Accumulation of Heavy Metals in Edible Organs of Different Meat Products Available in the Markets of Lahore, Pakistan

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Abstract. The present study assessed the accumulation of selected heavy metals (Cd, Cr, Pb and Cu) in different organs including brain, heart, lungs, liver, stomach, kidney and flesh (muscles) of several animals commercially available in the market of Lahore, Pakistan. The concentrations found in different organs of chicken, goat and cow ranged between 0.132-2.165 μ g/g for Cd, 0.768-2.335 μ g/g for Cr, 0.260-1.411 μ g/g for Pb and 0.092-1.195 μ g/g for Cu. In the absence of national safety standards in respect to the content of heavy metals in foodstuffs, the results obtained were compared with international guidelines and found concentrations considerably higher than the prescribed safe limits. Therefore, immediate attention must be paid to prevent public health risks associated with the presence of toxic heavy metals in the commercially available meat products.

Keywords: meat products, toxicity, heavy metals, bioaccumulation

Introduction

Meat and meat products are extensively consumed all over the world as they are substantial source of proteins, amino acids, and essential minerals, required for proper tissue formation, growth and repair (Alturigi and Albedair, 2012; Chowdhury et al., 2011). With population increase worldwide, the consumption of the meat products has also been increased. According to Worldwatch Institute (WWI, 2014), the global meat consumption has been increased 3 fold over last four decades and by 20% only in last decade, which is significantly more than the population rise. However, in recent times, the food security is considered a significant global concern due to the direct public health risks associated with it. In this context, heavy metals contamination of food products, especially the meat products has been broadly investigated worldwide because of their direct toxic effects on human health (Asegbeloyin et al., 2012; Oforka et al., 2012; Mariam et al., 2004). It is evident that human intake is the most common source of potentially deleterious heavy metals (Bennet, 1984).

Meat contamination with heavy metals is a serious threat because of their toxicity, bioaccumulation and biomagnification in the food chain being transferred to humans (Demirezen and Uruc, 2006; Demirezen and Aksoy, 2004; Abou-Arab, 2001). These heavy metals are stored in body tissues and often have direct physiological toxic effects (Mariam et al., 2004). The accumulation of toxic heavy metals may lead to organ failure, retarded mental development, and cancer (Asegbeloyin et al., 2012). Although trace amount of heavy metals occur due to natural geological activities including such as ore formation, weathering of rocks and leaching may occur. Heavy metals are transferred to the meat's source animals via polluted water, grazing crops on irrigated sewage and industrial wastewater and contaminated feed (Sabir et al., 2003). Moreover, the contaminated soil ingested by animals, upto 18% during grazing in some domestic ungulates is also another source of these toxic elements (Thornton and Abrahams, 1983).

Since heavy metals are bio-accumulative and in less developed countries, less preference is given on existence of these toxic metals in the food products due to limited resources and lack of proper legislative framework that leads to frequent prevalence of the fatal epidemics outbreaks. The heavy metals' contamination like Pb can effects the animals present in its surrounding that can be risky for

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human meat consumers (Pareja-Carrera *et al.* 2014). Therefore, determination of heavy metals associated with the consumption of the meat products, commercially available in local markets of less developed countries has become necessary. The aim of the present study was to assess the concentration of Cd, Cr, Cu and Pb in different organs of chicken, mutton (goat), and beef (cow) samples, commercially available in the markets of Lahore, (Pakistan) and to evaluate the public potential health risk. These results were compared with different available international safety standards, since no national food safety standards have been formulated to date for comparison.

Materials and Methods

Sample collection. A total of 30 fresh samples of different organs (brain, heart, lungs, liver, gizzard, kidney and muscle tissues) of chicken, goat and cow were collected from June to September in 2013. Age of these animals was 3-4 months, 2-3 years and 3-4 years old for chicken, goat and cow, respectively. For sampling five markets of Lahore city, including Tollinton Market, Icchra Market, Mozang Bazar, Gulberg Main Market, and Township Main Market were visited. All samples were collected in polyethylene bags, sealed, stored in ice box and transported to the laboratory and wet digestions were performed on the same day.

