FACTORS LEADING TO AROMA OF COOKED BEEF: EFFECT OF DIFFERENT SEXES AND AGEING DAYS

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Abstract – This study was conducted to compare effect of free amino acid contents fatty acid composition (sex and ageing effect), and their relationship on deriving volatile compounds in beef. For this study, a total of 30 *longissimus dorsi* muscles (10 bulls, 10 heifers, 10 steers) was collected and divided into 2 portions for ageing (day3 and 14). Free amino acid contents were significantly differed by different sex and ageing periods, and it tended to show the ageing effect was higher relationship than sex. While, fatty acid composition was significantly affected by different sexes. Almost volatile flavor compounds of aldehyde and ketone groups were affected by sex. As a result, variation in each free amino acid contents due to ageing influenced flavor compounds, however almost all levels of aldehyde and ketone flavor compounds were decided by fatty acid in beef. Additionally the interaction between sex and ageing on aroma of cooked beef was not significant.

Key Words – SPME, beef, aroma, sex, ageing.

I. INTRODUCTION

The main reason we eat meat is because of the proteins contained in meat, but nowadays, it change from just being provided protein source to getting delicious protein source. When consumer ate meat, taste of the meat is distributed by tenderness, juiciness and flavor-likeness. Actually many meat scientists focused their studies on meat tenderness. When Korean consumers eat beef, consumers' satisfaction is evaluated by tenderness, flavor and juiciness of the meat [1]. Several researchers reported that flavor of meat had similar impact to distribute meat taste like tenderness [2] . There are many factors influencing release of meat flavor and they are fatty acids, low molecular peptide , and water-soluble components such as free amino acids, nucleotides, vitamins, sugar and phosphates [3] [4]. Moreover scientists found different amounts of flavor precursors participating in these reactions (i.e., amino acids, peptides, intramuscular fat content and sugars) will create varied flavor attributes in a given cooking condition. To date, a large number of studies have reported pre-harvest factors such as breeds, sexes and diets; considerably affect flavor attributes. Indeed, the differences in volatile compound composition among the breeds were considerable and may contribute to the perception of flavor differences in cooked beef [5] [6]. Thus we studied the effects of sexes (pre-harvest factor) and ageing periods (post-harvest factor) on aroma of cooked beef.

II. MATERIALS AND METHODS

Animals, sample collection

We used a total of 30 Korean Hanwoo cattle (10 bulls, 26 months age; 10 heifers, 27 months age; 10 steers, 31 months age) for this experiment. All animals were born and raised on a feedlot (Dan-poongmein farming org.) at Chonbuk province in South Korea. Animals were conventionally slaughtered and chilled for 24 hours. Longissimus dorsi muscle (LD) samples were taken from the right side of the carcasses and moved to the meat science laboratory. Each sample was divided into two portions, vacuum packed, and was assigned to one of the two ageing groups, either 3 days or 14 days. Ageing was done at 4 ^{o}C.

According to Aristoy and Toldra [7], free amino acid was extracted with 0.01N HCl. 300µL of extracted sample was mixed with 10µL of internal standard (L-Citrulline) and 690µL of acetonitril, incubated for 30min at 4^{o}C, and centrifuged at 10,000xg for 15min at 4^{o}C. The supernatant was filtered through a 0.45µm filter. The filtered sample was analyzed with OPA (O-phthalaldehyde) and FMOC (9-fluorenylmethyl chloro-formmate) derivatization using HPLC (Agilent, USA).

Fatty acids were extracted following the direct trancesterification method of Rule [8] using a gas chromatograph (Agilent, 6890N).

The volatile components were analyzed using solid phase microextraction combined with gas chromategra-phy and mass spectrometry following the method as described by Ba et al [9] (2010) with suitable modifications. A gas chromategraphy (Agilent, 6890N) and a mass spectrometer (Ag ilent, 5973) were used for all analyses. The identification of the volatile compounds was performed according to Wiley Registry of Mass Spectral Data 7th edition [10].

