TITLE PAGE
- Food Science of Animal Resources -
Upload this completed form to website with submission

ARTICLE INFORMATION	Fill in information in each box below
Article Type	Research article
Article Title	Distinguishing aroma profile of highly-marbled beef according to quality grade using electronic nose sensors data and chemometrics approach
Running Title (within 10 words)	Aroma profile of highly-marbled beef with different quality grades
Author	Dicky Tri Utama1,2, Aera Jang1, Gur Yoo Kim1, Sun Moon Kang3, and Sung Ki Lee1
Affiliation	1 Department of Applied Animal Science, College of Animal Life Sciences, Kangwon National University, Chuncheon, Korea 2 Department of Animal Product Technology, Faculty of Animal Husbandry, Universitas Padjadjaran, Jatinangor, Indonesia 3 Department of Animal Products Development and Utilization, National Institute of Animal Science, Rural Development Administration, Wanju, Korea
Special remarks – if authors have additional information to inform the editorial office	
ORCID (All authors must have ORCID) https://orcid.org	Dicky Tri Utama (https://orcid.org/0000-0003-2344-8548) Aera Jang (https://orcid.org/0000-0003-1789-8956) Gur Yoo Kim (https://orcid.org/0000-0001-9973-3367) Sun Moon Kang (https://orcid.org/0000-0003-3947-4337) Sung Ki Lee (https://orcid.org/0000-0002-2989-4787)
Conflicts of interest List any present or potential conflict s of interest for all authors. (This field may be published.)	The authors declare no potential conflict of interest.
Acknowledgements State funding sources (grants, funding sources, equipment, and supplies). Include name and number of grant if available. (This field may be published.)	This research was supported by Korea Institute of Planning and Evaluation for Technology in Food, Agriculture, Forestry and Fisheries (IPET) through Agri-Bioindustry Technology Development Program funded by Ministry of Agriculture, Food and Rural Affairs, Republic of Korea (Project No. 315017-05).
Author contributions (This field may be published.)	Conceptualization: Utama DT, Lee SK. Data curation: Utama DT. Formal analysis: Utama DT. Methodology: Utama DT. Software: Utama DT. Validation: Jang A. Investigation: Jang A, Kim GY, Kang SM, Lee SK. Writing - original draft: Utama DT. Writing - review & editing: Utama DT, Jang A, Kim GY, Kang SM, Lee SK
Ethics approval (IRB/IACUC) (This field may be published.)	This article does not require IRB/IACUC approval because there are no human and animal participants.

CORRESPONDING AUTHOR CONTACT INFORMATION

For the corresponding author (responsible for correspondence, proofreading, and reprints)	Fill in information in each box below
First name, middle initial, last name	Sung Ki Lee
Email address – this is where your proofs will be sent	skilee@kangwon.ac.kr
Secondary Email address	d.utama@unpad.ac.id
Postal address	Department of Applied Animal Science, College of Animal Life Sciences,

	Kangwon National University, Chuncheon 24341, Korea
Cell phone number	-
Office phone number	+82-33-250-8646
Fax number	+82-33-259-5574

7

8

Abstract

Fat deposition in animal muscles differs according to the genetics and muscle anatomical locations. Moreover, different fat to lean muscle ratios (quality grade) might contribute to aroma development in highly marbled beef. Scientific evidence is required to determine whether the abundance of aroma volatiles is positively correlated with the amount of fat in highly marbled beef. Therefore, this study aims to investigate the effect of quality grade on beef aroma profile using electronic nose data and a chemometric approach. An electronic nose with metal oxide semiconductors was used, and discrimination was performed using multivariate analysis, including principal component analysis and hierarchical clustering. The *M. longissimus lumborum* (striploin) of quality grade (QG) 1++, 1+, 1, and 2 of Hanwoo steers (n = 6), finished under identical feeding systems on similar farms, were used. In contrast to the proportion of monounsaturated fatty acids (MUFAs), the abundance of volatile compounds and the proportion of polyunsaturated fatty acids (PUFA) decreased as the quality grade increased. The aroma profile of striploin from carcasses of different quality grades was well-discriminated. QG1++ was close to QG1+, while QG1 and QG2 were within a cluster. In conclusion, aroma development in beef is strongly influenced by fat deposition, particularly the fat-to-lean muscle ratio with regard to the proportion of PUFA. As MUFA slows down the oxidation and release of volatile compounds, leaner beef containing a higher proportion of PUFA produces more volatile compounds than beef with a higher amount of intramuscular fat.

Keywords: Hierarchical clustering; Lipid oxidation; Marbling; Multivariate analysis; Principal component analysis; Volatile compounds

Introduction

Studies on the effect of fat content on the volatile composition of meat have focused on processed meat products, such as meat batter, frankfurter, and ham (Jo et al., 1999; Domínguez et al., 2017; Sirtori et al., 2021). Meanwhile, studies on the effect of carcass quality grade or the intramuscular fat (IMF) level on the volatile profile of beef are still limited. Fat content in beef is positively associated with taste preference (Frank et al., 2016). Further study is necessary in order to provide more scientific evidence to clarify whether the abundance of aroma volatiles is positively correlated with the fat content in highly marbled beef.

In Korea, Hanwoo steers are finished on a high-energy diet and slaughtered at the age of 30–32 months, so that the marbling score and fat content of the highest quality grade loin can reach above 7% and 20%, respectively (Koh et al., 2019). The quality grade, which is determined by the marbling score, influences the generation of beef volatile compounds. Piao et al. (2017) reported that the release of some volatile compounds is affected by the quality grade of Hanwoo beef. The deposition of fat to muscle is affected by genetic factors; even though the fat amount is similar, beef from different breeds have different aroma profiles (Utama et al., 2018). Moreover, IMF content could influence the generation of volatile compounds and the release of such compounds from the matrix of the meat (Echegaray et al., 2021).