Sample preparation. The samples were prepared for heavy metals determination in the laboratory using a wet digestion method. One gram of chicken, meat and beef samples was dried in oven at 105 °C for 1h. The digestion was then carried out using 5 mL of conc. HNO₃ and 1 mL of HClO₄ in a digestion flask. The flask was then heated at 200-250 °C on a hot plate untill the digest became colourless and volume was raised up to 50 mL. The digest was then filtered through Whattman Filter Paper No. 42, preserved in polyethylene bottles to avoid contamination and stored at 4 °C until analysis.

Standards preparation. Six working standards of 0.2, 0.5, 1, 1.5, 2, and 5 ppm for Cd, Cr, Pb, and Cu were prepared using stock solutions of 1000 ppm, prepared in accordance to the ASTM Standards. Stock solutions (1000 ppm) of selected metals were prepared by dissolving appropriate amounts of Cd(NO₃)₂, K₂Cr₂O₇, Pb(NO₃)₂ and CuSO₄.5H₂O in 1000 mL of doubly distilled deionised water. Further dilutions were made from these stock solutions when required.

Analysis of metals. The digests were analysed for trace metal (Cd, Cr, Cu, and Pb) in chicken, mutton and beef

using a Perkin Elmer Analyst 800 Atomic Absorption Spectrophotometer using air-acetylene flame. Concentrations were determined by WinLab32 software operated.

Statistical analysis. Statistical Package for Social Sciences (SPSS) 16.0 was used for the data analyses. Descriptive statistics $\bar{x} \pm$ SD was of main concern. Student's t-test was applied to study the significance (p<0.05) of mean values with the permissible limits of the selected heavy metal in the organs of mutton (goat), chicken and beef (cow).

Results and Discussion

Concentration of Cd, Cr, Pb and Cu in different organs of mutton (goat), chicken and beef (cow) are presented in Table 1. Among the selective heavy metals of the study, the highest and lowest mean concentrations were recorded for Cd in liver (2.165 μ g/g) and Cu in brain (0.241 μ g/g) in mutton (goat). Moreover, it was revealed that the mean heavy metals concentrations in different organs of chicken and beef (cow) samples, ranges from 0.097 μ g/g (Cu) to 2.335 μ g/g (Cr) and 0.092 μ g/g (Cu) to 1.421 μ g/g (Cr), respectively.

Cadmium (Cd). Cadmium is a non-essential, toxic element for human and food is reported to be an important source of human exposure to Cd (Baykov et al., 1996). The Agency for Toxic Substances and Disease Registry (ATSDR, 2013) reported Cd as seventh most toxic substance. The high dose of Cd may lead to kidney dysfunction, liver and testicles damage, hypertension, lung damage and hepatic injury (John and Jeanne, 1994). Among different organs of mutton, chicken and beef samples, the highest mean concentration was recorded in kidney of mutton $(2.165 \pm 0.070 \ \mu g/g)$ and lowest in heart of chicken $(0.132 \pm 0.088 \,\mu\text{g/g})$ (Table 1). The concentration of Cd in the samples showed significant variability (p<0.05) among the brain, liver, gizzard, kidney and flesh of mutton; and lungs, liver, kidney and flesh of beef. This indicates the high Cd exposure risk associated with their consumption. It has no statistical significance in the meat of chicken (p>0.05). Cadmium mean concentration in all the chicken organs was found to be within the permissible limit of 0.5 ppm set by FAO/WHO (2000). However, the mean concentrations in brain, liver, kidney, gizzard, and flesh samples of goat, and lungs, liver, kidney and flesh samples of cow exceeded this limit, indicating high risk associated with their consumption. Compared with other studies, the Cd concentrations in beef were found lower than in some previously reported studies (Abd EI-Salam et al.,

