Research Article

Breed and Age Effects on Quality Traits of Pakistani Buffalo Beef

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Abstract | Pakistani buffaloes beef were analyzed for various quality characteristics in relation to breed and age. A total of one hundred and eight beef samples from Longissimus dorsi muscle were collected from three age groups (i.e. group 1= 12-18 monts, 2 =18.1-36.0 months and 3= > 36 months) of *Kundi* and *Nili Ravi breeds of buffalo*. Samples were obtained from freshly preselected slaughtered buffaloes at local municipal abattoir Peshawar. The effect of breed and animal age on marbling, water holding capacity (WHC) and tenderness and sarcomere lenth of the two buffalo breed beef samples were investigated. A two x three factorial design was used to analyze the beef quality characteristic data in Genstat software. Breed and animal age had significant and their interaction has significant ($P < 0.05$) effects on marbling. Animal age group and their (breed x age) interaction had significant effect on the Water holding capacity (WHC). Breed effect on WHC was found not significant at P 0.05. WHC increased significantly ($P < 0.05$) with increase in age of the buffaloes however *Nili Ravi* had slightly higher mean WHC relationship to *Kundi*, however, breed effects were not significant ($P > 0.05$). Tenderness in the buffalo beef samples were significantly ($P < 0.05$) affected by animal age and their breed X age interaction. However the breed had no significant ($P > 0.05$) effect on tenderness. In both breeds, the sarcomere lengths (SL) were significantly decreased ($P < 0.05$) by animal age and age x breed interaction. Nevertheless breed shows no significant ($P > 0.05$) effect on SL. *Kundi* showed low mean (1.79±0.033µm) SL compared to *Nili Ravi* (1.81±0.039µm). It was concluded that beef from *Kundi* buffalo is relatively better in beef quality characteristics (Marbling, Water holding capacity, Sarcomere length, and Tenderness) than *Nili Ravi* buffalo. Kundi buffalo may be further selected for improvement in beef quality traits and could be a potential source of quality beef for export in future.

Received | February 23, 2015; **Accepted** | July 08, 2016; **Published** | August 27, 2016

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Citation | Khan, A., A. Rahman, S. Akhtar, S. Uddin, Z. Ullah and N. Ahmad. 2016. Breed and age effects on quality traits of Pakistani buffalo beef. *Sarhad Journal of Agriculture,* 32(3): 223-229.

DOI | http://dx.doi.org/10.17582/journal.sja/2016.32.3.223.229

Keywords | Buffalos, Meat Quality, Breed, Slaughter age

Introduction

Meat is an affluent source of nutrients for human beings and provides good quality animal protein, fats, minerals and vitamins with nice flavour which dominate many other dishes (Romans et al., 2000). Meat and meat products contain beneficial amount of Magnesium, Zinc, Copper, Phosphorus, Iron, Selenium etc. which are required for the growth and maintenance of human health (Lawrie and Ledward, 2006; Wood et al., 1999).

Meat sector is not well developed in Khyber Pakhtunkhwa (KP) province. Animals slaughtered for

meat are mostly brought from rural areas of Punjab, Sindh and also KP. Such animals are usually obtained from culled dairy, traction, male young stock and old breeding cattle of subsistence farming system. On other hand, meat quality characteristics such as marbling, water holding capacity, tenderness, texture, aroma etc., are not properly investigated. Due to better information technology, consumers become aware of meat quality, hygiene in meat animal slaughtering and processing and bio-risk free meat and demand respectively (Guerrero, 2001). In the recent years, the importance of buffalo meat is increasing day by day because of its relatively more nutrients and better flavours (Kandeepan et al., 2009). Buffalo beef has high protein level, low fat content and cholesterol in comparison to cattle beef (Murthy and Davidson, 2003). It has been studied that breed, age, sex, feeding, growth rate and animal handling are the major factors that affect the quality of beef (Beltran et al., 1997). Likewise, post-mortem aging also affects meat quality (Monson et al., 2005). Diet and breed are the major factors affecting the eating quality of cattle beef (Melton, 1990). Both breeds of buffaloes were reared for multipurpose such as milking, beef, emergency cash and traction. However, in past, no planned research was conducted to investigate the role of Pakistani breeds and animal age in beef quality characteristics enhancement. Keeping in view the prevailing situation, present study was designed to determine effects of breed and animal age on beef quality and to investigate the relationships among different quality characteristics of buffalo beef presently slaughtered in Peshawar Khyber Pakhtunkhwa Pakistan.

