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Abstract

This study was carried out to assess the quality properties, components associated with taste and aroma of beef as a function of breed. For this purpose, steers from four Korean native cattle breeds: Hanwoo (n=10), Chikso (n=10), black Hanwoo (n=12, BHW) and Jeju black cattle (n=12, JBC) were used. The steers all were raised under identical conditions and finished at a similar age of around 30-months old. Following 24 h of slaughter, all *longissimus lumborum* (LL) muscles were collected and used for analysis of meat quality, fatty acids, and flavor-related components (metabolic compounds, free amino acids, and aroma volatiles). The Hanwoo presented a significantly higher intramuscular fat content (IMF, 22.85%) than the BHW (11.78%), Chikso (9.25%), and JBC (9.14%) ($p < 0.05$). The meat of Hanwoo breed showed lighter and redder color, and lower shear force value ($p < 0.05$). The JBC presented a "healthier" fatty acid profiles as it had a higher total unsaturated fatty acids content ($p < 0.05$). With regard to flavor-related components, Hanwoo also had higher total contents of free amino acids and metabolites associated with umami and sweet tastes, and fat-derived volatile compounds (aldehydes, alcohols, and ketones) associated with fatty aroma. It may be concluded that there was a considerable difference in the meat quality properties among breeds. The variations of IMF content and flavor-related components may be the main factors contributing to the typical flavors of beef among the four Korean native cattle breeds.

Keywords: Cattle, breed, meat quality, taste, aroma

Introduction

Quality is an utmost important element determining the success of the beef industry. Beef quality is a complex definition, comprised of multi-aspects such as: appearance, texture, marbling, water holding capacity, sensory properties (flavor, tenderness and juiciness) and credence quality (nutritional value, safety, and animal welfare) (Liu et al., (2022). All of these aspects are related and correlated with each other, which finally determine the beef eating quality (Gotoh et al., 2018; Lee et al., 2022; Schumacher et al., 2022).

Flavor, tenderness and juiciness all are the most important determinants of beef eating quality, however, flavor is considered as a stronger driver for overall liking by consumers (Kerth and Miller, 2015). Flavor of cooked meat is constituted of aroma and taste which are detected by smell and taste receptors on the nose and tongue, respectively (Chandrashekar et al., 2006). Tastes (e.g., sweetness, bitterness and umami etc.) of cooked meat are created by a variety of non-volatile constituents (e.g., free amino acids, sugars, and salts etc.) and nucleotides (inosine, inosine 5'-monophosphate, and guanosine 5'-monophosphate etc.) (Khan et al., 2015). While, aroma is contributed by a variety of volatiles (e.g., aldehydes, alcohols, and pyrazines etc.), which are formed via the thermal oxidation of fatty acids and Maillard reaction of amino acids and sugars during cooking (Mottram, 1998).

Among the ante-harvest factors, breed is recognized as an important element affecting growth performance and quality characteristics of beef (Cafferky et al., 2019; Vazquez-Mosquera et al., 2022). Researchers have found that under identical raising conditions, different cattle breeds exhibit a wide variation in growth rate, fat deposition (marbling degree), and quality (chemical composition and eating quality) of beef (Aviles et al., 2015; Shahrai et al., 2015). Literatures have also reported that breed affects the precursors of beef flavor (e.g., fatty acids, free amino acids, and metabolites) (Koutsidis et al., 2007), which subsequently influence the flavor characteristics of the beef after cooking (Conanec et al., 2021).

Until now, four main Korean native cattle breeds: brown (Hanwoo), brindle (Chikso), Jeju black (JBC) and black Hanwoo (BHW) cattle have already been registered with the Food and Agriculture Organization (Park et al., 2016). According to data reported by Korea Institute of Animal Products Quality Evaluation (KAPE, 2022), there were about 4 million cattle being raised for beef production in Korea. Hanwoo, with a population size of approximately 3.7 million, is the most predominant and important cattle breed in the Korea beef industry (KAPE, 2022). Hanwoo beef is known worldwide as a highly-marbled, tender and palatable meat type (Cho et al., 2017; Chung et al., 2018). In contrast to the Hanwoo, the other three remaining breeds are only reared in several locals with a limited population size (hundreds to thousands of heads per breed) (Alam et al., 2021; Haque et al., 2023; Park et al., 2020). Recently, beef producers have paid more attention to these cattle breeds (Song et al., 2018). Additionally, Korean consumers usually believe that beef derived from native breeds is a “healthier” meat type (Lee et al., 2019).

To the best of our knowledge, however, no study was conducted to compare the meat quality among these four cattle breeds under a same commercial raising condition. The aim of this study, therefore, was to compare the quality properties and flavor-related components of beef among the Korean native cattle breeds under identical raising conditions.

Materials and Methods

Animal and sample preparation

Forty-four steers from four Korean native cattle breeds including: Hanwoo (n=10), Chikso (n=10), black Hanwoo (n=12, BHW) and Jeju black cattle (n=12, JBC) were randomly selected and used in this study. The animals of each breed were raised in separate feedlots of farms under identical feeding condition. Over the fattening periods, a similar ration was applied to all the steers, following the standardized cattle feeding program (Lim et al., 2013). The animals were finished at around 30 months old. Following transporting to a practical plant of the National

Institute of Animal Science (Wanju-gun, Korea) with a journey of around 2 h (except for the JBC which were transported with a journey of approximately 6 h). After arrival, all the animals were kept in lairages where they were fasted from feed but had free access to water for approximately 3 h. To minimize pre-slaughter-related stress (e.g., fighting caused by unfamiliar breeds), different slaughter batches were carried out for each the cattle breed. The slaughter was carried out following an industry-accepted procedure. After overnight chilling at 2°C, *longissimus lumborum* (LL) muscles were collected and used. The analysis of meat quality properties was done at 48 h post-mortem; for each the analysis, the cutting manner (e.g., anatomical position) was fixed for all the muscle samples of breeds.

Chemical composition and meat quality

Moisture, fat, protein and collagen were determined with a Food Scan (Foss Tecator Co., Ltd., DK). Briefly, following trimming of outer fat and connective tissues and blending with a mixer (Hanil Co., Chungcheongnam-do, Korea), each sample (about 100 g) was placed on a petri dish that was then loaded on the device.

The pH values were measured in triplicate using a pH meter (pH*K 21, NWK-Technology GmbH, Kaufering, Germany) after calibrating with standard solutions (pH 4.00 and pH 7.00).

The meat color, cooking loss and Warner-Bratzler shear force (WBSF) were determined on a same steak (2.54-cm thickness) taken from each the sample, following the procedure as described in our previous study (Hoa et al., 2022).

Fatty acid composition

The fatty acids composition of beef samples was extracted and analyzed using the procedure of Folch et al. (1957). Briefly, duplicate aliquots (10 g) per sample were taken, placed in tubes containing 150 mL of chloroform: methanol (2:1, v/v) solution and homogenized at 11,000 ×rpm for 3 min, for lipid extraction. Following filtration with Whatman filter paper, the filtrates were collected, added with 20 g of Na₂SO₄ and then vortexed for 1 min. The upper lipid layer was

carefully collected and concentrated at 55°C. Thereafter, fatty acids were methylated and analyzed using a gas chromatography (GC)/flame ionization detector (FID, Varian Technologies, Palo Alto, CA, USA) under conditions same to those as described by Hoa et al. (2022). The results were expressed as relative percentage (%) of total fatty acids.

