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ASSESSMENT OF HEAVY METALS IN BROILER CHICKENS OF DIFFERENT FARMS AND THEIR SOURCE OF TRANSMISSION IN DISTRICT PESHAWAR

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Abstract

Background: Assessments of heavy metals in the food supply are crucial. Heavy metal contamination is a major environmental hazard that may negatively impact organisms above their threshold levels.

Methods: 45 samples were collected from three areas: A (Naguman), B (Badhaber), and C (Phandu), which included 27 broilers, 9 feed and 9 water samples. Atomic absorption spectrometry (AAS) was used for heavy metal detection.

Results: The result shows that feed of area C was reported to have significantly ($P<0.05$) higher Cd concentration, while Cr was significantly ($P<0.05$) reported in feed of area B, and Pb was significantly ($P<0.05$) reported in feed of area B. Further, higher concentrations of Ni were detected in feed from area C as compared to feed from areas B and A. There was no significant ($P = 0.40$) difference in Cd concentration in water in areas A, B, and C. Cr and Ni were not detected in all areas. However, significantly ($p<0.05$) higher concentrations of Pb were reported in the water of area A. The broiler meat analysis of heavy metals revealed that significantly ($P<0.05$) higher concentrations of Cd were reported in meat from area A. There was no significant difference in concentrations of Cr, Pb, and Ni in broiler meat from distinct areas.

Conclusion: To summarize farm water was the source of heavy metals in broiler chickens in area A, while feed was the source of heavy metals in broiler chickens in areas B and C.

Key words: Broiler chicken, heavy metals, wet-acid digestion, atomic absorption spectrometry

Introduction

The consumption of meat products has expanded along with the growth of the global population. World Watch Institute reports that the worldwide meat consumption has climbed threefold over the past forty years and by merely 20% in the past ten years, which is much greater than the increase in population. Meat and meat products are widely utilized all over the world to meet the protein needs for healthy growth, repair, and tissue creation. They are a significant source of minerals, amino acids,

and proteins. However, due to the direct threats to public health that are connected to it, food security is now seen as a significant worldwide concern [1]. Every year, more than 50 billion chickens are grown for food, including for their meat and eggs. Layers refer to a chicken produced for eggs, whereas broilers refer to a chicken reared for meat [2]. To address the demand for animal protein during the past 20 years, numerous emerging nations have shifted to extensive poultry farming. Nutritional supplementation of poultry diets is also necessary to improve feed quality, weight gain, and disease prevention, which raises the concentration of trace elements (including heavy metals). However, it has been noted that the presence of heavy metals in chicken diets, whether they are produced locally or naturally, can impact their quality [3]. The word heavy metals are said to be those elements or compounds (metals / metalloids) having more than $5gcm^{-3}$ density and atomic number higher than 20, are excessively harmful and toxic to the environment [4]. These are metals with a density greater than five times that of water [5] . Cadmium (Cd), Nickel (Ni), Arsenic (As), Iron (Fe), Lead (Pb) and Mercury (Hg), are the poisonous heavy metals that are most commonly used in products. Poultry feed, drinking water, and processing are some of the numerous ways that heavy metals can contaminate chicken meat. Some minerals are supplied to poultry feed as mineral supplements, but when their concentration exceeds the advised limit, they can build up in chicken tissues and release into the litter, which in turn pollutes the environment [6]. According to the Agency for Toxic Substances and Disease Registry (ATSDR), lead is ranked number two on the "Top 20 List." Most pediatric heavy metal poisoning cases are caused by lead. It produces acute and long-term toxicity, dysfunctions in physiology, biochemistry, and behavior in people, terrestrial and aquatic organisms [7]. Cadmium is at number seven on the ATSDR's list. Chromium Cr (VI) has been ranked as 17th most toxic substance (ATSDR, 2013). Heavy metal long-term exposure above their threshold values have adverse consequences on human body leading to both non-carcinogenic and carcinogenic dangers [8] . Therefore, the current study urges to carryout heavy metals assessments in broiler meat, feed and water source.

