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Characterization of Beef from Cross-bred Cows Using Principal Component Analysis

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ABSTRACT

The current study was undertaken to evaluate various quality attributes of beef from cross-bred dairy cows and to characterize it using principal component analysis (PCA). Ten different muscles each from six culled cross-bred cows (Holstein Friesian x Jersey, four to six years old) were analysed for 22 variables including physico-chemical, compositional and sensory attributes. The coefficients of variation of different attributes were found to range from 0.9 to 58.41 per cent. PCA transformed the variables into eight principal components (PCs) which explained more than 79.53 per cent of total variability. PC1 accounted for 19.37 per cent of total variability and it comprised of sensory attributes (excluding appearance and flavour), shear force, collagen content and collagen solubility. PC2 was characterized by b* and chroma. Loading plots of the first two PCs revealed high correlation between most of the eating quality attributes. Shear force, myofibril fragmentation index and collagen content formed another group of highly correlated variables. The study has revealed that PCA can be effectively used for interpretation of large amount of data generated in studies like quality profiling of beef from cross-bred dairy cows.

Keywords: *Beef, Principal component analysis, Sensory attributes, Shear force*

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INTRODUCTION

India has 190.9 million cattle population (DAHD 2012) comprising of about 40 breeds and distributed to different agro-climatic conditions. The country has produced about 0.33 million tonnes of beef out of 7.3 million tonnes of total meat production during 2016-17 (DAHD 2017). Meat quality attributes can be characterized by various physico-chemical, sensory, nutritional and microbiological parameters. Thus, comprehensive evaluation of beef quality characteristics can generate a large set of complex data which may be very complicated to interpret. Though all characteristics could be relevant to the quality of beef, identifying a smaller set of variables that can effectively explain the total variation in the data would be a very worthwhile methodology. Classical methods of statistical processing of large set of data deliver an important methodology to study every single variable. However, these methods may not provide holistic information on the relationships between the variables selected and also do not allow grouping of samples with identical characteristics.

Principal component analysis (PCA) is one of the most basic methods of data compression developed to analyse large data matrices (Naes *et al*. 1996). PCA explains the variance-covariance structure of a large set of data through few linear combinations of the variables. The general objectives of PCA are data reduction and interpretation (Johnson and Wichern 2007). PCA linearly transforms the original set of variables into a substantially smaller set of uncorrelated variables that represent the whole information in the original set of variables. The linear composites are ordered with respect to their variations, so that the first few principal components (PC) account for most of the variation present in the original variables (Duntemann 1989). In order to interpret the new composite variables, we need to study the directions of different components. These directions represent the relationship between the principal components and the original variables. The plots of such directions are two or three-dimensional scale plots, called PC loading plots. In the loading plots, variables close together are positively correlated, while variable lying opposite to each other tend to have negative correlation. The more a variable is away from the axis origin, the better it is appreciated in the considered plane (Naes *et al*. 1996).

The present study was undertaken to apply PCA to analyse various physico-chemical, structural and sensory attributes of meat from mature cross-bred cows.

MATERIALS AND METHODS

Sample collection: Six cross-bred culled dairy cows (Holstein Friesian x Jersey) from Cattle Breeding Farm, Thumburmuzhy, Kerala were utilized in this study. All animals were in the age group of four to six years. The reasons for culling included loss of production, infertility and production diseases. They were reared intensively under similar management practices with occasional periods of grazing. The animals were slaughtered at the Meat Technology Unit, Kerala Veterinary and Animal Sciences University, Mannuthy after 12-24 h fasting as per scientific slaughter procedures. Ante-mortem and post-mortem inspections were conducted for each animal. The carcasses were electrically stimulated (100-110 V, 1.5 to 2 min) and the following muscles were immediately harvested from each carcass by hot deboning. Ten muscles were selected generally based on yield. *viz. serratus ventralis cervicis, supraspinatus, infraspinatus, triceps brachii, longissimus thoracis et lumborum, psoas major, vastus lateralis, rectus femoris, semimembranosus* and *biceps femoris*, and separable fat and the connective tissues were removed.

