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Assessment of Heavy Metals in Tannery Waste-Contaminated Poultry Feed and Their Accumulation in Different Edible Parts of Chicken

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Abstract: *A large amount of leather solid waste is generated daily from tanning industries. In recent years shaving dust – a major solid waste of tanning process, is being used in poultry feed production as a protein source because of its costless availability. However, leather is treated with many chemicals during its tanning process. Thus it may be one of the major routes of heavy metals assortment in poultry feed and may accumulate into human body through metal contaminated chicken meat. An experiment was conducted to determine heavy metals in poultry feed made of shaving dust that was collected from tannery. Assessment of deposition of heavy metal on tissues of broilers reared from day one old to 42 days by feeding this feed was also carried out. Concentration of heavy metals in shaving dust, powder (treated shaving dust with acid), poultry feed and in different edible organs of chicken like thigh, breast, liver, bones were analyzed using Inductively Coupled Plasma-Optical Emission Spectrometry (ICP-OES, Optima 7000DV, USA).Excess amount of Cr, Cu, Ni, Pb and Cd were found in poultry feed as well as in the different organs of chicken. The result found in this study crosses the limit stipulated by the joint FAO/WHO Expert Committee and EC on poultry.*

Keywords: *Carcinogenic effect, Chicken organs, Heavy metals, Shaving dust, Poultry feed.*

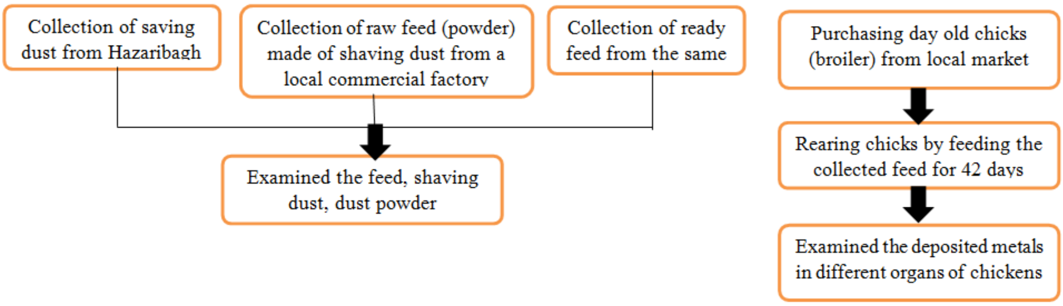
I. Introduction

Over the years, poultry meat and eggs has become an important source of protein in Bangladesh as well as in many parts of the world. All over the world poultry meat food like crispy spicy fried chicken, spicy chicken roll, tandoory chicken, burger, chicken nuggets, chicken ball have been attracted a considerable attention of young generation. From the nutritional perspective, chicken meat delivers essential vitamins, protein, minerals and fulfills a healthy eating pattern in Bangladesh diet. In Bangladesh – a densely populated country–for the demand of non-vegetable protein, people mainly relay on poultry meat and eggs in a large scale as it is affordable. With this huge demand of poultry products poultry firming has expanded to a significant extent which in turn lead to increase in poultry feed production. At present this poultry firming and poultry feed production is playing an important role in the economic growth and employment opportunities in Bangladesh. It also plays an important role on people's health (nutrition) in general. However, it is reported that poultry feed production system has been affected due to the use of heavy metals [1]. Although metals like iron, copper, zinc and manganese are essential and they play an important role in biological system but the metals like cadmium, lead, mercury are toxic even in trace amount [2]. All mineral elements, whether it is theorized as essential or potentially toxic, may have an adverse effect if included in the diet at excessively high concentration [3]. Contamination of chicken meat [4] with heavy metals may cause severe health hazard because of their toxicity, bio-accumulation and bio-magnifications. Serious illness such as kidney disease, damage to the nervous system, diminished intellectual capacity, heart disease, gastrointestinal diseases, bone fracture, cancer and even death can also happen due to the absorption of heavy metals through food [5]. Chicken meat could accumulate heavy metals if the chicken feed is contaminated through the various raw materials used for its production [3]. High concentration of Cr was found in different parts of chicken in Lahore, Pakistan [7] and in Tamil Nadu, India [8].