Ethical approval

The study was conducted after receiving the approval from the Ethics Review Committee of the SBBWU. Furthermore, the samples were obtained after poultry farms owner's permission.

Site selection

The study was conducted in District Peshawar. Three different areas A (Naguman), B (Badhaber), and C (Phandu) were selected for heavy metals assessment. A total of nine broiler farms were selected, three farms from each locality.

Sample collection

A total of 45 samples were collected from each locality. All the samples were collected in triplicates. 9 samples of water (1 liter) were collected from area A, B and C in polypropylene clean bottles. The collected samples were filtered and then preserved in order to prevent adsorption of heavy metals on the inner side of container by adding 5ml of 55% HNO3 per liter of water and stored at 4 ˚C before further analysis. Additionally, 9 samples of feed were also collected in clean air tight polythene bags in order to avoid moisture by following the American Public Health Association protocol (APHA 1995). 27 healthy, diseased free broiler chicken of same age (40 days old) were collected from selected farms. After collection, they were slaughtered and internal organs (liver, gizzard, heart, intestine, lungs) were removed. Then they were washed with clean water and preserved at low temperature (4˚C) in lab until further procedure.

Sample preparation for metal estimation

Water sample

The acidified 100ml water samples were reduced to 20ml in a volumetric flask on a hotplate. By following the approach [9]. The solutions were then sent to Central Resource Lab and analyzed through AAS (Spectra-AA-700) for heavy metals determination

Meat sample

Wet-acid digestion method was used in order to complete dissolution of the samples [10]. The samples were then sent to Central Resource Lab for heavy metals analysis.

Feed sample

The feed samples were first weighted with the help of digital balance and 1g feed was grinded using pestle and mortar, to make it fine powdered. The powdered sample was then shifted to flask and the above mentioned procedure was followed.

Heavy metals detection

Chromium, nickel, cadmium and lead concentration were examined in the samples by applying the standard methods and Atomic Absorption Spectrophotometer (AAS).

Statistical Analysis

Statistical analysis was performed using software IBM-SPSS Statistics 22. One-way analysis of variance (ANOVA) was used to evaluate whether heavy metals varied significantly ($P<0.05$) and was presented as mean and standard deviation (mean ± S.D.).

RESULTS

The study was conducted to find out the concentration of heavy metals (Cd, Cr, Pb and Ni) in broiler's meat, feed and water of different poultry farms in district Peshawar as a result of bioaccumulation. For this purpose, samples were collected from different sites/areas and were digested by the process of wet-acid digestion process. The following results were obtained from atomic absorption spectrometry, which uses different wavelength for analysis of heavy metals (Figure 1).

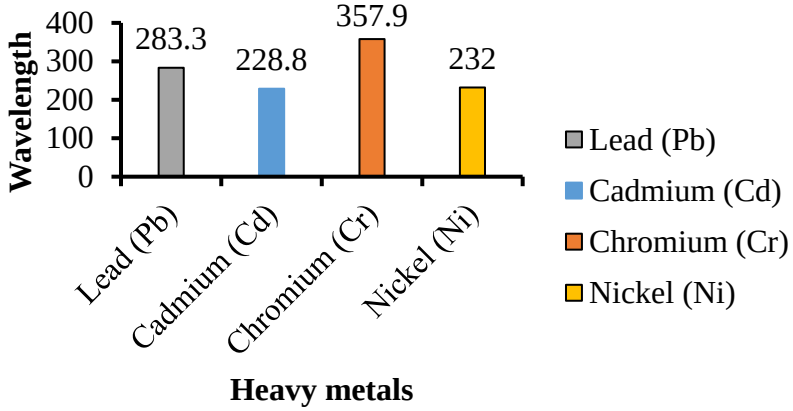

Figure 1: Wavelength of different heavy metals

Heavy Metals concentration in Feed

Our results indicated a significantly higher concentration of heavy metals in different feeds. The Arabic numerals (1-2-3) represents different farms. Feed-3 was reported to have significantly higher ($p<0.05$) Cd (0.081±0.002) and Ni (0.277±0.068) concentration as compared to feed-2 (0.058±0.002) and feed-1 (0.035±0.009). Correspondingly, Feed 2 shows a significantly higher Cr (0.49±0.00) concentration in comparison to feed 3 and feed 1. Additionally, feed 2 also reported significantly higher ($p<0.05$) Pb concentration (4.89±0.00) followed by feed 3 and feed-1 (Table 1).