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Each muscle was packed in high density poly ethylene (HDPE) pouches and aged for 72 h at 2-4^{o}C (Samsung Digital Inverter Technology, India). After ageing, each muscle was portioned parallel to the direction of muscle fibers for sensory evaluation, determination of Warner-Bratzler shear force and for analysis of physico-chemical and structural attributes. The muscle portions were again packed in HDPE pouches and transferred to deep freezer and maintained at -18^{o}C until further analysis which took place within one week of freezer storage. The samples were thawed at 4±1^{o}C for 12 h before assessment of the parameters. The samples were analysed for the following variables.

Physico-chemical characteristics

pH: Ultimate pH was measured 72 h of post slaughter as per the method of O'Halloran *et al.* (1997).

Water holding capacity (WHC): Water holding capacity (WHC) was estimated after 72 h of ageing using filter paper press method as per Grau and Hamm (1957).

Colour (Hunter L*, a*, b*, Hue and Chroma): Colour of the each muscle was objectively determined using Mini Scan XE Plus Spectrophotometer (Hunter Lab, Virginia, USA) with diffuse illumination.

Myofibril fragmentation index (MFI): MFI of each muscle sample was determined after 72 h of ageing by the procedure outlined by *et al.* (1980).

Warner-Bratzler shear force (WBSF): WBSF of each muscle sample was determined by the method outlined by Wheeler *et al.* (1997). Three cores of 1.27 cm diameter were taken from each cooked meat along the longitudinal orientation of muscle fibres. Each core was sheared perpendicular to the muscle fibre on a Texture Analyzer (Model EZ-SX, Shimadzu Corporation, Kyoto, Japan) at a cross head speed of 200 millimetre /min. WBSF was expressed in Newton (N).

Drip Loss (DL) and Cooking Loss (CL): DL at 24 h of each muscle sample was estimated as per the method outlined by Honikel (1987). CL of each muscle sample was calculated as per Boccard *et al.* (1981).

Structural characteristics

Sarcomere length (SL): SL of each muscle fibres was measured as per the method outlined by Hostetler *et al.* (1972). Sarcomere lengths of 25 randomly selected fibre fragments were measured under Trinocular Research Microscope (Leica DM 2000 LED, Germany) using 10X eye piece and 100X objective under 20 micrometer calibration and expressed in micrometer (μm).

Fibre diameter (FB): FB was measured as per the method outlined by Jeremiah and Martin (1977). Muscle fibre diameter was measured as the mean cross-sectional distance between exterior surfaces of sarcolemmae of 20 randomly selected muscle fibres and expressed in micrometer measured under Trinocular Research Microscope (Leica DM 2000 LED, Germany) equipped with 20X objective and 10X eye piece under 100 micrometer calibration and expressed in micrometer (μm).

Compositional characteristics

Fat content (FC): FC was estimated as per AOAC (1990).

Collagen content (CC): CC of each muscle sample was determined as per Stegman and Stadler (1967). Collagen content was determined by multiplying the hydroxyproline content with 7.25 and was expressed as per cent of fresh muscle weight.

Collagen solubility (CS): CS of each buffalo muscle was determined as per Hill (1996) which was determined from the soluble hydroxyproline content of the sample. The collagen solubility was calculated by dividing the soluble collagen with collagen content and expressed as per cent of total collagen.

Sensory evaluation: The sensory evaluation of each muscle was conducted by a semi-trained panel (n=10) consisting of faculty and post-graduate students from the Department of Livestock Products Technology, College of Veterinary and Animal Sciences, Mannuthy. They were briefly told about the nature of the experiment without disclosing the identity of samples. Meat samples used were cut into approximately equal sizes (1.5 x 1.5 x 1.9 cm) and were cooked by indirect pressure cooking in small stainless-steel boxes.

All panelists received two cubes each of cooked beef muscles coded with three random digit numbers along with a score card. The panelists were asked to rate the samples for appearance, tenderness, juiciness, flavour, amount of connective tissue and overall acceptability on an eight-point hedonic scale 8 and 1 are respectively for maximum and minimum sensory scores (AMSA 1983). Two fore-noon sessions were scheduled with a gap of 30-45 min between the sessions. Panelists were provided with filtered water to cleanse their palate between samples during sensory evaluation.

