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Research Article

Assessing Levels of Heavy Metals in Broiler Chickens Reared in Crude Oil Exploration Areas of Delta State, Nigeria

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ABSTRACT

Introduction: Heavy metals (HM) commonly contaminate water, food materials, and the environment in industrial and oil exploration areas, exposing humans and animals to health risks. The present study aimed to assess the levels of HM, such as Cadmium (Cd), Iron (Fe), Nickel (Ni), Vanadium (V), and Mercury (Hg), in the meat of broiler chickens raised in areas impacted by crude oil exploration in Delta State, Nigeria.
Materials and methods: A total of 75 broiler chickens of mixed sex and breed were randomly selected from 15 intensively managed poultry farms. Three farms were chosen from each of the five zones, including Urhobo, Isoko, Ijaw, Itsekiri, and Ukwani, with five chickens selected from each farm. Blood samples were taken from the chickens for haematological and serum biochemical analysis. The concentration of heavy metals (ppm) in the breast meat of the broiler chickens was determined using an Atomic Absorption Spectrophotometer. The findings were compared to the acceptable limits of international standards and the World Health Organization (WHO).
Results: The present study's findings revealed that the HM concentration in broiler meat was significantly higher than the WHO's tolerable limits. No significant variations were recorded in Mercury concentration in the broiler chickens across all the zones. The Packed Cell Volume (PCV) for chicken from Ijaw zone was significantly higher (48.33), followed by chicken from Urhobo zone (44.78), both were significantly higher than the standard values for a normal healthy chicken, which ranged from 31.50 to 36.70, and the least PCV value was observed in Itsekiri zone (35.11) compared to other zones. Haemoglobin values were highest in the Isoko zone (16.96) and least in the Urhobo zone (12.25). The Red Blood Cell (RBC), Mean Corpuscular Hemoglobin (MCH), and Mean Corpuscular Volume (MCV) values from all the groups in this study were significantly lower than the standard range of values for normal chickens, except for the MCH value of 34.60 in Isoko.
Conclusion: Crude oil exploration activities significantly influenced the heavy metal concentration in broiler meat and certain haematological indices of chicken, and thus, chicken exposed to heavy metal contamination could be a risk factor for consumers.

1. Introduction

In Nigeria, humans and animals are exposed to heavy metal contamination from different sources, such as water, feeds, food, and oil exploration operations[1]. Environmental and animal contamination by heavy metals due to crude oil exploration in the Niger Delta area has raised concerns about the potential health risks associated with prolonged exposure[2]. The harmful effects of these heavy metals on chickens include low feed intake, low hatchability, low digestibility, retarded growth, weight loss, and liver and kidney damage[3]. In humans, heavy metal toxicity can lead to central nervous system defects, liver and kidney damage, depression, cancer, respiratory distress, and in extreme cases, death[4].

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However, livestock production remains crucial for providing nutrition, supporting the economy, and meeting the protein needs of the population through the production of eggs, milk, and meat[5]. Poultry meat is a valuable source of animal protein, containing all the essential amino acids, minerals, and vitamins needed for human nutrition[6]. Nevertheless, the meat can become contaminated if the chickens consume water contaminated with heavy metals. In such cases, consumers who eat this meat are at risk of heavy metal poisoning, which is a serious threat to their health and well-being.

Concerns about food safety and public health have prompted research on the assessment of levels of contaminants, including heavy metals, in poultry products[7]. Furthermore, studies have shown widespread contamination of drinking water, food crops, livestock, and the entire food chain with chemicals used in the petrochemical industry[8]. It is important to evaluate poultry meat for heavy metals such as Nickel (Ni), Cadmium (Cd), Mercury (Hg), Iron (Fe), and Vanadium (V), especially in industrialized areas, due to their functions in the body and their potential toxicity to consumers. However, there is a paucity of information regarding heavy metal concentrations in poultry. Therefore, the present study aimed to evaluate the effect of heavy metal contamination on the well-being of broiler chickens and their deposits in the meat, including the potential implications on the consumers' health.