Table 1: Concentration of heavy metals in feed samples at broiler farm in district Peshawar

Heavy Metals	Feed			Maximum Permissible limit(ppm) WHO/FAO	P-value
	1	2	3		
	Mean ± Standard Deviation				
Cd	0.035^{c}±0.009	0.058^{b}±0.002	0.081^{a}±0.002	0.5	<0.05
Cr	0.000^{b}±0.000	0.049^{a}±0.000	0.000^{b}±0.000	0.5	<0.05
Pb	0.000^{c}±0.000	4.890^{a}±0.000	0.245^{b}±0.065	0.5	<0.05
Ni	0.032^{b}±0.002	0.038^{b}±0.014	0.277^{a}±0.068	0.5	<0.05

Note: Means within same column having different lowercase letters differ significantly ($p<0.05$)

Heavy Metals Concentration in Water

It has been indicated that water has no significant effect ($p= 0.40$) on prevalence of Cd. There was no significant difference of Cd concentration among water-1 (0.045±0.005), water-2 (0.0413±0.005) and water-3 (0.044±0.0017). Similarly, Cr (0.00±0.00) and Ni (0.00±0.00) detected in all the water samples collected from different farms. However, Water-1 (0.016±0.005) was reported to have significantly higher ($p<0.05$) Pb concentration as compared to water-2 (0.00±0.00) and water-3 (0.00±0.00). Table 2 shows heavy metals concentration in water samples.

Table 2: Heavy metals concentration in water samples of different farms of district Peshawar

Heavy metals	Water			Maximum permissible limit(ppm) WHO/FAO	P-value
	1	2	3		
	Mean ±Standard Deviation				
Cd	0.045±0.005	0.0413±0.0015	0.044±00173	0.05	0.40
Cr	0.000 ±0.000	0.000±0.000	0.000±0.000	0.05	0.30
Pb	0.017^{a}±0.006	0.000±0.000^{b}	0.000±0.000^{b}	0.05	<0.05
Ni	0.000±0.000	0.000±0.000	0.000±0.000	0.05	0.20

Means within same column having different lowercase letters differ significantly ($p<0.0$)

Heavy Metals concentration in Broilers Meat

Our results indicated that Meat 1 was reported to have significantly higher ($p<0.05$) Cd (0.0417±0.245) concentration as compared to Meat-2 and Meat-3. There was no significant difference ($p=0.14$), reported in concentrations of Cr, Pb and Ni in meat-1, meat-2, and meat-3. The heavy metal concentration is illustrated in details in table 3.

Table 3: Heavy Metals concentration in Broilers meat in different farms of district Peshawar

Heavy Metals	Meat			Maximum Permissible Limit WHO/FAO	P-value
	1	2	3		
	Mean ±Standard Deviation				
Cd	0.0417^{a}±0.2454	0.057^{a}±0.00379	0.002^{b}±0.00346	0.5	<0.05
Cr	0.0307±0.2517	0.002±0.00346	0.000±0.000	0.5	0.075
Pb	0.5433±0.4865	0.04±0.0346	0.1333±0.0057	0.5	0.145
Ni	0.127±0.08750	0.0647±0.0398	0.0363±0.2601	0.5	0.222

Note: Means within same column having different lowercase letters differ significantly ($p<0.05$)