Statistical analysis: Data recorded were analysed statistically by using PCA for identifying the underlying structure of the variables. PCA with varimax rotation was used for identifying unrelated components in the PCA. Data analysis was done by the dimension reduction procedure of SPSS Software (Version 21.0) (Snedecor and Cochran 1994).

RESULTS AND DISCUSSION

Coefficient of variation of physico-chemical attributes: The coefficient of variation for some of the variables like pH, L*, WHC, appearance, amount of connective tissue and hue was less than 10 per cent, while CC, CS, DL and FC showed more than 30 per cent coefficient of variation. Highest coefficient of variation was recorded for DL (43.96 per cent) followed by FC (58.41 per cent) (Table 1). Similar observations were reported by the Destefanis *et al.* (2000) and Kopuzlu *et al.* (2011) for beef and Prajwal *et al.* (2017) for buffalo meat.

Correlation coefficients of physico-chemical and sensory attributes: WBSF and MFI were significantly ($p<0.01$) negatively correlated to collagen solubility and all the sensory attributes of beef, and were positively correlated with collagen content (Table. 2). Silva *et al.* (1999), Whipple *et al.* (1990), Rhee *et al.* (2004) and Destefanis *et al.* (2000) reported similar results with beef and Rajagopal and Oommen (2015) and Prajwal *et al.* (2017) for buffalo meat. The a* and b* values were significantly ($p<0.01$) correlated with hue and chroma. Karamucki *et al.* (2006) reported significant correlation for a* and b* with hue and chroma of pig longissimus muscle. Fat content was significantly ($p<0.01$) correlated to flavour, juiciness and hue of beef. Juiciness of muscle has an important fat component (Savell and Cross 1988).

Table 1: Mean, standard error and coefficients of variation of variables

Attributes	Mean	Std. Error	Coefficient of variation
pH	5.53	0.01	0.99
L* (Lightness)	41.86	0.51	9.46
a*(Redness)	18.01	0.33	14.02
b* (Yellowness)	18.02	0.28	12.21
Warner-Bratzler shear force (N)	35.68	1.38	29.86
Myofibril fragmentation index	797.60	10.34	10.04
Water holding capacity	0.35	0.00	9.98
Cooking loss (per cent)	26.47	0.55	16.06
Sarcomere length (µm)	16.22	0.41	19.74
Fiber diameter (µm)	41.21	1.55	29.12
Fat content (per cent fresh weight)	2.18	0.16	58.41
Collagen content (per cent fresh weight)	0.54	0.02	32.81
Collagen solubility (per cent of collagen)	3.84	0.19	37.84
Drip loss (per cent)	3.17	0.18	43.96
Appearance	6.28	0.06	7.24
Tenderness	5.76	0.13	17.07
Juiciness	5.63	0.08	10.56
Flavour	5.37	0.09	13.41
Amount of connective tissue	5.14	0.07	9.86
Overall acceptability	5.89	0.08	10.18
Hue angle	45.09	0.58	9.90
Chroma	25.55	0.35	10.57

Principal components: Results of PCA of the 22 variables which gave eight PCs are represented in Table 3. Out of the variables, the eight PCs were extracted using the Kaiser criterion (Johnson and Wichern 2007) to determine the number of components, retaining only those components which had Eigenvalue greater than one (Figure 1). The eight PCs could explain a cumulative variance of 79.53 per cent. Similar observations were reported by the Destefanis *et al.* (2000) and Kopuzlu *et al.* (2011) for beef and Prajwal *et al.* (2017) for buffalo meat.

