Determination of heavy metals in various tissues of locally reared (country) chicken in major districts of Karnataka, India: Assessment of potential health risks.

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Abstract: Food is one of the most prevalent ways that humans are exposed to metals. Heavy metals including cadmium, iron, zinc, lead, and mercury are harmful to humans and have a detrimental impact on health because they accumulate in biological organs. The concentration levels of these heavy metals were tested in different edible parts of the country (locally raised) chicken from various districts in Karnataka, India, namely Bengaluru, Tumakuru, Mangaluru, and Udupi, using an Atomic-Absorption Spectrophotometer (AAS). Heavy metal concentrations in various chicken parts were found to be below detectable limits (BDL)-0.0062, 0.027-3.178, and 0.262-2.103 ppm for Cd, Fe, and Zn, respectively, whereas Hg and Pb were BDL. The content of Zn was found to be significantly higher in all chicken samples from all examined districts, followed by Fe and Cd. Hg and Pb concentrations, on the other hand, were below the detection level in all samples. The estimated daily intakes (EDIs) of the observed metals from country chicken consumption were found to be lower than their respective FAO/WHO reference oral doses (RfD). The non-carcinogenic health hazards posed by the tested metals to the target population were estimated using the Hazard Quotient (HQ) and Hazard Index (HI) values. The HQ and HI values observed in this estimation were less than one, indicating that exposure to these heavy metals through the consumption of country chicken is unlikely to provide possible health concerns to the examined region's human population.

Keywords: Metal exposure; Country chicken; Food safety; Toxicity; Health hazards; Permissible Limit.

1. Introduction

The chicken industry (Poultry) is one of India's most rapidly increasing agro-based industries, and it is a major supplier of critical nutrients, namely protein (Burel and Valat, 2009). In India, two chicken varieties, broiler and country chicken (village-reared), are consumed. Country chicken is raised locally by people in their homes and is identifiable by its more diminutive stature and different colours. Country chicken consumption by the native population is substantial, particularly in our research area. However, it is lower than broiler chicken consumption due to the latter's ease of availability and low cost. Chicken meat is relatively less expensive than other meats, making up about 45% of the total meat consumed in India (Russell et al., 2001; Chatterjee and Rajkumar, 2015). Recently, the high demand for chicken meat has substantially affected production, leading to increased production and extensive modification of poultry feeds to meet the increased demand. It has been reported that poultry feeds, whether local, natural or improved modifications from unique manufacturing processes, are affected by the content of heavy metals (Chatterjee and Rajkumar, 2015). The primary way that heavy metals bioaccumulation in chicken is via consuming contaminated food.

Heavy metal food poisoning is a significant health hazard globally, especially in developing nations like India (Mohammed et al., 2013). Heavy metals commonly poison chickens through contaminated water, poultry feed, industrial effluents, sewage water, agricultural activities, and aerial spraying in chicken breeding zones. Several studies on the heavy metal level in meat samples indicate that bioaccumulation of heavy metals in animal feeds is the main source of intake (Das and Das, 2018; Korish and Attia, 2020). Heavy metals like Cadmium (Cd), Iron (Fe), Zinc (Zn), Mercury (Hg) and Lead (Pb) are plentiful in the environment and can also be found in small amounts in fishes and chickens. Humans are exposed to these heavy metals through the consumption of contaminated food, particularly livestock (Khan et al., 2015; Mathiyan et al., 2021). Furthermore, other heavy metals such as Hg, Cd, and Pb are not required for the body's health and operation (Ayar et al., 2009). In general, increasing toxic element intake endangers all living organisms (Qin et al., 2009). The International Research Agency on Cancer and the United States National Toxicology Program have both classified increased human Cd consumption as a carcinogen (Congeevaram et al., 2007). Cd is a typical additive toxic compound that can build up in the kidneys over the years. It poses acute toxicity in humans due to taking more than the limit from contaminated food and beverages (Khan et al., 2015). Fe and Zn are useful and necessary elements in human development; Zn deficiency will cause abnormal growth, skin eruption, hair loss, etc., and excessive intake will Pb to nausea, cramps, and diarrhoea (Sharma and Agarwal, 2005). A deficiency of Fe causes anaemia in humans and the high accumulation leads to dangerous problems with the liver, pancreas, heart diseases, and diabetes (Whittaker, 1998). Hg and Pb are non-essential metals and both will affect human beings seriously, even in the presence of low concentrations to humans (Park and Zheng, 2012; Assi et al., 2016). In children, low level of Hg exposure leads to depression, rashes, and pleading of hands and legs, in adult memory loss and more Hg intake leads to Minamata disease. Pb disturbs many metabolic activities in humans, like protein folding and apoptosis, and causes an increase in Reactive Oxygen Species and a decrease in antioxidant activity (Rice et al., 2014; Bharti and Sharma, 2022).