Multivariate analysis can help interpret the data for classification. Principal component analysis (PCA) and cluster analysis (CA) are often used to simplify large amounts of data for a better understanding. However, as these tools are unsupervised statistical methods, it is inappropriate to correlate the content of bioactive compounds with in vitro functional properties (Nunes et al., 2015: Granato et al., 2018). PCA has been widely applied as an adaptive descriptive data analysis tool to investigate the authenticity of food and to determine some intrinsic and extrinsic effects on food quality based on their chemical traits, including

the aroma volatile compounds (Procida et al., 2005; Suslick et al., 2010; Wang et al., 2014; Kebede et al., 2018;). In addition, hierarchical clustering, a part of CA, helps to identify the origin of the food, the diversity of microorganisms in the food, and ensures the authenticity and quality of the food (Danezis et al., 2016; Granato et al., 2018). Therefore, the objective of this study was to investigate, using sensor data from an electronic nose and a chemometrics approach, whether the differences in the fat to lean muscle ratio (carcass quality grade) of highly marbled beef contribute to the distinct aroma profile.

Materials and Methods

Sample preparation

The *M. longissimus lumborum* (striploin) of grade 1++, 1+, 1, and 2 Hanwoo steers (n = 6), finished under identical feeding systems on a similar farm, were removed from the left side of the carcasses after 24-h of chilling. The striploin was chosen because this cut is usually used for roasts and grills. Samples were vacuum-packed and distributed to the laboratory in an icebox. Proximate composition, pH, color, and fatty acid analyses were performed on day 4 after postmortem. The remaining sample was lyophilized using a benchtop freeze dryer (Eyela FDU-1200, Tokyo Rikakikai Co., Ltd., Tokyo, Japan) and stored at –24 ℃ for analysis of volatile compounds and aroma patterns. The dry sample was used to avoid the effect of different moisture contents among quality grades.

Proximate composition analysis

Samples were ground using a food blender at minimum speed for 10 s (HMF-1600PB, Hanil Electric, Korea). The proximate composition was determined using the AOAC official methods (AOAC, 2002). Moisture content was determined by dry-heating the samples at 105 ℃ for 24 h and calculating the proportion of weight loss during heating per fresh weight.

Crude fat content was determined by ether extraction using a Soxhlet system. Nitrogen content was determined using the Kjeltec system (2200 Kjeltec Auto Distillation Unit, Foss, Sweden), and crude protein was calculated by multiplying the nitrogen content by 6.25. The ash content was determined by burning the samples in a muffle furnace at 550 ℃ for 16 h.

Fatty acid composition analysis

Meat fat was extracted from the samples using a chloroform-methanol (2:1 v/v) solution and prepared in triplicate (Folch et al., 1957). Fatty acid methyl esters (FAMEs) were prepared in hexane by mixing saponified fat (added with 1 N KOH) with boron trifluoride at 80℃. The fatty acid composition of beef fat was determined using an Agilent gas chromatography system (6890N, Agilent Technologies, USA). The sample (1 μL) was injected into the GC port using an autosampler (7683, Agilent Technologies, USA). A split ratio of 100:1 was programmed for the inlet and the temperature was set to 250 ℃. FAMEs were separated using a WCOT-fused silica capillary column (100 m × 0.25 mm i.d., 0.20 μm film thickness; Varian Inc., USA) with a 1.0 mL/min helium flow. The oven temperature and holding-time were programmed as follows: 150 ℃/1 min, 150–200 ℃ at 7 ℃/min, 200 ℃/5 min, 200–250 ℃ at 5 ℃/min, and 250 ℃/10 min. The temperature of the detector was set to 280 ℃. The peaks were identified as fatty acids using the retention time of the fatty acid standards (47015-U, Sigma-Aldrich Corp., LLC., USA). The peak area of each identified fatty acid was used to calculate the proportion (%) of the total identified peak area.

Volatile compound identification and aroma profiling

The volatile compounds from heated samples were separated and identified by gas chromatography-mass spectrometry (GC-MS) using a modified version of the method described by Ba et al. (2010). Approximately 1 g of dry sample (prepared in duplicate) was

immediately placed in a 50-mL headspace vial and heated at 105 °C in a drying oven for 10 min to release the volatile compounds. Prior to extraction, the sample was calibrated to 60 °C in a drying oven for 10 min. The carboxen®/polydimethylsiloxane fiber (Supelco, Sigma-Aldrich Corp., LLC., USA) with a diameter of 75 µm was injected into the vial for extraction for 30 min. Following extraction, the fiber was injected into the inlet, which was set to 250 °C. The split ration of 1:5 was used for desorbing the volatile compounds for 5 min. Helium was used as the carrier gas at a flow rate of 1 mL/min. Separation of the individual compound was performed using a DB5 fused silica column (30 m x 0.25 mm inner diameter, 0.25 µm film thickness, J&W Scientific, USA) in a gas chromatograph (7890A Agilent Technologies, USA). The GC oven was set to operate at an initial temperature of 40 °C for 2 min, increased to 160 °C (at rate of 5 °C/min), then to 180 °C (at rate of 6 °C/min, holding time of 5 min), and finally to 200 °C (at rate of 10 °C/min, holding time of 5 min). The interface and quadruple temperatures were set at 280 °C and 150 °C, respectively. Volatile compounds were detected using a mass spectrometer (5975C, Agilent Technologies, USA). The ion source temperature of the MS was set to 280 °C with an electron impact of 70 eV. A scanning mass range of 50–450 m/z with a scan rate of 1 scan/s was used. Identification was performed by comparing the experimental spectra with the National Institute of Standards and Technology (NIST) mass spectral library. Data are presented as area units (AU) × 10^6/g.

