Master's Thesis of Agriculture

Establishment of aging indicator for dry-aged beef

건식 숙성 소고기를 위한 숙성 지표 확립

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List of abbreviations

ADP	Adenosine Diphosphate
ATP	Adenosine Triphosphate
DC	Direct Current
pH_u	Ultimate pH

Chapter 1. General Introduction

The meat quality can be defined by various categories: organoleptic, ethical and social, symbolic and cultural, nutritional, functionality, and many other factors including safety, availability, and price (Bekhit *et al.*, 2014b). Organoleptic (sensory or eating) quality consists of color, flavor, juiciness and tenderness. It is mainly considered in the purchase of consumers (Aaslyng *et al.*, 2003).

Meat color is the first criterion for consumers to judge the quality of meat. Consumers use meat color as an indicator of spoilage and freshness (Faustman and Cassens, 1990; Mancini, 2009). The myoglobin, major meat pigment, and their redox state of meat determine the meat color (Mancini and Hunt, 2005). As food flavor is a principle factor of consumer purchasing, the flavor of meat is also one of the important determinant of meat quality (Spanier *et al.*, 2004). It is reported that cooked meat flavor is very complex and can involve a lot of factors: free amino acids, fatty acids, reducing sugars, and nucleotides and the volatile compounds generated through Maillard reaction (Kook *et al.,* 2009: Mottram, 1998). Meat juiciness means the feeling of moisture in

the mouth when chewing meat which shows a close relationship with water holding capacity (Moon, 2002). It is highly related to tenderness by helping the chewing process as well as to flavor by bring the flavor component to the taste buds (Aaslyng *et al.*, 2003).

Above all, tenderness of meat is the most important factor. Consumers feel satisfaction of eating meat when their meat is tender, thus consider tenderness as a preferred criterion when repurchasing meat (Bekhit *et al.*, 2014b). There are a number of factors that make meat tough, but the following factors are mainly mentioned.

The tenderness of meat depends on the structures and composition of skeletal muscle (muscle fibers and intramuscular connective tissues). Form of myofibrils is called 'actomyosin toughness' and composition of intramuscular connective tissues are called 'background toughness' (Nishimura, 2010). As an example, the fiber hypertrophy by maturing causes the non-reducible cross links between the collagen molecules that increase the toughness of meat (Fang *et al.*, 1999). The cold shortening of muscle during postmortem causes toughening of meat. There are a lot of metabolic and structural changes through rigor mortis after slaughter. If carcass temperature falls below 10-15℃ before ATP

is completely depleted in muscle, the rigor bond of myosin head and actin retained by residual ATP is associated with an increase in toughness (Geesink *et al.*, 2000). The pH_u and tenderness are highly correlated. The meat with a high pH_u is dark, firm and dry as a result of low glycogen and lactate (Wulf *et al.*, 2002). And a low pH_u meat has poor eating quality because the low pH_u inhibit the postmortem tenderization by internal enzymes (Van Laack and Kauffman, 1999).

Since the meat tenderness is an important factor of consumer buying decision, the meat industry is trying to develop technology to improve tenderness of meat (Bekhit *et al.*, 2014b). According to Bekhit *et al.* (2014b), the tenderness improvement techniques are mainly classified into physical, chemical, and enzymatic practice. Physical tenderness improvement techniques include electrical stimulation of carcasses, aging, freeze-thaw cycles, pressure treatments, mechanical tenderization, and contraction-prevention. These techniques cause structural change in the meat through applying physical force (Bekhit *et al.*, 2014a). Chemical tenderness improvement techniques include infusion, marinating or injection with chemical compounds such as salts. These techniques can improve the tenderness of meat by improving proteolysis,

solubility of myofibrillar proteins and water holding capacity (Bekhit *et al.*, 2014a). Enzymatic tender improvement techniques use plant proteases (ficin, bromelain, papain, actinidin, and zingibain) instead of chemical compounds. These enzymes act on myofibrillar proteins and connective tissues to break down muscle structures (Bekhit *et al.*, 2014a).

Many studies have attempted to explain and improve the tenderness of meat. Among the techniques to tenderize meat, the meat aging received the attention of consumer recently because there are no changes in appearance of meat and no use of artificial additives. The aging is also a state-of-art technology by strictly controlled and specialized facilities with time, temperature, and humidity factors. Some commercial aging processors also control microorganisms and drying rate to maximize efficiency and quality level. The meat aging is a technique to improve the eating quality using only protein hydrolytic enzymes of meat. However, because the meat aging technique requires a lot of experience and trained skills, there are needs to simplify the meat aging processing. Recently, small meat benders start to see the aged-meat market, but encounter difficulties for indication of completion of aging.

Chapter 2. Literature Review

1. Aging

Aging is the technique for improving the eating quality of meat by storing at refrigerated temperatures for specific period. This improvement has been known and studied for decades (Koohmaraie *et al.*, 1994). According to Khan *et al.* (2016), aging is the process during or after the muscle is converted into meat, accompanied by various changes such as proteolysis, lipolysis and oxidation. The many changes during postmortem affect the meat quality; tenderness, juiciness and flavor. In order to have competitiveness in the market, the meat industry has used the aging technique that improves the eating quality which has the greatest impact on consumer meat purchasing.

1.1. Meat toughness

The conversion of muscle to meat begins immediately after slaughter. In the early postmortem, muscle glycogen metabolized by anaerobic glycolysis phosphorylates ADP to supply ATP. Due to the accumulation of lactate formed by anaerobic glycolysis, the

intracellular pH is decreased and reached about 5.4−5.7 by 24 h postmortem (Maltin *et al.*, 2003). During pH decrease, the muscles continue to consume ATP as the myosin heads and actin repeat their binding and separation. The ATP decreases to a level that locks myosin heads to actin and form inextensible actomyosin causing the sarcomere shorten. This stiffness of muscles is rigor mortis (Jiang, 1998).

During the development of rigor mortis, the postmortem changes and condition of meat determine meat texture (Bekhit *et al.*, 2014b). If the muscle temperature falls below 10−15℃ in the early postmortem period with 6.0−6.4 pH and sufficient residual ATP, the muscle will shorten and lead toughening (Maltin *et al.*, 2003). Meat tenderness is affected by both slaughter condition and the postmortem storage condition.

1.2. Postmortem aging

Postmortem aging is initiated by the endogenous proteolytic systems in muscle including the calpain−calpastatin system, the cathepsin−lysosomal system and the matrix metalloproteinases (Maltin *et al.*, 2003).

The calpain system is consisted of μ− and m−calpain (Goll *et*

al., 1992: Huff-Lonergan and Lonergan, 1999). The μ-calpain is active in the early stage of proteolysis, acting on the costameres, intermediate filaments and the sarcolemmal membrane in the first 1-2 day postmortem. And m-calpain becomes activated, leading to further degradation of cytoskeletal proteins over the subsequent days (Table 1). The role of calpastatin remains to be elucidated, but the calpastatin is activated to control the action of m-calpain through inhibition (Doumit and Koohmaraie, 1999).

Differing that the calpains proteolysis certain proteins of the Z-line (such as desmin, filamin, and nebulin) and connectin (Huff-Lonergan and Lonergan, 1999: Lusby *et al.*, 1983), cathepsins is consider to proteolysis myosin and actin directly (Jiang *et al.*, 1992, 1996). In addition, they can act on various contractile proteins (Table 1).

The MMP system, including collagenases, stromelysins, and gelatinases, degrades connective tissue proteins. Collagen denatured by collagenases can be degraded to small peptides by gelatinase activity (Seltzer *et al.*, 1981; 1990). According to Sylvestre *et al.* (2002), by increasing active MMP at slaughter or by increasing postmortem conversion of latent MMP into their active forms, the proteolytic activity is increased and the

intramuscular connective tissue is degraded.