The contamination of trace elements in various tissues of poultry chicken has been studied and reported by various authors (Mathaiyan et al., 2021; Pappuswamy et al., 2021; Karabasavanar et al., 2020; Kumar et al., 2007). However, due to a lack of information on contamination of heavy metal levels and their possible negative effects on the population of consumers, the country (village-reared) chicken species cultivated in India is one of the items that is disregarded in this respect. As far as our understanding, not many studies on the quantification of heavy metals in various edible tissues of country chicken in Karnataka have been published. Hence, this research aims to identify the concentrations of heavy metals such as Cd, Fe, Zn, Hg, and Pb, in various organs of edible country chicken, including the muscle, bone, gizzard, intestine, and liver, collected from four different Karnataka cities (Bangalore, Tumakuru, Mangalore, and Udupi).

2. Materials and Methods

2.1. Reagents and instruments

In this study, all the chemicals and reagents used were of analytical quality. Our lab used a Milli-Q water purification equipment (Millipore, USA) was used to create double deionized water for dilutions. Before being cleaned with deionized water, the glass and plastic dinnerware were immersed in weak HNO_3 (10%) for the night. The stock solutions (1000 mg/l) of Ultra-Scientific were used to create standard solutions for all the heavy metals evaluated.

2.2. Study Area

Country chicken samples were taken from four major districts of Karnataka state, such as Bengaluru (BLR), Tumakuru (TKM), Mangaluru (MLR), and Udupi (UDU), to assess the existence of heavy metals such as Cd, Fe, Pb, Zn, and Hg, in different edible parts of country chickens. The sample areas are presented in Fig. 1. Enormous industrial and agricultural activities across these cities are considered to be the major contributors to environmental pollution.

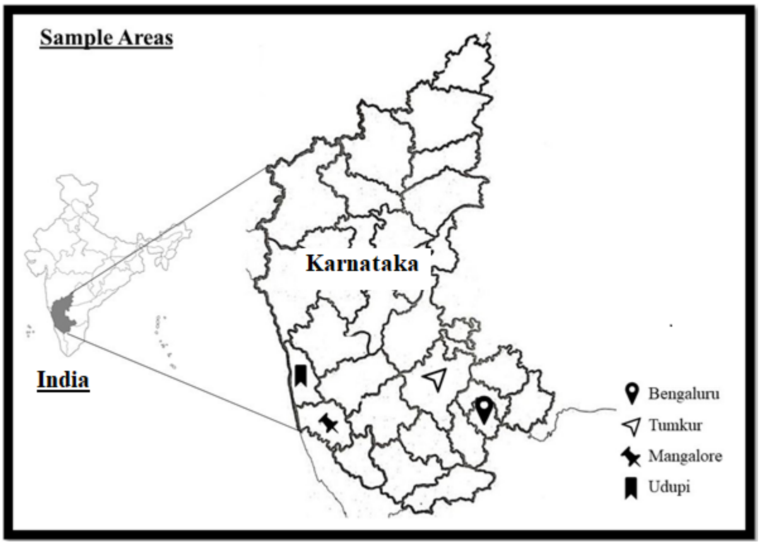

Figure 1 - Map indicating major districts where country chickens were sampled from Karnataka, India

2.3. Sample Collection

Throughout the selected districts, 1000 live adult country chickens were collected, individually, from August to September 2022. Each district gathered 250 chicken samples, which included 50 muscle, bone, gizzard, intestine, and liver samples (50×5×4=1000). To avoid metal contamination, the chicken samples were cut and removed from the live chicken with a clean ceramic knife, then packed, tagged, and stored in the cooled container with ice bags before being transported to the laboratory and kept in the -20 °C freezer for further investigation.

2.4. Sample Preparation

All the glassware used for the sample digestion was cleansed with double distilled water before the sample digestion. They were then immersed in a 5% HNO_3 solution overnight and rinsed several times in distilled water before use. The chicken samples were chopped into small pieces using a clean ceramic knife after thawing at room temperature. Then the samples were dried at 80 ºC for 48 hr in a hot air oven and were ground using a porcelain pestle and mortar. Each powdered chicken sample was kept for incubation for 10 hr, by mixing 1 g with HNO_3 and HCl in a 3:1 ratio. Then it was digested for two hours at 40 °C in a fume hood on a hot plate, with the beakers stirred every 30 minutes to ensure thorough digestion. The samples were then transferred to a sample vial for analysis after being filtered in a 50 ml volumetric flask using Whatman filter paper and filled to the appropriate level with distilled water (Beutler et al., 2014).