Recently in Bangladesh, some poultry feed producers using shaving dust from tannery industries as a protein source because of its costless availability. It imparts threatening to human health as tannery waste obtains large amount of toxic metals when the hides and skins are treated with different chemicals and salts in various stages of tanning process [9, 10]. About 40 heavy metals and various acids are used during the processing of raw hides [11]. Generally basic chromium sulphate salt is used in conventional chrome tanning of which about 60% to 70% of chromium compound is consumed by hides and skins [12] and chrome shaving dust contain 3.204% of chromium [13]. Use of these chrome shaving dust in poultry feed production as protein source without appropriate treatment would create a potential health risk of heavy metals in food chain [9] as they may be stored or incorporated in body tissues and have physiological and biochemical interruption in cellular levels [14]. There is a high possibility of transportation of these heavy metals from tannery waste contaminated poultry feed to chicken and then to the human body. Under such situation this research work was done to determine the amount of heavy metals present in contaminated poultry feed made of shaving dust and their response to the different edible parts of chicken after having such contaminated feed. In general, to use the shaving dust as a protein concentrate for poultry feed, it is boiled with a mixture of H_2SO_4 and water in a big container for 3 to 4 hours. After cooling, liquid is removed from the container and the concentrate of treated shaving dust is dried in the sunlight for few days and grinded to make powder. Powder is then mixed with other feed ingredients likes soya oil cake, corn grain and dried fish to make final feed.

II. Materials and Method

Experiment lay out: Current study was conducted according to the lay out below-



Scheme 1: Chart of experimental lay out

Sample Collection

Poultry feed and powder (modified shaving dust) were collected from a local feed factory while raw shaving dust was collected from a tannery of Hazaribagh area in polyethylene packets and analyzed as received. The collected shaving dust, powder and feed are shown in fig 1.

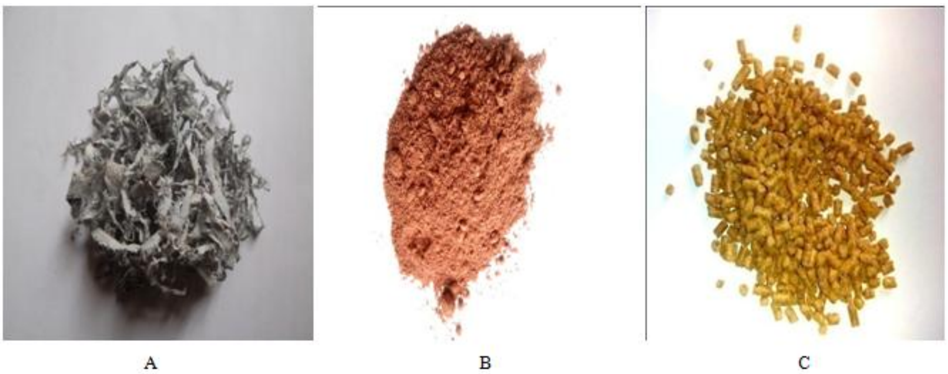


Figure no. 1: (A) shaving dust, (B) Powder, (C) Feed.

Collection of day old chicks and rearing

From a local poultry farm four day one old chick were collected for the present research. Rearing of chickens was done using the collected feed. At the beginning, feed was given after a little bit mashing to the day old chicks. As the chicks were growing older, they started taking whole grain feed. Sufficient amount of water was supplied in a bowl which was refreshed in regular intervals. The litter of the chicks was cleaned daily and they were put under the sunlight at day time. The day old chick's weight was around 40 g and after 42 days their weight was around 1.2 kg. Around 18 kg feed required for 42 days of rearing of four chickens. After 42 days, chickens were slaughtered and prepared for digestion.

III. Digestion of Samples

Shaving dust, powder and feed samples (about 200 g) were dried in an oven at 105^{o}C for two hours to remove all the moisture content from the samples and kept in desiccators for cooling. One gram of each sample was digested with a mixture of concentrated HNO_3, H_2SO_4 and $HClO_4$ in 5:1:1 ratio (15 mL) at 80^{o}c until the solutions become clear [15]. Then it was filtrated by Whatman no. 42 filter paper and diluted to 50 mL with de-ionized water.