Metabolic profiles

The procedure of Hoa et al. (2023b) was used for analysis of metabolic compounds in beef of four cattle breeds. Briefly, after chopping and cooking for about 2 min at around 180 °C, duplicate aliquots (20 mg) of each sample were weighted and homogenized with acetonitrile/water (1:1, v/v) mixture on ice for 10 min. The samples were centrifuged for 10 min (at 3000 x g and 4°C), and the resultant supernatant was collected and lyophilized. Thereafter, each the sample was added with 20 µL of D₂O containing 2 mM 3-trimethylsilyl-2,2,3,3-tetradeuteropropionicacid-d₄ in a 5-mm NMR nano tube. The metabolites were analyzed with a Nuclear Magnetic Resonance (NMR) spectrometer (Agilent Technologies, Palo Alto, CA, USA). ¹H-NMR spectra were acquired at a ¹H frequency of 599.93 MHz, and were Fourier transformed using Vnmrj (version 4.2, Agilent). The identification of metabolic compounds was done using the MHz library database (Chenomx Inc., Canada). The identified metabolites were quantified using the known concentration of the internal standard (TSP-d₄).

Free amino acids (FAA)

The FAAs content in the meat samples (5 g each) were extracted with 10 mL distilled water as described in our previous study (Cho et al., 2020). The resultant FAAs were then analyzed with an Ultra Performance Liquid Chromatography (Waters Co. Milford, MA, USA). The separation was carried out on an amino acid column (diameter: 2 ×50 mm, 3µm) using two different solvents: A [acetonitrile: 100 mM ammonium formate; 20:80 v/v] and B [acetonitrile: trifluoroacetic acid: 25 mM ammonium formate: formic acid: 9:75:16:03 v/v/v]. The FAAs were detected at 254 nm with a photodiode array detector (Waters Co. Milford, MA, USA). The

FAAs identification was carried out by using the amino acid standards which were separated under the same condition. The results were calculated and expressed as milligram per 100 g meat (mg/100 g meat).

Aroma volatiles

Aroma volatiles were analyzed using the method of Hoa et al. (2010). Briefly, the chopped meat samples were cooked at around 180 °C on a frying-pan, with continuous turning for about 2 min. Next, proximately 1.0 g of each the sample was taken, placed into 20-mL vial and tightly capped with a magnetic cap. One microliter of 2-methyl-3-heptanone (816mg/mL) was also added into the sample vial as an internal standard. Thereafter, the vials containing samples (each) were inserted with a 75- μ m carboxen–polydimethylsiloxane fiber (Supelco) held by an auto-sampler robot, and the extraction was conducted at 60 °C for 50 min. The fiber with volatiles were then desorbed at 250 °C for 5 min into a capillary column (Agilent) connected to a gas chromatography (GC) with mass spectrophotometry (5977B MS, Agilent Technologies) system. The volatiles were identified by using Wiley registry library (Agilent Technologies) and a series of standards which were separated under the same condition. Concentration of volatiles was quantified by using the concentration-known internal standard and expressed as microgram per gram sample.

Statistical analysis

In the present investigation, to minimize the effect of ante-and post-slaughter factors that may influence the experimental results, all the animals were raised under an identical feeding condition, and then handled and slaughtered using a same process. Data was analyzed using a SAS (version 7.1; SAS Institute, Inc., Cary, NY, USA). One-way ANOVA was used where cattle breed was set as a fixed main factor, and the data (color, quality traits, FAA, metabolites,

aroma volatiles, fatty acids) were set as random variables. The differences among the means were compared using Duncan's Multiple Range Test. Significance was set at $p < 0.05$.

Results and Discussion

Chemical composition and meat quality traits

As presented in Table 1, all the chemical composition varied among the breeds; JBC had a lower protein content, while Hanwoo exhibited the highest fat content (almost two times greater) compared to those of all the other three remaining cattle breeds ($p < 0.05$). However, the fat contents ranging from 9% to 11% among the remaining breeds in the present study, were higher compared to those reported in LL muscles of other foreign cattle breeds such as Limousine (2.05%), Angus (2.78%), Charolais (2.05%), Belgium Blue (1.70%), Hereford (2.13%) and Wangus (6-7%) (Cafferky et al., 2019; Vazquez-Mouquera et al., 2022). According to the fat level corresponding to each beef quality grade in the Korean carcass grading system (Jo et al., 2012), and based on our results the Hanwoo beef could be classified into the 1++ grade while BHW, and Chikso and JBC could be classified into the 1 and 2 grade, respectively. IMF is the most important contributor of beef eating quality (Gotoh et al., 2018). Literatures have reported a significant effect of breed on IMF content in which early maturing cattle breeds usually exhibit a faster fat deposition, resulting in a higher IMF content compared to late maturing breeds (Coleman et al., 2016). Indeed, Hanwoo cattle generally possess a better growth performance and could be finished at around 22 to 28 months of age (Chung et al., 2018; Jo et al., 2012). Otherwise, Hanwoo is recognized as cattle breed with a high-fat accumulating potential, as a result of long-term pure-breeding and strict selection program for improving economic traits (e.g., marbling) over the last decades (Chung et al., 2018). In a study conducted by Hoa et al. (2023a), the Hanwoo beef exhibited a higher expression of lipid-metabolism-related genes compared to Chikso cattle. With regard to moisture content, Hanwoo beef had a lower level than other remaining breeds ($p < 0.05$). In fact, it is generally found that fat and moisture in meat is

inversely related with each other; the higher the fat the lower the moisture content (Jayasena et al. (2015). This is the case of our study as the highest fat and lowest moisture content was observed in the Hanwoo cattle. Collagen is known to be the most abundant structural protein in all animal's connective tissues (Shoulder and Raines, 2009). In meat, collagen plays an important role in maintaining its acceptable texture. The cross-links (together binding of collagen molecules), rather than the concentration of collagen content, increase meat toughness (Weston et al., 2002). Our results depict that Hanwoo had a higher collagen content compared to the other breeds ($p < 0.05$).

The quality properties of LL muscles of four cattle breeds are presented in Table 2. Color, reflecting the visual appearance (e.g., freshness and wholesomeness), is considered as an important factor affecting meat purchasing decision by consumers (Purslow et al., 2019). We observed that there was a significant effect of breed on all color traits. Significantly higher L^* , a^* and b^* values were found in Hanwoo than in the other breeds ($p < 0.05$). JBC also exhibited a higher a^* value than Chikso and BHW ($p < 0.05$). Our results are in agreement with those of Avilés et al. (2015) and Xie et al. (2012), who reported a significant breed effect on color traits of beef. According to the classification of beef groups using L^* (lightness) values by Hughes et al. (2017), the Hanwoo beef belonged to the light meat group while meat of other remaining breeds was considered as medium-light meat group. Meat color is directly influenced by chemical composition such as; fat content and pigment proteins (Faustman et al., 2010). We assumed that when the ante- and post-slaughter factors are kept constant, the IMF content might play the most important role in determining the meat color. The mechanism underlying this phenomenon still remains unknown. However, previous studies on beef have also reported a similar trend: Xie et al. (2012) reported no differences in color traits (L^* and a^*) of beef among 5 cattle breeds which had a similar fat content. Yim et al. (2015) reported a significantly higher L^* and a^* values in beef with higher fat content compared to beef with a lower fat content.

Similarly, Zhang et al. (2022) reported that beef with a higher fat content is associated with lighter, redder and yellower color.

Tenderness is undoubtedly the most important contributor of eating quality of beef. There was a significant effect of breed on the WBSF value ($p < 0.05$). The lowest values were found in the Hanwoo, followed by BHW, Chikso and JBC. It is demonstrated that IMF in meat is negatively correlated with WBSF value (Cafferky et al., 2019; Zhang et al., 2022; Yim et al., 2015). Hoa et al. (2023b) also reported a significant effect of breed on WBSF value, beef containing a higher fat content had a lower shear force value. Similarly, Xie et al (2012) found that beef from cattle breeds with similar fat contents did not differ in WBSF values. In the present study, the variations of WBSF values among the breeds could directly be linked to their corresponding fat contents (Table 1). According to a consumer evaluation study conducted by Boleman et al. (1997), beef LL muscle with shear force values of 2.27-3.58, 4.08-5.40, and 5.90-7.21 (kgf) are categorized into "tender", "intermediate" and "tough" meat groups, respectively. Based on the WBSF values in this study, the Hanwoo beef could be considered as the "tender" meat category while, meat from other three remaining breeds could be considered as the "intermediate" meat group.