The postmortem aging for the conversion of muscle to meat is a complex process in which various endogenous proteolytic systems operate. The proteolytic systems weaken the shortened muscle structure which causes meat toughness and results in a tenderization of the meat (Huff-Lonergan and Lonergan, 1999: Jiang *et al.*, 1992).

Table 1. Sites of proteolytic enzyme action during postmortem aging

Proteolytic Enzyme	Site
Calpains	1. Troponin T (In case of pH > 6)
	2. Z-line (desmin)
	3. Connectin (gap filaments)
	4. M-line proteins
	5. Tropomyosin
Lysosomal enzymes (including cathepsins B, D, H and L)	1. Troponin T, I
	2. C-proteins
	3. Myosin Heavy Chain and Light Chains
	4. Actin
	5. Tripomyosin
	6. Nebulin
	7. Titin
	8. α-Actinin
	9. Collagen
	10. Mucopolysaccharides

(Taken from Jiang, 1998)

2. Aging methods

There are two different aging methods used in beef processing today, wet aging and dry aging (Table 2). In the meat industry, wet aging is commonly used, but dry aging which causes a unique flavor attract the attention of consumer. And this trend have increased the production of dry-aged meat (Bekhit *et al.*, 2014b; Khan *et al.*, 2016: Marchello and Marchello, 2015).

2.1. Wet aging

In the late 1960s, the method which stored carcasses processed into primal or sub-primal in vacuum packaged bags was developed (Gazalli *et al.*, 2013). Because the vacuum packaging of beef is improving sanitation and reduction of shrink, many studies tried to apply the advantages of vacuum packaging to meat aging (Bowling *et al.*, 1977; Gutowski *et al.*, 1979: Minks and Stringer, 1972). In this method, called vacuum aging or wet aging, meat is vacuum packed in an oxygen impermeable packaging bag and stored at refrigerated temperature (Obuz *et al.*, 2014). As well as the wet aging increases the shelf life of meat through the microbial safety, it has low ageing loss and

convenience during storage and transport (DeGeer *et al.*, 2009; Li *et al.*, 2014).

2.2. Dry aging

Dry aging is an ancient food technology of preserving fresh meat. It is still used today for improving meat quality but not for preservation purpose (Marchello and Marchello, 2015). Dry aging is the tenderizing process which keeps the meat at refrigerated temperature and controlled humidity without vacuum packaging (Dikeman *et al.*, 2013; Khan *et al.*, 2016). Therefore, the storage environment such as temperature and humidity have a great influence on the aged meat quality, so that this aging method requires cautions for environmental control (DeGeer *et al.*, 2009; Li *et al.*, 2014). In addition, the dry-aged meat that is unprotected and exposed to air has a high risk of microbial contamination. During dry aging process, a large amount of water evaporation occurs in the meat which increases weight loss at aging and trimming (Stenström *et al.*, 2014). Despite these disadvantages, some consumers prefer the dry-aged beef than the wet aged beef because they feel that dry-aged beef have a more beefy, brown roasted flavor than wet aged one (Dikeman *et*

al., 2013: Warren and Kastner, 1992). As consumer demand for and interest in dry-aged beef increases, the production and investment of dry-aged beef in the meat industry is increasing (Marchello and Marchello, 2015).

Table 2. Summary of aging methods

	Aging methods	
	Dry aging	Wet aging
Packaging	Un-packaged	Vacuum-packaged
Environmental condition	In controlled temperature, humidity, and air flow	In controlled only temperature
Microbial safety	Possible risk of contamination	Excellent safety
Yield	Low	High
Cost	High	Low

(Modified from DeGeer *et al.*, 2009; Dikeman *et al.*, 2013; Li *et al.*, 2014; Marchello and Marchello, 2015: Stenström *et al.*, 2014)

3. Quality of aged meat

The main purpose of aging is improving the tenderness, flavor and juiciness of meat (Irurueta *et al.*, 2008; Khan *et al.*, 2016). During the aging period, the activated enzymes of the endogenous proteolytic systems in muscle improve tenderness of meat by breaking down the muscle structure (Jiang *et al.*, 1992; Maltin *et al.*, 2003). Many studies have shown improvement of meat tenderness through both dry aging and wet aging (Table 3) (Campbell *et al.*, 2001; Parrish *et al.*, 1991; Sitz *et al.*, 2006: Warren and Kastner, 1992).

As the aging processed, the flavor related substances of meat, such as sugars, free fatty acids, free amino acids, and nucleotides, increased (Koutsidis *et al.*, 2008; Lee *et al.*, 2015). Proteolytic and lipolytic enzymes activated during aging not only break down muscle structures but also produce various by-products, which are flavor-related substances (Gorraiz *et al.*, 2002). Campo *et al.* (1999) confirmed that sensory evaluation panelists felt the intensity of the meat flavor increased as the aging period increased. Especially, Dry-aged meat has a more intensified flavor, such as a beefy and roasted flavor, than wet aged meat (Warren and Kastner, 1992). The reason for the stronger flavor

of dry-aged meat can be explained by the oxidation by exposing to air during the aging period (Khan *et al.*, 2016). Kim *et al.* (2016) confirmed that glutamate and other free amino acids of dry-aged meat were higher abundance than wet-aged and suggested that this result play a more important role in influencing consumer preference in beef flavor.

The color of wet aged meat has brighter than unaged meat (Caldwell *et al.*, 2017; Gašperlin *et al.*, 2001). According to Gašperlin *et al.* (2001), this is because the proteolytic enzymes of aging affect certain proteins associated with color. And the physicochemical changes of muscle by aging are attributed to the oxygenation of myoglobin. As a result, meat aging inhibits the meat color stability. However, dry-aged meat had a darker color than wet aged meat or unaged meat (Kim *et al.*, 2016: Obuz *et al.*, 2014). The less moisture content of dry-aged meat due to evaporation during aging has a low light reflection ability and dark color (Kim *et al.*, 2016).

Table 3. Quality comparison of dry- and wet-aged beef

Parameters	Dry aging	Wet aging
Tenderness	Improved (Campbell *et al.*, 2001; Sitz *et al.*, 2006: Warren and Kastner, 1992)	Improved (Parrish *et al.*, 1991; Sitz *et al.*, 2006; Warren and Kastner, 1992)
Flavor	Flavorsome (King *et al.*, 1995; Campbell *et al.*, 2001: Warren and Kastner, 1992)	Mostly unchanged (King *et al.*, 1995)
Color	Dark red (Kim *et al.*, 2016: Obuz *et al.*, 2014)	A little bright red (Caldwell *et al.*, 2017: Gašperlin *et al.*, 2001)

(Modified from Khan *et al.*, 2016)

Chapter 3. Evaluation of aging parameters for dry-aged meat

1. Abstract

The purpose of this study is to screen and validate the indicators that can determine the aging degree of dry-aged beef during dry aging periods. Two experiments were performed as below.

Experiment I: In order to select the candidate of indicator, various physicochemical changes occurred in the meat during the wet aging and the dry aging period were measured. With moisture content, wet aging did not have a significant relationship, but dry aging decreased significantly with increasing aging period ($P<0.05$). The pH did not show any tendency in both dry and wet aging depending on the aging period. Because only in dry aging, the mold grew and the distinctive aging odor was stronger, the mold distribution and odor were analyzed only in dry-aged beef. The quantified mold distribution of dry-aged beef was

significantly increased with the dry aging period. The dry aging flavor analyzed by electronic nose was also classified according to the aging period by principal component analysis (PCA).

Experiment II: The moisture content, the electric resistance, and the mold distribution on dry-aged beef surface were selected as candidates for indicator of dry aging. The relationship between selected aged indicator candidates and the quality change of dry-aged beef during aging period was confirmed. The selected indicator candidates showed a high correlation with aging period ($P<0.05$). The moisture content, pH, color (L^*, a^*, and b^*), and shear force of internal meat were, the remainder after removing external crust, measured to determine the change of beef quality according to aging period. As the aging period increased, the moisture content and shear force tended to decrease and the pH tended to increase ($P<0.05$). But the color had no particular tendency.

