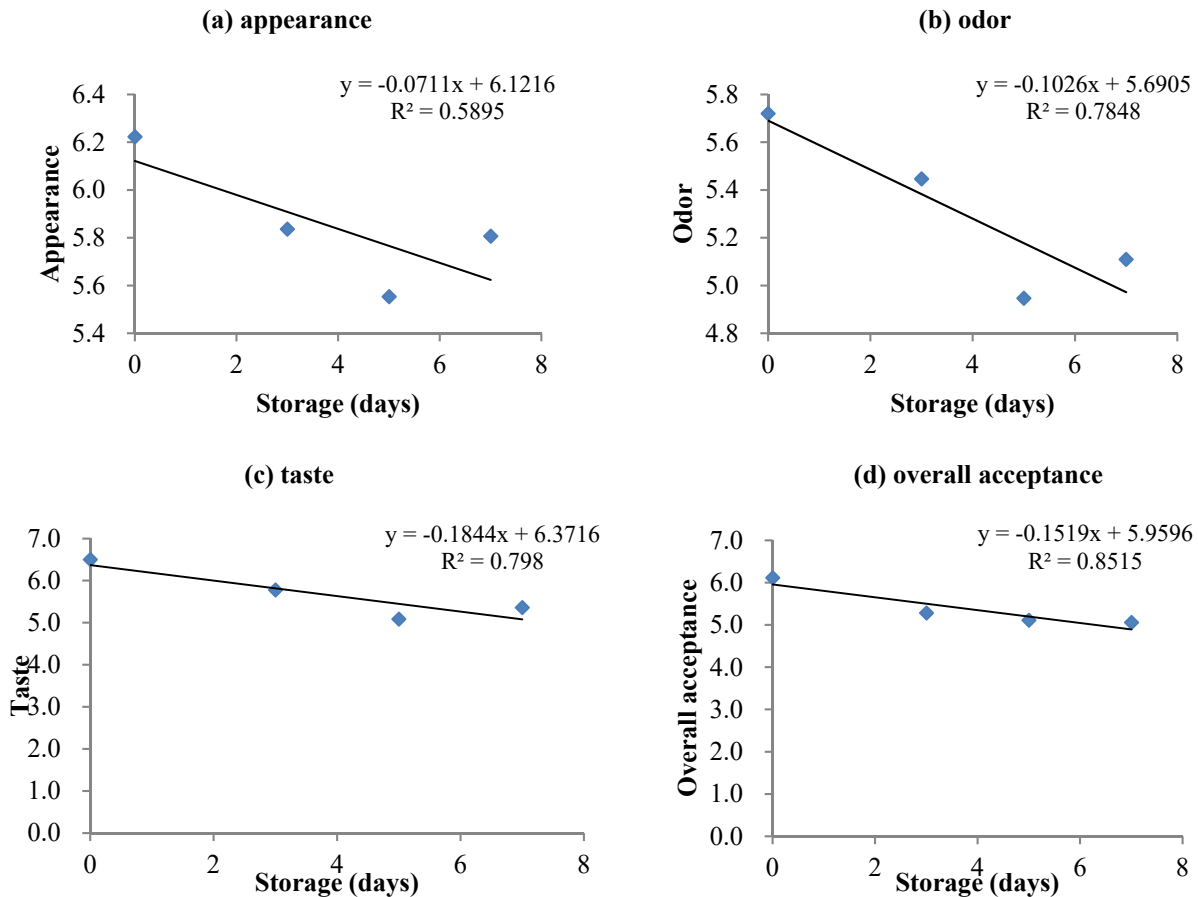


Fig. 8. Regression equation for estimated shelf-life (days) of wrap-packaged dry-aged beef based on (a) appearance, (b) odor, (c) taste, and (d) overall acceptance.

How calculated:

- 1) $y = 5$ (considered as acceptable).
- 2) substitute 5 (y) to the each formula (see Fig. 8) and calculate estimated shelf-life (x).