Table 3: Eight principal components

Components	Eigenvalues	Percent of variance	Percent cumulative Variance
1	4.26	19.37	19.37
2	2.60	11.84	31.21
3	2.05	9.31	40.52
4	1.92	8.71	49.23
5	1.76	8.02	57.25
6	1.72	7.82	65.06
7	1.68	7.66	72.72
8	1.50	6.81	79.53

Table 2: Correlation coefficients of physico-chemical and sensory attributes of beef

	pH	L*	a*	b*	WBSF	MFI	WHC	CL	SL	FB	FC	CC	CS	DL	AP	TA	JU	FL	OA	ACT	H	CH
pH		0.17	0.09	0.11	-0.07	0.02	-0.02	0.02	0.09	-0.02	0.19	-0.14	0.22	-0.07	-0.06	-0.02	-0.15	-0.30*	-0.16	0.18	0.03	0.18
L*			-0.14	0.34**	0.11	-0.20	0.23	0.16	0.07	0.27*	-0.37**	-0.02	0.06	0.17	0.35**	0.02	-0.03	-0.06	0.02	-0.13	0.35**	0.08
a*				0.29*	-0.11	-0.22	-0.17	-0.20	0.20	-0.17	0.19	-0.30*	0.28*	0.21	0.03	0.13	-0.03	0.05	0.06	0.07	-0.68**	0.81**
b*					-0.08	-0.18	-0.20	-0.03	0.15	-0.10	0.08	-0.30*	0.41**	0.17	0.23	0.16	0.00	0.10	0.01	0.14	0.42**	0.76**
WBSF						0.30*	0.34**	0.03	-0.11	-0.14	0.00	0.32*	-0.37**	0.00	-0.29*	-0.77**	-0.52**	-0.44**	-0.54**	-0.63**	0.06	-0.19
MFI							0.10	-0.06	-0.13	-0.07	0.12	0.30*	-0.43**	-0.09	-0.36**	-0.32*	-0.24	-0.32*	-0.22	-0.30*	0.06	-0.22
WHC								-0.23	0.09	0.20	0.01	0.32*	-0.31*	-0.06	0.00	-0.26*	-0.03	-0.05	-0.05	-0.10	0.05	-.27*
CL									-0.17	0.06	-0.30*	0.16	-0.09	-0.33*	0.06	-0.13	-0.16	0.01	-0.08	-0.13	0.11	-0.10
SL										-0.15	0.04	0.00	0.04	0.08	0.07	0.13	0.20	0.14	0.24	0.26*	-0.11	0.25
FB											-0.15	0.18	-0.13	-0.03	0.09	0.06	0.04	0.12	0.07	0.04	0.15	-0.17
FC												-0.04	0.18	0.09	-0.40**	0.05	0.11	-0.04	-0.11	0.27*	-0.07	0.18
CC													-0.75**	-0.24	-0.26*	-0.47**	-0.21	-0.07	-0.25	-0.13	0.06	-0.38**
CS														0.03	0.20	0.41**	0.21	0.15	0.22	0.29*	0.08	0.43**
DL															0.26*	0.23	0.30*	0.14	0.26*	0.02	-0.06	0.24
AP																0.47**	0.41**	0.47**	0.56**	0.29*	0.17	0.17
TA																	0.75**	0.51**	0.77**	0.63**	-0.01	0.24
JU																		0.62**	0.83**	0.576**	0.04	0.03
FL																			0.56**	0.50**	0.05	0.11
OA																				0.53**	0.00	0.10
ACT																					0.05	0.18
H																						-0.21
CH																						

Note: WBSF-Warner-Bratzler shear force, MFI-Myofibril fragmentation index, WHC-Water holding capacity, CL-Cooking loss, SL-Sarcomere length, FB-Fiber diameter, FC-Fat content, CC-Collagen content, CS-Collagen solubility, DL-Drip loss, AP-Appearance, TD-Tenderness, JU-Juiciness, FL-Flavour, ACT-Amount of connective tissue, OA-Overall acceptability, H-Hue and CH-Chroma, L*-lightness a*- redness and b*-yellowness: **$p<0.01$ and *$p<0.05$.