Materials and Methods

The present **s**tudy was conducted in the Livestock Management, Breeding and Genetics department, The Agricultural University Peshawar. Buffalo beef samples of *Longissimus dorsi* muscle from the 12-13 rib area, were collected from the three age groups 1= < 18 months, age group 2 = 18.01 to 36.0 months and age group 3 = > 36 months) of the buffalo breeds (*Nili Ravi and Kundi)* from the municipal slaughter house ring road Peshawar, Pakistan during October 2013 to January 2014. A total of one hundred and eight samples were collected from males of two buffalo breeds (fifty four samples per breed). Within a buffalo breed eighteen samples per age group were used in the research. For breed identification in buffaloes, individual animals brought to the slaughter house were identified on the basis of breed characteristics. Dentitions were checked for the age laboratory of the faculty for further analysis following the guidelines of Church and Wood (1990).

Collected beef samples were analyzed for water holding capacity using the press method with slight modification as prescribed by Whiting and Jenkins (1981). Weight of two empty Whatsman filter paper # 42 were initially recorded before each sample placement. A thin flake of 1.00 g beef sample was placed in the center filter paper between the two filter papers and placed it on a rigid, flat surface of glass (3.0 mm) and covered from top with similar glass. The pressure of 2.8 kg was applied on it for 15 min. After completion of pressing time, the buffalo beef sample was removed and discarded. The moisture absorbed by the filter papers were weighed after removal of sample and any tissue residue is a measure of water holding capacity (WHC) was expressed as percent using the below formula;

$$\textbf{Percent WHC} = \frac{\textbf{post press wight of meat flake}}{\textbf{pre press weight of flake}} \textbf{X 100}$$

Tenderness

A 3.0 cm core from the buffalo beef samples were cut and sealed it in polyethylene bag. After sealing it was placed in the water bath for 40 minutes at 80°C, after completion of time the polyethylene bag was taken out from the water bath and placed in the refrigerator for overnight chilling. After chilling the sample was placed for equilibration, after equilibration to room temperature a 1.25 cm core was cut, shear force was measured by Warner–Bratzler shear force (WBSF) machine. The power which is used for shearing the meat was recorded in kg and converted the force into the Newton unit. Tenderness is calculated in Newton (N) unit of force, where 1 kg = 9.8 Newton using the Kandeepan et al. (2009) formula.

Visual Examinations for Marbling

The visual examination of the marbling in buffalo beef samples were performed in the lab. According to the United State Department of Agriculture following standard score card for meat marbling, The *Longissimus dorsi* muscles of 12th and 13th vertebrae were examined. Marbling Score has divided marbling score into 10 points. These are: 10. Abundant; 9. Moderate Abundant; 8. Slight Abundant; 7. Moderate; 6. Modest; 5. Slight; 4. Small; 3. Traces; 2. Partial devoid; 1.

Devoid (USDA, 1994). Each sample was examined by authors and average marbling values was calculate dfor each sample.

Sarcomere Length (SL)

The sarcomere length of buffalo beef samples were determined using the Cross (1979) method. The 5.0 g beef was homogenized in a kitchen blender twice for 15 seconds at low speed in 30 ml of 0.25 M sucrose solution for 60 seconds. The homogenate stuff was transferred to the slide and examined under microscope at 100 x under oil immersion for the sarcomere length (Cross, 1979).