An electronic nose (FOX3000, Alpha MOS, France) was used for analyzing the aroma pattern. Dry and heated samples (0.5 g) were placed in a 10 mL headspace vial and prepared in duplicate. The vial was sealed with a rubber septa cap (Supelco 29176-U, Sigma-Aldrich Corp., LLC, USA). The samples were heated at 60 °C for 600 s at an agitation speed of 500 rpm. The 2.5 mL of headspace gas was extracted with an automatic sampler syringe (HS 100, Alpha MOS, France) at 65 °C, flow-injected into the port of the electronic nose with synthetic air as the carrier gas (pressure was set to 0.5 bar with 150 mL/min flow rate) and

detected by a metal oxide sensor (MOS) array system with an acquisition time of 150 s. The following sensors were chosen (T30/1, P10/1, P10/2, P40/1, T70/2, PA2) as the sensitivity against fat-derived volatile compounds are high. The sensor resistance ratio $(R\text{-}R_0)/R_0$ was calculated (R is the real-time resistance and R_0 is the sensor's resistance baseline). The time taken to return to baseline after acquisition was 1,080 s. The maximum resistance ratio was considered as the data value of a single measurement.

Statistical analysis

The statistically significant difference between the mean values from different quality grades was determined using a one-way analysis of variance (ANOVA). The mean values were then separated by Duncan's multiple range test at a 5% significance level. Correlation coefficients between the resistance ratio of the six metal sensors of the electronic nose and the peak area of the volatile groups were determined using Pearson's method. Multiple regression analysis was also performed to determine the multiple correlations between the resistance ratio of the six metal sensors of the electronic nose and the peak area of the volatile groups. Two-dimensional principal component analysis (PCA) and cluster dendrograms were used to discriminate the aroma profile according to the sensor resistance ratio. Analyses were performed using R-version 3.3.3 (R Core Team, 2018) with the "agricolae" package for Duncan's multiple range test (De Mandiburu, 2017) and with the "dendextend," "ggfortify," and "ggplot2" packages for plotting the PCA and cluster dendrogram (Galili, 2015; Tang et al., 2016; Wickham, 2016).

Results and Discussion

The proximate composition of beef striploins of different quality grades is presented in

157 Table 1. Among the quality grades, moisture and protein content decreased as the quality
158 grade increased. In contrast, the crude fat content increased as the quality grade increased.
159 Different carcass quality grades showed different fat-to-lean muscle ratios, and the ratio
160 increased linearly as the quality grade increased. No differences were found in ash content
161 among the quality grades. These findings are in accordance with those of previous reports by
162 Piao et al. (2017) and Koh et al. (2019).

163 The fatty acid composition of Hanwoo beef, categorized by different quality grades, is
164 shown in Table 2. No differences were found in the proportions of saturated fatty acids
165 (SFA). However, quality grade 1++ had the lowest proportion of palmitic acid (C16:0) (P =
166 0.04). The highest proportion of monounsaturated fatty acids (MUFAs) was found in beef
167 with the highest quality grade (1++). A higher oleic acid (C18:1n9) proportion was observed
168 in grade 1++ striploin than in lower quality grades, contributing to the increased proportion of
169 MUFAs. In contrast with MUFA, the PUFA proportion was found to be lower in higher
170 quality grades. This is mainly attributed to the higher proportion of linoleic acid (C18:2n6)
171 and arachidonic acid (C20:4n6) in lower-grade striploin. The ratio of omega-6 to omega-3
172 was found to be higher in beef with higher quality grade as the α-linolenic acid (C18:3n3)
173 content decreased. Wood et al. (2008) mentioned that neutral lipids are predominantly
174 deposited into intramuscular adipose tissue to build marbling, whereas PUFAs are mostly
175 deposited into the membrane of muscle cells as cell membranes are built by phospholipids.
176 Cho et al. (2020) reported that coarsely marbled Hanwoo beef loins contain higher
177 proportions of PUFAs than the finer ones, which corresponds to linoleic acid (C18:2n6) and
178 eicosapentaenoic acid (C20:5n3). In other words, the proportion of PUFA increases as the
179 meat cut has more muscle area or tends to be coarse in appearance. PUFAs have a lower
180 melting point and are stable in liquid form at ambient temperature, thus establishing the
181 elasticity of muscle cells to contract and relax (Abbott et al., 2012). Previous studies have

reported that oleic acid is the major fatty acid in highly marbled Hanwoo beef, and this fatty acid may contribute to a more acceptable flavor (Jo et al., 2013). Furthermore, this study confirms that the proportion of oleic acid in Hanwoo striploin increases with an increase in intramuscular fat content or carcass quality grade, as previously reported (Lim et al., 2014; Joo et al., 2017; Piao et al., 2017).