	Organ $(n = 30)$	Cd	Cr	Рb	Cu
Mutton	Brain	$1.784 \pm 0.088 **$	1 ± 0.135**	$1.281 \pm 0.087*$	0.241 ± 0.071
	Heart	0.266 ± 0.032	$1.053 \pm 0.275*$	0.559 ± 0.216	0.315 ± 0.066
	Lungs	0.341 ± 0.100	$0.967 \pm 0.160*$	0.728 ± 0.093	0.319 ± 0.039
	Liver	$2.074 \pm 0.212 **$	$1.029 \pm 0.149 **$	$1.411 \pm 0.139*$	1.195 ± 0.077
	Gizzard	$1.023 \pm 0.162*$	$1.342 \pm 0.067 **$	0.974 ± 0.197	0.898 ± 0.059
	Kidney	$2.165 \pm 0.070 **$	$0.884 \pm 0.096^{**}$	0.788 ± 0.069	0.524 ± 0.024
	Flesh	$0.741 \pm 0.064*$	$1.148 \pm 0.260 *$	0.683 ± 0.122	0.491 ± 0.055
Chicken	Brain	0.337 ± 0.070	$0.768 \pm 0.069 **$	0.504 ± 0.224	0.154 ± 0.179
	Heart	0.132 ± 0.088	$0.769 \pm 0.143*$	0.697 ± 0.297	0.174 ± 0.115
	Lungs	0.137 ± 0.191	$0.902 \pm 0.278*$	0.261 ± 0.188	0.088 ± 0.049
	Liver	0.156 ± 0.119	$1.048 \pm 0.229^*$	0.705 ± 0.072	0.168 ± 0.069
	Gizzard	0.178 ± 0.083	$2.335 \pm 0.697 *$	0.783 ± 0.324	0.097 ± 0.063
	Kidney	0.182 ± 0.092	1.211 ± 0.699 *	0.846 ± 0.367	0.354 ± 0.208
	Flesh	0.211 ± 0.149	$1.912 \pm 0.458*$	0.962 ± 0.417	0.409 ± 0.201
Beef	Brain	0.407 ± 0.083	$1.421 \pm 0.060 **$	0.698 ± 0.023	0.453 ± 0.074
	Heart	0.398 ± 0.089	$1.206 \pm 0.351*$	0.504 ± 0.139	0.354 ± 0.106
	Lungs	$0.574 \pm 0.105 *$	$1.411 \pm 0.635*$	0.688 ± 0.059	0.211 ± 0.082
	Liver	$0.619 \pm 0.060 *$	$1.086 \pm 0.326*$	0.634 ± 0.185	0.181 ± 0.089
	Gizzard	0.367 ± 0.092	$1.212 \pm 0.314*$	0.773 ± 0.279	0.092 ± 0.039
	Kidney	$0.634 \pm 0.191 *$	$1.219 \pm 0.332*$	0.714 ± 0.088	0.191 ± 0.105
	Flesh	$0.597 \pm 0.140 **$	$0.898 \pm 0.256 *$	$1.122 \pm 0.250 *$	0.125 ± 0.060

Table 1. Mean concentration \pm SD (µg/g) of the selective heavy metals in different organs of mutton (goat)(n=210), chicken (n=210) and beef (cow)(n=210) samples

*and ** shows statistically significant and highly significant mean values (p<0.05), respectively.

2013; Alturiqi and Albedair, 2012; Chowdhury et al., 2011; Asegbelovin et al., 2010; Mariam et al., 2004), except in the study by Akan et al. (2010) which reported higher Cd concentration in beef than in this study. In chicken samples, Cd levels were reported lower than some previous studies (Abd EI-Salam et al., 2013; Alturigi and Albedair, 2012; Chowdhury et al., 2011; Mariam et al., 2004) but were higher than those reported by Mohammed et al. (2013) and Akan et al. (2010). However, the Cd levels detected in mutton were found higher than those previously reported (Table 2). Therefore, consumption of local meat with these high Cd content may cause serious public health concerns such as kidney dysfunction, liver and testicles damage, hypertension, lung damage and hepatic injury in the consumers (Maurice et al., 1994). Moreover, Cd accumulates in the liver and kidney where it interacts with essential minerals such as Zn, Cu, Fe, and Se and competes for binding sites (McLaughlin et al., 1999) and also affects the calcium and phosphorus metabolism in human (Jarup et al., 1998).

