Research Article

Heavy Metals and Mineral Contents of Beef Sold at University Campus, Peshawar, Khyber Pakhtunkhwa, Pakistan

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Abstract | The objective of this study was to examine the heavy metals (Cu, Zn, Pb and Ni) and electrolytes (Na, K, Ca, Mg and P) contents of cow and buffalo muscle meat obtained from University Campus, Peshawar. The data showed that Cu, Zn, Pb and Ni concentrations in cow meat ranged from 7.27 to 14.81, 64.00 to 112.12, 3.30 to 6.25 and 0.73 to 2.53 mg/kg whereas the concentrations of these heavy metals in buffalo meat varied from 3.15 to 7.71, 72.88 to 88.47, 1.41 to 13.28 and 0.38 to 1.93 mg/kg, respectively. The average Na, K, Ca, Mg and P contents in cow's and buffalo's meat were found to be 54.75 and 67.45, 351.67 and 333.75, 4.61 and 6.31, 21.50 and 26.76, and 212.75 and 181.32 mg/kg, respectively. The average contents of Cu, Zn, Ni, K and P were higher in cow's meat whereas the concentrations of Pb, Na, Ca and Mg were higher in buffalo's meat. The toxic heavy metal Pb concentration was greater than the maximum permissible limit (0.1 mg/kg) of World Health Organization (WHO) in both types of meats. It was concluded that beef is a valuable source for essential mineral elements in human nutrition. However, the increased level of Pb should be of utmost concern while consuming excessive amount of beef.

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Introduction

Essential minerals are required by living organisms for vital body functioning. Soil and river water are the two natural sources of all mineral elements. Human beings obtain these inorganic substances from drinking water and by eating plants and animals as food. About twenty-one essential minerals are needed by human body, but the prominent minerals include calcium (Ca), potassium (K), sodium (Na), phosphorus (P), iron (Fe), copper (Cu), zinc (Zn) and magnesium (Mg). Their deficiency or excess cause chronic metabolic disturbances in the body (Chowdhury et al., 2011). Since these minerals cannot be synthesised by the body itself, therefore, they must be constantly supplied in diet to maintain a healthy life. This can be achieved by consuming such foods which contain sufficient concentration of these minerals. Contrary to this, the presence of heavy metals such as nickle (Ni), lead (Pb), cadmium (Cd), chromium (Cr) and arsenic (As) cause sever biochemical and neurological disturbances in the body (Ogwok et al., 2014).

Food security and safety are the two major challenges

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faced by all nations to meet the basic requirements of spiraling world population. Among the various foods, meat is one of the most important item as it contains many micro and macro nutrients. It is widely consumed and is the most delicious part of food. However, it is the most sensitive food in term of perishability and contamination with toxic substances. Meat can be contaminated with toxic agents via many sources such as animal drugs, pesticides, water, feed and agricultural or industrial effluents. The polluted feed and water may be the principal cause of heavy metals accumulation in animals' meat. As the shopkeepers in developing world retail most of the meat in open bazars and on this way many toxic metals pollute the meat via vehicular emission and road side dust.

Pollution of meat by heavy metals is a trouble of great alarm for food safety and human health. Even in a small amount in food, they may cause hostile effects. Lead, arsenic, nickel, and cadmium are those heavy metals, which are linked to human harming at definite level. Whereas cobalt, zinc, manganese and chromium are vital in small quantity for human body functioning but in larger amount can be poisonous. Living organisms acquire them in minor quantity from their surrounding because these cannot be produced by themselves. Most of the microelements are moved into animal and human body as they are used as enzyme cofactor. These essential minerals include Fe, Zn, Co, Cr, Cu, Mn, Se and Mo (Shar et al., 2013). The long-term exposure to heavy metals may cause poisoning to the central nervous system, impaired normal brain functioning, and may result in slow progression of physical, muscular and neurological degenerative conditions including cancer (Jaishankar et al., 2014).