Three major groups of volatile compounds were identified from different quality grades of Hanwoo beef striploin (Table 3). Pyrazine and aldehyde were the two predominant volatile compounds, as the peak areas of these volatile groups were higher than those of hydrocarbons. Lyophilized samples (dry, with low water activity) were used in this study, and the occurrence of the Maillard reaction, which produces pyrazines, was high. Water activity is one of the many factors affecting the rate of the Maillard reaction. The maximum reaction can occur under low water activity conditions (Labuza and Saltmarch, 1981). The proportions of pyrazine and aldehyde ranged from 41–58% and 32–48%, respectively. The hydrocarbon content was the third most abundant, ranging from 10–11%.

Lower grade (QG1 and QG2) striploin released more fatty and meaty flavor aldehydes and hydrocarbons (in area units), such as 2- and 3-methyl butanal, hexanal, heptanal, nonanal, dodecane, and pentadecane, although the proportion of aldehyde groups was higher in higher quality grades (QG1++ and QG1+). The proportion of aldehydes increased as the quality grade (intramuscular fat content) increased. The fat content in emulsion systems and meat products slows down the release of polar volatile compounds, such as aldehydes, ketones, and alcohols (Jo et al., 1999; Jo and Ahn, 1999). Thus, the present results confirm previous findings (Jo et al., 1999; Jo and Ahn, 1999). Aldehyde is also one of the products of the Maillard reaction at high temperatures and is derived from the thermal degradation of unsaturated fatty acids, such as linoleic and linolenic acids (Elmore et al., 2004). Some aldehydes possess pleasant flavors, such as fatty, roasted meat, and an almond-like aroma

based on olfactory analysis (Xie et al., 2008). Ba et al. (2012) found that the *longissimus* tissue of Hanwoo released high amounts of aldehydes. Furthermore, Frank et al. (2016) reported that the proportion of most aliphatic aldehydes increases as the polar lipid content increases. These results indicate that the major aldehydes from leaner striploins were mainly derived from lipid oxidation of muscle cell membrane phospholipids.

Among pyrazines, 2,5-dimethylpyrazine was the most abundant volatile in leaner striploin, comprising more than 30% of the total volatile compounds, followed by styrene, a hydrocarbon, which was remarkably higher than that of higher quality grades. Pyrazines are generally the products of the Maillard reaction between free amino acids and reducing sugars (Yu et al., 2021). The flavor characteristics of pyrazines are roasted and nutty, and are mostly found in roasted beef, coffee beans and nuts (Mortzfeld et al., 2020). This suggests that the roasted aroma from aldehydes in lower-quality grade Hanwoo striploin was obtained from pyrazines. Mottram and Edwards (1983) reported that the amount of pyrazines is negatively associated with the presence of the lipid fraction in beef. Therefore, the present results are in line with those of previous reports. Hydrocarbons, which are the main products of the oxidation of polyunsaturated fatty acids through thermal degradation, were higher in leaner striploins. This can also be associated with the higher proportion of PUFAs in lower quality grade striploin than in higher quality grades. Legako et al. (2015) and Hunt et al. (2016) reported that higher quality grade beef is associated with more neutral lipids (MUFA) than polar lipids (PUFA).

From electronic nose sensor data, the findings from gas chromatography can be associated with the highest intensity of beef volatile compounds released from the lowest quality grade group, wherein a significant proportion of pyrazines was observed. The sensor resistance ratios of the volatile compounds in the headspace derived from the heated samples are shown in Figure 1. The resistance ratios of T30/1, P10/1, P10/2, P40/1, T70/2, and PA2 were

232 significantly higher in the lower quality grade, indicating significant differences in the
233 intensity of volatile compounds. The clustering is clear, indicating statistical discrimination
234 (Utama et al., 2017). Among the volatile groups, aldehydes were positively correlated with
235 the resistance ratio of the T30/1, P10/2, T70/2, and PA2 sensors, while hydrocarbons and
236 pyrazines were positively correlated with the resistance ratio of all sensors (Table 4). Multiple
237 regression models revealed that the combination of all volatile groups showed significant
238 regression with the resistance ratio of each sensor (Table 5). However, each volatile group
239 independently affected the resistance ratio of all sensors. Although the regression model is
240 significant, the linearity or accuracy ($0.57 < R^2 < 0.62$) shows that the model is not good
241 enough to predict the response of the sensor using the abundance of volatile groups.
242 The principal component analysis plot (Fig. 2) and cluster dendrogram (Fig. 3) revealed
243 that the aroma profile differed according to quality grade. The loading plots and the resistance
244 ratio of the sensors led to a group with a high intensity of volatile release. The aroma profile
245 of striploin with different quality grades was well-discriminated, indicating that marbling or
246 the fat to lean muscle ratio affects the release of volatile compounds. However, the cluster
247 dendrogram shows that the aroma profile between the higher quality grades (QG1++ and
248 QG1+) and the lower quality grades (QG1 and QG2) is close to each other with a smaller
249 distance than that between the higher quality grade group and lower quality grade groups.
250

251 ## Conclusion

252 The aroma profile of beef according to carcass quality grade can be discriminated using
253 chemometrics approach. The higher the quality grade, the less abundant volatile compounds
254 released from the beef. The chemometrics approach helps to confirm the effect of fat
255 deposition on the differences in the aroma profiles of beef. The correlation between the

sensor resistance ratio or the response of the electronic nose and the abundance of volatile compounds is strongly dependent on the intensity of the volatile compounds. Therefore, to predict the abundance of individual volatile compounds using the response of each sensor, pre-treatments, such as temperature adjustment prior to the extraction of volatile compounds, should be considered.

References

Abbott SK, Else PL, Atkins TA, Hulbert AJ. 2012. Fatty acid composition of membrane bilayers: Importance of diet polyunsaturated fat balance. Biochim Biophys Acta Biomembr 1818:1309-1317.