2.5. Analytical Method

The sample solutions were analyzed using an atomic absorption spectrophotometer (AAS) (Shimadzu AA6880, Japan) for the quantification of Cd, Fe, Pb, Zn, and Hg. The analytical wavelengths of investigated elements were 228.8 nm for Cd, 248.3 nm for Fe, 213.9 nm for Zn, 283.3 nm for Pb, 220.353 nm for Pb, and 253.7 nm for Hg. All the samples were injected three times and the data were monitored using WizAArd software. The concentrations of all heavy metals were calculated in 'ppm' on the fresh weight basis of the chicken tissue samples.

2.6. Potential health risk assessment

The approach established by the US Environmental Protection Agency (USEPA, 1989) was used to assess the non-carcinogenic risk of heavy metals exposure through the consumption of country chicken parts for people (male and female) in the study area. The calculated estimated daily intake (EDI) values of heavy metals from the eating of edible chicken parts (muscle, bone, gizzard, liver) were based on the mean concentrations of heavy metals in the national chicken samples and the average daily consumption in these regions. The EDI (mg/kg/day) has been calculated using the equation (Eq. 1)

$$EDI = Cm \times \frac{ADC}{BW} \tag{1}$$

Where, Cm (mg/kg on a fresh weight basis) – the average metal concentrations in country chicken samples observed in this study, ADC (kg/person/day) – average daily basis consumption of country chicken in the region and BW – body weight of the target population, 65 kg for adult males and 55 kg for adult females (ICMR-NIN, 2020)

During sample collection, a survey was done to determine the aforementioned study region, and the average daily consumption of country chicken was supposed to be 0.08 kg and 0.06 kg for adult males and females, respectively. Furthermore, because these components are frequently not consumed by the target population, the gut was not included in the risk assessment based on this survey.

The non-carcinogenic risks of trace metals in country chicken consumed by the local population are expressed as the HQ (Hazard Quotient), which is the ratio of a contaminant's EDI (estimated in this study) to the reference oral dose (RfD), which is an estimated amount of tolerable daily intake of contaminants by humans over a lifetime. The HQ was calculated using the USEPA guidelines (USEPA, 1989) and the following mathematical equation (Eq. 2)

$$HQ = \frac{Cm \times EF \times ADC \times ED}{RfD \times BW \times At} \times 10^{-3} \tag{2}$$

Where, Cm (mg/kg on a fresh weight basis) – the average metal concentrations in chicken samples observed in this study,
ADC (kg/person/day) – average daily basis consumption of country chicken;
BW – body weight of the target population,
ED – exposure duration (70 years),
EF is exposure frequency (365 days/year), At is the average exposure time (ED×356 days) and RfD is oral reference dose (mg/kg/day).

The RfD (ingestion) values used in this assessment were selected as per the guidelines of Joint, F.A.O., WHO Expert Committee on Food Additives, & World Health Organization (2011). If the HQ value is less than unity (<1), the exposure of pollutants to the population is unlikely to cause obvious adverse effects.

The degree of the exposed population's negative impacts was expected to be proportionate to the total amount of various pollutants. The Hazard Index (HI) was subsequently determined by adding the Hazard Quotients (HQs) of all the metals evaluated in this study. According to the USEPA (1989), HI is regarded as an assessed possible risk of exposure to certain heavy metals. The following equation (Eq. 3) was used to determine the HI.

$$HI = \sum_{HQ} = HQ_{Cd} + HQ_{Fe} + HQ_{Zn} + HQ_{Hg} + HQ_{Pb} \tag{3}$$

2.7. Statistical Analysis

All obtained data were analyzed statistically using IMB SPSS 22.0 software; mean results obtained from the work were subjected to ANOVA (one-way analysis of variance). Tukey's technique was used to compare means, and a difference of 5% (p <0.05) was declared significant. The data is provided with mean and standard errors with a p-value superscript given based on concentrations (EPA, 2000). The Pearson correlation test was used to assess the association between heavy metal levels in various tissue samples.

3. Results

In the present study, the concentrations of various heavy metals such as Cd, Fe, Zn, Pb, and Hg were assessed in country chicken tissues (muscle, bone, gizzard, intestine, and liver) collected from Bengaluru, Tumakuru, Mangalore, and Udupi are depicted in table 2.