Chromium (Cr). Chromium (VI) has been ranked as 17th most toxic substance (ATSDR, 2013) and has been classified into the group A: "Human Carcinogen" by USEPA (1999), due to its carcinogenic impacts. The

mean concentration of Cr (VI) determined in different organs of mutton, chicken and beef were ranged from $0.768 \pm 0.069 \ \mu$ g/g in brain samples of chicken to $2.335 \pm 0.697 \ \mu$ g/g in gizzard samples of chicken (Table 1). The concentration of Cr was statistically significant (p<0.05) in all the organs of mutton, chicken and beef, particularly a high significance was reported in beef samples.

The mean Cr concentrations in all the studied organs of chicken, mutton, and beef samples exceeded the permissible limit of 0.1 ppm set by ANZFA (2008). These high concentrations of Cr (VI) in meat samples are probably due to its uncontrolled release from industrial discharges; where it has been used in leather tanning, mining, cement and construction industries, electroplating, dyeing, paints and pigments, rust inhibitors, fungicides, alloys manufacturing and glass manufacturing industries (Fahim et al., 2006). Moreover, fascinatingly, Mahmud et al. (2011) reported a fact that in Pakistan to meet the high chicken consumption demand, the poultry chicken is fed with the feed containing small leather pieces from leather tanneries, contaminated with Cr (VI) during chrome tanning process. In the present study, the Cr concentrations in the meat products were higher as compared to previous studies (Abd EI-Salam *et al.*, 2013; Chowdhury *et al.*, 2011; Mahmud *et al.*, 2011), except for beef samples in comparison with previously reported data by Abd EI-Salam *et al.* (2013) (Table 2).

Lead (Pb). Lead concentrations assessed in different organs of mutton, chicken and beef varied from lowest in lungs samples of chicken (i.e. $0.261 \pm 0.188 \, \mu g/g$) and highest in liver samples of mutton (i.e. $1.411 \pm$ $0.139 \mu g/g$ (Table 1). On comparison with permissible limit of 1 ppm set by ANZFA (2008), it was revealed that the mean Pb concentration in brain and liver samples of mutton and flesh samples of beef exceeded the limit, indicating the potential risk to human from the second most hazardous substance worldwide (ATSDR, 2013). In comparison with previously reported data, the Pb concentrations in this study were lower than previous reported data (Abd EI-Salam et al., 2013; Alturiqi and Albedair, 2012; Chowdhury et al., 2011; Mariam et al., 2004), and higher than those reported by Akan et al. (2010). However, results were comparable with data reported by Asegbelovin et al. (2010). The Pb exposure to the meat consumer in the less developed country may impart toxic impacts on haemopoietic, nervous, renal and gastrointestinal systems (Baykov et al., 1996).

Copper (Cu). Copper is an essential element in trace amount for the production of heamoglobin and haemocyanin in the vertebrates. It also plays a vital role in bone formation, integrity of the connective tissues, and skeletal mineralisation (Akan et al., 2010). However, its concentration in excess to the permissible limits may cause adverse impacts such as liver and kidney damage (Brito et al., 1990). Macrae et al. (1993) reported that dizziness, intestinal discomfort and headaches, hepatitis or cirrhosis, and/or hemolytic crisis in human were associated with ingestion of copper in food. The highest Cu level in this study was determined in liver samples of mutton $(1.195 \pm 0.077 \,\mu g/g)$, while the lowest mean Cu concentration was recorded in lungs tissues of chicken samples $(0.088 \pm 0.049 \,\mu\text{g/g})$ (Table 1). As Cu is an essential nutrient, a recommended dietary allowance (RDA) of 0.9 mg/day (0.013 mg/kg/day) has been set by ATSDR (2004). Thus, the Cu content in all the meat samples were in excess to recommended nutrient requirements by human through diet. Moreover, the Cu concentration in all the studied organs of the mutton, chicken and beef was found to be well within the permissible limits of 200 ppm set by ANZFA (2008). The copper concentrations were also found lower than those reported previously in other publications