In order to confirm the improvement of the beef quality by dry aging, the sensory evaluation of consumer acceptability was carried out by aging period and it was confirmed that the odor, taste, flavor, tenderness, juiciness, and overall acceptance were

significantly improved with increasing aging period ($P<0.05$). Moisture content, electrical resistance and mold distribution on the surface of dry-aged beef surface were highly correlated with moisture content, pH, and shear force of internal meat during dry aging ($P<0.05$). In the sensory evaluation, the moisture content of the dry-aged beef surface showed a close relationship with odor, taste, flavor, tenderness, juiciness, and overall acceptance ($P<0.05$). The electrical resistance was closely related to tenderness, and mold distribution was significantly related to flavor and tenderness ($P<0.05$). From the results, it can be concluded that moisture content, electrical resistance, and mold distribution of surface of beef could be good candidates of the indicator for measurement of doneness of dry-aged beef.

2. Introduction

The tenderness of meat is an important criterion for consumers to purchase meat (Shackelford *et al.*, 2001). Thus, many meat researchers and producers have applied several techniques to improve the meat tenderness (Bekhit *et al.*, 2014b). One of the techniques, meat aging, is usually divided into two methods, wet and dry aging. Dry aging is a traditional aging method that has been used before wet aging (Marchello and Marchello, 2015). In the last decades, due to the convenience of storage and transportation, the wet aging is used more than the dry aging in the meat industry (Minks and Stringer, 1972). However, in recent years, the unique flavor of dry-aged beef has caused consumers' interest and consumption. Especially, there is a great interest and popularity in dry-aged beef in Asia such as Korea, Japan, Singapore, Taiwan and China (Dashdorj *et al.*, 2016). Due to the fact, the number of restaurants and butcher shops implementing dry aging technique to their products is increasing.

On the other hand, the dry aging technique requires careful management because the beef is exposed to the air during aging

and has the quality changes according to the external environmental parameters. During aging, the external environmental parameters, such as temperature, relative humidity, and airflow, should be carefully controlled to inhibit microbial growth and produce the optimal meat quality (Baird, 2008; Dashdorj et al., 2016; Kim et al., 2016; Perry, 2012; Savell, 2008). However, there is no prescribed environmental condition for dry aging. Many studies have conducted dry aging using the environmental conditions including temperature, relative humidity, and airflow (Table 4).

Table 4. Previous results for the conditions of temperature, relative humidity, and air flow for dry aging

	Temperature (°C)	Relative humidity (%)	Air flow (m/s)
Parrish *et al.* (1991)	0−1	80−85	0.5−2.5
Warren and Kastner (1992)	3.1−3.6	78	NR[1]
Campbell *et al.* (2001)	2	75	NR[1]
Kim *et al.* (2016)	3	49	0.2

[1]Not reported.

Because the dry aging depends on the experience of the expert and must be managed with extreme care, many dry aging performers experience aging failure and difficulty in uniform quality control of dry-aged beef. Therefore, it is helpful to propose an easy and optimal method to guarantee suitable degree of dry-aged beef for producers.

During dry aging, microorganisms grow on the surface of the beef. In this circumstance, the growth of pathogenic or spoilage bacteria in the microorganisms should be limited and the growth of beneficial fungi should be encouraged (Dashdorj *et al.*, 2016). Several mold species, *Thamnidium elegans*, *Mucor mucedo* and *Chaetostylum fresnii*, were found on the surface of dry-aged beef (Kotula *et al.*, 1982). In particular, *Thamnidium elegans*, which can grow at refrigeration temperature, is known to improve the tenderness and flavor of the aged beef because it has proteolytic and lipolytic enzymes which break down the muscle structure (Dashdorj *et al.*, 2016). During dry aging, *Thamnidium elegans* grows in the visible form of patches with grayish-white hairs, called "whiskers" (Kotula *et al.*, 1982; Dashdorj *et al.*, 2016). The area where white mold grow on the surface of meat is significantly increased with increasing dry aging period.

According to Garcia *et al.* (2001), a mold coating on dry fermented sausages is considered as an indicator of good quality and completion of the ripening process. Therefore, the area of white mold can be considered as an indicator for dry aging.

Dry aging causes moisture loss due to evaporation on the surface of meat and the surface becomes dry and hard. This inedible surface is called “crust” (Adegoke and Falade, 2005). As the dry aging period increases, the surface of the meat is more hardened and shrunk. Therefore, the nondestructive measurement of significant moisture change on the aged meat surface during dry aging can be an effective indicator for dry aging. In addition, there is an electrical moisture measurement method through measuring the DC resistance which is nondestructive and easy to use (Gillespie and Kidd, 1978). In wood industry, the measurement of moisture contents through the DC resistance of wood become a common method since it has been confirmed that the DC resistance of wood increases with decrease of the wood moisture content (Fredriksson, 2010). In recent years, the moisture measurement has been attempted through measuring the DC resistance in wheat flour, potato starch and raw pork loin by Domagała and Rywotycki (2000). Therefore,

the resistance value for measuring the moisture content of the aged meat surface, which is expected to have a close relationship with the dry aging degree, can be used as an indicator for dry aging.

Dry aging not only changes the appearance of beef, such as the growth of microorganisms or the formation of crust, but also changes the flavor of beef. According to Campbell *et al.* (2001), many dry aging practitioners believe that dry aging enhances the flavor of beef, and his research has also found that the dry-aged beef had stronger beef flavor and brown roasted aromas than wet aged beef. The flavor changes of aged beef during dry aging were expected to be an indicator for the degree of aging. The electronic nose is mentioned as an effective device to analyze flavor or aroma (Schaller *et al.*, 1998). There were various attempts to evaluate the quality of meat products using electronic nose (Blixt and Borch, 1999; Ghasemi-Varnamkhasti *et al.*, 2009: Winquist *et al.*, 1993).

The indicators should be able to express the degree of dry aging clearly nondestructive to aged meat, and easy to use for unskilled people. Considering these criteria, the selected dry aging indicators for present study are the diffusion degree of

mold on the surface of dry-aged beef, the changes in electrical resistance of the crust, and the flavor changes of untrimmed beef surface during dry aging. The purpose of this study is to confirm whether the use of selected aging indicators in possible and to investigate that the selected aging indicators can express the degree of dry aging by confirming a meaningful relationship with the quality of dry-aged meat.

3. Materials and Method

3.1. Search for dry aging parameters

3.1.1. Sample preparation and aging process

A total of 30 strip loins (*longissimus lumborum*) were obtained from Holstein steers (quality grade 3) in Korea. Half of them were used for wet aging and the other half were used for dry aging (n=15 for each aging method). During aging, 3 strip loins which aged in different aging methods were used for analysis at aging days 0, 7, 14, 21 and 28. For the wet aging, the strip loin samples were vacuum packaged in oxygen impermeable nylon bags (2 mL $O_2/m^2/24$ h at 0°C, 0.09 mm thickness; Sunkyung Co. Ltd, Seoul, Korea) and stored at 2 ± 1°C. The strip loins for dry aging were unpackaged and stored in the dry aging condition as follows; 2.5 m/s of air velocity, 2 ± 1°C of temperature and 75 ± 10% of humidity. For analyzing the distribution of mold, the surfaces of aged strip loins were photographed. For the other analyses, the crust or excessive fat of aged strip loins were trimmed off and samples stored at −70°C

in vacuum packaging.

3.1.2. Moisture contents

The moisture contents were determined as outlined by AOAC (2005). The ground meat samples (3 g) from the surface of dry-aged beef were dried in a dry oven (DS-5020L, Daewon sci., Bucheon, Korea) at 110℃ for 16 hours. And the moisture contents were measured at a ratio of the weight difference before drying and after drying.