Statistical Analysis of Data

The buffalo beef quality parameters data was recorded in Microsoft Excel spreadsheet and exported to Genstat Discovery Edition -3 (Wim et al., 2004) for further analysis. A 2 X 3 factorial design was used for analysis of variance. The means and standard error of means of beef quality parameters in both Buffalo breeds across the age groups were compared.

Table 1: *Breed and age effects on marbling of buffalo beef samples collected from slaughtered animals at Municipal Abattoir Peshawar*

Age groups	Mean ± SE Marbling[1] of Kundi	Mean ± SE Marbling[1] of Nilli Ravi	P-value
1	3.60±0.80	1.60±0.13	0.001
2	5.60±0.35	4.00±0.22	
3	7.60±0.510	4.10±0.67	
P-value for age group	0.003		0.178*

*: *P- Value of the breed x animal age interaction at the significance level α 0.05, SE for standard error of mean. Marbling of each samples was examined using USDA standard score card i.e. 1.0- 10 scores. These scores are 10 for. Abundant marbling 9. Moderate Abundant 8. Slight Abundant 7. Moderate 6. Modest 5. Slight 4. Small 3. Traces 2. Partial devoid and score 1 for devoid of marbling*

Results and Discussion

Marbling

The effects of breed and animal age on marbling characteristics of buffalo beef are shown in Table 1. Breed and animal age group and their interaction had significant ($P < 0.05$) effect on the intramuscular fat content (marbling) in buffalo beef samples. The *Nili Ravi* buffalo beef was less marbled as compared to *Kundi*. Result showed that within age group the above three years old buffalo (animal age group 3) has highest marbling in both breeds, however, animal of age group 3 of *Nili Ravi* has almost 43 less marbling than Kundi breed (Table 1). Breed X age interaction effect on marbling was not significant ($P > 0.05$). Moreover, in both breeds of buffalo, the marbling in beef muscle also increased with age. Among many factors which affected the marbling of beef were; breed, age, environment, genetics, nutrition, handling, slaughtering procedures and pre-slaughter fasting stress period (Shackelford et al., 1995; Lawrie, 1985). Paredi et al. (2012) reported that marbling is a major indicator of the juiciness and flavour in beef. Factors that regulate marbling were influenced by environmental conditions and genetic variability and by processes occurring during conversion of muscle to meat that may alter the adipose tissue composition (Ueda et al., 2007). Ziauddin et al. (1994) reported that the buffalo male calves were slaughtered in young age (1–2 years of age) and female were slaughtered at the end of their reproductive life (12 years of age). Generally, in subsistence farming system, the young buffalo in a herd did not receive any concentrate; therefore, the marbling in such beef or veal was found negligible (Ziauddin et al., 1994). Forrest et al., (1975) stated that the development in muscle and bone takes place in early post-natal life in animals. As often inadequate nutrients are offered to the animal at this part of life under subsistence farming, the animals will have lesser marbling score. Moreover, Qiang et al. (2011) reported that age had no significant effect on marbling. Qiang et al. (2011) further explained that two factors are responsible for intramuscular fat content i.e. Leptin and fatty acid synthase (FAS). Leptin is a protein produced by fat cells that are involved in controlling the amount of adipose tissue laid down in the body. Fatty acid synthase (FAS) catalyzes fatty acid synthesis from acetyl coenzyme A and malonyl co-enzyme A and play important role in fat deposition in animals.

Results from the present study revealed that the age had significant effect on marbling in buffalo beef which are similar with the findings of Albrecht et al. (2006). Albrecht et al. (2006) revealed that marbling scores increased significantly during growth in all investigated breeds. Breed, age and the interaction between breed and age influenced quantity, structure, and distribution of intramuscular fat to various degrees.