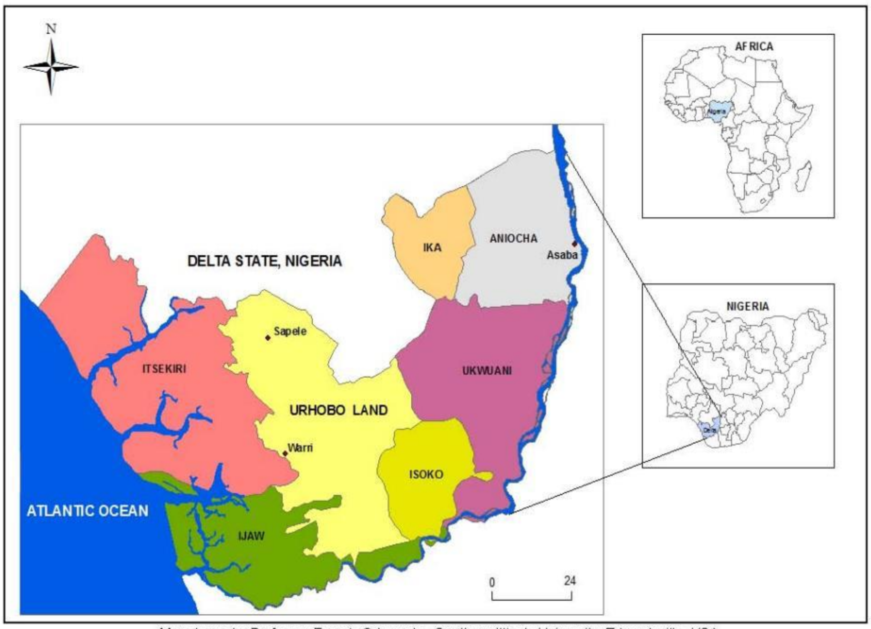
2. Materials and Methods

2.1. Ethical approval

The animals used for the present study were treated and handled humanely, and all procedures were carried out following the guidelines approved by the Animal Welfare and Ethics Committee of the University of Ibadan, Nigeria (AWE2018/0013478).

2.2. Study area and location

Fifteen Local Government Areas (LGA) in Delta State were purposively selected based on the presence of crude oil exploration, extraction, and refining companies. Additional criteria for selection included a history of gas flaring, the presence of crude oil pipelines, and incidents of crude oil spillage. The selected areas were chosen from the five zones, including Urhobo, Isoko, Ijaw, Itsekiri, and Ukwani (Figure 1). In each zone, three intensively managed poultry farms were sampled for the present study following the approval of the farm managers.



Map drawn by Professor Francis Odemerho, Southern Illinois University, Edwardsville, USA

Figure 1. The map of the Delta State showing the study areas, Nigeria

2.3. Sample size and collection

A total of 75 broiler chickens of mixed sex and eight weeks old, weighing 3.00 ± 0.71 kg, were randomly sampled from 15 intensively managed poultry farms in five zones. Five chickens were randomly selected from three farms in each of the selected zones of Delta State, Nigeria.

2.3.1. Blood sampling

Blood samples (five from each farm) were collected in the morning from the large veins (2 mL) of the chicken wing using a 5 mL sterile syringe and needle. The samples were collected into ethylenediaminetetraacetic acid bottles for haematology and plain bottles for serum analysis. The haematology samples were used for the determination of red blood cells (RBC), white blood cells (WBC), packed cell volume (PCV), mean corpuscular hemoglobin (MCH), mean corpuscular volume (MCV), mean corpuscular haemoglobin count

(MCHC), and leukocyte count. Serum parameters such as Albumin, Alkaline phosphatase (ALP), Alanine aminotransferase (ALT), Aspartate aminotransferase (AST), and uric acid were also determined according to the process described by Ewuola and Egbunike[9].

2.3.2. Meat sampling

The selected chickens were first stunned by neck dislocation to render them unconscious; thereafter, they were bled by severing the jugular vein with a sharp knife and allowing the blood to drain. The chickens were de-feathered, eviscerated, and dressed. The broiler chicken breast cuts were obtained from the five chickens per farm and transported in an ice box to the laboratory for determination of heavy metal concentration (HMC, ppm). The determination was done following standard analytical procedures, AOAC[10], and using an atomic absorption spectrophotometer (AAS) as per USEPA[11] guidelines.