Regarding pH and cooking loss, the effect of breed was observed. BHW beef exhibited the lowest cooking loss (16.20%) whereas, the Chikso beef had highest cooking loss (21.01%) ($p < 0.05$). Similar to our findings, Xie et al. (2012) and Cafferky et al. (2019) found a significant effect of cattle breed on cooking loss of beef. Compared with our results, however, these authors reported a relatively higher cooking loss (26-31%) under a same cooking temperature (internal temperature of 70°C). Chikso and Hanwoo showed a higher pH value than BHW and JBC ($p < 0.05$). The pH values ranging from 5.45 to 5.67 among the breeds in this study generally fell within the pH ranges of normal beef (Barrasso et al., 2022). It is well demonstrated that short-or long-term stresses (e.g., fear, transport exhaustion, fighting caused by unfamiliar animals, overloading etc.)

before slaughter significantly affect the extent of glycogen depletion which subsequently influences the post-mortem rate of pH decline (Reiche et al., 2019). Nevertheless, in the present study, all animals were handled under the same condition (e.g., transport condition and fasting time etc.) before slaughter. The variations of pH, therefore, could be related to the muscle glycogen-reserving capacity differences among the breeds.

Effects on fatty acids profile

Fatty acids content in meat is considered as an important index indicating the nutritional value and how it affects flavor characteristics of the meat after cooking (Dinh et al., 2021). As shown in Table 3, a considerable effect of breed on the fatty acids composition was observed. The JBC beef presented a "healthier" fatty acid profiles as it had a lower total saturated fatty acids (SFA) and higher polyunsaturated fatty acids (PUFAs) content as well as PUFA/SFA ratio value compared to the Hanwoo ($p < 0.05$). Similar to the current results, Lee et al. (2019) reported a lower SFA and higher PUFAs contents in JBC beef compared to commercial Wagyu cattle breed. Oleic acid is known to be the most predominant fatty acid in beef, and it has recently been demonstrated to improve flavor and overall palatability of beef (Smith, 2016). In the present study, the difference in oleic acid content was only observed between JBC and BHW ($p < 0.05$). In meat animals, two well-known main pathways of fatty acids synthesis are: (i) adipogenesis: this process involves the proliferation and differentiation of pre-adipocytes to pre-mature adipocytes and subsequently to mature adipocytes induced by elevated hormones and diet-derived fatty acids under the regulation of adipocyte protein 2, lipoprotein lipase and peroxisome proliferator activated receptor gamma etc., and (ii) de novo fatty acids biosynthesis: this process involves the conversion of glucose into triglycerides in the glycolysis pathway under regulation of fatty acid synthase and stearoyl-CoA desaturase-1 etc. (Malgwi et al., 2022). Furthermore, in the multi-gastric animals such as cattle, the diets-derived unsaturated fatty acids are dehydrogenated largely to saturated fatty acids by ruminal microorganisms. In cattle, a large proportion of UFAs are formed from the conversion of

SFAs by delta-9 desaturase enzyme in the de novo fatty acids biosynthesis (Smith et al., 2006). It is reported that fatty acids content in beef is affected by a number of pre-harvest factors such as feeding diet, age, gender and genetics (Joo et al., 2017; Wood et al., 2008; Xie et al., 2012). In this study, the gender, slaughter age and feeding regime all were kept constant for the selected animals, therefore, the variations of fatty acids content especially the UFAs could be related to the breed that might influence the rate of de novo fatty acids biosynthesis.

Effect on taste-related components

Taste, as a part of flavor, is mainly contributed by water-soluble active compounds such as free amino acids (FAAs), metabolites and degraded products of nucleotides (Khan et al., 2015; Mateo et al., 1996). The concentration of FAAs in the LL muscles of 4 breeds are presented in Table 4. With exception for the cases of Pro, Thr, Ile, and Arg, the breed affected all the FAAs ($p < 0.05$). Based on the similarity of taste qualities, the FAAs were grouped into several different taste groups including: umami (Glu, Asp, Asn, and Gln), sweetness (Gly, Ala, Ser, Pro, and Thr) and bitterness (Lys, Val, Leu, Ile, Arg, Phe, and Tyr) (Kato et al., 1989; Sasaki et al., 2007). Results showed that the level of umami-associated FAAs was found to be the highest in Hanwoo, followed by Chiko, JBC and HHW ($p < 0.05$). Regarding sweetness-associated FAAs, its level was also higher in Hanwoo and Chikso compared to the BHW and JBC. No differences in bitterness-associated FAAs content were found among Hanwoo, Chikso and BHW ($p < 0.05$). In agreement with our result, Dashmaa et al. (2013) reported a considerable effect of breed on FAAs in beef, with a majority of FAAs differed between Hanwoo and Angus breeds. While, Koutsidis et al. (2007) reported that Gly and Arg were only amino acids which differed between Aberdeen Angus and Holstein-Friesian cattle breeds. In meat, FAAs content is affected by rate of protein synthesis (in animals, a higher rate of protein synthesis results in a fast depletion FAAs), and the post-mortem proteolysis by endogenous proteases (Koutsidis et al., 2007).

After slaughter, there are many bio-chemical events (post-mortem glycolysis and proteolysis) occurring in animal carcasses (Chauhan and England, 2018). As a result of these processes, a significant number of metabolites (final or intermediate products) are formed, which subsequently contribute to development of meat flavor during cooking (Kim et al., 2016). A total of thirty-four metabolites was identified in the beef LL muscles of 4 cattle breeds (Table 5). Among them were amino acids, derivatives of amino acids, lipids, acids, sugars and nucleotides etc. As aforementioned, Ala and Gly are associated with sweetness, while Gln contributes to umami, their concentrations were higher in Hanwoo ($p < 0.05$). Inosine is an intermediate product of enzymatic reaction of inosine monophosphate (IMP), that is transformed from adenosine triphosphate (ATP) during the post-mortem glycolysis or phosphagen system mechanism (Chauhan and England, 2018). Excepting hypoxanthine that contributes to bitter taste, almost all the APT-degraded products are important compounds as they possess umami taste (Tikk et al., 2006). Results showed that Hanwoo beef had a higher inosine content compared to other three breeds ($p < 0.05$).

Carnosine, creatine, carnitine, and betaine have been considered as bioactive substances (e.g., antioxidant) from animal sources (Jayasena et al., 2015; Jung et al., 2013). In muscle, creatine is present in a form of creatine phosphate, which is subsequently broken-down to form ATP molecules. Our results showed that the betaine content was higher in the Chikso and BHW while, a higher creatine content was found in the Hanwoo and Chikso compared to other breeds ($p < 0.05$). No effect of beef on carnosine and carnitine content was observed ($p > 0.05$). Creatinine, known as a creatine-derived waste product, was higher in Hanwoo ($p < 0.05$). Out of detected sugars, glucose has been report to be the important precursor of meat flavor as it participates the Mallard reaction with amino acids to directly or indirectly generate a large number of aroma-active compounds (Dashdorj et al., 2015). In the present study, no effect on breed on the glucose content was found ($p > 0.05$). In general, it was observed that, Hanwoo cattle

generally exhibited a higher content of ATP-broken-down products (creatine, creatinine and inosine) and proteolysis-derived products (e.g., Glu, Gln, Gly and Ala etc.), signifying a greater degree of glycolysis, proteolysis and ATP degradation in this breed compared to the other remaining breeds after slaughter.