Harvested tissues of chickens were cleaned with de-ionized water and cut into small pieces with a stainless steel knife. About 100 g of each sample was dried at 80^{o}c for 24 hours and then at 105^{o}c for 2 hours to remove all the moisture content. 0.5 g of each dried thigh, breast, bone and liver samples were digested with 15 mL of concentrated HNO_3, H_2SO_4 and $HClO_4$ in 5:1:1 ratio at 80^{o}c until the solutions become clear [15]. Filtrations of digested samples were done with Whatman no.42 filter paper and the filtrate was transferred into the volumetric flux and finally diluted to 25 mL with de-ionized water. All the tools were soaked in 10% nitric acid solution for 24 hours and rinsed with de-ionized water before using.

All reagents used were Merck, analytical grade, including standard stock solutions of known concentrations of different metals. All samples were stored in acid-leached polyethylene bottles at ambient temperature before analysis. Metal concentrations were determined by using ICP-OES (Optima-7000 DV, USA). Detection limits were 0.07, 0.25, 0.9, 0.4 and 1.4 ppb for Cd, Cr, Cu, Ni and Pb respectively. Wavelengths used for metals were 214.440, 267.716, 224.700, 231.604 and 220.353 nm for Cd, Cr, Cu, Ni and Pb respectively. Concentrations were calculated on a dry weight basis.

IV. Results and Discussion

Concentration of heavy metals in shaving dust, powder and feed

The average concentration (mg/kg) of heavy metals found in chrome shaving dust were 27267 for Cr, 29.54 for Cu, 16.81 for Pb, 8.84 for Ni and 0.28 for Cd. Chromium content in shaving dust is extremely high compared to other heavy metals which reflects the fact that in the tanning process large quantities of basic chrome sulphate is used; however, the Cr content in shaving dust found in the present study is lower than the previous study 32037 mg/kg done by Ferrera et al. [13] but higher than 14085 mg/kg obtained by Hossain et al. [16].

In the production process (acid treatment) of shaving dust powder the chromium content is found to reduce from 27267 mg/kg to 17678 mg/kg. A similar result 24901 mg/kg was found after acid boiling of chrome shaving dust in a previous report [13]. However, to our surprise, all other metal content in shaving dust powder is found to be higher than those of the chrome shaving dust. The plausible explanation of the increase in heavy metal concentration may lie in the fact that the dust powder producer uses metal (copper) container for acid treatment of the raw chrome shaving dust. Under hot condition leaching of heavy metals from the container occur which then adsorbed by dust powder. Again the relative higher concentration of lead compared to copper (in spite the fact that the container is a copper-container) can be correlated to the relative solubility of $CuSO_4$ and $PbSO_4$; $CuSO_4$ being more soluble it is washed out whereas lead adheres the solid content.

The concentration of the heavy metals in poultry feed was found to decrease in order Cu < Cr < Ni < Pb < Cd. From in Table 1, it can be noted that the concentration of the heavy metals in poultry feed is diluted with other feed ingredients and thus their concentration is much reduced compared to their concentration in dust powder. The chromium content is diluted to a significant amount from 27267 mg/kg to 85.29 mg/kg. The present finding of Cr content in poultry feed is higher than the previous studies (0.0926 to 5.788 ppm) [1] and 0.98 mg/kg [17]. As an essential micro element, Cr plays an important role in nutrition of animal and human being [18]. But the Cr depositions in the form of Cr (VI) depress hatchability and have also toxic effect on liver [19]. As shown in Table (1), the mean concentration of Cadmium was found in feed is 0.45 mg/kg which is lower than the maximum acceptable limit (1 mg/kg) in feed by European Union [20]. However, Cd concentration found in the present study is lower than 3.8-33.6 mg/kg obtained by Mahesar et al. [21] but comparable to 0.463 mg/kg reported by Okoye et al. [3].

Mean concentration of Pb obtained was 14.36 mg/kg which exceeded the limit 5 mg/kg of Pb in feed stipulated by European Union [20]. However the value obtained in this study is lower than the previous results 23.2–32.6 mg/kg [21], 37.57 mg/kg [1] and higher than 6.52 to 14.20 mg/kg [3] and 10.32 mg/kg [17]. Mean copper concentration was 85.42 mg/kg which is within the maximum acceptable concentration of 100 mg/kg for Cu in feed as stipulated by EU [20].

Amount of nickel obtained was 24.7 mg/kg in feed sample which is higher than 2.250 to 4.875 mg/kg [3], 0.0125 to 5.163 mg/kg [1] and lower than the previous result 11.33 to 64.33 mg/kg [22].