2.4. Sample preparation

Meat samples were oven-dried at 65°C until constant weights were achieved. The dried samples were then crushed and pulverized using a ceramic mortar and pestle. Approximately 10 grams of the ground meat samples were placed in polyethylene vials and then placed in envelopes before being stored in air-tight containers[12].

2.5. Heavy metals determination

The meat samples were chemically analyzed for heavy metals, including Ni, Hg, V, Fe, and Cd. The concentration of heavy metals in the digested samples was determined using an AAS. To prepare the standards, a 10% aliquot of a newly made standard solution (1000 ppm) of each element was added onto Whatman's filter paper. One g of pulverized meat samples was weighed into beaker containers and 10 mL of a 5:1 ratio of HNO_3 and $HClO_4$ solutions was added. 2 mL of H_2O_2 acid was further added, followed by the introduction of 5 mL of a 1:1 ratio of distilled water and HNO_3. The beaker was heated and allowed to boil slowly in a fume cupboard until the mixture was properly digested. After the solution reached room temperature, it was transferred into a 100 mL flask through filter paper and filled to the mark with distilled water. The digestion of meat samples followed the AOAC[10] guidelines. The samples digested were subjected to analysis for heavy metal concentrations using the spectrophotometry model ACCUSYS-211, BUCK Scientific, in an air-acetylene flame. Starting with the blank then followed by the samples to evaluate the metal ions of V, Fe, Ni, Cd, and Hg in the chicken meat samples.

The absorbance of the test solution was compared against the standard of each metal ion, and the level of heavy metal residues from the samples was calculated by interpolation, using standard curves plotted by a computer program. The absorbance obtained was thereafter input to calculate the level of heavy metal residues in each sample. Thus, the slope was the absorbance against concentration. The metal concentration values were compared with the WHO tolerable limits for meat samples.

2.6. Statistical analysis

Analysis of the data was carried out using ANOVA, while means were separated using Fisher's Least Significant Difference (LSD) at 0.05 significance in SAS version 9[13].

3. Results and Discussion

3.1. Heavy metal in broiler chicken meat

Heavy metal concentrations (ppm) in broiler chicken meat samples in different zones with crude oil exploration activities within Delta State are presented in Table 1. There were significant variations in the HMC from one location to another ($p < 0.05$). The concentrations of V in broiler chicken samples ranged from 0.07 ppm (Ukwani) to 0.48 ppm (Isoko), and Cd levels ranged from 0.12 ppm (Ukwani) to 1.49 ppm (Urhobo) in broiler chickens. This exceeded the international tolerable limits[14]. The cadmium concentrations detected in this study aligned with the average concentrations stipulated by the international standard[15] for beef (1.90 µg/kg), mutton (1.22 µg/kg)[16], and pork (1.68 µg/kg). The cadmium levels in this study were above the permissible limit of 0.05mg/kg for poultry recommended by the European Commission (EC) Number 1881/2006[17].

Table 1. Heavy metal concentrations (ppm) in meat samples of broiler chickens reared in oil exploration areas of Delta State, Nigeria

Parameters	Urhobo	Isoko	Ijaw	Itsekiri	Ukwani	SD	P. Limits
Vanadium (V)	0.23^{ab}	0.48^{a}	0.20^{ab}	0.18^{ab}	0.07^{b}	0.033	0.009-0.34
Cadmium (Cd)	1.49^{a}	0.23^{b}	0.17^{b}	0.19^{b}	0.12^{c}	0.109	0.00-0.005
Iron (Fe)	1.61	1.75	1.68	1.43	1.46	0.06	0.30
Nickel (Ni)	0.003	0.002	0.002	0.001	0.001	0.0002	0.00-1.12
Mercury (Hg)	0.34^{a}	0.23^{ab}	0.17^{ab}	0.11^{b}	0.12^{b}	0.023	1.00-2.68

[abc] means that within the same row, with different superscript letters, are significantly different ($P < 0.05$). P. Limits: Permissible limits, SD: Standard deviation