Effect on aroma volatile compounds

Aroma, as a part of flavor sensed by smell receptors, is an important determinant of sensory quality of cooked meat (Arshad et al., 2018). During cooking, the oxidation of fatty acids, Maillard reaction of amino acids with sugars, and the interaction between their intermediate products are the major pathways for formation of volatile compounds which are the main contributors of the cooked meat aroma (Bassam et al., 2021). Under the current experimental condition, fifty-one volatiles were detected and quantified in the cooked LL muscles of four breeds (Table 6). Aldehydes, with 23 compounds, were the most predominant aroma class, followed by sulfur-and nitrogen-containing compounds (10), alcohols (7), ketones (6) and hydrocarbons (5). Aldehydes are mainly formed through the thermal oxidation of fatty acids, a few may be formed from the Strecker degradation of amino acids (Mottram, 1998). Aldehydes are well recognized as the major contributors to meat aroma due to their low-odor detection threshold (Bassam et al., 2021). Otherwise, the quality and quantity of aldehydes, have been considered as an important indicator to discriminate the characteristic aroma of different species (Arshad et al., 2018). Our results showed that 2-methylbutanal, pentanal, hexanal, heptanal, E,E,2,4-decadienal, E,2-octenal, nonanal, E,2-nonenal, decanal and E,2-dodecanal exhibited a breed effect ($p < 0.05$). The majority of these aldehydes are known as the oleic acid-derived products (pentanal, octanal, nonanal, E,2-nonenal and decanal) associated with desirable odors (e.g., fatty aroma) (Aaslyng and Meinert, 2017). The Hanwoo beef exhibited a significantly higher level of almost these aldehydes compared to the other breeds regardless of its oleic acid

content that was not different from the other remaining breeds (Table 3). Likewise, Hanwoo beef also had a higher total aldehydes content compared to other breeds ($p < 0.05$).

Together with aldehydes, other classes such as; alcohols, ketones and hydrocarbons, are also formed as a result of thermal oxidation of fatty acids (Chang et al., 2020). However, alcohols, ketones and hydrocarbons are lesser important in cooked meat aroma development as they possess a high odor detection threshold (Bassam et al., 2021). We observed that almost all alcohols, ketones and hydrocarbons showed a breed effect, in which the Hanwoo beef had a significantly higher amount compared to the other breeds ($p < 0.05$). It has been proposed that when exposed to heat during cooking, not only UFAs but also the SFAs are thermally oxidized (Mottram, 1998). Contrasting to the mono-gastric animals, the IMF content of ruminant meat contains more SFAs than UFAs (Dinh et al., 2021). In the present study, the higher IMF content of Hanwoo beef (Table 1) seemed to be the main factor responsible for the higher concentration of the fat-derived volatile compounds associated with fatty aroma in this beef breed. In a study conducted by Frank et al. (Frank et al., 2016), Wagyu beef having a higher IMF content exhibited a higher level of fat-derived compounds associated with fatty aroma compared to lower-IMF beef breed. Recent studies have also found an increase in level of fat-derived aldehydes with increased IMF content in beef (Hoa et al., 2022). Bassam et al. (2021) stated that the higher the fat content the higher the level of fat-derived aroma compounds (e.g., hydrocarbons) in grilled beef.

Sulfur- and nitrogen-containing compounds are known to be the Maillard reaction-derived products associated with desirable aromas (e.g., roasted and meaty) in cooked meat (Aaslyng et al., 2017). The breed did not affect all the sulfur- and nitrogen-containing compounds, with exception of 3-ethyl-2,5-dimethylpyrazine whose level was higher in Chikso compared to the other remaining breeds ($p < 0.05$). Based on the analysis of aroma compounds, it may be said that

breed showed a significant effect on aroma volatiles component of cooked beef, especially the fat-derived compounds.

Conclusion

In the present study, four Korean native cattle breeds reared under identical condition were used to study the effect of breed on meat quality properties. There was a considerable effect of breed on chemical composition, color and technological quality of beef *longissimus lumborum* muscles. Hanwoo (brown cattle), with the highest IMF content indicated its highest potential of fat deposition compared to all the other remaining breeds. The breed also exhibited an effect on almost all flavor-related components such as; free amino acids and metabolites and aroma compounds such as; thermal oxidation of fat-derived products (aldehydes, alcohols and ketones). Out of four breeds, Hanwoo beef possessed a significantly higher amount of taste (umami and sweetness) and aroma (fatty)-active compounds which, therefore, could be used as biomarkers to discriminate the characteristic flavor of this beef breed with those from the other remaining cattle breeds.

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References

- Aaslyng MD, Meinert L. Meat flavor in pork and beef - From animal to meal. *Meat Sci* 132: 112–117.
- Alam MZ, Lee YM, Son HJ, Hanna LH, Riley DG, Mannen H, Sasazaki S, Park SP, Kim JJ. 2021. Genetic characteristics of Korean Jeju Black cattle with high density single nucleotide polymorphisms. *Anim Biosci* 34:789-800.

- Arshad MS, Sohaib M, Ahmad RS, Nadeem MT, Imran A, Arshad MU, Kwon JH, Amjad Z. 2018. Ruminant meat flavor influenced by different factors with special reference to fatty acids. *Lipids Health Dis* 17:223.
- Aviles C, Martínez AL Domenech V, Peña F. 2015. Effect of feeding system and breed on growth performance, and carcass and meat quality traits in two continental beef breeds. *Meat Sci* 107:94–103.
- Barrasso R, Ceci E, Tufarelli V, Casalino G, Luposella F, Fustinoni F, Dimuccio MM, Bozzo G. 2022. Religious slaughtering: Implications on pH and temperature of bovine carcasses. *Saudi J Biol Sci* 29:2396–2401.
- Bassam SM, Noleto-Dias C, Farag MA. 2022. Dissecting grilled red and white meat flavor: Its characteristics, production mechanisms, influencing factors and chemical hazards. Review. *Food Chem* 371:131139.
- Boleman SJ, Boleman SL, Miller RK, et al. 1997. Consumer evaluation of beef of known categories of tenderness. *J Anim Sci* 75:1521-4.
- Cafferky J, Hamill RM, Allen P, O’Doherty JV, Cromie A, Sweeney T. 2019. Effect of breed and gender on meat quality of m. *longissimus thoracis et lumborum* muscle from crossbred beef bulls and steers. *Foods* 8:173.
- Chandrashekar J, Hoon MA, Ryba NJP and Zuker CS. 2006. The receptors and cells for mammalian taste. *Nature* 444:288–294.
- Chang C, Wu G, Zhang H, Jin Q, Wang X. Deep-fried flavor: 2020. Characteristics, formation mechanisms, and influencing factors. *Crit Rev Food Sci Nutr* 60:1496–1514.
- Chauhan SS, England EM. 2018. Postmortem glycolysis and glycogenolysis: insights from species comparison. *Meat Sci* 144:118-126.

- Cho SH, Kang SM, Seong PN, Kang GH, Kim Y, Kim JH, Chang SS, Park BY. 2017. Effect of aging and freezing conditions on meat quality and storage stability of 1++ grade Hanwoo steers: Implication for shelf-life. *Korean J Food Sci Anim Resour* 37:440–448.
- Cho SH, Seol KH, Kang SM, Kim YS, Seo HW, Lee W, Kim JH, Hoa VB. 2020. Comparison of tastes-related components and eating quality between Hanwoo steer and cow *longissimus thoracis* muscles. *Food Sci Anim Resour* 40:908-923.
- Chung KY, Lee SH, Cho SH, Kwon EG, Lee JH. 2018. Current situation and future prospects for beef production in South Korea—A review. *Anim Biosci* 31:951–960.
- Coleman LW, Hickson RE, Schreurs NM, Martin NP, Kenyon PR Lopez-Villalobos N, Morris, ST. 2016. Carcass characteristics and meat quality of Hereford sired steers born to beef-cross-dairy and Angus breeding cows. *Meat Sci* 121:403–408.
- Conanec A, Campo M, Richardson I, Ertbjerg P, Failla S, Panea B, Chavent M, Saracco J, Williams JL, Ellies-Oury MP, Hocquette JF. 2021. Has breed any effect on beef sensory quality?. *Livest Sci* 250:104548.
- Dashdorj D, Jang J, Ba HV, Ryu KS, Hwang IH. 2013. The differences in taste-active compounds between Hanwoo *longissimus dorsi* and *semitendinosus* muscles and its comparison with Angus *longissimus dorsi* beef muscle. *J Food Sci Anim Resour* 33:508-514.
- Dashdorj, D., Amna, T., & Hwang, I. (2015). Influence of specific taste-active components on meat flavor as affected by intrinsic and extrinsic factors: An overview. *Eur Food Res Technol* 241: 157-171.
- Dinh TTN, To KV, Schilling MW. 2021. Fatty acid composition of meat animals as flavor precursors. *Meat Mus Biol* 5:34.
- Faustman C, Sun Q, Mancini R, Suman S. 2010. Myoglobin and lipid oxidation interaction: Mechanistic bases and control. *Meat Sci* 86:86–94.