Heavy metals such as lead, copper, cadmium, zinc, arsenic and nickel are among the major threats to human health. The international bodies such as WHO/FAO have widely studied the effects of these heavy metals on human health and have set maximum permissible limits for their intake in human diet. The maximum levels set by WHO/FAO for Pb, Cu and Zn are 0.1, 40 and 60mg/kg, respectively (WHO, 1982; Zhu et al., 2011) in meat and other food items. The Codex Alimentarium and WHO/FAO have no set standard for Ni concentration in meat, however, the Russian authority has a permissible limit of 0.5 mg/kg for nickel in meat and meat products (WHO, 1991). Since heavy metals contamination in diet can cause chronic health complications in consumers, therefore, it is obligatory to trace the severity of their contamination in commonly consumed food items such as meat and meat products. This study was, therefore, conducted to examine the concentration of heavy metals Pb, Ni, Zn and Cu in beef meat commercially available at University Campus, Peshawar, Khyber Pakhtunkhwa, Pakistan. The study also aimed to assess the major electrolytes such as Na, K, Ca, Mg and P concentrations of beef in the study area.

Materials and Methods

Study site

University Campus, Peshawar Khyber Pakhtunkhwa, Pakistan. was selected as the study site. The campus is comprised of four universities i.e. University of Peshawar, University of Engineering and Technology, Islamia College University and The University of Agriculture, Peshawar. The campus area is predominantly occupied by students, staffs, faculty and their families where majority of the inhabitants obtain their groceries from local markets located on the campus.

Samples collection

About 200 g each of fresh cow and buffalo meat samples were purchased from local meat shops at four locations viz. Lalazar (A), Forest bazar (B), Coffee Shop (C) and Madina Market (D) of the University Campus, Peshawar Khyber Pakhtunkhwa, Pakistan. Three samples of each meat type were obtained from each location. All the samples were packed in clean zip-lock polythene bags and labeled according to their type and shifted to the laboratory for heavy metals and electrolytes analysis.

Mineral analysis

For mineral analysis the samples were acid digested (AOAC, 2000). The digests were then used for analysis of Na, K, Ca, Mg, P, Cu, Zn, Pb and Ni. Flame photometer (Jenway PFP 7) was used for the determination of Na and K following the method of Khan and Zeb (2007) whereas P was determined by spectrophotometer (UV 1700 Shimadzu) by the method of Cottenie (1980). Atomic Absorption Spectrophotometer (Perkins Elmer A Analyst 200) was used for the assessment of Ca, Mg, Pb, Cu, Zn and Ni (Sattar and Chaudhary, 1978). The instruments were calibrated with the standards of respective elements before the sample digest was introduced. All the analyses were carried out in triplicate. The concentrations of

$$\text{Heavy metal concentrtion} \left(\frac{mg}{kg}\right) = \frac{\text{Absorbance reading} \times \text{Dilution factor} \times 100}{\text{Weight of sample}} \ldots (1)$$

heavy metals were calculated by the formula given above: (Equation 1)

Statistical analysis

Statistical analysis of the data was carried out with statistical software *Statistix* version 8.1. Analysis of variance (ANOVA) was performed using completely randomized design (CRD) with two factors i.e. beef type and locations. The significant differences in means were calculated by Least significant difference (LSD) test at 0.05% probability. All the means were calculated from triplicate values.

Table 1: *Copper content (mg/kg) of cow and buffalo meat collected from four different sites of University Campus, Peshawar, Khyber Pakhtunkhwa, Pakistan.*

Location	Beef type		Mean
	Cow meat	Buffalo meat	
A	9.37±1.20	4.49±0.20	6.93±0.70 c
B	7.27±1.01	3.15±1.12	5.21±1.06 d
C	14.81±1.82	6.31±1.20	10.56±1.51 a
D	9.08±2.01	7.71±1.20	8.39±1.61 b
Mean	10.13±1.51a	5.41±0.93 b	

Means followed by similar letters are not significantly different at $p \leq 0.05$; **LSD value for location:** *0.27;* **beef type:** *0.19;* **location× beef type:** *0.38*

Table 2: *Zinc content (mg/kg) of cow and buffalo meat collected from four different sites of University Campus, Peshawar.*

Location	Beef type		Mean
	Cow meat	Buffalo meat	
A	74.50±2.11	88.47±3.25	81.48±2.68 b
B	80.02±5.02	73.79±1.32	76.90±3.17 b
C	112.12±4.18	72.88±2.23	92.50±3.21 a
D	64.00±3.23	77.02±2.61	70.51±2.92 c
Mean	82.66±3.64 b	78.04±2.35a	

Means followed by similar letters are not significantly different at $p \leq 0.05$; **LSD value for location:** *6.34;* **beef type:** *4.48;* **location× beef type:** *8.97*