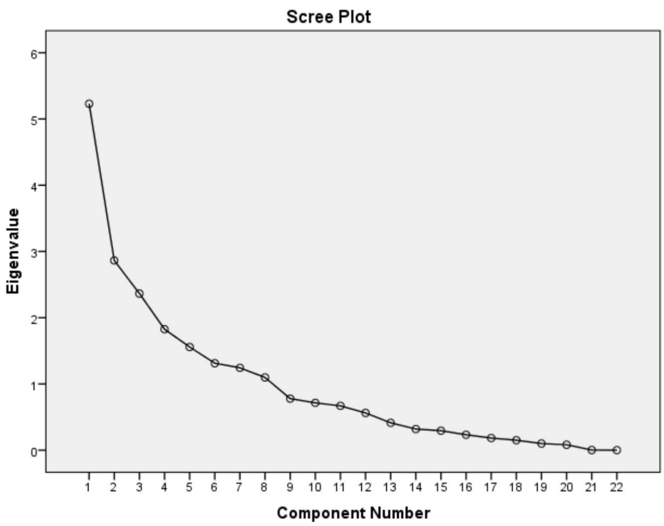

Figure 1: Scree plot

The first PC accounted for 19.37 per cent of the variation. In a study to characterize beef from Holstein Friesian young bulls using PCA, Kopuzlu *et al.* (2011) observed the first PC to be explaining 28.66 per cent of variation. Destefanis *et al.* (2000) observed that the first PC accounted for 33.90 per cent of variation in beef and Prajwal *et al.* (2017) observed the first PC to be explaining 24.40 per cent of variation for buffalo meat. The first PC was represented by significantly high component loading of WBSF, CC, CS and sensory attributes of beef except appearance and flavour. The first PC seemed to explain the maximum of 'eating quality attributes' in beef. The second component explained 11.84 per cent of total variance with high component loading of b* and chroma. Third PC explained 9.31 per cent of total variance and showed high component loading for L*, MFI, FC and sensory appearance. The fourth PC accounted for 8.71 per cent for total variability, with higher loading for a* and hue of beef.

Loading plot of first two principal components: The loading plot for the first two PCs is shown in (Figure 2). The Principal component loadings has been tabulated in Table 4.

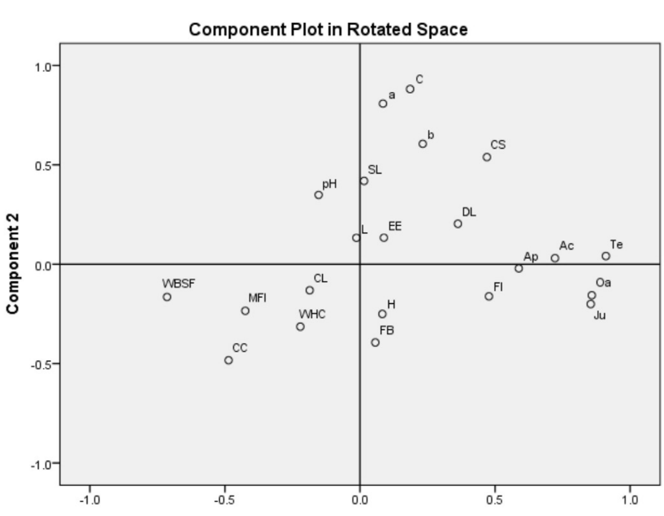

Figure 2: Component loading plot

Note: WBSF-Warner-Bratzler shear force, MFI-Myofibril fragmentation index, WHC-Water holding capacity, CL-Cooking loss, SL-Sarcomere length, FB-Fiber diameter, FC-Fat content, CC-Collagen content, CS-Collagen solubility, DL-Drip loss, AP-Appearance, TD- Tenderness, JU-Juiciness, FL-Flavour, ACT-Amount of connective tissue, OA-Overall acceptability, H-Hue C-Chroma, L-lightness a- redness and b-yellowness.