- Folch J, Lees M, Stanley GHS. 1957. A simple method for the isolation and purification of total lipid from animal tissue. *J Biol Chem* 226: 497-500.
- Frank D, Ball A, Hughes J, Raju K, Udayasika P, Stark J, Warner R. 2016. Sensory and flavor chemistry characteristics of Australian beef: Influence of intramuscular fat, feed, and breed. *J Agric Food Chem* 64:4299–4311.
- Gotoh T, Nishimura T, Kuchida K, Mannen H. 2018. The Japanese Wagyu beef industry: Current situation and future prospects—A review. *Asian-Australas J Anim Sci* 31: 933–950.
- Haque MA, Iqbal A, Bae H, Lee SE, Park S, Lee YM, Kim JJ. 2023. Assessment of genomic breeding values and their accuracies for carcass traits in Jeju Black cattle using whole-genome SNP chip panels. *J Anim Breed Genet* 140:519–531.
- Hoa VB, Oliveros MC, Ryu KS, Hwang I. 2010. Development of analysis condition and detection of volatile compounds from cooked Hanwoo beef by SPME-GC/MS analysis. *Korean J Food Sci Anim Resour* 30:73-86.
- Hoa VB, Song DH, Seol KH, et al. 2022. Half-castration is a newly effective method for increasing yield and tenderness of male cattle meat. *Anim Biosci* 35:1258-69.
- Hoa VB, Song DH, Seol KH, Kang SM, Kim HW, Bae IS, Kim ES, Park YS, Cho SH. 2023a. Comparative study on the carcass and meat chemical composition, and lipid-metabolism-related gene expression in Korean Hanwoo and brindle Chikso cattle. *Curr Issues Mol Biol* 45: 3279–3290.
- Hoa VB, Song DH, Seol KH, Kang SM, Kim HW, Bae IS, Kim ES, Park YS, Cho SH. 2023b. A Comparative study on the meat quality, taste and aroma related compounds between Korean Hanwoo and Chikso cattle. *Foods* 12:805.
- Hughes J, Clarke F, Purslow P, Warner R. 2017. High pH in beef *longissimus thoracis* reduces muscle fibre transverse shrinkage and light scattering which contributes to the dark color. *Food Res Int* 101:228–238.

- Jayasena DD, Jung S, Bae YS, Park HB, Lee JH, Jo C. 2015. Comparison of the amounts of endogenous bioactive compounds in raw and cooked meats from commercial broilers and indigenous chickens. *J Food Compos Anal* 37: 20-24.
- Jayasena DD, Nam KC, Kim JJ, Jo C. 2015. Association of carcass weight with quality and functional properties of beef from Hanwoo steers. *Anim Prod Sci* 55:680–690.
- Jo C, Cho SH, Chang J, Nam KC. 2012. Keys to production and processing of Hanwoo beef: A perspective of tradition and science. *Anim Front* 2:32–38.
- Joo ST, Hwang YH, Frank D. 2017. Characteristics of Hanwoo cattle and health implications of consuming highly marbled Hanwoo beef. *Meat sci* 132:45-51.
- Jung S, Nam KC, Lee KH, Kim JJ, Jo C. 2013. Meat quality traits of *longissimus dorsi* muscle from carcasses of Hanwoo steers at different yield grades. *Food Sci Anim Resour* 33: 305-316.
- Kato H, Rhue MR, Nishimura, T. 1989. Role of free amino acids and peptides in food taste. In: Flavor chemistry. Teranishi R, Buttery RG, Shahidi F (Eds). Trends and Development. pp. 158-174.
- Kerth CR, Miller RK. 2015. Beef flavor: a review from chemistry to consumer. *J Sci Food Agric* 95:2783–2798.
- Khan MI, Jo C, Tariq MR. 2015. Meat flavor precursors and factors influencing flavor precursors-A systemic review. *Meat Sci* 110: 278–284.
- Kim YHB, Kemp R, Samuelsson LM. 2016. Effects of dry-aging on meat quality attributes and metabolite profiles of beef loins. *Meat Sci* 111:168–176.
- Korean Institute for Animal Products Quality Evaluation (KAPE). Animal Products Grading Statistical Yearbook (17th Ed) 2022, Sejong, Korea.
- Koutsidis G, Elmore JS, Oruna-Concha MJ, Campo MM, Wood JD, Mottram DS. 2007. Water-soluble precursors of beef flavor: I. Effect of diet and breed. *Meat Sci* 79:124–130.

- Lee HJ, Koh YJ, Kim YK, Lee SH, Lee JH, Seo DW. 2022. MSENNet: Marbling score estimation network for automated assessment of Korean beef. *Meat Sci* 188:108785.
- Lee SH, Kim CN, Ko KB, Park SP, Kim HK, Kim JM, Ryu YC. 2019. Comparison of beef fatty acid and amino acid characteristics between Jeju black cattle, Hanwoo and Wagyu breeds. *Food Sci Anim Resour* 39:402-409.
- Lim DH, Han HK, Kim JH, et al. 2013. Annual report for livestock animal. National Institute of Animal Science, Suwon, Korea. Report No. 11-1390906-000217-10.
- Liu J, Ellies-Oury MP, Stoyanchev T, Hocquette JF. 2022. Consumer Perception of Beef Quality and How to Control, Improve and Predict It? Focus on Eating Quality. *Foods* 11:1732.
- Malgwi IH, Halas V, Grünvald P, Schiavon S, Jócsák I. 2022. Genes related to fat metabolism in pigs and intramuscular fat content of pork: A focus on nutrigenetics and nutrigenomics. *Animals* 12:150.
- Mateo J, Domínguez M, Aguirrezábal MM, Zumalacárregui JM. 1996. Taste compounds in Chorizo and their changes during ripening. *Meat Sci* 44:245–254.
- Mottram DS. 1998. Flavor formation in meat and meat products: A review. *Food Chem* 62: 415–424.
- Park B, Choi TJ, Park MN, Oh SH. 2020. Estimation of environmental effects and genetic parameters of carcass traits on Chikso (Korean brindle cattle). *Asian-Australas J Anim Sci* 33: 525–530.
- Park MN, Choi TJ, Park B, Lee SS, Choi JG, Cho KH, Yang CB, Lee JG, Cho HJ, Mahboob, A, et al. 2016. Distribution of Chikso (Korean brindle cattle) in south Korea and their coat color expression. *Korean J Int Agric* 28:407–413.
- Purslow P, Warner RD, Clarke FM, Hughes JM. 2020. Variations in meat color due to factors other than myoglobin chemistry; a synthesis of recent finding. Review. *Meat Sci* 159: 107941.