Table 1: Heavy metals concentration in collected shaving dust and poultry feed from Industrial area of Hazaribagh and Gazipur respectively, Bangladesh. (N= 4), no. of samples

Metals	Shaving Dust (mg/kg)				Powder (mg/kg)				Feed (mg/kg)			
	Max.	Min.	Mean	SD	Max.	Min.	Mean	SD	Max.	Min.	Mean	SD
Cd	0.33	0.22	0.28	0.08	1.57	1.42	1.50	0.11	0.48	0.41	0.45	0.05
Pb	18.37	15.24	16.81	2.21	290	281.66	285.83	2.95	16.01	12.70	14.36	2.34
Cu	30.72	28.35	29.54	1.68	146.96	144.95	145.96	1.42	86.65	84.18	85.42	1.75
Ni	9.95	7.72	8.84	1.58	53.06	50.97	52.02	1.48	24.92	24.48	24.7	0.31
Cr	27964	26570	27267	985.71	17863	17841.6	17852.3	15.13	88.34	82.25	85.29	4.31

Fig. 2 : (A) Day old chicks, (B) 10 days old chicken, (C) 42 days old chicken, (D) Different parts of chicken body.

Table 2: Mean Concentration (mg/kg) of heavy metals in different part of chicken after rearing

Metal	Safe limit[a]	Safe limit[b]	Safe limit[c]	Breast				Thigh				Liver				Bone (Leg)			
				Max.	Min.	Mean	SD	Max.	Min.	Mean	SD	Max.	Min.	Mean	SD	Max.	Min.	Mean	SD
Cd	0.05	0.05	0.1	1.34	0.89	1.14	0.32	0.66	0.57	0.63	0.06	2.23	0.45	1.15	1.26	1.02	0.88	0.97	0.10
Pb	0.	0.1	0.2	33.86	28.41	31.2	3.85	18	12.61	15.42	3.81	25.74	5.33	15.68	14.43	55	5.93	25.76	20.68
Cu	1	1	10	16.54	12.29	14.73	3.00	30.06	9.66	19.11	14.42	50.88	33.16	42.21	12.53	36.2	4.2	18.74	22.63
Ni	0.5			7.32	5.05	6.24	1.61	6.65	1.76	3.70	3.46	12.32	3.11	7.41	6.51	3.88	0.39	2.22	2.47
Cr				3.55	1.92	2.78	1.15	2.88	1.1	1.79	1.26	7.6	1.81	4.56	4.09	5.8	1.73	4.08	2.88

[a]FAO/WHO 2002 Codex Alimentarius, Schedule 1 of the proposed draft Codex general standards for contaminants and toxins in food. Joint FAO/WHO Food Standards Programme, Codex Committee, Rotterdam. Reference CX/FAC 02/16.
[b]EC (The Commission of the European Communities) 2006 Commission Regulation No. 1881/2006 of 19 December 2006 setting maximum levels for certain contaminants in foodstuffs. Official Journal of the European Communities, L 364/ 18-19.
[c]MHPRC (Ministry of Health of the People's Republic of China) the limits of pollutants in foods (GB 2762-2005 for Pb and Cd, GB 15199-1994 for Cu) Beijing, China: MHPRC

The heavy metals content in different parts of chicken is shown in Table 2 which largely reflects their concentrations in the feed consumed by the chickens. The current results (Table 2) revealed that the deposition of heavy metals in liver is higher than the other edible parts of chicken. The average concentrations of analyzed metals found to follow the order Cu > Pb > Ni > Cr > Cd.

The maximum amount of Cd accumulation was recorded in liver and the lowest amount of Cd content was found in thigh meat. Cadmium concentrations found in the present study (0.45 - 2.23 mg/kg) in different tissues are higher than the previous results (0.01 to 0.031 mg/kg) [17]; (0.019 to 0.061 mg/kg) [23] and lower than (0.059 to 9.36 mg/kg) [24]. The maximum permissible level of Cd for chicken samples is 0.5 mg/kg according to Saudi Arabia standards. However, the lowest average amount of Cd was found 0.63 mg/kg in thigh in the present study which is also higher than the Saudi Arabia standard. The obtained result is also above the highest permissible hygienic limits for Cd in muscles 0.1 mg/kg and in internal organs 0.5 mg/kg according to Codex 1996, FAO/WHO, 2000. Cadmium has a deadly effect at a very low concentration and responsible for cancer and potential mutation causing [25]. Cadmium accumulation in the human body is responsible for kidney dysfunction, skeletal damage, reproduction deficiencies, prostate cancer, mutations and fetal death [26].