Moisture content (%)

$$= \frac{[\text{Weight before drying (g)} - \text{Weight after drying (g)}]}{\text{Weight before drying (g)}} \times 100$$

3.1.3. pH

Each meat sample (1 g) from surface of dry-aged beef was homogenized with 9 mL of distilled water using a homogenizer (T10 basic, IkaWorks, Staufen, Germany) for 30 s. The homogenates were centrifuged (Continent 512R, Hanil Co., Ltd, Incheon, Korea) at 2,265×g for 10 min and filtered by filter paper (Whatman No. 4, Whatman PLC., Buckinghamshire, UK). The pH value of each filtrate was measured using a pH meter (SevenGo, Mettler-Toledo International Inc., Schwerzenbach,

Switzerland), which was calibrated by standard buffers (pH 4.01, 7.00, and 9.21).

3.1.4. Mold distribution

The surface of aged strip loin were photographed in a camera obscura made of 58×40×40 cm size. The digital camera used for photographing was CMOS 16.0 MP, Samsung Galaxy S6 smartphone and the resolution of camera was 16M (screen ration, 16×9; pixel number, 5,312×2,988). A LED lighting (MS-273, Myung Sung, Daegu, Korea) with 108 lux was installed in the camera obscura for constant illumination. The Adobe Photoshop CC 2015 software (Adobe Systems Inc., California, USA) was used to analyze the mold distribution from digital photographs of aged meats. A total of two steps were carried out for the analysis of mold distribution (Figure 1). The first step was to remove unnecessary background and fat from the surface of the meat that can interfere the analysis. The second step was to determine the range of pixels to be analyzed and perform analysis using the histogram function. The ratio of pixels over 128 levels in the blue channel of histogram was considered as the distribution of mold.

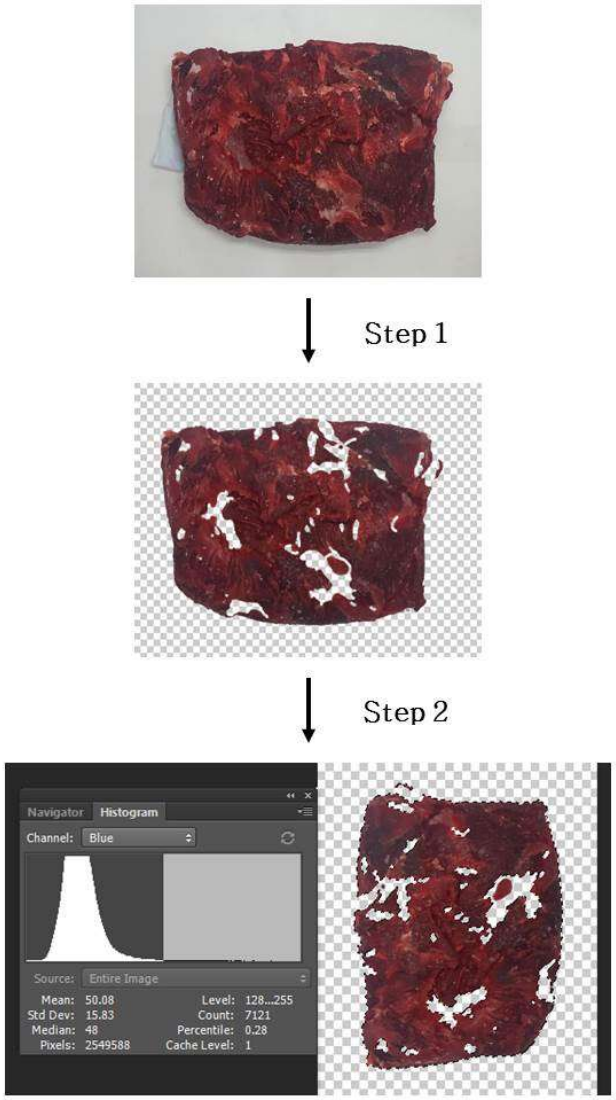


Figure 1. Steps for analyzing mold distribution using Adobe Photoshop CC 2015 software.

3.1.4. Electronic nose

The crust of dry-aged beef (5 g) from dry-aged beef surface was ground, and placed in 20 mL headspace vials. The vials were placed in an electronic nose instrument and analyzed. The instrument used to analyze the aroma of aged meat crust was Heracles II electronic nose (Alpha M.O.S., Toulouse, France) based on ultrafast gas chromatography. The instrument consists of an auto sampler (HS100), an injector containing two columns (MXT-5 and MXT-1701), and a main body containing two flame ionization detectors. The analyzing conditions were follows: incubation temperature, 80℃; incubation time, 600 s; injected volume, 500 μL; injected speed, 250 μL/s; injector temperature 200℃; trap initial temperature, 40℃; split, 10 mL/min; trapping duration, 30 s; trap final temperature, 240℃; column initial isotherm, 40℃(5 s); column temperature program, up to 150℃ (0.5℃/s, 5 s) and up to 260℃ (5℃/s, 30 s); column acquisition duration, 282 s. The analysis was repeated three times for the principal component analysis (PCA), and retention indices of the constituents were determined by the Kovats index based on the AroChembase (Alpha M.O.S., Toulouse, France).

3.1.6. PCA analysis

PCA was used to analyze the results of electronic nose and was performed in Alpha Soft (V12.4, Alpha M.O.S, Toulouse, France). Principal component is a statistical procedure that uses orthogonal transformation to convert a set of observations of possibly correlated variables into a set of values of linearly uncorrelated variables called principal components (Peng et al., 2015). Principal components are guaranteed to be independent if the data set is jointly normally distributed. PCA is sensitive to the relative scaling of the original variables (Abdi and Williams, 2010).

3.1.7. Statistical analysis

All experiments were performed in triplicate. The data were analyzed using the SAS statistical software program (SAS, Release 9.4, SAS Institute Inc., Cary, NC, USA). The general linear model was applied to investigate the changes of aging indicator candidates (mold distribution, moisture, and pH) during aging period. The results were reported as mean values with standard error of the means (SEM). Significant differences among the mean values were determined on the basis of the Student

Newman-Keuls multiple comparison test at a level of $P<0.05$. The relationship between aging day and aging indicator candidates (mold distribution, moisture, and pH) was evaluated using correlation coefficients.

3.2. Analysis of correlation between dry aging parameters and dry-aged beef quality factors

3.2.1. Sample preparation and aging process

A total of 18 strip loins (*longissimus lumborum*) were obtained from Holstein steers (quality grade 3) in Korea. All of them were used for dry aging. During aging, 3 strip loins were used for analysis at aging days 0, 7, 14, 21, 28 and 35. The strip loins for dry aging were unpackaged and stored in the dry aging condition as follows; 2.5 m/s of air velocity, 2 ± 1°C of temperature and 75 ± 10% of humidity. Before the strip loins were trimmed off, the mold distributions, the DC electrical resistances and the surface color were measured. Then for the other analyses, the crust or excessive fat of aged strip loins were trimmed off. The trimmed strip loins and the crust produced by trimming were separately stored at −70°C after vacuum

packaging.

3.2.2. Moisture of the surface crust and internal meat of dry-aged beef

The moisture measurement of dry-aged beef surface crust and internal meat was performed in the same method as 'Experiment I. Search for dry aging parameters'. See section 3.1.2.

3.2.3. Mold distribution

The mold distribution measurement was performed in the same method as 'Experiment I. Search for dry aging parameters'. See section 3.1.4.

3.2.3. Electrical resistance

The electrical resistance of aged meat surface was measured by using an electric resistance meter (SH-3234, Saehan tester co., Busan, Korea). The measurement site was on the surface of lean meat excluding fat and connective tissue and the distance between (+) electrode and (-) electrode was set at 1 cm.

3.2.4. pH

The pH measurement was performed in the same method as 'Experiment I. Search for dry aging parameters'. See section 3.1.3. Samples were taken from internal meat of the dry-aged beef.