AOAC. 2002. Official methods of analysis of AOAC International. 19th ed. AOAC International, Gaithersburg, MD, USA.

Ba HV, Oliveros MC, Ryu K, 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.

Ba HV, Hwang IH, Dawoon J, Amna T. 2012. Principle of meat aroma flavor and future prospect. In Latest Research into Quality Control. Akyar I (ed.). pp 145-176. In Tech, Rijeka, Croatia.

Cho S, Lee W, Seol KH, Kim Y, Kang SM, Seo H, Jung Y, Kim J, Van Ba H. 2020. Comparison of storage stability, volatile compounds and sensory properties between coarsely-and finely-marbled 1+ grade Hanwoo beef loins. Food Sci Anim Resour 40(4):497-511.

Danezis GP, Tsagkaris AS, Camin F, Brusic V, Georgiou CA. 2016. Food authentication: Techniques, trends & emerging approaches. TrAC Trends Anal Chem 85:123-132.

Domínguez R, Pateiro M, Agregán R, Lorenzo JM. 2017. Effect of the partial replacement of pork backfat by microencapsulated fish oil or mixed fish and olive oil on the quality of

282 frankfurter type sausage. J Food Sci Technol 54(1):26-37.
283 Echegaray N, Domínguez R, Cadavez VA, Bermúdez R, Pateiro M, Gonzales-Barron U,
284 Lorenzo JM. 2021. Influence of feeding system on *M. longissimus thoracis et lumborum*
285 volatile compounds of an Iberian local lamb breed. Small Rum Res. 201:106417.
286 Elmore JS, Warren HE, Mottram DS, Scollan ND, Enser M, Richardson RI, Wood JD. 2004.
287 A comparison of the aroma volatile compounds and fatty acid compositions of grilled beef
288 muscle from Aberdeen Angus and Holstein-Friesian steers fed diets based on silage or
289 concentrates. Meat Sci 68:27-33.
290 Folch J, Lees M, Sloaney Stanley GH. 1957. A simple method for the isolation and
291 purification of total lipides from animal tissues. J Biol Chem 226:497-509.
292 Granato D, Santos JS, Escher GB, Ferreira BL, Maggio RM. 2018. Use of principal
293 component analysis (PCA) and hierarchical cluster analysis (HCA) for multivariate
294 association between bioactive compounds and functional properties in foods: A critical
295 perspective. Trends Food Sci Technol 72:83-90.
296 Jo C, Ahn DU. 1999. Fat reduces volatiles production in oil emulsion system analyzed by
297 purge-and-trap dynamic headspace/gas chromatography. J Food Sci 64:641-643.
298 Jo C, Ahn DU, Lee JI. 1999. Lipid and cholesterol oxidation, color changes, and volatile
299 production in irradiated raw pork batters with different fat content. J Food Qual 22:641-
300 651.
301 Jo C, Jayasena DD, Lim DG, Lee KH, Kim JJ, Cha JS, Nam KC. 2013. Effect of
302 intramuscular fat content on the meat quality and antioxidative dipeptides of Hanwoo beef.
303 Korean J Food Nutr 26:117-124.
304 Joo ST, Joo SH, Hwang YH. 2017. The relationships between muscle fiber characteristics,
305 intramuscular fat content, and fatty acid compositions in M. *longissimus lumborum* of
306 Hanwoo steers. Korean J Food Sci Anim Resour 37:780-786.

307 Kebede B, Lee PY, Leong SY, Kethireddy V, Ma Q, Aganovic K, Eyres GT, Hamid N, Oey I.
308 2018. A chemometrics approach comparing volatile changes during the shelf life of apple
309 juice processed by pulsed electric fields, high pressure and thermal pasteurization. Foods
310 7:169-180.
311 Koh KC, Chung KY, Kim HS, Kang SJ, Choi CB, Jo C, Choe J. 2019. Determination of point
312 of sale and consumption for hanwoo beef based on quality grade and aging time. Food Sci
313 Anim Resour 39(1):139-150.
314 Labuza TP, Saltmarch M. 1981. The nonenzymatic browning reaction as affected by water in
315 foods. In Water Activity Influences on Food Quality. Rockland L, Stewart GF (ed). pp
316 605-650. Academic Press, New York, NY, USA.
317 Legako JF, Dinh TTN, Miller MF, Brooks JC. 2015. Effects of USDA beef quality grade and
318 cooking on fatty acid composition of neutral and polar lipid fractions. Meat Sci 100:246-
319 255.
320 Lim DG, Cha JS, Jo C, Lee KH, Kim JJ, Nam KC. 2014. Comparison of physicochemical
321 and functional traits of Hanwoo steer beef by the quality grade. Korean J Food Sci Anim
322 Resour 34:287-296.
323 Mortzfeld FB, Hashem C, Vranková K, Winkler M, Rudroff F. 2020. Pyrazines: Synthesis
324 and industrial application of these valuable flavor and fragrance compounds. Biotechnol J
325 15(11), 2000064. https://doi.org/10.1002/biot.202000064
326 Mottram DS, Edwards RA. 1983. The role of triglycerides and phospholipids in the aroma of
327 cooked beef. J Sci Food Agric 34:517-522.
328 Nunes CA, Alvarenga VO, Sant'Ana A, Santos JS, Granato D. 2015 The use of statistical
329 software in food science technology: Advantages, limitations and misuses. Food Res Int
330 75:270-280.
331 Piao MY, Yong HI, Lee HJ, Fassah DM, Kim HJ, Jo C, Baik M. 2017 Comparison of fatty

acid profiles and volatile compounds among quality grades and their association with carcass characteristics in longissimus dorsi and semimembranosus muscles of Korean cattle steer. Livest Sci 198:147-156.