Water Holding Capacity (WHC)

The effect of breed and age group of buffalo on the water holding capacity of beef samples are presented in Table 2. Animal age and age into breed interac-

tion affected significantly (P < 0.05) the WHC. In present research, the WHC was slightly higher in *Nili Ravi* buffalo beef belonging to age group 1 and 3 compared with *Kundi* buffalo beef. However, breed effect in the present study were not significant. Water holding capacity is the ability of meat proteins to retain its own water content when subjected to pressure treatment (Lonergan et al., 2005). There are many factors which influence WHC quality characteristics in buffalo beef. Sami et al. (2006) revealed that breed and age are among those factors. Sex of the animals may also affect WHC (Rao et al., 2009). Beef WHC is affected by protein content in the muscle however post mortem pH decline and transportation stress also causes reduction in water holding capacity (Offer, 1991). Similarly, Nuraini et al. (2013) found different result in swamp buffaloes' beef in Banten province of Indonesia. Swamp buffaloes are genetically different (having 48 chromosomes) than south Asian riverine buffaloes (having 50 chromosomes) which might have resulted differences in meat quality characteristics. The decline in water holding capacity of buffalo beef depended upon: age of buffalo at slaughter time, body condition of the animal and beef processing and storage conditions at slaughterhouse, etc.

Table 2: *Effect of breed and age on water holding capacity of buffalo beef collected from slaughtered animals at Peshawar slaughtered house*

Age groups	Mean ± SE Water holding capacity[1] of Kundi	Mean ± SE Water holding capacity[1] of Nilli Ravi	P-value
1	83.00±1.82	83.80±2.15	0.0621
2	68.60±1.03	62.40±1.99	
3	50.20±1.65	53.40±1.88	
P-value for age group	0 .001		0.039*

*: *P- Value of the breed x age interaction at the significance level α 0.05;* **SE:** *standard error of mean; WHC is calculated in percentage*

Tenderness

Breed and age effects on tenderness of buffalo beef are shown in Table 3. The study results showed that age of the animal had a significant effect (P < 0.05) on tenderness of the beef. Similarly Interaction effect of breed X age had also significantly effect on tenderness of buffalo beef however the breed effect was not significant (P < 0.05). Tenderness in buffalo beef was significantly affected with increase in age of buffalo. The tenderness of buffalo beef decreased as the age of animal increased.

Table 3: *Effect of breed and age on tenderness[1] of buffalo beef collected from slaughtered animals at Peshawar slaughtered house*

Age groups	Mean ± SE Tenderness[1] of Kundi	Mean ± SE Tenderness[1] of Nilli Ravi	Breed P-value
1	57.40±1.631	60.80±1.428	0.950
2	72.80±1.655	79.20±2.289	
3	106.80±3.839	117.00±3.450	
Age P-value	0. 001		0.008*

*: *P- Value of the breed x age interaction at the significance level α 0.05;* **N:** *Newton;* **SE:** *standard error of mean;* [1]**:** *Tenderness is calculated in Newton (N) unit of force;* **1 kg:** *9.8 Newton*

Rao et al. (2009) revealed that age of the animals influences the tenderness in buffalo beef. Increased value of shear force in older buffaloes may be due to the development of connective tissue after maturity. The old animals change the structure of the connective tissue and meat becomes tough. In the present study, it was observed that age had significant effect on tenderness in buffalo beef which are similar with the findings of Rao et al. (2009). On the other hand, Nuraini et al. (2013) concluded that age did not influence tenderness in buffalo beef. Another factor that could affect the tenderness in beef was degradation of myofibril, connective tissue, sarcomere length, muscle fibers, and a decreased of pH (Neath et al., 2007) and lack of dissipation of temperature from carcass during long post slaughter interval. Tenderness of meat is influenced by several factors such as feed, livestock activities and treatment before and after slaughter (Nuraini et al., 2013). It is extrapolated that the other management and post slaughter factors were changed in the study of Nuraini et al. (2013) which turned their results different from present study. Holing et al. (2007) revealed that correct animal handling at farm and transportation to the abattoir also influence the tenderness of meat in the animals. Similarly, Plessis and Hoffman (2007) investigated the lack of significant effect of age and breed on meat quality characteristics of cattle. Beef was tender at 18 months of age than at 30 months of age.