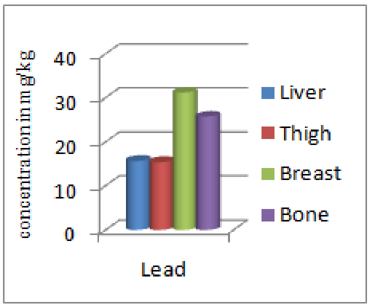
The highest amount of Pb (31.2-33.86 mg/kg) deposition was recorded in breast meat and the lowest (12.61-18.0 mg/kg) was found in thigh meat. Lead concentration in all analyzed parts of chicken contained higher than the permissible limit of FAO/WHO and EC. The current data shows the average concentration of Pb in liver (15.68 mg/kg) and muscle (15.42 mg/kg) which are higher from 0.73 for liver and 0.52 for muscle reported by Zhuang et al. [24]. The amount of Pb found in this study is also higher than (0.498 to 0.749 mg/kg) [17]. On the other hand, mean concentration of Pb and Cd were found in breast meat, liver and thigh are comparatively higher than (0.012, 0.062 and 0.023 mg/kg) and (0.156, 0.215 and 0.152 mg/kg) in breast, liver and thigh for Cd and Pb respectively reported by Abdolgader et al. [27]. The maximum Pb levels stipulated for chicken samples is 1.0 mg/kg according to Saudi Arabia standards but Pb levels in present study in chicken samples were much more higher than the Saudi Arabia standards. Lead is toxic to the blood and the nervous, urinary, gastric and genital system and it is also coated in causing carcinogenesis mutagenesis in experimental animals [28].

Concentrations of Cu varied in different parts of tissues and the highest amount of Cu was found in liver and the lowest amount was observed in breast meat. Assortment of Cu was found in the increasing order Liver > Thigh > Bone > Breast. Cu contents found in tissues were higher than the results found in previous report conducted by Zhuang et al. (2.34 mg/kg for liver and 1.43 mg/kg for muscle) [24] and comparing with standard reference of FAO/WHO and EC, the results are extremely higher than those standards. The copper concentration in liver and breast meat were found 9.67 and 1.55 mg/kg respectively [29] which are also lower than present study (14.73-50.88 mg/kg). Copper is an essential micronutrient for body but very high intakes can cause health problems such as liver and kidney damage. FAO/WHO Expert Committee concluded that no injurious effects can be expected in humans whose copper intake is 0.5 mg/kg body weight per day (FAO/WHO, 1971). The Recommended Dietary Allowance (RDA) for adult men and women is 900 µg/day and the median intake of copper from food in the United States is approximately 1.0 to 1.6 mg/day for adult men and women [30].

Ni concentration was high in liver (7.41 mg/kg) followed by breast meat (6.24 mg/kg), thigh meat (3.70 mg/kg) and bone (2.22 mg/kg). Mean concentration of nickel in present research were higher than the previous study (1.909 mg/kg for liver, 1.19 mg/kg for breast meat) [29]. Current result was also higher than the concentration found in Bangladeshi [22] and in Indonesian chicken [31]. According to WHO standard, the permissible limit of Ni in food is 0.5 mg/kg. The nickel concentration in all analyzed organ of chicken were extremely higher than the permissible limit. According to the New York University School of medicine, chronic exposure of nickel increases risk of lung cancer, cardiovascular disease, neurological deficits and developmental deficits in childhood and high blood pressure [32].