- Reiche AM, Oberson JL, Silacci P, et al. 2019. Pre-slaughter stress and horn status influence physiology and meat quality of young bulls. *Meat Sci* 158:1–12.
- Sasaki K, Motoyama M, Mitsumoto M. 2007. Changes in the amounts of water-soluble umami-related substances in porcine longissimus and *biceps femoris* muscles during moist heat cooking. *Meat Sci* 77:167-72.
- Schumacher M, DelCurto-Wyffels H, Thomson J, Boles J. 2022. Fat deposition and fat effects on meat quality—A Review. *Animals* 12:1550.
- Shahrai NN, Babji AS, Maskat MY, Razali AF, Yusop SM. 2021. Effects of marbling on physical and sensory characteristics of ribeye steaks from four different cattle breeds. *Anim Biosci* 34:904-913.
- Smith SB, Lunt DK, Chung KY, Choi CB, Tume RK, Zembayashi M. 2006. Adiposity, fatty acid composition, and delta-9 desaturase activity during growth in beef cattle. *Anim Sci J* 77:478-486.
- Smith SB. 2016. Marbling and its nutritional impact on risk factors for cardiovascular disease. *Korean J Food Sci Anim Resour* 36:435-444.
- Shoulder MD, Raines RT. 2009. Collagen structure and stability. *Annu Rev Biochem* 78: 929-958.
- Song JS, Seong HS, Choi BH, Lee CW, Hwang NH, Lim D, Lee JH, Kim JS, Kim JD, Park YS, et al. 2018. Genome-wide analysis of Hanwoo and Chikso populations using the BovineSNP50 genotyping array. *Genes Genom* 40:1373–1382.
- Tikk M, Tikk K, Torngren MA, Aaslyng MD, Karlsson AH, Karlsson AH, Andersen HJ. 2006. Development of inosine monophosphate and its degradation products during aging of pork of different qualities in relation to basic taste and retronasal flavor perception of the meat. *J Agric Food Chem* 54:7769-7777.

- Vázquez-Mosquera JM, de Mercado E, Fernández-Novo A, Gardón JC, Pesántez-Pacheco JL, et al. 2022. Comparison of pure and crossbred Japanese black steers in growth performance and metabolic features from birth to slaughter at a Spanish fattening farm. *Animals* 12: 1671.
- Weston AR, Rogers RW, Althen TG. 2002. The role of collagen in meat tenderness. A review. *Appl Anim Sci* 18:107-111.
- Wood JD, Enser M, Fisher AV, et al. 2008. Fat deposition, fatty acid composition and meat quality: A review. *Meat Sci* 78: 343-58.
- Xie X, Meng Q, Cui Z, Ren L. 2012. Effect of cattle breed on meat quality, muscle fiber characteristics, lipid oxidation and fatty acids in China. *Asian-Australas J Anim Sci* 25:824–831.
- Yim DG, Kim YJ, Chung KY. 2015. Effect of quality grade and storage time on the palatability, physicochemical and microbial quality of Hanwoo striploin beef. *Korea J Food Sci Anim Resour* 35:449–459.
- Zhang X, Liu C, Kong Y, Li F, Yue X. 2022. Effects of intramuscular fat on meat quality and its regulation mechanism in Tan sheep. *Front Nutr* 9:908355.

Table 1. Proximate composition of *longissimus lumborum* muscle by cattle breeds

	Cattle breed			
	Chikso	BHW	JBC	Hanwoo
Protein (%)	16.92±2.15 ^a	18.21±1.44 ^a	15.5±1.14 ^b	16.98±1.28 ^a
Moisture (%)	67.78±2.47 ^a	63.72±2.29 ^a	68.3±1.35 ^a	55.01±4.46 ^b
Fat (%)	9.25±3.92 ^b	11.78±2.95 ^b	9.14±1.89 ^b	22.85±5.91 ^a
Collagen (%)	1.27±0.39 ^c	1.59±0.12 ^b	1.11±0.19 ^c	2.10±0.44 ^a
Ash (%)	3.44±0.61 ^a	3.77±0.02 ^a	3.49±0.53 ^a	2.12±0.67 ^b

Means in a same row with different superscripts (a,b,c) differ significantly (p<0.05).

BHW: Black Hanwoo, JBC: Jeju black cattle

Table 2. Meat quality properties of *longissimus lumborum* muscle by cattle breeds

Items	Cattle breed			
	Chikso	BHW	JBC	Hanwoo
L* (Lightness)	34.67±1.04 ^b	33.31±2.70 ^b	33.60±1.50 ^b	38.13±3.05 ^a
a* (Redness)	19.36±1.54 ^c	18.90±1.61 ^c	20.89±1.64 ^b	23.46±1.66 ^a
b* (Yellowness)	7.79±1.07 ^c	8.40±0.78 ^{bc}	9.09±0.71 ^b	11.1±1.17 ^a
Shear force (kgf)	5.71±1.63 ^a	4.48±0.56 ^b	5.93±0.69 ^a	3.90±0.69 ^b
Cooking loss (%)	21.01±4.15 ^a	16.20±3.59 ^b	19.87±2.82 ^a	18.22±2.07 ^{ab}
pH	5.67±0.10 ^a	5.45±0.06 ^b	5.54±0.13 ^b	5.66±0.13 ^a

Means in a same row with different superscripts (a,b,c) differ significantly ($p < 0.05$).

BHW: Black Hanwoo, JBC: Jeju black cattle

Table 3. Relative percentage of fatty acids of beef *longissimus lumborum* muscle by cattle breeds

Items	Cattle breed			
	Chikso	BHW	JBC	Hanwoo
C14:0	3.32±0.74 ^b	3.10±0.41 ^b	2.32±0.42 ^c	3.81±0.62 ^a
C16:0	28.31±1.83 ^{bc}	29.45±2.49 ^{ab}	26.94±1.47 ^c	29.93±1.46 ^a
C16:1n7	3.87±1.28 ^b	5.31±0.97 ^a	3.65±0.88 ^b	4.25±0.71 ^b
C18:0	12.16±2.13	11.37±1.87	11.74±1.35	11.00±1.15
C18:1n9	48.91±2.41 ^{ab}	47.36±3.04 ^b	50.41±2.97 ^a	48.86±2.29 ^{ab}
C18:1n7	0.42±0.15	0.47±0.13	0.40±0.16	0.35±0.12
C18:2n6	2.39±0.91 ^{ab}	2.18±0.24 ^{ab}	3.15±2.85 ^a	1.49±0.30 ^b
C18:3n6	0.05±0.04 ^c	0.12±0.04 ^a	0.09±0.03 ^b	0.01±0.01 ^d
C18:3n3	0.09±0.04 ^b	0.16±0.02 ^a	0.16±0.08 ^a	0.04±0.02 ^c
C20:1n9	0.16±0.06 ^b	0.32±0.15 ^a	0.34±0.11 ^a	0.18±0.08 ^b
C20:4n6	0.27±0.28 ^b	0.11±0.04 ^b	0.71±1.60 ^a	0.06±0.02 ^b
C22:4n6	0.05±0.02	0.04±0.02	0.08±0.09	0.03±0.01
SFA	43.80±2.34 ^a	43.92±3.24 ^a	41.00±2.77 ^b	44.74±1.98 ^a
UFA	56.20±2.34 ^b	56.08±3.24 ^b	59.00±2.77 ^a	55.26±1.98 ^b
MUFA	53.37±2.53	53.46±3.27	54.80±3.58	53.64±2.04
PUFA	2.84±1.20 ^{ab}	2.62±0.29 ^{ab}	4.21±4.63 ^a	1.62±0.31 ^b
n3	0.09±0.04 ^b	0.16±0.02 ^a	0.17±0.12 ^a	0.04±0.02 ^b
n6	2.75±1.19 ^{ab}	2.45±0.28 ^{ab}	4.03±4.52 ^a	1.58±0.30 ^b
n6/n3	32.75±12.21 ^{ab}	14.95±1.01 ^b	29.00±30.36 ^b	48.10±23.84 ^a
MUFA/SFA	1.22±0.12 ^b	1.23±0.17 ^b	1.34±0.13 ^a	1.20±0.10 ^b
PUFA/SFA	0.07±0.03 ^{ab}	0.06±0.01 ^{ab}	0.11±0.13 ^a	0.04±0.01 ^b

SFA: Saturated fatty acid; UFA: Unsaturated fatty acid; MUFA: Monounsaturated fatty acid; PUFA: Polyunsaturated fatty acid. BHW: Black Hanwoo, JBC: Jeju black cattle
Means in a same row with different superscripts (a,b,c) differ significantly (p<0.05).