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 Table 2. Concentrations (ppm) of heavy metals in meat samples reported in other studies

Meat sample	Metal	Concentration	Region	Reference
Chicken	Cd	1.36-1.68	Saudi Arabia	Alturiqi &
	Pb	7.61-10.49		Albedair (2012)
	Cu	2.31-7.88		
Beef	Cd	1.56-2.02		
	Pb	5.85-7.93		
	Cu	9.59-13.10		
Chicken	Cd	0.03-0.019	Nigeria	Mohammed et al.
	Pb	0		(2013)
Beef	Cu	25-30	Azad	Sabir <i>et al.</i> (2003)
Mutton	Cu	68-71	Kashmir,	
Chicken	Cu	11-13	Pakistan	1
Beet	Cd	0.17-0.22	Nigeria	Akan <i>et al.</i> (2010)
	Pb	0.15-0.25		
	Cu	0.54-0.87		
Mutton	Cd	0.34-0.76		
	Pb	0.08-0.16		
C1 · 1	Cu	0.34-0.98		
Chicken	Ca	0.16-0.27		
	Pb	0.16-0.22		
Deef	Cu	0.01-1.44	T -1	Maniana et al
Beer	Ca Dh	0.33-0.909	Lanore,	Mariam $et al.$
	PD	2.02-2.19	Pakistan	(2004)
Mutton	Cu	5.42-93.24		
Mutton	Ca Dh	0.37-0.45		
	PD	5.85-4.25		
Chielen	Cu	5.01-518.82		
Chicken	Dh	0.51-0.49		
	PU	5.1-5.15		
Poof	Cd	0.91-12.80	Panaladash	Chowdhum at al
Deel	Cu Cr	0.05-8.04	Bangiadesn	(2011)
	Dh	0.67.24.9		(2011)
	Cu	0.07-24.9		
Goat	Cd	0.15		
Goat	Cr	0.08		
	Ph	1 35		
	Cu	3.92		
Chicken	Cd	5 20		
chienen	Cr	0.69		
	Pb	41.94		
	Cu	10.33		
Beef	Cd	0.28-1.50	Nigeria	Asegbelovin et al.
	Pb	0.80-1.42	U	(2010)
Mutton	Cd	0.04-0.93		
	Pb	0.02-1.36		
Beef	Cd	0.3-1.23	Kohat,	Abd EI-Salam et al.
	Cr	0.3-15.76	Pakistan	(2013)
	Pb	2.5-11.83		
	Cu	4.6-8.58		
Goat	Cd	0.37-1.58		
	Cr	0.41-0.46		
	Pb	1.85-2.7		
	Cu	3.22-82.83		
Chicken	Cd	0.86-1.51		
	Cr	0.07-0.53		
	Pb	1.95-3.25		
	Cu	0.41-20.86		
Chicken	Cr	0.233-1.266	Lahore, Pakistan	Mahmud <i>et al.</i> (2011)

(Abd EI-Salam *et al.*, 2013; Alturiqi and Albedair, 2012; Chowdhury *et al.*, 2011; Mariam *et al.*, 2004; Sabir *et al.*, 2003) (Table 2). Therefore, Cu contents in the local meat samples could be considered with insignificant risk to public.

Conclusion

This study revealed that the concentrations of Cd, Cr and Pb in different organs of meat (chicken, goat and cow) samples, commercially available in local markets of Lahore, (Pakistan) exceed to the permissible limits set by international health organizations. The high concentrations of non-essential metals have identified the high risk vulnerability of the local population on consumption of the contaminated meat products. Therefore, this study suggests a critical need to formulate and implement national food safety standards in Pakistan to ensure the availability of safe meat products in Pakistan. Further studies are needed to investigate the risk associated with bioaccumulation of these trace metals in locals due to consumption of the contaminated meat products.

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