Residues of Fe in the broiler meat ranged from 1.43 ppm (Itsekiri) to 1.75 ppm (Isoko), all of which were higher than the WHO[14] recommended tolerable limits. High levels of Fe in animal tissues can increase the risk of myocardial infarction[18]. The concentration of Nickel in broiler chicken meat ranged from 0.001 (Itsekiri and Ukwani) to

0.003 mg/kg (Urhobo). Similarly, Surtipanti et al.[19] reported traces of Ni in meats, gizzard, and liver from chickens raised in Indonesia. Nickel is known to cause respiratory challenges and is classified as carcinogenic. The tolerable range of Nickel in food is 0.5 mg/kg, as recommended by the WHO[20] and the USSR standards.

The concentration of Hg in broiler chicken meat ranged from 0.11 ppm (Itsekiri) to 0.34 ppm (Urhobo). These values were lower than the internationally recommended safe limits. This finding aligned with the findings of Pribilincov et al.[21], Marettova et al.[22], and Cabanero et al.[23], who detected low Hg concentrations in blood tissues, organs, and chicken muscle when supplied with a mercury-fortified diet. Although these values were not higher than the international tolerable limits[14], the presence of these heavy metals in chicken meat could be attributed to the intense crude oil exploration activities and their subsequent transmission into the poultry value chain through the environment, affecting the soil, air, and water.

The low levels of heavy metals detected in broiler chicken meat from Ukwani zone indicated no adverse effects on broiler chickens and subsequently on the final consumer.

3.2. Haematological indices in broiler chickens

Table 2 shows the haematological indices of chickens from different zones with crude oil exploration activities. The PCV of broiler chickens ranged from 35.11 % (Itsekiri) to 48.33 % (Ijaw), with values obtained from Urhobo (44.78 %) and Ijaw (48.33 %) being significantly higher ($p < 0.05$) than the range of values for a normal chicken, which ranges from 31.5 to 36.70 %. This could be attributed to dehydration. Haemoglobin (Hb) concentration ranged from 12.25 g/dL (Urhobo) to 16.96 g/dL (Isoko), which was significantly higher ($p < 0.05$) than the range of values for a normal chicken (7.40 to 12.20 g/dL). Hemoglobin concentration indicates the amount of oxygen present in the blood. The increased concentration observed in this study could be due to dehydration resulting from poor water intake and chronic pulmonary disease. Red blood cell counts ranged from 2.93 g/dL (Itsekiri) to 5.63 g/dL (Ijaw) and were significantly lower ($p < 0.05$) than the range of values for a normal chicken (10.30 to 12.90 g/dL). Abnormal RBC values indicate an anaemic condition[24].

Table 2. Haematological indices of broiler chickens reared in oil exploration areas of Delta state, Nigeria

Parameters	Urhobo	Isoko	Ijaw	Itsekiri	Ukwani	SD	Standard values
PCV (%)	44.78^{b}	37.22^{c}	48.33^{a}	35.11^{c}	35.33^{c}	0.55	31.5-36.70
Hb (g/dL)	12.25^{c}	16.96^{a}	14.02^{b}	13.83^{b}	12.33^{c}	1.315	7.40-12.20
RBC (x10^{6}ul)	5.33^{a}	3.84^{b}	5.63^{a}	2.93^{c}	4.49^{b}	0.06	10.30-12.90
Platelets (x10^{3}mm^{3})	25.11^{a}	19.89^{b}	26.33^{a}	18.22^{bc}	14.67^{c}	14.58	15.6-28.6
WBC (x10^{3}ul)	8.62^{b}	7.67^{b}	8.99^{b}	7.50^{b}	14.94^{a}	0.10	4.90-9.70
MCH (%)	28.29^{c}	34.68^{a}	29.83^{bc}	30.16^{b}	26.57^{d}	0.24	31.9-40.7
MCHC (%)	34.79^{a}	34.18^{b}	34.84^{a}	34.37^{ab}	33.73^{c}	46.77	25.9-33.9
MCV (fl)	84.49^{c}	95.31^{a}	89.72^{b}	85.35^{c}	81.66^{d}	0.75	102.0-129.0

^{abc} means that within the same row, with different superscript letters, are significantly different ($P < 0.05$). SD: Standard deviation, PCV: Packed Cell Volume, Hb: Haemoglobin, RBC: Red Blood Cell, WBC: White Blood Cell, MCH: Mean Cell Haemoglobin, MCV: Mean Corpuscular Volume, MCHC: Mean Cell Haemoglobin Count.