3.2.4. Color

After trimming, the color values of internal surface (International Commission on Illumination L^*, a^*, and b^* values representing lightness, redness, and yellowness, respectively) of the aged strip loins were measured using a colorimeter (CR-5, Minolta Camera Co., Japan). Before analysis, a standard black and white plate was used for calibrating the colorimeter. The average of the measured values at three different positions in each sample was used.

3.2.5. Shear force

The trimmed strip loin (100 g) were vacuum-packaged and cooked in a water bath at 85℃ for 40 min to achieve a core temperature of approximately 72℃ or above. Samples were then cooled in the iced water for 15 minutes and 6 core samples (a

diameter of 1.27 cm) were taken in the longitudinal direction of muscle fibers. The shear force of each was measured using a Warner-Bratzler blade on a texture analyzer (AMETEK Lloyd Instruments Ltd, Fareham, UK) with the following parameters: maximum cell load, 10 kg; target load, 10 g; target value, 25 mm; and target speed, 2.0 mm/s. The samples were sheared perpendicularly to the direction of the muscle fiber.

3.2.8. Sensory evaluation

The sensorial quality of samples was evaluated by an untrained consumer panel (8 sensory panelists). The sensory evaluation was carried out three times by the same 8 panelists for the samples of each aged day. The sensory panelists evaluated the sensorial quality of aged strip loins by six aging periods (0, 7, 14, 21, 28, and 35 days). The frozen samples were thawed at 4℃ for 16 h before evaluation, cooked in a conventional oven set at 177℃ until the core temperature of samples reached 72℃, and cut into pieces of a similar size (30×30×10 mm). Each sample was scored on a single sheet using a 9 points hedonic scale (1 = dislike extremely, 9 = like extremely). The sensory evaluation was performed based on four individual traits: appearance, odor,

taste, flavor, juiciness, tenderness, and overall acceptance. The mean scores of the 8 panelists were used for statistical analysis.

3.2.9. Statistical analysis

All experiments were performed in triplicate. The data were analyzed using the SAS statistical software program (SAS, Release 9.4, SAS Institute Inc.). The general linear model was applied to investigate the changes of aging indicator candidates (moisture, mold distribution, and electrical resistance) and meat quality (moisture, pH, color, shear force, and sensory evaluation) during aging period. 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 $P<0.05$. The relationship between aging indicator candidates and meat quality (moisture, pH, color, shear force, and sensory evaluation) was evaluated using correlation coefficients.

4. Results and Discussion

4.1. Search for dry aging parameters

4.1.1. Moisture contents

The moisture contents of the beef surface, which is aged by different methods and different aging period, are present in Table 5. On all days except for day 0, the moisture contents of wet aged beef were significantly higher than those of dry-aged beef ($P<0.05$). In dry aging, as the aging period increased, the moisture content decreased significantly ($P<0.05$). In wet aging, the moisture contents were different according to aging period, but there was no specific trend. The dry aging method, which exposes meat to air, causes evaporation on the meat surface causes of continuous moisture loss during dry aging (Juárez *et al.*, 2011). Obuz *et al.* (2014) also confirmed the same results and tendency in his study about aging method and aging time interaction on moisture content of *Longissimus lumborum* cull cow steaks. There was no correlation between wet aging period and moisture content, but that between moisture content and dry

aging period was significant. (R^2=−0.856, P<0.001). This result showed that the change of moisture content has the possibility of an indicator to see the degree of aging when dry aging is applied.

4.1.2. pH

The changes of pH on beef surface according to aging methods and aging period are present in Table 5. At day 7 and 28, pH between dry−aged and wet−aged beef showed significant difference, but there was no specific trend. During dry aging, there were no significant pH changes. But in wet aging, pH increased up to 14 days and then decreased (P<0.05). According to Boakye and Mittal (1993), as the vacuum aging period increased, the pH was increased, which was attributed to proteolytic enzymes during aging. On the other hand, Parrish *et al.* (1991) confirmed that dry−aged and wet−aged beef did not differ in pH, similar to the current experimental results. Also, as a result of correlation analysis (Table 5), pH could not be used as an indicator for the aging degree in both dry and wet aging method.

Table 5. The change of aging indicator candidates on beef surface according to aging methods and aging period

	Aging method	Aging period (day)					SEM[1]	R^2	*P*-Value
		0	7	14	21	28			
Moisture (%)	Dry	72.80^{a}	52.38^{by}	51.03^{by}	46.06^{cy}	45.85^{cy}	0.385	−0.856	<0.001
	Wet	72.80^{a}	70.14^{bx}	68.35^{cx}	67.63^{cx}	72.52^{ax}	0.296	−0.202	0.470
	SEM[2]	0.159	0.351	0.288	0.494	0.339			
pH	Dry	5.51	5.50^{y}	5.58	5.53	5.55^{x}	0.018	0.473	0.075
	Wet	5.51^{c}	5.58^{bx}	5.61^{a}	5.51^{c}	5.44^{dy}	0.003	−0.469	0.078
	SEM[2]	0.009	0.008	0.017	0.019	0.005			

[1]Standard error of means (n= 15), [2](n=9), [3](n=6).

[a−d]The letters within the same row were significantly different (P<0.05).

[x, y]The letters within the same column were significantly different (P<0.05).

4.1.3. Mold distribution

In the case of wet aging, mold could not grow on the surface of beef because the beef was sealed in a vacuum package. Therefore, it was not possible to quantify the mold distribution in wet-aged meat. Figure 2 shows the growth and spread of mold on the beef surface during dry aging. On 28 day of dry aging periods, mold can clearly be seen covering the beef surface (Figure 2). The digital photographs of dry-aged beef were analyzed to quantify the mold distribution and the result expressed as a graph in Figure 3. As the dry aging period increased, the fungal distribution increased significantly ($P<0.05$). There have been no previous studies that have examined mold distribution changes during dry aging. In 1923, Brooks and Hansford summarized the molds that could grow in the cold storage meat and described one of the molds with greyish white tufts, *Thamnidium elegans*. Kotula *et al.* (1988) tried to enhance the rapid dry aging of beef by inoculating *Thamnidium elegans* on the beef surface. The mold with greyish-white tufts shown in this study is expected to grow easily on the surface of low temperature stored beef such as *Thamnidium elegans*. The degree of mold distribution has already been considered as a criterion for judging completion of dry fermentation in fermented sausages (Garcia *et al.*, 2001).

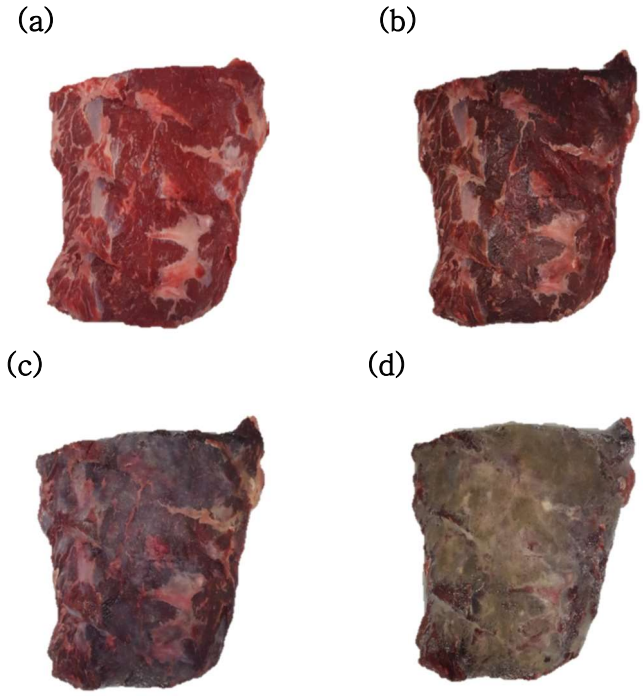


Figure 2. Appearance change of the mold distribution on dry-aged beef by aging period; (a) 7 day, (b) 14 day, (c) 21 day, (d) 28 day.