Cd levels in the investigated chicken sample were determined to be far below the maximum permitted levels for human consumption of 0.05 ppm in meat, 0.5 ppm in liver, and 0.5-1.0 ppm overall (EC, 2006). The mean amount of Cd found in the various edible parts of the country chicken samples, such as muscle, bone, gizzard, intestine, and liver, collected from all the different cities of Karnataka is presented in table 2. Cd concentration was registered high in the muscle sample of BLR (0.0046±0.0008 ppm) followed by TKU (0.0033±0.0006 ppm) and the lowest level was recorded in MLR and UDU and differed significantly among the cities (p <0.05). Similarly, the bone and intestine samples from TKM and gizzard samples from MLR, liver from all the towns, except UDU, registered a significantly high level of Cd (p <0.05) (Table 2). Cd concentrations for muscle, bone, gizzard, intestine, and liver samples were in the order of BLR > TKM > MLR = UDU; TKM > MLR > UDU = BLR; MLR > TKM > BLR = UDU; TKM > MLR = BLR = UDU; MLR > BLR > TKM = UDU respectively. Whereas the amount of Cd registered in different tissues was found in the order of Gizzard > Liver > Bone > Intestine > Muscle and all the samples varied significantly (p <0.05).

Agency	Cd	Fe	Zn	Hg	Pb
WHO/FAO 2002	0.05 for meat 0.5 for liver	30-150	20	NA	0.1
MHPR China	0.1 for meat 0.5 for liver	NA	100	NA	0.2
European Communities 2006	0.05 for meat 0.5 for liver	NA	20	NA	0.1
FSSAI 2011	0.05	NA	NA	NA	0.1

Table 1 – Permissible limit of selected metals in chicken samples (ppm).

Fe concentrations detected in the selected edible part of the country chicken samples, such as muscle, bone, gizzard, intestine, and liver, sampled from major cities of Karnataka. All the chosen samples that were recorded had Fe values that were within the maximum allowable range recommended by WHO/FAO (Table 1). The Fe concentrations (ppm) in muscle registered 0.5241±0.0352 (BLR), 0.0463±0.0236 (TKM), 0.3979±0.0820 (MLR) and 0.3166±0.1305 (UDU) all the samples are varied significantly (p <0.05), the bone samples did not register any significant variations among the cities. The Fe concentrations in liver samples from BLR, TKM, MLR, and UDU registered 2.1047±0.2370, 2.3673±0.7155, 0.4952±0.0597 and 0.6252±0.1862 ppm respectively, which were all good within the maximum permissible limits, the highest individual concentration was found in liver 3.1782ppm with the mean value of 1.3981±0.9336ppm. The samples of gizzard and intestine also registered low concentrations in all the cities of Karnataka with mean values of 1.1187±0.5602 and 1.0512±0.5412ppm. The order of the heavy metals in tissues found to be liver > gizzard > intestine > bone > muscle, except bone in all the samples, varied significantly with (p < 0.05).