Results and Discussion

Heavy metals contents of beef

The data showed that cow's meat contained significantly ($p \leq 0.05$) higher average content of Cu i.e. 10.13 mg/kg compared to buffalo meat, which contained 5.41 mg/kg Cu content (Table 1). The Cu content of beef also significantly ($p \leq 0.05$) varied with location. The highest average copper content (10.56 mg/kg) was found in beef samples collected from location C followed by 8.39 mg/kg Cu content in the samples collected from location D. The lowest average Cu content (5.21 mg/kg) was examined in beef samples collected from location B. The data regarding the Zn content of cow and buffalo meat were presented in (Table 2). The data showed that both the beef type and location individually as well as interactively significantly ($p \leq 0.05$) affected the Zn content of beef available at University Campus, Peshawar. The highest amount of Zn (112.12 mg/kg) was examined in cow meat collected from location C whereas the minimum concentration (64.00 mg/kg) was noted in cow meat obtained from location D. (Table 3) represented the Pb content of cow and buffalo beef samples collected from four locations at University Campus, Peshawar. Analysis of variance showed that there was no significant ($p > 0.05$) influence of beef type and location on the Pb content of beef meat. However, non-significant ($p > 0.05$) variation existed between the samples means. Overall, the average Pb content ranged from 1.41 to 13.28 mg/kg. Similarly, the data regarding the Ni content showed that beef samples contained significantly ($p \leq 0.05$) different amounts of Ni with respect to beef type and location (Table 4). The average content of Ni in cow's beef was found to be 1.55 mg/kg whereas that of buffalo meat was 1.15 mg/kg. Regarding locations, the highest Ni content (1.68 mg/kg) was examined in beef sample from location B whereas the lowest concentration (0.65 mg/kg) was noted in beef samples from location C.

The results of the present study were closely in agreement with Ahmad, (2016), Nawaz et al. (2015), Shehzad et al. (2014) and Chowdhury et al. (2011). Meat contamination with toxic heavy metals is a risk factor for public health safety because the consumption of contaminated meat may cause various toxic and adverse ailments in human population (Demirezen and Uruc, 2006). The concentration of Cu, Zn and Ni, in the present study, was within the safe limits recommended by international bodies such as WHO/FAO. The recommended safe limits for Cu, Zn and

Ni in food items are 40, 60 and 5 mg/kg, respectively (WHO, 1982; FAO/WHO, 2000; Zhu et al., 2011). However, the Pb concentration was above the permissible limit of 0.1 mg/kg as recommended by Codex Alimentarious Commission (FAO/WHO, 2003). This high concentration of Pb in the meat samples might be due to the grazing of buffaloes and cattle on contaminated soil. It may also be due to the age, place of animal, dietary habits, slaughtering, transportation condition and exposure to dust etc. Pb is a toxic heavy metal that causes a number of diseases in human beings. For example, certain enzymes which are essential for neurotransmission are inhibited by Pb that results in hyperactivity and convulsion. Similarly, Pb toxicity is one of the major causes of "Gout" disease, which is characterized by enhanced uric acid level and impairment of kidney functions (Hussain et al., 2006). On the other hand, Cu and Zn are essential heavy metals and perform key roles in normal body functioning. Zn is essential for the synthesis of nucleic acid and proteins and facilitate the absorption and utilization of nitrogen and phosphorus by the body (Wardlaw and Hampl, 2006). Copper is essential enzymatic element; however, it can be toxic at excessive level. The toxic effects of Cu include hypertension, insomnia, loss of skin tone and dark pigmentation of the skin around the face. Nickel is required in minute amount for body as it functions as a co-factor with a variety of enzymes such as insulin and those involved in the breakdown of branched chain amino acids and odd chain lengthy fatty acids. It is also involved in the metabolism of folic acid and vitamin B_{12} (Wardlaw and Hampl, 2006). However, increase amount of Ni has many health effects. For example, Ni tends to accumulate in the kidneys causing kidney damage. A steady exposure to Ni can cause cancer of the lungs and nasal sinus.