Procida G, Giomo A, Cichelli A, Conte LS. 2005. Study of volatile compounds of defective virgin olive oils and sensory evaluation: a chemometric approach. J Sci Food Agric 85:2175-2183.

R Core Team. 2018. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. http://www.R-project.org

Sirtori F, Aquilani C, Dimauro C, Bozzi R, Franci O, Calamai L, Pezzati A, Pugliese C. Characterization of subcutaneous fat of Toscano dry-cured ham and identification of processing stage by multivariate analysis approach based on volatile profile. Animals 11(1):13.

Suslick BA, Feng L, Suslick KS. 2010. Discrimination of complex mixtures by a colorimetric sensor array: coffee aromas. Anal Chem 82:2067-2073.

Utama DT, Lee SG, Baek KH, Chung WS, Chung IA, Jeon JT, Lee SK. 2017. High pressure processing for dark-firm-dry beef: effect on physical properties and oxidative deterioration during refrigerated storage. Asian-Australas J Anim Sci. 30(3):424-431.

Utama DT, Lee CW, Park YS, Jang A, Lee SK. 2018. Comparison of meat quality, fatty acid composition and aroma volatiles of Chikso and Hanwoo beef. Asian-Australas J Anim Sci 31(9):1500-1506.

Ventanas S, Estevez M, Andrés AI, Ruiz J. 2008. Analysis of volatile compounds of Iberian dry-cured loins with different intramuscular fat contents using SPME–DED. Meat Sci 79(1):172-180.

Wang Q, Jin G, Jin Y, Ma M, Wang N, Liu C, He L. 2014. Discriminating eggs from different poultry species by fatty acids and volatiles profiling: Comparison of SPME-GC/MS,

electronic nose, and principal component analysis method. Eur J Lipid Sci Technol 116:1044-1053.

Wood JD, Enser M, Fisher AV, Nute GR, Sheard RI, Richardson SI, Hughes FM, Whittington FM. 2008. Fat deposition, fatty acid composition and meat quality: a review. Meat Sci 78:343-358.

Xie J, Sun B, Zheng F, Wang S. 2008. Volatile flavor constituents in roasted pork of mini-pig. Food Chem 109:506-514.

Yu H, Zhang R, Yang F, Xie Y, Guo Y, Yao W, Zhou W. 2021. Control strategies of pyrazines generation from Maillard reaction. Trends Food Sci Technol 112:795-807.

Tables

Table 1. Proximate composition of beef striploin as affected by carcass quality grade

Variable	Quality grade				SEM	*P* value
	1++	1+	1	2		
Moisture (%)	61.4^{c}	64.2^{b}	66.8^{a}	68.0^{a}	1.11	<0.001
Crude fat (%)	24.5^{a}	19.7^{b}	14.4^{c}	12.2^{c}	1.53	<0.001
Crude protein (%)	12.9^{c}	15.0b^{c}	17.6^{ab}	18.7^{a}	0.75	0.01
Ash (%)	1.17	1.08	1.12	1.14	0.02	0.39

SEM, standard error of the means.
Sample size; each quality grade (n = 6).
Carcass quality grade (1++, 1+, 1 and 2) was assessed according to Korea Institute for Animal Products Quality Evaluation (KAPE, 2017).
Different superscripts (a-c) ~~a, b, c~~ in the same row indicate differences among quality grades (P <0.05).

Table 2. Fatty acid composition (%) of beef striploin as affected by carcass quality grade

Fatty acid	Quality grade				SEM	*P* value
	1++	1+	1	2		
C14:0	3.34	3.11	2.88	2.81	0.07	0.35
C16:0	27.9^{b}	29.3^{a}	29.6^{a}	29.2^{a}	0.21	0.04
C16:1n7	4.80	4.32	4.73	4.15	0.09	0.74
C18:0	11.08	12.24	12.2	13.6	0.30	0.42
C18:1n9	50.4^{a}	49.4^{ab}	48.9^{ab}	47.8^{b}	0.39	0.01
C18:2n6	1.16^{b}	1.21^{b}	1.22^{b}	1.78^{a}	0.08	0.02
C18:3n6	0.07	0.08	0.09	0.09	0.00	0.33
C18:3n3	0.10^{c}	0.11^{c}	0.15^{b}	0.25^{a}	0.02	<0.001
C20:4n6	0.09^{b}	0.09^{b}	0.10^{b}	0.13^{a}	0.01	0.03
C22:4n6	0.04	0.04	0.05	0.05	0.00	0.14
SFA	42.4	44.7	44.7	45.6	0.40	0.21
MUFA	56.2^{a}	53.8^{ab}	53.6^{ab}	52.0^{b}	0.49	0.02
PUFA	1.45^{b}	1.54^{b}	1.62^{b}	2.30^{a}	0.11	0.02
n6	1.36^{b}	1.43^{b}	1.46^{b}	2.05^{a}	0.09	0.03
n3	0.10^{c}	0.11^{c}	0.15^{b}	0.25^{a}	0.02	<0.001
n6/n3	14.3^{a}	13.3^{a}	9.60^{ab}	8.25^{b}	0.83	0.01

SEM, standard error of the means.
Sample size; each quality grade (n = 6).
Carcass quality grade (1++, 1+, 1 and 2) was assessed according to Korea Institute for Animal Products Quality Evaluation (KAPE, 2017).
SFA, saturated fatty acids.
MUFA, monounsaturated fatty acids
PUFA, polyunsaturated fatty acids.
Different superscripts (a-c) ~~a, b, c~~ in the same row indicate differences among quality grades (P <0.05).