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Chicken sample	Bengaluru	Tumakuru	Mangaluru	Udupi	P value	Min-Max	Overall mean
			Cadmium (Cd)				
Muscle	$0.0046^{a}\pm0.0008$	$0.0033^{b}\pm0.0006$	$0.0002^{c}\pm0.0002$	$0.0002^{c}\pm0.0001$	0.000	0-0.0058	0.0021
Bone	$0.0014^{b}\pm0.0002$	$0.0049^{a}\pm0.0019$	$0.0022^{b}\pm0.0009$	$0.0014^{b}\pm0.0005$	0.000	0.001-0.007	0.0025
Gizzard	$0.0027^{c}\pm0.0004$	$0.0064^{b}\pm0.0011$	$0.0135^{a}\pm0.0012$	$0.0021^{c}\pm0.0006$	0.008	0.0013-0.0151	0.0062
Intestine	$0.0019^{b}\pm0.0003$	$0.0044^{a}\pm0.0015$	$0.002^{b}\pm0.0004$	$0.0015^{b}\pm0.0003$	0.000	0.0012-0.0065	0.0025
Liver	$0.0022^{a}\pm0.0006$	$0.0019^{ab}\pm0.0003$	$0.0041^{a}\pm0.0055$	$0.0017^{b}\pm0.0001$	0.013	0.0007-0.019	0.0025
			Iron (Fe)				
Muscle	$0.5241^{a}\pm0.0352$	$0.0463^{d}\pm0.0236$	$0.3979^{b}\pm0.082$	$0.3166^{c}\pm0.1305$	0.000	0.0274-0.6169	0.3212
Bone	$0.3704^{b}\pm0.1073$	$0.3375^{b}\pm0.1622$	$0.7365^{a}\pm0.1159$	$0.3624^{b}\pm0.1433$	0.805	0.0453-0.877	0.4517
Gizzard	$0.7239^{c}\pm0.1176$	$0.9993^{b}\pm0.0677$	$2.0304^{a}\pm0.1021$	$0.7213^{c}\pm0.2009$	0.000	0.4501-2.1581	1.1187
Intestine	$0.966^{b}\pm0.0947$	$1.4547^{a}\pm0.3453$	$0.3355^{c}\pm0.1174$	$1.4486^{a}\pm0.4407$	0.000	0.1254-2.0497	1.0512
Liver	$2.1047^{a}\pm0.237$	$2.3673^{a}\pm0.7155$	$0.4952^{b}\pm0.0597$	$0.6252^{b}\pm0.1862$	0.000	0.4458-3.1782	1.3981
			Zinc (Zn)				
Muscle	$1.5174^{a}\pm0.0122$	$1.4866^{a}\pm0.2276$	$0.991^{b}\pm0.0267$	$1.1044^{b}\pm0.1341$	0.000	0.9574-1.7828	1.2749
Bone	$0.7041^{c}\pm0.0900$	$1.2328^{a}\pm0.2997$	$1.2321^{a}\pm0.1298$	$1.0266^{b}\pm0.1698$	0.000	0.5257-1.4944	1.0489
Gizzard	$0.6521^{bc}\pm0.1534$	$0.8326^{b}\pm0.0395$	$1.5783^{a}\pm0.3769$	$0.5087^{c}\pm0.1202$	0.000	0.3258-2.1025	0.8929
Intestine	$1.3731^{a}\pm0.1026$	$1.2978^{a}\pm0.0155$	$0.9318^{c}\pm0.0634$	$1.071^{b}\pm0.1991$	0.000	0.8573-1.5155	1.1684
Liver	$0.5065^{d}\pm0.1816$	$1.1527^{b}\pm0.1458$	$1.5784^{a}\pm0.0988$	$0.9089^{c}\pm0.1047$	0.012	0.2615-1.6836	1.0366
			Mercury (Hg)				
			Hg found BDL in all the samples				
			Lead (Pb)				
			Pb found BDL in all the samples				

Table 2 – Heavy metal concentrations (Mean ± SD ppm) in edible chicken parts of major cities in Karnataka.

Zn in different tissues such as muscle, bone, gizzard, intestine, and liver of country chicken collected from major cities of Karnataka. The analyzed values for muscle samples from BLR, TKM, MLR, and UDU were 1.517±0.012, 1.487±0.228, 0.991±0.027 and 1.104±0.134ppm respectively. Zn's concentrations for muscle, bone, gizzard, intestine, and liver samples were in the order of BLR > TKM >UDU= MLR; MLR>BLR = UDU =TKM; MLR > TKM = BLR = UDU; TKM =UDU> BLR >MLR; MLR >TKM>UDU >BLR respectively. Whereas the concentration of Zn registered in different tissues was found in the order of muscle>intestine > bone > liver > gizzard and all the samples varied significantly (p <0.05). However, Zn concentrations in all samples collected from Karnataka's major cities are within the maximum permissible limit of 0.05 ppm, as recommended by various organizations (Table 2). The Pb and Hg concentrations were below detectable in all the samples (Muscle, Bone, Gizzard, Intestine, and Liver) collected from Bengaluru, Tumakuru, Mangaluru, and Udupi.

The correlation analysis revealed that there was no strong correlation between any of the metals. A positive correlation was observed between Fe-Cd, Zn-Cd, Zn-Fe, Pb-Fe, Pb-Zn, Hg-Fe, Hg-Zn, and Hg-Pb with r values of 0.177, 0.044, 0.235, 0.061, 0.232, 0.094, 0.558 and 0.095 respectively (Table 3). However, a negative correlation was found only between Pb-Cd and Hg-Cd with Corresponding r values of -0.095 and -0.085 respectively. The positive correlation coefficient is probably controlled by other sources.

	Cd	*Fe*	*Zn*	*Pb*	*Hg*
Cd	1				
Fe	0.17736	1			
Zn	0.04407	0.235330	1		
Pb	-0.09568	0.061742	0.232675	1	
Hg	-0.08503	0.094165	0.558071	0.095604	1

Table 3 – Correlation between heavy metal in different tissues of country chicken from major districts of Karnataka, India.