Sources of Heavy Metals in different Areas

It has been shown that water is the major source of Cd in farm A. However, Cr was not detected in samples of feed and water although meat samples have a little amount of it, which may possibly come from the surrounding environment. Pb was not detected in feed samples although found in water and meat samples. Ni was found in feed samples but not detected in water; therefore, the source for this particular heavy metal is feed. From these results we can predict that the source of heavy metals in farm A broilers chicken could possibly be the water. Additionally, the source of Cd in farm B was feed, Cr was not detected in samples of water while found in feed and meat samples, which indicated the feed may be the possible reason of it. Pb was not detected in water samples although found in feed and meat samples, therefore we can say that it come from the feed. Ni was found in feed samples but not detected in water; therefore, we can say that it comes from feed samples. From these results we can predict that the source of heavy metals in farm B broilers chicken could possibly be the feed provided to them. The concentration of heavy metals in different sources of farm C was also detected. It was concluded that the source of Cd in farm C could be the feed. Cr was not detected in samples of feed, water and meat samples. Pb was not detected in water samples although found in water and meat samples. Ni was found in feed samples but not detected in water. From these results we can predict that the source of heavy metals in farm C broilers chicken could possibly be the feed.

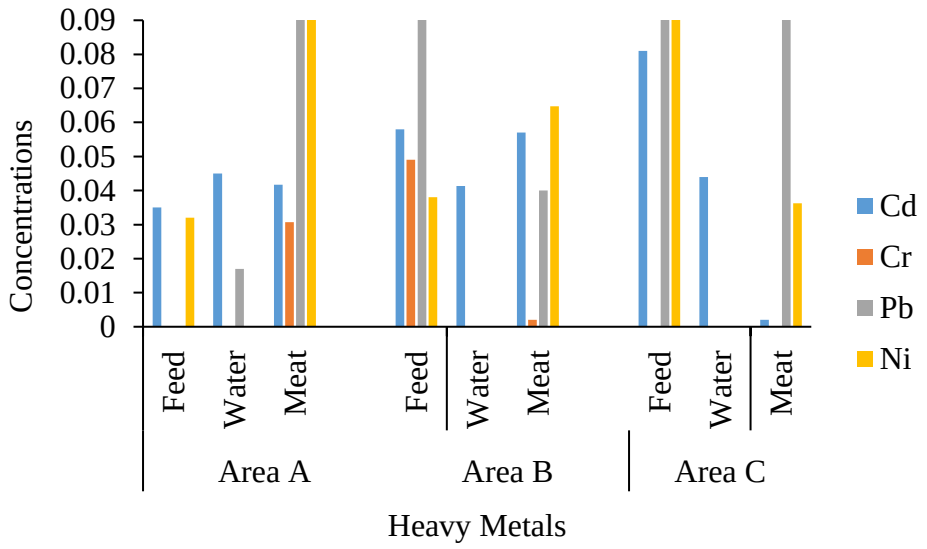

Figure 2: Sources of Heavy metals in feed, water and meat in different areas

Discussion

The global food supply depends heavily on poultry products. Animal husbandry is constantly striving to generate highest quality protein, to satisfy the world's food demand; however, it also confronts a distinctive set of issues, including environmental pollution, political turmoil, animal welfare, and industrial
mechanization and robotization [11]. Heavy metals impose a greater threat to feed industry as a result of bioaccumulation in tissues and various organs of chickens. The current study focusses on the identification of concentration of (Cadmium, Chromium, Lead and Nickel) in feed of broiler chickens, in the water, and meat and their sources.

The results revealed that the concentration of Cd in feed in our hands is lower than the previously reported ranges obtained at Hyderabad Sindh [12]. While the Pb concentration (0.313-0.49ppm) recorded was approximately in range of present obtained results. Cd concentration obtained by [13] is also higher than the present obtained data.

The current study's lower range of Cd and Pb were in accordance to the study of [14] who reported (0.52-0.06ppm) and (1.17-0.071ppm) for Cd and Pb respectively. The Pb concentration (4.9ppm) in our study is similar to the Pb ranges (3.54- 3.7ppm) obtained by [6]. However, the Cd, Ni and Cr

ranges were not in agreement to the obtained data that might be due to the difference in areas, feed type and other environmental factors.