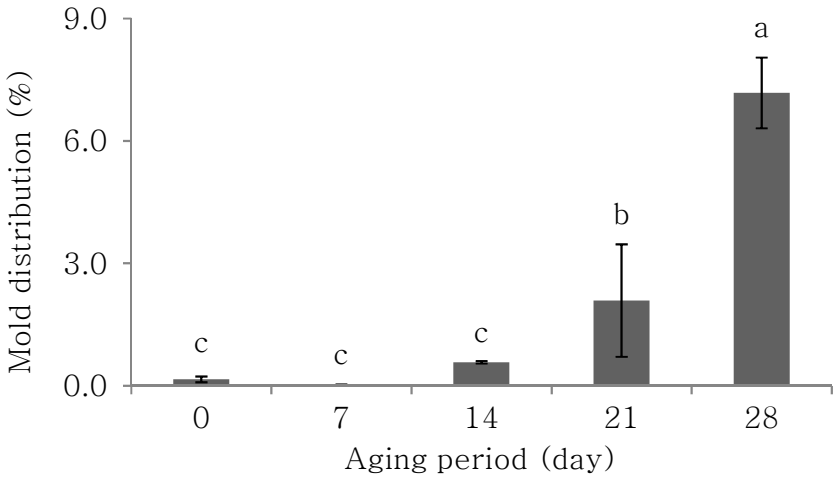


Figure 3. Change of the quantified mold distribution on dry-aged beef by aging period

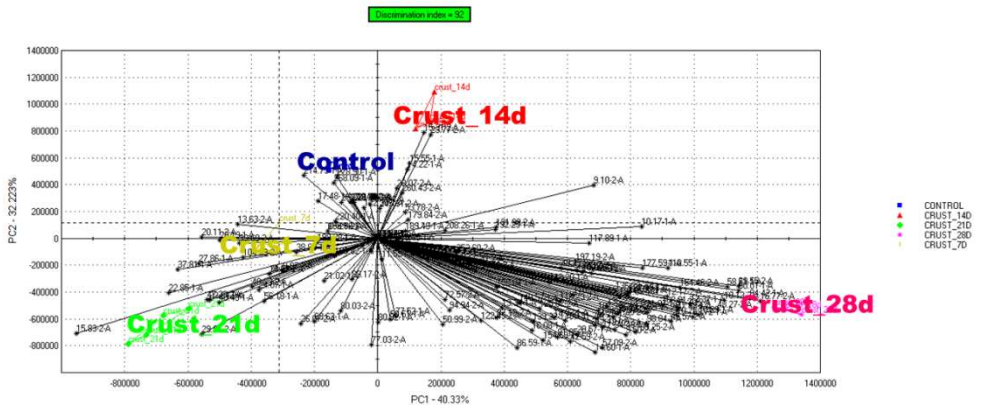
[a-c]Different letters means different significantly (P<0.05).

4.1.3. Electronic nose

The odor of dry-aged beef surface (crust) was analyzed by an electronic nose because it was observed the large changes in odor during dry aging. Then, PCA was performed to identify patterns of correlation with individual composition variables involved in the discrimination among aging periods of dry-aged beef. The odor map on PCA showed clear discrimination of 0, 7, 14, 21, and 28 day based on their all volatile composition, which were analyzed by the electronic nose [Figure 4(a)]. Figure 4(b) is the PCA odor map based on 11 volatile components selected from Figure 4(a) in consideration of reproducibility and specificity. Figure 4(b) shows clearer discrimination in volatile composition over the dry aging period. In particular, the odor of dry-aged beef aged for 28 days was clearly distinguished from the other aging periods. This result indicate that the change of dry-aged beef odor measured by electronic nose could be an indicator for dry aging degree. There have been attempts to measure the quality of fresh meat by electronic nose (Blixt and Borch, 1999; Winquist *et al.*, 1993), but no studies have attempted to analyze the aging degree of dry-aged meat using electronic nose. Eklöv *et al.* (1998) studied the fermentation

degree of fermented sausages by an electronic nose and confirmed the availability of the electronic nose for measurement in degree of fermentation, but there was a limitation in accuracy.

(a)



(b)

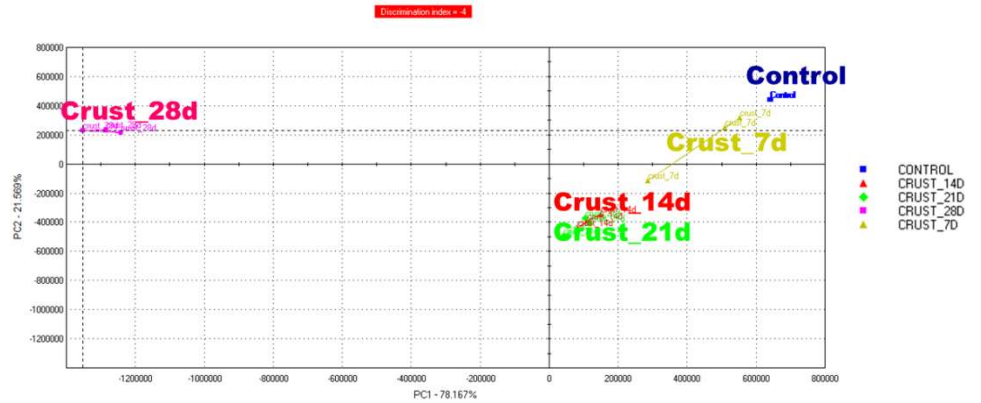


Figure 4. Plot of the first two PC score vectors; (a) all peaks which were found by electronic nose, (b) 11 peaks which were selected in consideration of reproducibility and specificity.

4.2. Analysis of correlation between dry aging parameters and quality factors of dry-aged beef

4.2.1. Moisture contents

Table 6 shows the moisture contents on dry-aged beef surface at 0, 7, 14, 21, 28, and 35 day. Experiment Ⅱ was performed only for dry aging method while studied for a longer aging period than Experiment 1. Similar to Experiment 1, the moisture contents decreased with the increase of aging period due to continuous water evaporation on beef surface ($P<0.05$, Table 6). Correlation coefficient of this result was higher than Experiment 1 and the results showed a linear tendency ($R^2=-0.945$, $P<0.001$). Thus, it is clearly shown that the moisture content of the beef surface in dry aging process can be an indicator of the degree of aging.

4.2.2. Mold distribution

There are results which quantify the fungal growth on beef surface during dry aging in Table 6. There were significant increases in the fungal growth at day 28 and 35 ($P<0.05$, Table 6).

The correlation between the aging period and the mold distribution showed a high linear trend (R^2=0.856, P<0.001). This study confirmed the possibility of mold distribution as a dry aging indicator of the aging degree.

4.2.3. Electrical resistance

The electrical resistance values of the beef surface at aging time of 0, 7, 14, 21, 28, and 35 days are shown in Table 6. As the dry aging period increased, the electrical resistance value of aged beef surface tended to increase (P<0.05, Table 6). The resistance value of dry-aged beef surface at 35 days was measured as 40 MΩ. However, since the maximum measurement value of the resistance the instrument was 40 MΩ, the actual resistance value is expected to be 40 MΩ or higher. According to Nelson (1991), the increase in the resistance of wheat samples as the moisture content of the grain decreased was confirmed in early in the 20th century. Stamm (1927) studied the availability of direct current resistance for measuring the moisture content of wood. This became the basic principle of an electrical-resistance type moisture meter, widely used today in the wood processing industry (Chen *at al.*, 1994). With the same principle, this study

confirmed that the resistance of aged beef surface was continuously increased during dry aging, which is expected to result from moisture evaporation. The correlation between aging period and electrical resistance was significant (Table 6, R^2=0.864, P<0.001). This result means that the electrical resistance of aged beef surface can be used as an indicator for the degree of dry aging. There have been attempts to measure the aging degree for wet-aged meat by measuring electrical resistance or impedance (Damez *et al.*, 2008; Gómez-Sánchez *et al.*, 2009). The electrical resistance during wet aging tends to decrease due to the breakdown of muscle structure by proteolytic enzymes (Damez *et al.*, 2008). However, the increase of electrical resistance in the dry-aged beef surface in this study seems not to be a result of the muscle structure destruction but a result of moisture loss due to evaporation of dry aging.