Table 3: *Lead content (mg/kg) of cow and buffalo meat collected from four different sites of University Campus, Peshawar.*

Location	Beef type		Mean
	Cow meat	Buffalo meat	
A	6.25±1.21	3.16±0.22	4.71±0.72
B	5.40±1.01	2.65±0.21	4.03±0.61
C	3.30±0.33	13.28±2.31	8.29±1.32
D	4.79±0.90	1.41±0.51	3.10±0.71
Mean	4.94±0.86	5.13±0.81	

Means followed by similar letters are not significantly different at p≤0.05; **LSD value for location:** *7.56;* **beef type** *: 5.35;* **location× beef type:** *10.70*

Table 4: *Nickel content (mg/kg) of cow and buffalo meat collected from four different sites of University Campus, Peshawar.*

Location	Beef type		Mean
	Cow meat	Buffalo meat	
A	2.53±0.59	0.38±0.04	1.46±0.32 b
B	1.67±0.09	1.70±0.10	1.68±0.10 a
C	0.73±0.10	0.57±0.04	0.65±0.07 c
D	1.26±0.06	1.93±0.07	1.60±0.06 ab
Mean	1.55±0.21a	1.15±0.06 b	

Means followed by similar letters are not significantly different at p≤0.05; **LSD value for location:** *0.19;* **beef type:** *0.13;* **location× beef type:** *0.27*

Electrolytes concentration of beef

The concentration of Na in the beef samples ranged from 50 mg/kg in cow meat from location A to 71.2 mg/kg in buffalo meat from location C of University Campus, Peshawar, Khyber Pakhtunkhwa, Pakistan. (Table 5). Analysis of variance indicated that both the location and meat type had significant (p≤0.05) influence on the Na content of beef. A similar trend of results was noted for K content (Table 6). The results showed that cow meat had significantly (p≤0.05) higher amount of average K content (351.67 mg/kg) as compared to buffalo meat, which contained an average amount of 333.75 mg/kg of potassium. A reversal of results was observed for calcium content (Table 7). The buffalo meat contained significantly (p≤0.05) higher average concentration of Ca (6.31 mg/kg) as compared to cow meat (4.61 mg/kg). Regarding location, the highest average concentration of K (367.83 mg/kg) was found in the samples collected from location D whereas the lowest (311.83 mg/kg) was recorded in the beef samples collected from location B. Similarly, the Ca content was maximum in beef samples from location C and minimum in the samples from location D.

Table 5: *Sodium content (mg/kg) of cow and buffalo meat collected from four different sites of University Campus, Peshawar.*

Location	Beef type		Mean
	Cow meat	Buffalo meat	
A	50.00±1.25	67.67±2.14	58.83±1.70 c
B	57.67±2.36	64.67±1.26	61.17±1.81 b
C	61.00±2.35	71.20±3.11	66.10±2.73 a
D	50.33±1.36	66.27±2.99	58.30±2.18 c
Mean	54.75±1.83 b	67.45±2.38 a	

Means followed by similar letters are not significantly different at p≤0.05; **LSD value for location:** *1.31;* **beef type:** *0.932;* **location × beef type:** *1.86*

Table 6: *Potassium content (mg/kg) of cow and buffalo meat collected from four different sites of University Campus, Peshawar.*

Location	Beef type		Mean
	Cow meat	Buffalo meat	
A	361.67±4.22	345.00±3.11	353.33±3.67 b
B	322.00±2.34	301.67±1.99	311.83±2.17 d
C	353.00±5.38	322.67±7.23	337.83±6.31 c
D	370.00±2.35	365.67±3.98	367.83±3.17 a
Mean	351.67±3.57 a	333.75±4.08 b	

Means followed by similar letters are not significantly different at p≤0.05; **LSD value for location:** *2.13;* **beef type:** *1.50;* **location × beef type:** *3.01*

Table 7: *Calcium content (mg/kg) of cow and buffalo meat collected from four different sites of University Campus, Peshawar.*

Location	Beef type		Mean
	Cow meat	Buffalo meat	
A	4.27±0.22	7.25±1.81	5.76±1.02 b
B	5.00±0.50	5.33±0.99	5.16±0.75 c
C	5.57±1.20	7.58±0.33	6.57±0.77 a
D	3.60±0.61	5.09±1.20	4.35±0.91 d
Mean	4.61±0.63 b	6.31±1.08 a	

Means followed by similar letters are not significantly different at p≤0.05; **LSD value for location:** *0.25;* **beef type:** *0.18;* **location × beef type:** *0.36*

Table 8: *Magnesium content (mg/kg) of cow and buffalo meat collected from four different sites of University Campus, Peshawar.*