Table 4. Free amino acids content (mg/100g) of *longissimus lumborum* muscle by cattle breeds

Items	Cattle breed			
	Chikso	BHW	JBC	Hanwoo
Glutamic acid	4.57±1.73 ^b	3.81±1.29 ^{bc}	3.03±0.36 ^c	6.78±0.83 ^a
Aspartic acid	1.81±0.42 ^b	2.28±0.20 ^a	2.18±0.02 ^a	1.33±0.00 ^c
Asparagine	1.02±0.44 ^b	1.65±0.20 ^a	1.45±0.09 ^a	0.53±0.22 ^c
Glutamine	13.91±5.96 ^a	6.66±2.78 ^b	7.70±2.56 ^b	18.86±7.95 ^a
Σ Umami taste	21.32±6.80 ^b	14.41±2.96 ^c	14.37±2.97 ^c	27.49±7.44 ^a
Glycine	3.07±2.66 ^b	0.70±0.42 ^c	0.56±0.26 ^c	4.96±2.14 ^a
Alanine	15.28±7.36 ^{ab}	9.20±3.00 ^c	11.04±2.37 ^{bc}	18.89±4.07 ^a
Serine	3.03±0.48 ^{ab}	3.46±0.82 ^a	2.56±0.25 ^b	3.15±0.57 ^{ab}
Proline	2.56±0.67	2.22±0.39	2.18±0.32	2.25±0.51
Threonine	3.14±0.74	2.96±0.53	2.58±0.23	2.92±0.53
Σ Sweet taste	27.09±11.33 ^a	18.54±3.90 ^b	18.91±3.05 ^b	32.17±7.62 ^a
Lysine	3.57±0.87	3.84±0.94	3.74±0.47	3.51±0.58
Valine	3.22±1.41 ^{ab}	2.68±0.82 ^b	2.26±0.40 ^b	3.93±0.93 ^a
Leucine	5.35±2.69 ^b	4.58±2.09 ^{bc}	2.94±0.54 ^c	7.74±2.17 ^a
Isoleucine	1.99±0.57	2.19±0.69	1.81±0.27	2.03±0.50
Arginine	5.56±1.53	5.41±0.97	6.15±0.57	5.84±1.40
Phenylalanine	3.51±0.76 ^{ab}	3.69±1.06 ^a	2.74±0.23 ^b	3.88±0.87 ^a
Tyrosine	4.02±0.66 ^{ab}	4.10±0.77 ^a	3.39±0.20 ^b	4.44±0.86 ^a
Σ Bitter taste	27.23±7.78 ^{ab}	26.49±6.57 ^{ab}	23.03±2.48 ^b	31.37±7.21 ^a

Means in a same row with different superscripts (a,b,c) differ significantly ($p < 0.05$).
BHW: Black Hanwoo, JBC: Jeju black cattle

Table 5. Concentration (mM) of metabolic compounds in *longissimus lumborum* muscle by cattle breeds

	Cattle breed			
	Chikso	BHW	JBC	Hanwoo
Acetate	2.31±0.89 ^{ab}	1.75±0.47 ^{ab}	1.60±0.73 ^b	2.39±0.63 ^a
Alanine	8.22±2.05 ^{ab}	8.58±1.38 ^b	7.87±1.62 ^b	10.12±1.47 ^a
Betaine	4.11±1.32 ^a	3.74±1.25 ^a	3.16±0.89 ^{ab}	2.61±0.40 ^b
Carnitine	10.68±3.42	11.44±1.74	11.86±2.91	11.58±1.56
Carnosine	39.16±7.22	36.13±5.87	37.51±9.81	43.27±2.27
Choline	1.53±0.70 ^a	0.98±0.20 ^b	0.72±0.30 ^b	1.57±0.38 ^a
Creatine	92.32±36.91 ^a	66.08±10.27 ^b	76.80±14.35 ^{ab}	95.60±4.43 ^a
Creatinine	3.81±0.96 ^{ab}	3.48±0.96 ^b	4.63±1.63 ^{ab}	5.25±2.51 ^a
Ethanolamine	1.96±0.77	1.60±0.21	2.06±0.69	3.38±0.97
Fructose 6-phosphate	8.43±1.46	10.15±3.67	10.17±3.90	ND
Glucose	19.97±7.39	16.88±3.29	16.97±3.96	17.31±5.39
Glucose-6-phosphate	19.62±3.28	19.76±4.87	18.76±3.69	ND
Glutamine	7.58±5.08 ^b	3.59±0.93 ^c	4.59±1.89 ^{bc}	11.02±2.79 ^a
Glutathione	1.47±0.68 ^b	1.17±0.24 ^b	1.04±0.28 ^b	2.27±0.33 ^a
Glycerol	14.01±5.11	10.52±2.25	11.42±3.13	15.09±1.60
Glycine	5.04±1.26 ^b	4.34±1.13 ^b	4.29±1.12 ^b	11.50±6.81 ^a
Inosine	1.74±0.28 ^b	1.68±0.28 ^b	2.06±0.30 ^b	3.45±0.88 ^a
Isoleucine	0.99±0.40 ^{ab}	0.82±0.26 ^{bc}	0.72±0.16 ^c	1.14±0.12 ^a
Lactate	275.40±30.40	260.18±36.82	280.63±45.56	282.72±28.95
Leucine	1.80±0.68 ^b	2.10±0.64 ^b	1.09±0.40 ^b	2.51±1.06 ^a
Malonate	7.77±4.39	8.82±2.35	10.69±5.85	6.18±3.69
Mannose	4.19±2.24	5.17±2.00	5.40±2.55	ND
N,N-Dimethylglycine	0.31±0.12 ^a	0.22±0.03 ^b	0.27±0.05 ^{ab}	0.33±0.02 ^a
Niacinamide	0.83±0.37	0.80±0.31	0.87±0.25	ND
O-Acetylcarnitine	4.70±1.29	3.94±0.67	4.56±0.92	3.79±0.44
O-Phosphocholine	1.95±0.64	1.75±0.77	1.60±0.59	2.35±0.45
Phenylalanine	0.73±0.08	0.57±0.16	0.34±0.11	0.91±0.01

Succinate	3.07±1.02	3.35±0.74	3.62±0.66	4.13±0.37
Taurine	7.92±1.75	6.03±1.58	6.60±2.37	ND
Tyrosine	0.71±0.17 ^b	0.49±0.16 ^c	0.40±0.18 ^c	0.89±0.23 ^a
Valine	1.37±0.39 ^{ab}	1.15±0.33 ^{bc}	0.97±0.27 ^c	1.62±0.25 ^a
myo-Inositol	2.15±0.51	2.04±0.97	1.35±0.56	3.84±0.81

Means in a same row with different superscripts (a,b,c) differ significantly (p<0.05).

BHW: Black Hanwoo, JBC: Jeju black cattle

ND: Not detectable.