The effects of sex and ageing were analyzed using SAS PROC GLM, and the least square means of the two main effects and their interaction were presented (SAS Institute, Cary, NC).

III. RESULTS AND DISCUSSION

Free amino acid contents of LD muscles from Hanwoo bull, heifer and steer by ageing are shown in Table 1. Total free amino acid of bull was significantly higher amount than heifer and steer at 3 and 14 ageing days. Almost free amino acid components was higher amount on bull than heifer and steer, but there was no significant sex affect on histidine, arginine, cysteine, methionine and phenylalanine, whereas, total amount of the free amino acids was increased during ageing. All free amino acids of bulls, heifers and steers beef meat of the 14 ageing days group were significantly higher than the day3 ageing group. Interaction between sex and ageing were observed for serine, phenylalanine, isoleucine, leucine and lysine contents.

Table 2 shows fatty acid composition of LD muscles from bulls, heifers and steers. Stearic acid and linoleic acid were significantly higher in bulls than in heifers and steers. Otherwise, oleic acid was significantly higher in heifers than in steers and bulls. Palmitoleic acid was lower in bulls than in heifers and steers. Several researchers found fatty acids have roles on flavor release in meat; polyunsaturated fatty acid levels had not only played positive effects but also negative effects on flavor characteristics. As a report of Larick et al [11], linolenic acid derived 4-heptanal, 2,4-heptadienal and 2,6-nonal, and linoleic acid derived 2,4-decadial, the report showed that the aldehyde group on volatile flavor compounds was depended animal feeding system like grass or grain fed. Although Elmore et al [12] presented many volatile flavor compounds of aldehyde family were derived by oleic acid, linoleic acid and linolenic acid. In our study, clearly different fatty acid compositions produced by different sex conditions led to various level of flavor compounds of the aldehyde family as well as the ketone family (Table 3). In this study, 2-methyllbutanal, which was key aroma compound in Japanese and Korean high salt soy sauces, and benzenaceta-ldehyde, which was high odor activity value in Japanese high salt say soy sauce [13], were higher in heifer and steer meat than bull meat at both 3 and 14 days of ageing. Levels of 3-methylbutanal, 2-methylbutanal, methanal, octanal, benzenacetald ehyde, 2-decenal, 1,3-dimethylben, methanethiol, methap- yrazine, demethydiulfide were affected by ageing, we expected that this result was derived from that free amino acid contents were changed with ageing. During cooking beef, many flavor precursors (proteins, lipids and sugars, etc.) in meat play their roles in flavor development on heating condition [14].

IV. CONCLUSION

Briefly, free amino acid contents and fatty acid composition varied significantly with different sex and ageing day. Aldehyde and ketone flavor compound group were very affected by sex. As a result, variation in each free amino acid contents by ageing influenced flavor compounds, however almost all flavor compound levels from the aldehyde and ketone families were driven by fatty acid composition of beef meat. Additionally the interaction between sex and ageing on aroma of cooked beef was not significant.

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Table 1. Free amino acid content of *longissmus dorsi* muscles from Hanwoo bulls, heifers, and steers at 3 and 14 days of ageing (μmol/g)