Table 6. The change of moisture, electrical resistance, and mold distribution on dry-aged beef surface according to aging period

	Aging period (day)						SEM[1]	R^2	*P*-Value
	0	7	14	21	28	35			
Moisture (%)	70.37^{a}	57.26^{b}	46.25^{c}	44.76^{d}	39.04^{e}	37.00^{f}	0.348	−0.945	<0.001
Mold distribution (%)	1.22^{c}	0.56^{c}	1.04^{c}	3.93^{c}	8.04^{b}	11.67^{a}	1.114	0.856	<0.001
Electrical resistance (MΩ)	0.94^{d}	2.47^{d}	8.09^{c}	9.82^{c}	14.10^{b}	40.00^{a}	0.870	0.864	<0.001

[1]Standard error of means (n= 18).

[a-f]The letters within the same row were significantly different (P<0.05).

4.2.4. Meat quality characteristics

4.2.4.1. Moisture contents

After the surface of aged beef was removed, the moisture content of the internal beef was measured (Table 7). The moisture contents of internal beef decreased significantly with the increase of the aging period ($P<0.05$, Table 7). The results of this study show a tendency similar to previous studies (Campbell *et al.*, 2001; Juárez *et al.*, 2011; Obuz *et al.*, 2014). When compared with the moisture content of aged meat surface (Table 6), the internal meat showed higher moisture content (Table 7). This difference occurs because the surface of dry-aged beef is more affected by moisture loss due to evaporation than inside of that. As the surface of the beef was dried rapidly, it has a hard layer called case hardening, which makes it difficult for the moisture inside the beef to move or evaporate (Md Atiqure Rahman *et al.*, 2017).

4.2.4.2. pH

Guignot *et al.* (1994) confirmed that as the pH was increased, the cooking loss decreased, and the tenderness, juiciness and

flavor were improved in veal. Because the proteolytic enzyme destroys the muscle structure and the resulting degradation products causes a change in pH, the pH is closely related to the quality of the meat (Boakye and Mittal, 1993; Guignot *et al.*, 1993). In this study, as the aging period increased, the pH was significantly increased ($P<0.05$, Table 7). Similar trend is found from previous results (Boakye and Mittal, 1993; Obuz *et al.*, 2014).

4.2.4.3. Color

Meat color is an indicator for consumers to judge the freshness of meat (Faustman and Cassens, 1990; Mancini, 2009). Immediately after slaughter, the myoglobin in the meat is in a deoxymyoglobin state, and the meat is reddish purple. When myoglobin becomes an oxymyoglobin by contacting with oxygen, the meat becomes bright red. When the meat is exposed to low oxygen pressure or after a long storage, the meat myoglobin becomes metmyoglobin and the color of the meat becomes brown (Mancini and Hunt, 2005). The values of L^*, a^*, and b^* showed differences according to aging period, but results are fluctuating and there was no tendency found (Table 7). Lee *et al.* (2015)

reported the reduction of L^* and b^* values of beef by dry aging, Obuz *et al.* (2014) reported the reduction of a^* and b^* values of dry-aged beef. Further studies are needed to explain the color change of beef due to dry aging.

4.2.4.4. Shear force

Meat tenderness is the most important quality for consumer evaluation, and shear force is a reliable measure of tenderness (Destefanis *et al.*, 2008). Aging of meat causes degradation of myofibrillar and cytoskeletal proteins by the activation of proteolytic enzymes. As a result, the structural weakness of muscle leads to increased tenderness (Nowak, 2011). As observed for the aging effect, it could be confirmed that the shear force of dry-aged beef significantly decreased with the increase of aging period ($P<0.05$, Table 7). The trend of decreasing shear force with increasing aging period can be found in the results of previous studies (Campbell *et al.*, 2001; Lee *et al.*, 2017; Obuz *et al.*, 2014; Wheeler *et al.*, 1999).

Table 7. Quality characteristics of dry-aged beef according to aging period

	Aging period (day)						SEM[1]
	0	7	14	21	28	35	
Moisture (%)	70.65[a]	70.75[a]	69.66[a]	70.18[a]	67.06[b]	65.26[c]	0.390
pH	5.67[c]	5.65[c]	5.77[b]	5.77[b]	5.80[b]	5.90[a]	0.012
L^*	31.85[bc]	34.66[ab]	36.02[ab]	35.17[ab]	36.64[a]	30.59[c]	1.043
a^*	24.89[a]	26.83[a]	18.45[d]	22.56[b]	25.26[a]	20.63[c]	0.524
b^*	18.33[a]	20.35[a]	14.45[b]	18.02[a]	18.37[a]	15.59[b]	0.646
Sher force (N)	33.49[a]	29.45[b]	28.96[b]	19.90[c]	19.22[c]	19.08[c]	0.741

[1]Standard error of means (n= 18).

[a-c]The letters within the same row were significantly different ($P<0.05$).

4.2.4. Sensory evaluation

Sensory evaluation of dry-aged beef was conducted through consumer acceptability assessment and a 9 point hedonic scale method was used. The scores of all sensory parameters except for appearance increased generally as the aging period increased (Table 8, $P<0.05$). Campbell *et al.* (2001) confirmed that the flavor traits (aged, beefy, and brown-roasted), tenderness, and juiciness of dry-aged beef became stronger as the dry aging period increased. And Li *et al.* (2014) reported that not only the taste traits (umami, sweet, bitter, and salty) became stronger as the dry aging period increased but also juiciness became stronger. Juárez *et al.* (2011) also reported that juiciness and tenderness of dry-aged pork became stronger as the dry aging period increased.

Table 8. Sensory evaluation of dry-aged beef according to aging period

	Aging period (day)						SEM[1]
	0	7	14	21	28	35	
Appearance	5.75	5.58	5.67	5.19	5.88	5.67	0.203
Odor	5.00^{b}	5.25^{ab}	5.63^{ab}	5.25^{ab}	6.08^{a}	5.75^{ab}	0.229
Taste	4.88^{b}	5.00^{b}	5.58^{ab}	5.83^{ab}	6.13^{a}	5.75^{ab}	0.274
Flavor	4.58^{b}	5.04^{ab}	5.42^{ab}	5.79^{a}	6.04^{a}	5.75^{a}	0.277
Tenderness	4.40^{b}	4.79^{b}	4.88^{b}	6.00^{a}	5.67^{a}	6.29^{a}	0.232
Juiciness	4.83^{b}	5.08^{ab}	5.50^{ab}	5.63^{ab}	5.79^{a}	5.92^{a}	0.231
Overall acceptance	4.63^{c}	4.88^{bc}	5.63^{ab}	5.75^{ab}	6.00^{a}	5.63^{ab}	0.268

[1]Standard error of means (n= 18).

$^{a-c}$The letters within the same row were significantly different ($P<0.05$).

4.2.5. Correlation between quality of dry-aged beef and aging indicator candidates

The change of moisture content in aged beef surface during aging showed a significant correlation with meat quality of internal beef. The change of surface moisture showed a high correlation with the quality characteristics measured by the device including moisture content, pH, and shear force ($P<0.05$, Table 9). All sensory parameters except for appearance also showed high correlation with the change of surface moisture ($P<0.05$, Table 9).

Electrical resistance was measured without damaging the surface of aged beef. Moisture content and shear force showed a negative correlation and pH showed a positive correlation with the electrical resistance ($P<0.05$, Table 9). Among the traits of sensory evaluation, only tenderness showed high correlation with the change of electrical resistance ($P<0.05$, Table 9).

The correlation between mold distribution quantified by digital photograph analysis and the quality of dry-aged beef is shown in Table 9. The mold distribution had a highly significant relationship with moisture content, pH, and shear force ($P<0.05$, Table 9). In relation to the sensory quality, the mold distribution

had a significant correlation with the tenderness and the flavor ($P<0.05$, Table 9).