The platelet counts of broiler chicken from Urhobo (25.11 x 10^{3}mm^{3}) and Ijaw (26.33 x 10^{3}mm^{3}) were significantly higher ($p < 0.05$) than those of Isoko (19.89 x 10^{3}mm^{3}), Itsekiri (18.22 x 10^{3}mm^{3}), and Ukwani (14.67 x 10^{3}mm^{3}). The platelet count of broiler chickens from Ukwani (14.67 x 10^{3}mm^{3}) was significantly lower ($p < 0.05$) than all the others. The WBC count of broiler chickens ranged from 7.50 x 10^{3}ul (Itsekiri) to 14.58 x 10^{3}ul (Ukwani), with Ukwani (14.58 x 10^{3}ul) being significantly higher ($p < 0.05$) than the range of values for normal chickens (4.90 to 9.70 x 10^{3}ul). High levels of WBC indicated a response to a stressful condition, resulting in more production of antibodies. The MCH values ranged from 26.57 (Ukwani) to 34.68 x 10^{3}ul (Isoko), which were within the normal range for chickens. The MCHC obtained from broiler chickens ranged from 33.73 % (Ukwani) to 34.84 % (Ijaw). Values obtained from all other zones except Ukwani were significantly higher ($p < 0.05$) than the range of values for a normal chicken (25.9 to 33.9 %). Significant variations were observed for the mean cell volume in broiler chickens, with values ranging from 81.66 % (Ukwani) to 95.31 % (Isoko).

3.3. Serum biochemical response in broiler chickens

Table 3 presents the serum biochemical indices of broiler chickens in different zones with crude oil exploration activities in Delta State. The albumin content in chickens from Urhobo (4.06 g/dL) and Ukwani (3.91 g/dL), was the highest ($p < 0.05$) and lowest ($p < 0.05$) in chickens from Isoko (1.40 g/dL). The indirect bilirubin of broiler chickens from Urhobo (0.87) and Ukwani (0.72) was similar, though significantly higher ($p < 0.05$) than those from other zones, while the least bilirubin was observed in Isoko (0.45). The total protein of broiler chickens in Urhobo (13.19 g/dL) and Isoko (13.47 g/dL) were not significantly different ($p < 0.05$) from each other; and significantly higher ($p < 0.05$) than the values obtained for broiler chickens in Ijaw (12.25 g/dL), Ukwani (12.60 g/dL), and Itsekiri (8.10 g/dL), with the least significant values ($p < 0.05$) recorded in Itsekiri (8.10 g/dL). The uric acid values of chicken from Urhobo (3.07 mg/dL) and Ukwani (2.61 mg/dL) were similar ($p > 0.05$) to those of Isoko (2.18 mg/dL) and Itsekiri (2.04 mg/dL) but significantly higher ($p < 0.05$) than the values obtained for Ijaw (1.36 mg/dL) which was the least. The AST value of broiler chickens in Ukwani (101.51 IU/L) was significantly higher ($p < 0.05$) than that observed in Urhobo (77.00 IU/L). Still, AST value of broiler chickens showed no significant variation ($p < 0.05$) compared to the values obtained in Isoko (84.53 IU/L), Ijaw (80.25 IU/L), and Itsekiri (81.71 IU/L). The lowest value was observed in Urhobo (77.00 IU/L). The ALT values of broiler chickens ranged from 21.62 (IU/L; Isoko) to 52.99 (IU/L; Ukwani), with those from Urhobo (27.70 IU/L), Ijaw (29.50 IU/L), and Itsekiri (28.73 IU/L) not showing significant ($p < 0.05$) differences among them. The results of the present study revealed normal and increased levels of serum ALT and AST beyond the upper limits of the normal range for healthy chickens. These results are in line with findings of Cabanero et al.[23], Borg et al.[25], Jagadeesan and Pillaiwhich[26], and El-Enaen and Reda[27] who reported significant improvement in serum ALT and AST levels of rats as a result of exposure to $HgCl_2$ for a prolonged period of 30 days. In a separate report, a meaningful increase in serum AST and ALT has also been reported due to rat exposure to mercury[28]. Hence, the activity of ALT in serum may be used as an enzyme marker to assess the liver's functional status, as recommended by Sobutskii et al.[29].