	3days ageing			14days ageing			SEM[1]	F-value[2]		
	Bull	Heifer	Steer	Bull	Heifer	Steer		Sex	Ageing	Sex ×Ageing
Glutamic acid	0.421	0.280	0.254	0.614	0.494	0.614	0.117	6.44**	72.01***	2.99
Asparagine	0.309	0.157	0.272	0.373	0.305	0.348	0.065	15.59***	33.35***	2.49
Serine	0.438	0.309	0.346	0.675	0.658	0.770	0.164	3.19*	150.14***	3.88*
Glutamine	5.835	3.024	4.017	7.581	4.188	5.007	1.113	41.06***	20.46***	0.63
Histidine	0.164	0.098	0.099	0.290	0.280	0.321	0.094	0.8	52.68***	1.3
Glycine	1.412	0.782	0.773	1.559	0.982	1.158	0.308	22.48***	9.41**	0.81
Threonine	0.141	0.207	0.324	0.385	0.360	0.459	0.153	4.05*	20.16***	0.73
Arginine	0.384	0.331	0.345	0.532	0.617	0.616	0.124	0.17	52.21***	1.81
Alanine	20.040	14.981	17.324	22.876	21.123	23.758	3.419	5.32*	33.87***	1.71
Tyrosine	0.252	0.256	0.281	0.371	0.429	0.490	0.080	4.26*	64.79***	1.62
Cysteine	0.106	0.093	0.122	0.135	0.162	0.151	0.036	1.05	20.51***	2.01
Valine	0.230	0.314	0.324	0.417	0.558	0.606	0.116	8.32***	62.74***	0.84
Methionine	1.044	0.872	1.022	1.143	1.087	1.135	0.226	1.51	5.97*	0.39
Phenylalanine	1.072	0.862	0.809	1.460	1.644	1.558	0.145	1.85	291.29***	11.29***
Isoleucine	0.487	0.235	0.256	0.664	0.747	0.708	0.063	14.49***	594.81***	43.86***
Leucine	0.328	0.332	0.371	0.399	0.597	0.709	0.121	10.64***	51.45***	6.47**
Lysine	0.337	0.275	0.270	0.421	0.631	0.752	0.121	5.86**	95.44***	13.96***
Proline	1.142	0.901	1.094	1.870	1.379	1.813	0.221	16.05***	126.4***	2.07
Total amount	34.140	24.309	28.302	41.767	36.239	40.970	4.88	12.45***	72.63***	1.55
Df[3]	5/59									

[1] SEM: Standard error of the mean

[2] *, p<0.05; **, p<0.01; ***, p<0.001

[3] Df: Degrees of freed

Table 2. Fatty acid contents of *longissmus dorsi* muscles from Hanwoo bulls, heifers, and steers (%)

	Bull	Heifer	Steer	SEM[1)]	F-value[2)]
Caprylic acid (C8:0)	0.001^{b}	0.006^{a}	0.006^{a}	0.003	8.96***
Capric acid (C10:0)	0.041^{b}	0.042^{b}	0.060^{a}	0.017	3.91*
Lauric acid (C12:0)	0.311^{a}	0.150^{b}	0.244^{a}	0.099	6.68**
Myristic acid (C14:0)	5.311^{ab}	5.108^{b}	6.153^{a}	0.932	3.54*
Palmatic acid (C16:0)	27.855^{b}	27.675^{b}	29.801^{a}	2.054	3.29*
Palmitoleic acid (C16:1)	4.354^{b}	5.887^{a}	5.699^{a}	0.921	8.23**
Stearic acid (C18:0)	17.465^{a}	10.883^{b}	11.520^{b}	2.154	28.41***
Oleic acid (C18:1)	38.894^{c}	47.910^{a}	44.142^{b}	3.775	14.39***
Linoleic acid (C18:2)	5.433^{a}	2.068^{b}	2.175^{b}	1.840	10.81***
Linolenic acid (C18:3)	0.298^{a}	0.228^{ab}	0.156^{b}	0.116	3.72*
Aricidic acid (C20:0)	0.037	0.043	0.033	0.028	0.32
Behenic acid (C22:0)	0.000	0.000	0.013	0.023	1.00
Df[3)]	2/29				

1) SEM: Standard error of the mea

2) *, p<0.05; **, p<0.01; ***, p<0.001

3) Df: Degrees of freedom

a-c Means in the same row having different superscript letters are significantly different

Table 3. Aldehyde and ketone groups on volatile compounds of *longissmus dorsi* muscles from Hanwoo bulls, heifers and steer sat 3 and 14 ageing days of ageing