Regarding Cr accumulation in chicken organs, highest amount of Cr was found in liver (1.81-7.6 mg/kg) and minimum amount was found in thigh meat (1.10-2.88 mg/kg). The average concentrations of Cr in breast meat and in bone were found 3.17 and 3.76 mg/kg respectively. The Cr concentration in the current research were higher than the previous studies where Cr concentrations in meat, bone and liver were found 0.35 mg/kg, 1.99 mg/kg and 0.61 mg/kg respectively [16] and 0.24 to 0.72 for meat and 0.16 to 1.26 for liver obtained by Bari et al. [17]. According to the International Program on Chemical Safety (IPCS), chromium concentrations in chicken are in the range of 10-60 µg/kg [33]. Cr III is a micro-nutrient and required in a very small amount for normal growth and development of animal and human but Cr VI is carcinogenic according to many research. The present study shows the total amount of Cr in different parts of chicken where Cr III and Cr VI might be present. Comparison of heavy metals (Cd, Pb, Cu, Ni and Cr) deposited in liver in the present study is comparatively higher than previous studies might be the reason of having tannery waste contaminated feed.

In addition, the accumulation of Cd and Cr in different parts followed the order Liver > Breast > Bone > Thigh and Liver > Bone > Breast > Thigh. The Pb and Ni followed the order of assortment Breast > Bone > Liver > Thigh and Liver > Breast > Thigh > Bone respectively (Chart 1, 2, 4, 5). The heavy metals concentration in present study is too much higher than the safe limits of Cd, Pb and Cu proposed by FAO/WHO, EC and China showing in Table 2. Prolonged intake of these metals contaminated meat can have adverse effect on human body.



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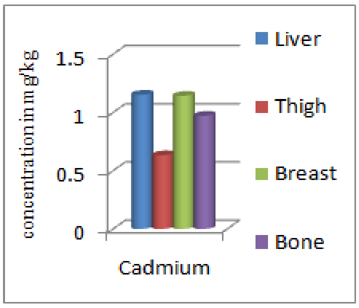

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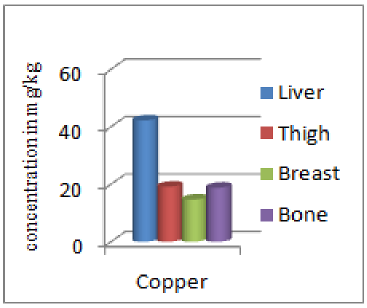

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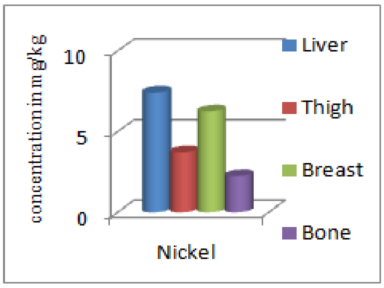

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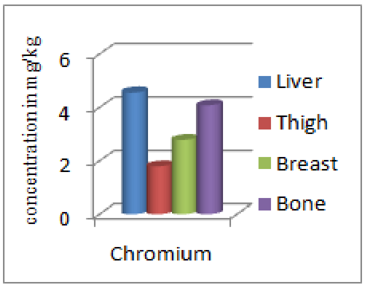

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The chart of metals shows that soft part of chicken body deposits more metal comparing other parts. Liver contained highest amount of metals than bone, breast and thigh.

The transfer factor (TF) of metals from feed to different parts of chicken was calculated as the ratio of the metals content in tissues to that in the feed. The TF range of metals for various parts of chicken was 1.4 to 2.55 for Cd, 1.07 to 2.17 for Pb, 0.17 to 0.49 for Cu, 0.09 to 0.3 for Ni and 0.02 to 0.05 for Cr. The TF values of Cd and Pb was higher than 1 which indicates that accumulation of these metals in tissues of chicken was very high. On the hand, the TF value of Cr is very low. Metal accumulation in analyzed tissues of chicken was in the following order: Cd > Pb > Cu > Ni > Cr.

V. Conclusion

Worldwide food safety is an important issue and has become challenging to maintain it in Bangladesh. It is an important issue which needs preventive measures throughout the food chain from farm to table. Concentrations of all metals studied in different tissues of chicken not only exceeded the limit recommended by FAO/WHO but also too much higher comparing with previous studies because of using contaminated poultry feed. Chicken meat being one of the most popular and affordable protein source, people of Bangladesh takes it regularly. Through contaminated chicken meat the toxic metals can easily be entered into food chain and ultimately entered into the human body. The excess amount of these heavy metals may have carcinogenic effect on human body and can bring a serious health hazard. To maintain the safety of food chain and to minimize the heavy metals contamination, it is mandatory for the feed producers to always observe and maintain standards for heavy metals in chicken feeds. Feed companies should carry out heavy metal assessment in their feed products to keep them at a safe limit.

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