In feed of area A, the concentration of Cd, Cr, Pb and Ni lie under the prescribed WHO permissible range of heavy metals indicating that the feed is safe for use. In feed of area B, the concentration of Cd, Cr and Ni lie under the WHO prescribed permissible range of heavy metals while containing high amount of Pb indicating that the feed is not safe or unfit for use. In feed of area C, the concentration of Cd, Cr and Ni is in permissible range while the Pb concentration is near to the limit value indicating a threat that feed is unfit

The concentration of Cd in water samples range from 0.040-0.045ppm while the results obtained by [6] was 2.62-3.87ppm which is higher than the findings of the present study. There is no Cr and Ni found in present water samples while the results of obtained in above study mentioned shows Ni 12.37-27.00ppm. The Cd, Cr, Pb and Ni concentrations in water sample in our study was dissimilar to the concentrations recorded for these heavy metals by [9]. Correspondingly, [15] detected the concentrations of Cr, Cd and Pb (0.007mg/l, 0.00, 0.00) accordingly which is nearly similar to the present data. While, our study's results are contrary to the findings of [16]. This shows that the difference in results may be due to the different location, water supply source or any other environmental factors.

The concentration of Cd in water samples of all areas was significantly higher in comparison to other metals. The order of heavy metals concentration was found to be Cd>Pb> Cr> Ni. In area A, B and C the concentration of heavy metals in water samples lies within the WHO prescribed permissible limits, therefore indicating that the water supplied to this area is safe.

The concentration of Cd in broiler chicken meat samples ranges from 0.00 to 0.06. Cr concentration ranges between 0.00-0.03. The concentration of Pb ranges between 0.04-0.5 while the concentration of Ni ranges from 0.04 to 0.12. which contradicted the previous study [17]. In Nigeria [18] obtained the following results: Cd 0.03-0.019, Pb 0.00ppm. The concentration of Cd range is similar with recent results while Pb concentration range is not similar. In Bangladesh [19] obtained the following concentrations: Cd 5.20, Cr 0.69, Pb 41.94 which were higher than the results of the present study. The results obtained in a previous study carried out by [6] in district Charsadda KP were Pb 0.17ppm, Cd 0.016, Ni 0.38, which is not similar with the present obtained data. In Karachi the study reported: Pb 1.79ppm Cd 0.366 Ni 4.13ppm [20], shows that this data is higher than the findings of the recent study. The concentrations of all heavy metals in broiler meat lies under the WHO prescribed permissible limits. Therefore, the broiler meat is safe for consumption.

Conclusion

It was concluded that the concentration of Cd, Cr, Pb and Ni was found to be high in feed respectively. Further, the concentrations of all heavy metals was found to be in normal range in farm water except the water of area A, indicating that the farms might be constructed in industrial or irrigated zone. Therefore, avoid making farms in these areas or make healthy water supplies free from any kind of toxic elements. The concentration of all heavy metals in broiler meat was found under permissible levels suggesting that the chicken meat is safe for usage. From the results it was concluded that the main source of heavy metals in area A farms was water while in area B and C, feed was considered to be the main source of toxic elements. Therefore, care must be taken by poultry farmers in selection and processing of poultry feed in order to maintain its nutritional values for animals. Government should not allow farms constructions in industrial and polluted zones to control water quality. Government incentives must be taken into consideration to control environmental pollution in district Peshawar.

Authorship credit

Iram Maqsood, Muneeza Sher: Concept and design of study

Muneeza Sher: Conduction of Experiment and Acquisition of data.

Aishma Khatak, Muhammad Mohsin Ahsan: Analysis and interpretation of data

Rabea Ejaz, Asima Azam, Ayesha Hidayat: Drafting the article or revising it critically for intellectual content

All authors approved the Final version of the article to be published.

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