Effect of Breed and Age on Sarcomere Length (SL)

Effect of breed and age on the SL in buffalo beef were presented in Table 4. Age has significant (P < 0.05) effects on SL in both breeds. However, current results revealed that the SL was not significantly affected (P > 0.05) by breed. Nevertheless, interaction of age X breed has significant effect (P < 0.05) on SL of buffalo beef (Table 4). The sarcomere length of buffalo beef

decreased as the age of the animal increased. Sarcomere length of *Kundi* buffalo of age group 1 was 6.81 % lower than the *Nili Ravi* counterpart. Kandeepan et al. (2013) stated that reduction in the SL of meat increases its toughness. The Sarcomere Length of buffalo beef decreased with advancement of age and resultantly increased the toughness of beef. Sarcomere length was found long (1.83 µm) in young age group animals slaughter as compared to mature buffalo beef (1.56 µm). Present values of young animals were greater than the previous study however the trend in the same and in accordance with the results of Kandeepan et al. (2009).

Table 4: *Effect of breed and age on sarcomere length of buffalo beef collected from slaughtered animals at Peshawar slaughtered house*

Age groups	Mean ± SE Sarcomere length[1]	Mean ± SE Sarcomere length[1] of Kundi of Nilli Ravi	Breed P-value
1	2.19±0.037[a]	2.35±0.055[a]	0.499
2	1.79±0.032[b]	1.76±0.022[b]	
3	1.41±0.030[c]	1.34±0.042[c]	
Age P-value	0 .001		0.016 *

*: *P- Value of the breed into age interaction at the significance level α 0.05, µm stands for micrometre;* **SE:** *standard error of mean;* [1]: *Sarcomere length is calculated in micrometre (µm); Different alphabetic superscript along the mean value showing the LSD at α = 95% confidence interval*

Elsa-be (2011) reported that breed and interaction (breed X production system) had significant effect ($P < 0.05$) on SL in cattle. Differences in SL in various breeds were due to the shortening of sarcomeres. During muscular contraction the contractile units of the myofibrils, the sarcomeres shorten. Shortening is induced by the release of Ca^{2+} ions from the sarcoplasmic reticulum into the myofibril space. The movement of Ca^{2+} ions within the muscle cells post mortem depends on temperature, pH and ATP concentration present. They also observed that animal reared on pastures had shortest sarcomere length (1.62 µm) and those from feedlot production system had longest sarcomere length (1.79 µm). However, effect of production system on meat quality was not investigated. Results obtained in present study that did not agreed with the findings of Elsa-be (2011). Present study findings differ from the results of Elsa-be (2011) and that might be due to difference in animal species. Similarly, Honikel et al. (1968) reported that sarcomere length is affected by temperature and pH fall in bovine muscle. They observed a SL of 1.9 µm – 2.0 µm at pH 6.8. As the pH fall (from 6.8) the length of sarcomere shortens. After 24 hrs at pH below 5.9 sarcomere shortened by 50%. Sarcomeres shortening occur by storing at various temperatures for 24 hrs. By storing it at 0°C at pH 6.7-6.8 the reduction in SL occurs by 10% within 30 min.

Conclusions

Quality traits of buffalo beef such as; marbling, water holding capacity and sarcomere lengths were significantly better in *Kundi* buffalos in comparison with N. Ravi, however tenderness of the beef was not significantly affected by breed. *Kundi* potential for beef need to further explore.

Recommendations

Biochemical parameters (fatty acid profile, micro minerals etc.) and sensory evaluation of *Kundi* beef need further research studies and should be examined for better quality meat production in the Pakistan.

Authors' Contribution

Arif Khan and Abdur Rahman planned the research experiment. Sohail Akhtar and Nazir Khan facilitated in the manuscript editing and facilitating in the statistics of the data. Whereas Siraj and Zahid helped in the field to properly identify the breed and age of the animals before the beef samples were collected by Arif and Rahman.

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