	3 days ageing			14 days ageing			SEM[1)]	F-value[2)]		
	Bull	Heifer	Steer	Bull	Heifer	Steer		Sex	Ageing	Sex*Ageing
Aldehyde group										
Acetaldehyde	0.064	0.085	0.083	0.055	0.083	0.088	0.024	7.23**	0.09	0.45
2-methylpropanal	0.012	0.019	0.021	0.017	0.044	0.030	0.032	1.51	2.58	0.51
2-butenal	0.001	0.003	0.005	0.003	0.001	0.003	0.006	0.61	0.14	0.44
3-methylbutanal	0.051	0.076	0.099	0.099	0.156	0.206	0.048	12.89***	39.84***	1.95
2-methylbutanal	0.031	0.062	0.070	0.054	0.090	0.112	0.037	8.55***	10.14**	0.32
Pentanal	0.129	0.269	0.244	0.135	0.241	0.256	0.084	13.79***	0.02	0.34
Hexanal	0.806	1.167	1.080	0.881	1.103	1.163	0.337	4.75*	0.13	0.3
Furfural	0.002	0.000	0.000	0.017	0.026	0.016	0.013	0.81	31.1***	1.05
Heptanal	0.330	0.587	0.511	0.244	0.459	0.418	0.214	6.53**	3.39	0.06
Methional	0.000	0.000	0.000	0.004	0.005	0.011	0.012	0.54	4.16*	0.54
E-2-heptenal	0.023	0.051	0.044	0.012	0.047	0.037	0.024	9.17***	1.23	0.09
Benzaldehyde	1.108	0.887	1.017	1.181	0.881	1.112	0.308	3.77*	0.46	0.15
Octanal	0.524	0.635	0.618	0.371	0.513	0.485	0.254	1.4	4.29*	0.02
Benzenacetaldehyde	0.000	0.012	0.011	0.005	0.043	0.046	0.021	9.12***	18.34***	2.82
2-octenal	0.041	0.071	0.067	0.033	0.076	0.067	31	7.61**	0.02	0.2
Nonanal	0.564	0.543	0.644	0.511	0.514	0.490	0.266	0.12	1.3	0.31
2-nonenal	0.030	0.070	0.068	0.019	0.057	0.055	0.032	8.69***	2.18	0.01
Decanal	0.036	0.019	0.025	0.032	0.019	0.020	0.013	6.41**	0.6	0.13
E-2-decenal	0.049	0.094	0.103	0.040	0.083	0.070	0.051	4.75*	1.75	0.33
Benzeneacetaldehyde alpha	0.000	0.000	0.000	0.003	0.000	0.002	0.004	0.8	3.01	0.8
Undecanal	0.020	0.009	0.012	0.023	0.012	0.006	0.011	6.79**	0	0.77
E,E,2,4-decadienal	0.019	0.027	0.030	0.019	0.024	0.021	0.012	1.77	1.64	0.71
2-undecenal	0.036	0.049	0.063	0.028	0.034	0.037	0.028	2	4.92*	0.46
Tridecanal	0.016	0.002	0.003	0.019	0.000	0.002	0.007	28.07***	0.05	0.46
Tetradecanal	0.159	0.066	0.070	0.204	0.044	0.063	0.086	13.16***	0.06	0.83
Ketone group										
2-propanone	0.030	0.038	0.039	0.034	0.041	0.050	0.012	5.42**	3.5	0.63
2,3-butanedione	0.005	0.016	0.013	0.002	0.014	0.014	0.009	10.18***	0.28	0.28
2-butanone	0.053	0.076	0.078	0.055	0.072	0.086	0.023	7.99***	0.15	0.38
3-hydroxy-2-butanone	0.009	0.033	0.024	0.004	0.028	0.035	0.025	5.77**	0.01	0.72
1-(acetyloxy)-2-propanone	0.004	0.014	0.013	0.005	0.011	0.010	0.007	5.97**	0.62	0.54
2-heptanone	0.038	0.016	0.025	0.028	0.024	0.014	0.019	3.3*	0.73	1.5
Df[3)]	5/59									

1) SEM: Standard error of the mean

2) *, p<0.05; **, p<0.01; ***, p<0.001

3) Df: Degrees of freedo