The estimated Daily Intake for males and females through consumption of the country chicken parts from major districts of Karnataka (BLR, TKM, MLR, and UDU) were calculated and presented in Table 4. The average metal concentrations estimated in various edible parts of country chicken (muscle, bone, intestine, gizzard, and liver) were used to compute the EDIs of all the heavy metals. The calculated EDIs for all the heavy metals in this study are found below the proposed doses (RfDs) set by USEPA (United States Environmental Protection Agency). However, the EDIs estimated in this research for women were higher than their male counterparts. The non-toxic risks of tested heavy metals via consumption of country chicken parts from major districts of Karnataka were calculated based on Hazard Quotient (HQ). The calculated HQ values of individual males and females were found less than 1 (Table 4), indicating that ingestion of these country chicken parts from major districts of Karnataka will not likely cause the target population to experience any negative health effects. The HQ values for male and female populations due to exposure to tested heavy metals were found safe. Hazard Index (HI), regarded as the total HQ values for each metal, has been computed to quantify the cumulative risk of exposure to multiple toxic elements. The HI values for both men and women (0.0416 and 0.0915, respectively) were found to be below 1 (Table 5), suggesting that there is no significant health effect of tested heavy metals to the target population through consumption of country chicken parts from major districts of Karnataka.

		Estimated Daily Intake (EDI) Male					Estimated Daily Intake (EDI) Female				
Place	Organ	Cd	Fe	Zn	Hg	Pb	Cd	Fe	Zn	Hg	Pb
Bangalore	Muscle	0.0056	0.645	1.8675	NA	NA	0.0066	0.7623	2.2071	NA	NA
	Bone	0.0004	0.1139	0.2166			0.0005	0.1346	0.25600		
	Gizzard	0.0004	0.1113	0.1003			0.0004	0.1316	0.1185		
	Liver	0.0006	0.6476	0.15558			0.0007	0.7653	0.1841		
Tumkur	Muscle	0.0040	0.0569	1.8296			0.0048	0.0673	2.1623		
	Bone	0.0015	0.1038	0.3793			0.0017	1.2272	0.4482		
	Gizzard	0.0009	0.1537	0.128			0.0011	0.1816	0.1513		
	Liver	0.0005	0.7284	0.3546			0.0006	0.8609	0.4191		
Mangalore	Muscle	0.0002	0.4897	1.2196			0.0002	0.5787	1.4414		
	Bone	0.0006	0.2266	0.3791			0.0008	0.2678	0.4480		
	Gizzard	0.0020	0.3123	0.2428			0.0024	0.3691	0.2870		
	Liver	0.0012	0.1523	0.4856			0.0014	0.1800	0.5739		
Udupi	Muscle	0.0002	0.3896	1.3592			0.0002	0.4605	1.6063		
	Bone	0.0004	0.1115	0.3158			0.0005	0.1317	0.3733		
	Gizzard	0.0003	0.1109	0.0782			0.0003	0.1311	0.0924		
	Liver	0.0005	0.1923	0.2796			0.0006	0.2273	0.3305		
WHO/FAO		1	17	20	0.71	3.57	1	17	20	0.71	3.57

Table 4 – Average metals concentrations (Cm, mg/kg fresh weight basis in Muscle, Bone, Gizzard and Liver samples), estimated daily intake (EDI, mg/kg/day) evaluated for heavy metal exposure to male and female in the major districts of Karnataka, India. Intestine was not considered in this risk evaluation, as it typically not consumed by the target population.

		Hazardous Quotient (HQ) Male					Hazardous Quotient (HQ) Female				
Place	Organ	Cd	Fe	Zn	Hg	Pb	Cd	Fe	Zn	Hg	Pb
Bangalore	Muscle	0.0056	0.0379	0.0933	NA	NA	0.0066	0.0448	0.0933	NA	NA
	Bone	0.0004	0.0067	0.0108			0.0005	0.0316	0.0433		
	Gizzard	0.0004	0.0065	0.005			0.0009	0.0154	0.0100		
	Liver	0.0006	0.038	0.0077			0.0008	0.1800	0.0311		
Tumkur	Muscle	0.004	0.0033	0.0914			0.0048	0.0039	0.0914		
	Bone	0.0015	0.0061	0.0189			0.0017	0.0288	0.0758		
	Gizzard	0.0009	0.009	0.0064			0.0023	0.0213	0.0128		
	Liver	0.0005	0.0428	0.0177			0.0006	0.2025	0.0709		
Mangalore	Muscle	0.0002	0.0288	0.0609			0.0002	0.0340	0.0609		
	Bone	0.0006	0.0133	0.0189			0.0008	0.0630	0.0758		
	Gizzard	0.002	0.0183	0.0121			0.0049	0.0434	0.0242		
	Liver	0.0012	0.0089	0.0242			0.0014	0.0423	0.0971		
Udupi	Muscle	0.0002	0.0229	0.0679			0.0002	0.0270	0.0679		
	Bone	0.0004	0.0065	0.0157			0.0005	0.0310	0.0631		
	Gizzard	0.0003	0.0065	0.0039			0.0007	0.0154	0.0078		
	Liver	0.0005	0.0113	0.0139			0.0006	0.0534	0.0559		
Hazardous Index (HI)	Male				0.0416						
	Female				00915						

Table 5 – Health risk assessment data (HQ and HI) calculated for heavy metal exposure to male and female in the major districts of Karnataka, India.