The PCs are interpreted according to the correlations between each attribute and each PC, thus measurements close to each other are positively correlated, measurements separated 180° are negatively correlated, whereas those separated by 90° are independent (Kopuzlu *et al.* 2011). All the eating quality attributes are closely clustered and placed away from the origin on the right-hand side of the plot. The WBSF, MFI and CC are closely placed and situated away from the origin on the left-hand side of the plot. Destefanis *et al.* (2000) also reported closely located eating quality attributes on the right side and hydroxyproline content and WBSF on the left side of the PC loading plot for beef and Prajwal *et al.* (2017) reported WBSF, MFI, CC and CS and eating quality attributes of buffalo meat to be closely placed and situated away from the origin on the left-hand side and right-hand side of the plot, respectively. FC, SL and FB are also closely situated but are closer to the origin. It may thus follow that one parameter among MFI, CC or WBSF may explain the variation between muscles with respect to these attributes. These attributes were negatively correlated with the sensory scores as these two groups were placed opposite to each other. Similarly, as eating quality attributes were closely correlated, one attribute among these, say sensory tenderness may be used to explain the sensory variability in these muscles. The CS and sensory amount of connective tissue were only moderately correlated. The sensory attributes were placed away from pH and colour characteristics. The structural and compositional characteristics including FB, SL and FC were not sufficient to explain the variability between the muscles. Some physico-chemical attributes like cooking loss, water holding capacity and pH were also inadequate to explain the variability. Destefanis *et al.* (2000) stated that the more the variables are away from the origin in the loading plot; the better it is represented in the considered plane.

CONCLUSIONS

The study has shown that PCA can be effectively used for a general judgment of beef quality, by identifying the groups of variables that determine the nature and extent of variability in beef by the techniques of data reduction and visualization. However, for more analytical information intended to estimate any effects and explain differences between treatments, traditional statistical methods may be applied.

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Table 4: Principal component loadings

Attributes	PC 1	PC 2	PC 3	PC 4	PC 5	PC 6	PC 7	PC8
pH	0.01	0.16	-0.08	-0.02	0.09	-0.09	-0.81	0.05
L*	-0.07	0.38	0.63	0.20	0.30	0.01	-0.23	0.17
a*	0.04	0.57	-0.03	-0.78	-0.10	0.14	-0.05	-0.15
b*	0.09	0.90	0.06	0.33	-0.12	0.06	-0.05	-0.08
WBSF	-0.80	-0.08	0.03	0.12	0.28	0.13	0.18	-0.04
MFI	-0.45	-0.15	-0.48	0.24	0.09	0.19	0.05	0.14
WHC	-0.10	-0.11	0.07	0.04	0.88	0.06	-0.07	0.00
CL	-0.16	-0.01	0.34	0.11	-0.30	-0.61	0.19	0.09
SL	0.00	0.29	0.21	-0.12	0.16	-0.02	0.16	-0.78
FB	0.04	0.02	0.20	0.04	0.16	-0.10	0.09	0.86
FC	0.17	0.18	-0.84	-0.02	0.06	0.11	-0.02	0.02
CC	-0.34	-0.20	-0.10	0.02	0.55	-0.44	0.32	0.09
CS	0.42	0.37	0.10	0.05	-0.43	0.12	-0.41	-0.11
DL	0.09	0.14	0.04	-0.08	-0.12	0.82	0.17	-0.03
AP	0.44	0.17	0.61	0.10	0.04	0.20	0.21	0.03
TE	0.86	0.06	0.05	-0.02	-0.25	0.21	0.03	0.10
JU	0.76	-0.08	0.04	0.09	0.00	0.39	0.28	0.02
FL	0.44	0.21	-0.12	0.02	0.19	-0.03	0.59	0.03
OA	0.76	-0.06	0.19	0.03	-0.03	0.32	0.30	-0.03
ACT	0.86	0.12	-0.21	0.03	0.12	-0.08	-0.08	-0.06
H	0.02	0.18	0.08	0.96	0.00	-0.07	0.00	0.05
CH	0.08	0.90	0.00	-0.32	-0.13	0.11	-0.06	-0.14

Note: WBSF-Warner-Bratzler shear force, MFI-Myofibril fragmentation index, WHC-Water holding capacity, CL-Cooking loss, SL-Sarcomere length, FB-Fiber diameter, FC-Fat content, CC-Collagen content, CS-Collagen solubility, DL-Drip loss, AP-Appearance, TD- Tenderness, JU-Juiciness, FL-Flavour, ACT-Amount of connective tissue, OA-Overall acceptability, H-Hue, CH-Chroma, L*-lightness a*- redness and b*-yellowness.

COMPETING INTEREST: The authors have no known competing interests, either financial or personal, between themselves and others that might bias the work.

ETHICS STATEMENT: Not applicable

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