Table 3. Aroma volatile compounds (area unit x 10^6) released from beef striploin as affected by carcass quality grade

Compound name	Quality grade				SEM	*P* value
	1++	1+	1	2		
Aldehydes						
2-Methyl butanal	11.0^{ab}	15.5^{a}	8.06^{b}	16.9^{a}	1.26	0.03
3-Methyl butanal	14.4	19.3	14.5	21.9	1.54	0.23
Hexanal	18.7	25.6	27.0	28.5	1.68	0.18
Heptanal	3.55	3.19	3.19	4.26	0.19	0.17
Benzaldehyde	3.69^{c}	5.95^{b}	6.63^{b}	14.1^{a}	1.17	<0.01
Nonanal	3.32	3.34	3.30	3.44	0.16	0.99
Hydrocarbons						
Toluene	4.84^{c}	7.90^{bc}	9.50^{b}	13.9^{a}	0.91	<0.001
Styrene	5.02^{b}	5.75^{b}	8.40^{ab}	11.3^{a}	0.70	0.02
Dodecane	1.38	1.48	1.05	1.26	0.10	0.21
Pentadecane	0.87^{a}	0.82^{a}	0.43^{b}	0.50^{b}	0.05	0.03
Pyrazines						
Pyrazine	1.29^{bc}	1.88^{a}	1.01^{c}	1.80^{ab}	0.11	0.01
2-Methyl pyrazine	23.0^{b}	30.9^{b}	32.9^{b}	56.0^{a}	4.07	0.01
2,5-Dimethyl pyrazine	18.3^{b}	26.0^{b}	72.6^{a}	62.9^{a}	6.72	<0.01
2-Ethyl-6-methyl pyrazine	0.77^{b}	0.68^{b}	1.56^{a}	1.59^{a}	0.13	<0.01
2,3,5-Trimethyl pyrazine	2.14^{b}	2.89^{b}	2.93^{b}	6.80^{a}	0.54	0.03
3-Ethyl-2,5-dimethyl pyrazine	0.62^{c}	0.87^{c}	2.26^{b}	6.10^{a}	0.63	<0.01
Total	112.9	152.1	195.3	251.3	16.1	<0.01

SEM, standard error of the means.
Sample size; each quality grade (n = 6).
Carcass quality grade (1++, 1+, 1 and 2) was assessed according to Korea Institute for Animal Products Quality Evaluation (KAPE, 2017).
Different superscripts (a-c) ~~a, b, c~~ in the same row indicate differences among quality grades (*P* <0.05).

Table 4. Correlation coefficients between the volatile groups and resistance ratio of six metal sensors of electronic nose

Major volatile group	Sensor					
	T30/1	P10/1	P10/2	P40/1	T70/2	PA2
Aldehydes	0.34*	0.32	0.33*	0.33	0.34*	0.36*
Hydrocarbons	0.56***	0.55***	0.56***	0.56***	0.56***	0.58***
Pyrazines	0.61***	0.58***	0.59***	0.59***	0.61***	0.63***

Significance level; *** P <0.001, * P <0.05.
Sample size~~;~~ for each quality grade (~~1++, 1+, 1 and 2,~~ n = 6) ~~and each beef cut (striploin, brisket and chuck, n=6)~~.

Table 5. Multiple regression models for resistance ratio of six metal sensors of electronic nose using the measured peak area of volatile groups in study 2 as covariate

Sensor	Intercept	Covariate			R^2	*P* value
		Aldehydes	Hydrocarbons	Pyrazines		
T30/1	<0.001	<0.001*	<0.001***	<0.001***	0.60	<0.001
P10/1	<0.001	<0.001*	<0.001***	<0.001***	0.57	<0.001
P10/2	<0.001	<0.001*	<0.001***	<0.001***	0.59	<0.001
P40/1	<0.001	<0.001*	<0.001***	<0.001***	0.58	<0.001
T70/2	<0.001	<0.001*	<0.001***	<0.001***	0.60	<0.001
PA2	<0.001	<0.001*	<0.001***	<0.001***	0.62	<0.001

Significance level; *** *P* <0.001, * *P* <0.05.
Sample size~~;~~ for each quality grade (~~1++, 1+, 1 and 2,~~ n = 6) ~~and each beef cut (striploin, brisket and chuck, n=6)~~.