ACCEPTED

Table 6. Concentration ($\mu\text{g/g}$) of aroma volatile compounds in *longissimus lumborum* muscle by cattle breeds

Items	Retention Time (min)	Cattle breed			
		Chikso	BHW	JBC	Hanwoo
<i>Aldehydes</i>					
2-Methyl pentanal	1.611	0.03 \pm 0.002	0.02 \pm 0.008	0.01 \pm 0.006	0.03 \pm 0.002
2-Methyl propanal	1.860	0.01 \pm 0.009	0.004 \pm 0.000	0.004 \pm 0.000	0.008 \pm 0.000
Butanal	1.994	ND	0.001 \pm 0.000	ND	0.002 \pm 0.000
Butanal, 3-methyl-	2.435	0.02 \pm 0.005	0.008 \pm 0.000	0.005 \pm 0.000	0.029 \pm 0.003
Butanal, 2-methyl-	2.610	0.027 \pm 0.002 ^a	0.008 \pm 0.000 ^{bc}	0.002 \pm 0.000 ^c	0.032 \pm 0.000 ^{ab}
Pentanal	3.036	0.172 \pm 0.06 ^{ab}	0.104 \pm 0.005 ^b	0.021 \pm 0.008 ^b	0.324 \pm 0.024 ^a
Hexanal	5.654	2.071 \pm 0.253 ^{ab}	1.541 \pm 0.272 ^{bc}	0.231 \pm 0.096 ^c	3.104 \pm 0.154 ^a
E,2-Hexanal	7.388	0.002 \pm 0.000	0.001 \pm 0.000	0.001 \pm 0.000	0.006 \pm 0.000
Heptanal	8.808	0.038 \pm 0.004 ^b	0.136 \pm 0.008 ^b	0.071 \pm 0.009 ^b	0.478 \pm 0.094 ^a
E,2-Heptenal	10.291	0.002 \pm 0.000	0.003 \pm 0.000	0.002 \pm 0.002	0.025 \pm 0.005
Benzaldehyde	10.375	0.024 \pm 0.002	0.031 \pm 0.005	0.022 \pm 0.006	0.027 \pm 0.001
E,E-2,4-Decadienal	11.136	0.010 \pm 0.008 ^{bc}	0.032 \pm 0.003 ^a	0.003 \pm 0.000 ^c	0.028 \pm 0.002 ^{ab}
Octanal	11.448	0.166 \pm 0.033	ND	ND	0.242 \pm 0.093
Benzeneacetaldehyde	12.405	0.002 \pm 0.001	0.002 \pm 0.002	0.002 \pm 0.001	0.004 \pm 0.003
E,2-Octenal	12.728	0.002 \pm 0.000 ^b	0.005 \pm 0.000 ^b	0.003 \pm 0.000 ^b	0.012 \pm 0.000 ^a
Nonanal	13.712	0.021 \pm 0.001 ^b	0.042 \pm 0.001 ^b	0.082 \pm 0.005 ^b	0.187 \pm 0.013 ^a
E,2-Nonenal	14.834	0.015 \pm 0.007 ^{ab}	0.004 \pm 0.000 ^b	0.006 \pm 0.000 ^{ab}	0.021 \pm 0.004 ^a
Decanal	15.720	0.002 \pm 0.000 ^b	0.001 \pm 0.000 ^b	0.002 \pm 0.000 ^b	0.009 \pm 0.000 ^a
E,E,2,4-Nonadienal	15.872	0.001 \pm 0.000	0.001 \pm 0.002	0.001 \pm 0.000	0.002 \pm 0.000
E,2-Dodecenal	16.757	0.001 \pm 0.000 ^b	0.001 \pm 0.000 ^b	0.003 \pm 0.000 ^b	0.020 \pm 0.005 ^a
Undecenal	17.547	0.001 \pm 0.000	0.002 \pm 0.000	0.001 \pm 0.000	0.004 \pm 0.001
2-Undecenal	18.527	0.002 \pm 0.000	0.002 \pm 0.000	0.003 \pm 0.000	0.013 \pm 0.003
Pentadecanal	19.255	ND	0.001 \pm 0.000	0.001 \pm 0.000	0.001 \pm 0.000
Σ Aldehydes content		2.532 \pm 1.738 ^b	1.931 \pm 1.153 ^{bc}	0.473 \pm 0.320 ^c	4.497 \pm 2.732 ^a
<i>Alcohols</i>					
3-Methyl-2-butanol	3.772	0.005 \pm 0.000	ND	ND	0.047 \pm 0.001
1-Pentanol	4.601	0.093 \pm 0.001 ^{ab}	0.038 \pm 0.002 ^b	0.009 \pm 0.000 ^b	0.183 \pm 0.064 ^a
1-Hexanol	7.905	0.008 \pm 0.007 ^b	0.012 \pm 0.009 ^b	0.005 \pm 0.000 ^b	0.052 \pm 0.002 ^a
1-Heptanol	10.668	0.006 \pm 0.000 ^b	0.007 \pm 0.000 ^b	0.008 \pm 0.000 ^b	0.051 \pm 0.003 ^a
1-Octen-3-ol	10.892	0.014 \pm 0.001 ^{ab}	0.023 \pm 0.008 ^a	0.006 \pm 0.000 ^b	0.022 \pm 0.002 ^a
1-Octanol	12.999	0.003 \pm 0.000 ^b	0.003 \pm 0.000	0.007 \pm 0.000 ^b	0.033 \pm 0.003 ^a

1-Nonanol	15.056	ND	ND	ND	0.008±0.000
<i>Σ Alcohols content</i>		0.119±0.084 ^b	0.084±0.004 ^b	0.034±0.009 ^b	0.357±0.050 ^a
2,3-Butanedione	1.94	0.047±0.009	0.002±0.000	0.001±0.000	0.034±0.001
2- Butanone	2.0275	0.059±0.007 ^a	0.012±0.005 ^b	0.010±0.003 ^b	0.040±0.006 ^{ab}
2-Pentanone	2.839	0.001±0.000 ^b	0.001±0.000 ^b	0.001±0.000 ^b	0.006±0.001 ^a
3-Heptanone	8.392	0.007±0.000	0.010±0.002	0.001±0.000	0.002±0.000
2-Heptanone	8.507	0.014±0.003 ^b	0.029±0.007 ^{ab}	0.004±0.000 ^b	0.018±0.003 ^a
2-Nonanone	13.455	ND	ND	ND	0.003±0.000
<i>Σ Ketones</i>		0.097±0.000 ^a	0.028±0.007 ^b	0.015±0.005 ^b	0.086±0.007 ^a
<i>Sulfur and nitrogen compounds</i>					
Carbon disulfide	1.754	0.013±0.005	0.004±0.000	0.022±0.009	0.009±0.000
Dimethyl disulfide	3.956	ND	0.001±0.000	ND	ND
Methyl pyrazine	6.377	0.047±0.002	0.002±0.000	0.001±0.000	0.004±0.000
Methional	8.913	0.006±0.001	0.007±0.001	ND	ND
2,5-Dimethylpyrazine	9.158	0.012±0.000	0.006±0.001	0.005±0.000	0.014±0.006
2,3-Dimethyl pyrazine	9.238	0.018±0.024	0.012±0.018	0.002±0.001	0.005±0.000
Dimethyl trisulfide	10.570	0.001±0.000	0.002±0.000	0.003±0.000	0.002±0.000
2-Ethyl-6-methylpyrazine	11.357	0.001±0.000	0.002±0.000	0.001±0.000	0.003±0.000
3-Ethyl-2,5-dimethylpyrazine	13.187	0.015±0.007 ^a	0.003±0.000 ^b	0.002±0.000 ^b	0.006±0.000 ^b
2,5-Dimethyl-3-methylbutylpyrazine	17.717	ND	0.003±0.002	0.002±0.003	0.002±0.000
<i>Σ Sulfur and nitrogen compounds</i>		0.065±0.006	0.026±0.001	0.033±0.003	0.032±0.006
<i>Hydrocarbons</i>					
Ethyl acetate	2.145	0.006±0.001	0.008±0.002	0.008±0.004	0.012±0.009
Toluene	4.546	0.002±0.000 ^b	0.001±0.000 ^b	0.001±0.000 ^b	0.004±0.002 ^a
1,3-Dimethylbenzene	7.815	0.007±0.004 ^{ab}	0.013±0.008 ^a	0.002±0.000 ^b	0.013±0.003 ^a
2,4,6-Dimethyldecane	11.368	ND	0.001±0.000	0.001±0.000	0.008±0.000
2,6,6-Trimethylheptane	12.517	0.002±0.000	0.002±0.000	0.001±0.000	0.001±0.000
<i>Σ Hydrocarbons content</i>		0.010±0.009	0.019±0.002	0.011±0.003	0.020±0.004

Means in a same row with different superscripts (a,b,c) differ significantly (p<0.05).

ND: Not detectable. BHW: Black Hanwoo, JBC: Jeju black cattle.