According to the study, the computed risk parameters (EDI, HQ, and HI) were within the permitted limits (table 5). Therefore, eating country chicken from the selected Karnataka districts is not anticipated to have appreciable negative health consequences for either the male or female communities. Toxic contaminants can accumulate in the human body over time and cause significant harmful effects. Thus, the health hazards associated with exposure to them through eating country chicken cannot be disregarded. So, proper measures have to be taken to control the contamination of these heavy metals in the selected areas to prevent the risks in future.

4. Discussion

Generally, heavy metals have several functional and physiological impacts on cell metabolism, as well as an oxidative impact on biological macromolecules that negatively affect nuclear proteins and DNA. In modest concentrations, these metals are necessary for the preservation of certain physiological and biochemical activities in living organisms; however, they become detrimental when they surpass specific criteria, and it is recognized that heavy metals can cause cell malfunction and, ultimately, toxicity. While chickens themselves do not naturally contain high levels of heavy metals, contamination can occur through their feed, water, or the environment in which they are raised. Consuming chicken that is contaminated with heavy metals can negatively affect human health. Heavy metals are toxic substances that can accumulate in the body over time, leading to various health problems (Jaishankar et al., 2014). Exposure to Pb, for example, can cause neurological damage, particularly in children. It can impact cognitive function, hinder development, and impair the nervous system (Sanders et al., 2009). Cd, another heavy metal, can accumulate in the kidneys and Pb to kidney damage. Prolonged exposure to high levels of Cd is associated with kidney disease, bone damage, and an increased risk of lung cancer (Fatima et al., 2019). Although Hg contamination in chickens is less common, it can still be a concern if the chickens have been exposed to Hg-contaminated feed or the environment. Hg can cause neurological and developmental issues, particularly in fetuses and young children (Bose-O Reilly et al., 2010; Abedi et al., 2023). The maximum permissible limit of selected elements is presented in Table 1.

Cd serves no physiological purpose in the human organism. Even at low amounts, its slow elimination rate can cause hepatic damage, respiratory and pulmonary issues, renal system damage, and prostate tumors (Rahimzadeh et al., 2017). In the current research, it was discovered that Cd contents were reduced in all four cities examined and were also below allowable limits, making them potentially safe for ingestion. Similar results were obtained with the chicken meat from retail chicken meat shops of Shivamogga district of Karnataka (Karabasanavar et al., 2020), Bareilly Western part of Utter Pradesh and Coimbatore, Erode, Chennai, Madurai, Salem, and Trichy cities of Tamilnadu states (Mathaiyan et al., 2021; Pappuswamy et al., 2021) in India. However, the mean level of Cd found in the present research is lower than the level reported in Malaysia (Abduljaleel et al., 2017), Iran (Sadeghi et al., 2015), Iraq (Al-Zuhairi et al., 2015; Hozan and Hemin, 2013), Bangladesh (Ullah et al., 2022), Pakistan (Imran et al., 2015), and Nigeria (Ogbomida et al., 2018). On the other hand, higher Cd contents have been reported from Bangladesh (Mottalin et al., 2018; Islam et al., 2015), Egypt (Ismail and Abolghait, 2013), and Riyadh, Saudi Arabia (Alturiqi and Albedair, 2012). Cd build-up in chicken may be caused by contact with Cd-plated items; batteries, paints, electronic waste, plastics, and so on (Hossain et al., 2019). Pb and Cd can accumulate in the reproductive organs, leading to reduced egg production, poor egg quality, and increased embryonic mortality (Taslima et al., 2022).