Location	Beef type		Mean
	Cow meat	Buffalo meat	
A	15.67±1.23	28.15±2.11	21.91±1.67 bc
B	21.00±1.61	22.27±1.32	21.64±1.47 c
C	30.00±2.11	30.03±2.05	30.02±2.08 a
D	19.33±1.32	26.60±2.58	22.97±1.95 b
Mean	21.50±1.57 b	26.76±2.02 a	

Means followed by similar letters are not significantly different at p≤0.05; **LSD value for location:** *1.12;* **beef type:** *0.79;* **location × beef type:** *1.58*

The data regarding Mg content of beef samples collected from four locations of University Campus Peshawar is presented in (Table 8). The mean value of Mg concentration was higher in buffalo meat (26.76 mg/kg) as compared to cow meat that contained 21.50 mg/kg of Mg. Location also significantly (p≤0.05) affected the Mg content of the beef samples. The maximum mean value of magnesium (30.02 mg/kg) was found in beef samples from location C whereas the lowest (21.64 mg/kg) was found in the samples from area B. The P content of beef samples ranged from 169.58 mg/kg in buffalo meat from location C to 231.67 mg/kg in cow meat from location B (Table 9). Analysis of variance showed that both the meat type and location individually as well as interactively significantly (p≤0.05) affected the P content of beef. On the average, cow meat contained significantly (p≤0.05) higher amount of P than buffalo meat.

Table 9: *Phosphorus content (mg/kg) of cow and buffalo meat collected from four different sites of University Campus, Peshawar.*

Location	Beef type		Mean
	Cow meat	Buffalo meat	
A	214.00±2.59	194.03±3.58	204.02±3.09 a
B	231.67±5.11	170.20±6.25	200.93±5.68 b
C	206.67±3.18	169.58±2.84	188.12±3.01 d
D	198.67±4.24	191.47±3.67	195.07±3.96 c
Mean	212.75±3.78 a	181.32±4.09 b	

Means followed by similar letters are not significantly different at p≤0.05; **LSD value for location:** *1.32;* **beef type:** *0.93;* **location × beef type:** *1.87*

The present results are fairly supported by the work done by previous researchers. Shehzad et al. (2014) investigated the dietary mineral contents of beef and mutton samples collected from different locations of district Peshawar and reported similar results for Na, K, Ca, Mg and P content as we found in our study. The present values were also within the range of those reported by Doornenbal and Murray (1981). Meat is a good source of dietary minerals that play critical role in normal body functioning (Williams, 2007). For instance, sodium is a key electrolyte in extra cellular fluid and regulates the fluid balance of the body within and outside the cells. It also facilitates the absorption of other nutrients in small intestine. On the other hand, excessive amount of sodium is a potential source of problems like high blood pressure and shocks. The large amount of potassium in beef, in comparison to sodium, could be beneficial for hypertension, body fluid balance, nerve impulse transmission and contractility of smooth skeletal cardiac muscles (Wardlaw and Hampl, 2006). Phosphorus is an essential component for the formation and maintenance of bones, teeth and cartilages. The body need P for the metabolism and utilization of carbohydrates and lipids. It is required for protein synthesis and cells maintenance and repair to sustain a normal growth. It is also a component of the enzyme system, ATP, DNA, RNA and cell membrane (Wardlaw and Hampl, 2006). Similarly, magnesium is an essential activator of more than 300 enzymes that utilize ATP. In human beings, Mg is beneficial for heart and nerves functions, as well as insulin release from the pancreas (Wardlaw and Hampl, 2006). The present study, thus, warrants the regular inclusion of beef in the daily food diet of low energy and malnourished people in order to meet their nutritional requirements.

Conclusions

The findings of the present study showed that both the beef type and location individually as well as interactively significantly affected the heavy metals and essential electrolytes concentration of beef. The average contents of Cu, Zn, Ni, K and P were higher in cow's meat whereas the concentrations of Pb, Na, Ca and Mg were higher in buffalo's meat. The toxic heavy metal Pb concentration was greater than the maximum permissible limit (0.1 mg/kg) of World Health Organization (WHO) in both types of meats. Further studies are needed to evaluate the adulteration of other toxic heavy metals such as arsenic, cadmium and mercury in beef to provide comprehensive information for the awareness of consumers.

Author's Contribution

Gulsanga: Carried out samples collection, analytical work and manuscript writing.
Zaheen Anjum: Designed the study.
Sahib Alam: Supervised the analytical work and helped in the statistical analysis and interpretation of data.
Farhat Shehzad: Proof read the manuscript.

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