Figure Captions

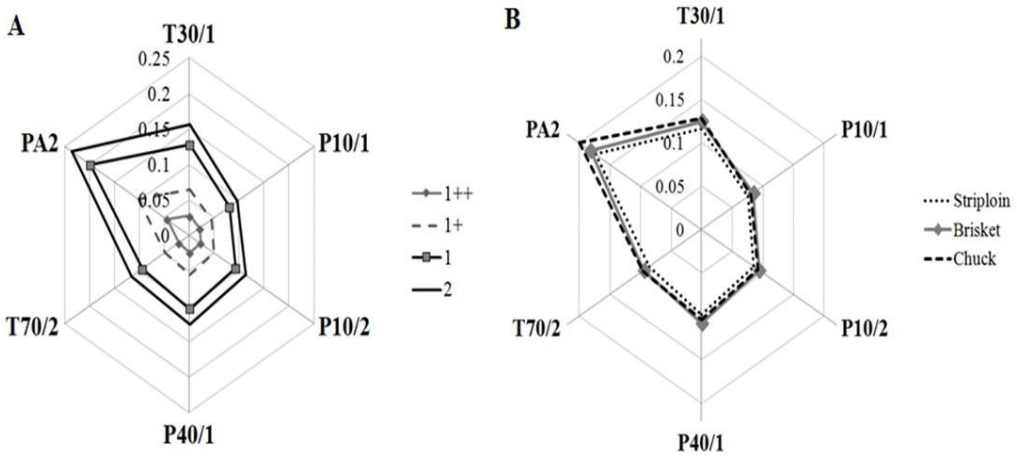

Fig. 1. Differences in aroma intensity among quality grades (A) and beef cuts (B). Data are shown as mean of each sensor's resistance ratio. Metal oxide sensors; T30/1, P10/1, P10/2, P40/1, T70/2, PA2. Sample size for each quality grade (n = 6). Carcass quality grade (1++, 1+, 1 and 2) was assessed according to Korea Institute for Animal Products Quality Evaluation (KAPE, 2017).

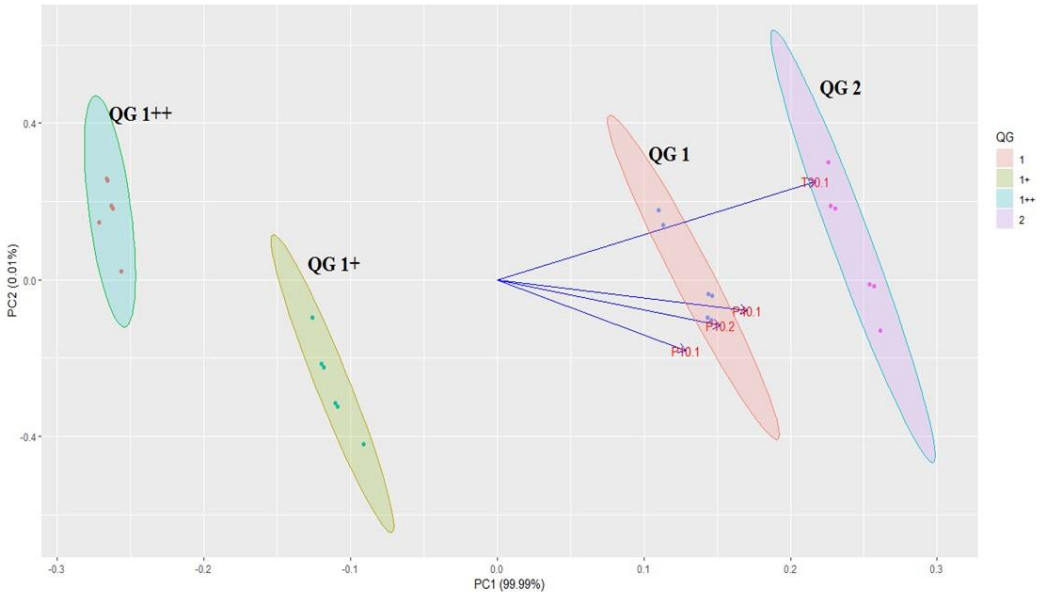

Fig. 2. Principal component analysis plot of the aroma profile of different quality grades (QG). Total contribution of principal component 1 and 2 (PC1 and PC2) is 100%, which means that 100% of data variance is explained. Loading plots; T30/1, P10/1, P10/2, P40/1, T70/2, PA2, are the intensity of the response of the sensor. Sample size for each quality grade (n = 6). Carcass quality grade (1++, 1+, 1 and 2) was assessed according to Korea Institute for Animal Products Quality Evaluation (KAPE, 2017).

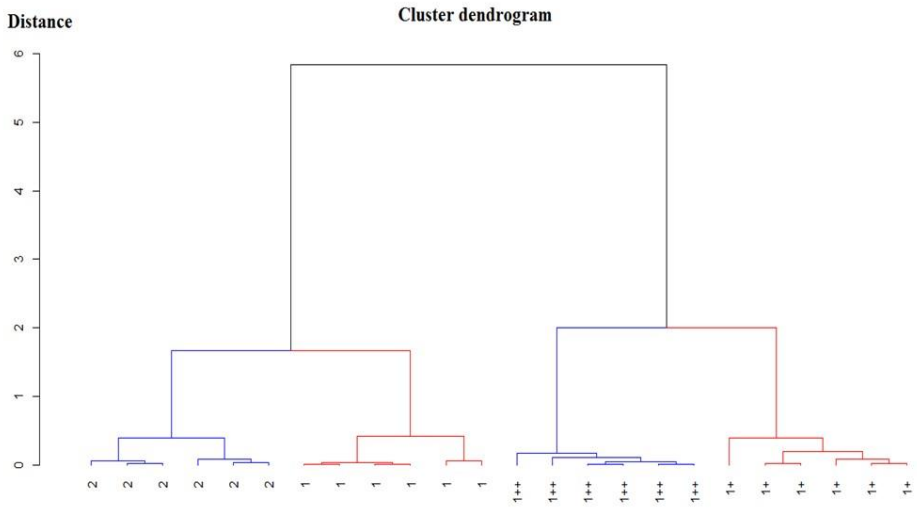

Fig. 3. Cluster dendrogram of the aroma profile of different quality grades. Sample size for each quality grade (n = 6). Carcass quality grade (1++, 1+, 1 and 2) was assessed according to Korea Institute for Animal Products Quality Evaluation (KAPE, 2017).