Table 3. Serum Biochemical Response of broiler chickens in different zones, Nigeria

Parameters	Urhobo	Isoko	Ijaw	Itsekiri	Ukwani	SEM	Standard values
Albumin (g/dL)	4.06^{a}	1.40^{c}	2.86^{b}	2.88^{b}	3.91^{a}	0.15	2.10-3.45
Direct bilirubin	0.66	0.34	0.53	0.42	0.58	0.45	______
Indirect bilirubin	0.87^{a}	0.45^{c}	0.60^{b}	0.57^{b}	0.72^{ab}	0.03	______
Total Protein (g/dL)	13.19^{a}	13.47^{a}	12.25^{b}	8.10^{c}	12.60^{b}	0.26	5.20-6.90
Cholesterol (mg/dL)	98.12	101.74	128.17	98.58	96.64	14.51	52.0-148.0
Uric acid (mg/dL)	3.07^{a}	2.18^{ab}	1.36^{b}	2.04^{ab}	2.61^{a}	0.12	2.47-8.08
AST (IU/L)	77.00^{b}	84.53^{ab}	80.25^{ab}	81.71^{ab}	101.51^{a}	2.23	88.0-208.0
ALP (IU/L)	20.44	13.78	16.67	13.00	17.67	2.979	24.50-44.40
ALT (IU/L)	27.70^{b}	21.62^{c}	29.50^{b}	28.73^{b}	52.99^{a}	1.71	9.50-37.20

^{abc} means that within the same row, with different superscript letters, are significantly different ($p < 0.05$). SD: Standard deviations, AST: Aspartate Aminotransferase, ALT: Alanine Aminotransferase, ALP: Alkaline Phosphatase.

5. Conclusion

The findings of the present study indicated that the activities of crude oil exploration influenced the concentrations of different heavy metals and haematological and biochemical parameters in chickens reared in crude oil exploration areas of Delta State, Nigeria. The findings revealed that the concentrations of V, Cd, and Fe in broiler chicken samples exceeded the recommended tolerable limits. However, the concentrations of Ni and Hg in broiler chicken meat were lower than the internationally recommended safe limits. The presence and concentrations of these heavy metals in chicken meat could be attributed to the intense crude oil exploration activities in the areas and their subsequent transmission into the poultry value chain through the environment, soil, air, and water, affecting the health of chickens. The PCV of broiler chickens obtained from Urhobo and Ijaw, and Hb concentration across the zones indicated increased concentration above that of normal chickens. The total protein of broiler chickens across the zones and ALT values of broiler chickens in Ukwani were higher than those of normal healthy chickens. The increased serum total protein and ALT levels might serve as potential health risk biomarkers in chickens exposed to heavy metals, which can result in changes in the structural integrity and functionality of the kidney and liver, referred to as nephrotoxicosis and hepatotoxicosis, which ultimately affect the overall health of the chickens. We suggest that further studies should analyse the liver and kidney tissues for confirmation of the real damage.

Declarations

Competing interests

The authors declare that there is no competing interest with the present study.

Ethical considerations

The authors of the current study confirm that this study is submitted for the first time, and the authors checked all the ethical criteria for publication.

Authors' contributions

Unukevwere Jerome U. experimented and wrote the draft, Kuka Timothy T. assisted in laboratory work and edited the manuscript. Obakanure Oghenbrorie assisted with data analysis, Okpara Oghensuvwe reviewed the manuscript, and Olatunbosun Odu supervised the experiment. All authors read the last edition of the manuscript and agreed to publication.

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Availability of data and materials

Data and materials for the study are available and shall be presented at the proper request from the corresponding author.

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