In this study, moisture content, electrical resistance, and mold distribution were selected as candidates for aging indicator in order to easily determine the degree of aging in dry-aged beef. These three aging indicators were confirmed to have a significant correlation with the beef quality change during dry aging period. There was no study so far to measure the aging degree of dry-aged beef. The results indicate that it is possible to use beef with a non-destructive manner like moisture content, electrical resistance and mold distribution as indicators of the degree of dry aging.

Table 9. Analysis of correlation between aging indicators and meat quality of dry-aged beef

		Aging indicators		
		Surface moisture	Electrical resistance	Mold distribution
Quality characteristics	Moisture	0.67**	-0.86***	-0.85***
	pH	-0.87***	0.91***	0.81***
	L^*	-0.21	-0.42	-0.27
	a^*	0.45	-0.43	-0.17
	b^*	0.42	-0.44	-0.25
	Sher force	0.886***	-0.723***	-0.731***
Sensory evaluation	Appearance	0.03	0.05	0.12
	Odor	-0.48*	0.37	0.40
	Taste	-0.68***	0.41	0.46
	Flavor	-0.76**	0.46	0.48*
	Tenderness	-0.73**	0.69**	0.65**
	Juiciness	-0.57*	0.46	0.46
	Overall acceptance	-0.65**	0.37	0.36

*, $P<0.05$; **, $P<0.01$; ***, $P<0.0001$

5. Conclusion

Dry aging, which is accompanied by various changes depending on the aging environment, is a difficult method and requires experience and skill of the performer. Therefore, there is a need for techniques that are easy to evaluate the degree of dry aging.

In this study, indicators that significantly changed during wet and dry aging of beef were selected and measured nondestructively. On dry-aged beef surface, moisture content, mold distribution, and flavor measured by electronic nose changed significantly according to aging period. Thus, the moisture content, electrical resistance, and mold distribution of dry-aged beef surface were selected as the aging indicator candidates for the degree of dry aging. Then, the candidates confirmed the significant relationship between the change of quality characteristics (moisture content, pH, and shear force) during dry aging and the sensory quality (flavor and tenderness) enhancement.

On the basis of results, moisture content, electrical resistance, and mold distribution showed the possibility of indicating the degree of dry aging as a nondestructive manner and this method

was proposed for easy evaluation of the degree of dry aging. However, this is the first attempt of nondestructively method to measure the degree of dry aging thus remains needs to improve, such as accuracy.

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Chapter 5. Summary in Korean

식육의 연도는 소비자들이 식육의 품질을 판단하는 중요한 기준이 된다. 때문에 식육 산업에서는 식육의 연도를 향상시키기 위한 다양한 노력을 하고 있다. 그 중 숙성(aging) 기술은 물리, 화학적 처리 없이 사후 활성화되는 식육 내 단백질 분해 효소를 이용한다. 식육을 적절한 저장 환경에서 단백질 분해 효소가 충분히 활동할 수 있는 기간을 제공함으로써 식육의 근육 구조를 파괴하고 연도가 향상된다. 이러한 숙성 기술은 일반적으로 습식 숙성과 건식 숙성으로 구분된다. 습식 숙성은 식육을 진공 포장하여 냉장 온도 조건에서 숙성을 진행하는 방법이고 건식 숙성은 식육을 포장 없이 공기 중에 노출시켜 적정 온도, 습도, 풍속 조건에서 숙성을 진행하는 방법이다.

건식 숙성은 최근 특유의 풍미 때문에 소비자들이 관심을 갖고 그 소비 역시 증가하고 있다. 식육 산업에서는 이러한 소비자들의 요구에 맞춰 건식 숙성을 실시하는 업자가 증가하고 있다. 하지만 규정된 온도, 습도, 그리고 풍속과 같은 건식 숙성 조건이 없고 그 조건에 따라서도 숙성육의 품질은 서로 다르다. 때문에 건식 숙성은 숙련된 경험을 요구하는 어려운 숙성 방법이다. 이를 개선하기 위해서 이번 연구는 비파괴적으로 건식 숙성의 숙성 정도를 나타내는 지표를 찾고자 하였다.

실험 1에서는 숙성 지표 후보를 선정하기 위해 습식 숙성과 건식

숙성 기간 중 식육에서 발생하는 다양한 변화를 확인하였다. 습식 숙성의 수분 함량은 숙성 기간과 유의미한 관계를 가지고 있지 않았지만 건식 숙성의 수분 함량은 숙성 기간이 증가함에 따라 유의미하게 감소하였다. pH는 건식 숙성과 습식 숙성 모두에서 숙성 기간에 따라 특정 경향을 보이지 않았다(Table 5). 건식 숙성의 경우 식육을 포장하지 않고 공기 노출시키기 때문에 증발로 인한 건조 표면의 발생뿐만 아니라 흰색 털을 가진 곰팡이가 생장하고, 숙성 기간이 증가함에 따라 특유의 숙성 향이 강해진다. 이러한 변화를 비파괴적인 방법으로 측정하고자 건식 숙성 기간 중 숙성 육의 외관을 디지털 사진으로 촬영하여 포토샵 프로그램을 이용하여 곰팡이 분포도 (%)를 정량화 하였고 건식 숙성 중 풍미 변화를 전자코로 분석하였다. 곰팡이 분포는 숙성 기간에 따라 유의미하게 증가하였고(Figure 3) 전자코로 분석된 건식 숙성의 풍미 역시 주성분 분석(PCA)에 의해 숙성 기간에 따라 구분할 수 있었다.

실험 2에서는 건식 숙성 육 표면의 수분 함량, 이를 비파괴적으로 측정할 수 있는 전기 저항, 그리고 곰팡이 분포를 건식 숙성 지표 후보로 선정하여 건식 숙성 기간 중 숙성 육 품질 변화와의 상관 관계를 확인하였다. 선정된 지표 후보들은 숙성 기간과 높은 상관 관계를 보였다. 숙성 기간에 따른 숙성 육 품질의 변화를 확인하기 위해 내부 육의 수분 함량, pH, 색도(L^*, a^*, b^*), 그리고 전단력을 측정하였다. 색도를 제외하고 수분 함량과 전단력은 숙성 기간에 따라

감소하는 경향을 보였고 pH는 증가하는 경향을 보였다. 건식 숙성에 의한 식육의 품질 증진을 확인하기 위해 숙성 기간 별 소비자 기호도 관능 평가를 실시하였고 숙성 기간이 증가함에 따라 냄새, 맛, 풍미, 연도, 다즙성, 그리고 종합적 기호도가 유의미하게 개선되는 것을 확인하였다. 건식 숙성 지표 후보인 숙성육 표면의 수분 함량, 전기 저항, 그리고 곰팡이 분포는 숙성 기간 중 숙성 육 내부의 품질인 수분 함량, pH, 그리고 전단력과 높은 상관관계를 가지고 있었다. 소비자 기호도 관능 평가에 있어서 숙성 육 표면의 수분 함량은 냄새, 맛, 풍미, 연도, 다즙성, 그리고 종합적 기호도와 유의미한 관계를 보였고, 전기 저항은 연도와 유의미한 관계를 보였고, 곰팡이 분포는 풍미와 연도에 유의미한 관계를 보였다.

이번 연구는 건식 숙성의 숙성 정도를 나태내기 위한 지표를 찾고 검증하기 위해 처음 시도되었다. 건식 숙성 육 표면의 수분 함량, 전기 저항, 그리고 곰팡이 분포를 건식 숙성 지표로 이용할 수 있음을 확인하였다. 하지만 관련 연구가 부족하며 정확성과 같은 문제점들을 개선하기 위한 더 많은 연구가 필요하다. 또 비파괴적으로 측정할 수 있는 다양한 숙성 지표가 고안되어야 할 것이다.