Since Fe is a necessary component of haemoglobin, both humans and animals need to consume Fe in their diets. The oxidation of carbohydrates, proteins, and lipids is facilitated by Fe, which helps maintain a healthy body weight. Low Fe levels make nasal bleeding, cardiovascular disease, and gastrointestinal disorders more likely by low Fe levels (Hunt, 2003). All plant and animal meals include Fe as a naturally occurring element. Water may also contain Fe. Toxic Fe levels in animals may cause cardiac arrest, convulsions, respiratory failure, depression, and paralysis. Intoxicated animals' post-examination revealed the effects of poisoning on the gastrointestinal system (Al-Ashmawy, 2013). A similar study registered a mean level of 2.3247±1.9029 ppm in Shivamogga (Karabasanavar et al., 2020), Karnataka. But, on the other side, the Fe concentration was found lower compared to earlier studies in Selangor Malaysia (Abduljaleel et al., 2012), Bangladesh (Ullah et al., 2022), Pakistan (Imran et al., 2015; Abdel-Salam et al., 2013), Benin City of Nigeria (Ogbomida et al., 2018), Iraq (Al-Zuhairi et al., 2015; Hozan and Hemin, 2013), and Saudi Arabia (Alturiqi and Albedair, 2012). Zn is the most abundant intracellular component required for various biochemical tasks such as gene expression, genetic stability, DNA repair, and apoptosis induction (Al-Bratty et al., 2018). On the other hand, Zn acts as a mediator for various physiological enzymes. However, excess Zn intake from food, drink, or nutritional supplements can harm one's health (Hunt, 2003). When large amounts of Zn are consumed, even for a short time, it will cause nausea, stomach cramps, and vomiting can occur. Anemia, pancreatic damage, and decreased high-density lipoprotein (HDL) cholesterol levels have all been linked to long-term Zn consumption (Al-Ashmawy 2013). Similar trends (within permissible limits) were reported in poultry chicken from Shivamogga, Karnataka, India (Karabasanavar et al., 2020), and Azad Kashmir, Pakistan (Sabir, 2003). High Zn concentration was reported in other countries like Bennin city Nigeria (Ogbomida et al., 2018), Enugu state (Okoye et al., 2011), Bangladesh (Ullah et al., 2022), Iraq (Hozan and Hemin, 2013; Hussain et al., 2012), Pakistan (Imran et al., 2015; Hossain et al., 2019), and Saudi Arabia (Alturiqi and Albedair, 2012).

Humans exposed to Pb and Hg can cause severe toxic effects, primarily affecting hematopoiesis, kidney function, reproductive and central nervous systems, and several other neurotoxins (Assi et al., 2016; Itodo et al., 2010; Khalafalla et al., 2011). The biological half-life of absorbed Pb in the human body is approximately 27 years in bone (Hanaa et al., 2011). It is worth noting that Pb and Hg concentrations in all country chicken samples collected from major Karnataka cities in the present study were found to be below detectable limits (BDL). However, high Pb concentrations in various edible parts of poultry chicken have been reported globally (Khan et al., 2015; Mathaiyan et al., 2021; Karabasanavar et al., 2020; Kumar et al., 2007; Ogbomida et al., 2018; Al-Bratty et al., 2018; Yayayuruk and Yayayuruk, 2017). The detection limits of these two heavy metals in the chicken are shown below, indicating that they are safe to include in the diet. Heavy metal in feed and drinking water is the primary source of heavy metals in chicken meat (Tuzen, 2009; Ghimpeteanu et al., 2012). Because the country chicken samples were not fed such feed, Pb and Hg may have been present in BDL and thus, it is safe to include in the human diet.

The immune system of chickens can also be suppressed by heavy metals, making them more susceptible to infections and diseases. This can influence to higher mortality rates and increased vulnerability to common poultry illnesses (Kim et al., 2019). Accumulation of heavy metals in organs such as the liver, kidneys, and lungs can cause significant damage. Chronic exposure to metals like Pb, Hg, and Cd can result in organ dysfunction, tissue degeneration, and impaired detoxification processes (Korish and Attia, 2020). Certain heavy metals, including Pb and Hg, can affect the central nervous system of chickens, leading to neurological symptoms such as reduced motor coordination, tremors, convulsions, and behavioral abnormalities (Fernandes et al., 2012). Additionally, heavy metals consumed by chickens can be transferred to eggs, resulting in contaminated eggs. This poses a potential health risk to humans if these eggs are consumed, as heavy metals can accumulate in human tissues and have harmful effects on human health (Korish and Attia, 2020; Aliu et al., 2021).

5. Conclusion

In conclusion, Samples were taken from four main districts in the Indian state of Karnataka to assess the presence of harmful heavy metals Cd, Fe, Zn, Hg, and Pb in several edible portions of country chicken, such as the muscle, bone, gizzard, intestine, and liver. The findings demonstrated that the Cd, Fe, and Zn levels in every sample taken from every location were within the allowable limits established by WHO/FAO and FSSAI. In contrast, Hg and Pb levels in all samples were under the detectable limit. This could imply that eating these country chickens is safe and has no negative impact on humans. This study also concludes that eating country chickens raised in villages is superior to eating chickens raised in different poultry forms. However, the study concluded that the calculated risk parameters (EDI, HQ, and HI) were within safe limits. Therefore, the consumption of edible parts of country chicken from major districts of Karnataka is not expected to cause any significant adverse health effects for both male and female populations.

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