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Sources and Health Risks of Inorganic Toxicants in *Gallus Gallus Domesticus* (Broilers) in Uyo, Nigeria

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ABSTRACT: *Poultry-related foods are consumed as good and cheap sources of protein globally. This research investigated the levels of Cd, Cr, Cu, Fe, Mn, Ni, Pb, and Zn in poultry feeds, water samples, and broilers obtained from poultry farms randomly sampled within Uyo Metropolis. These poultry-related products were obtained, treated, and analysed spectrophotometrically using standard analytical techniques. Results showed that, the concentrations of all the metals in poultry feeds and broiler meat were within their recommended limits however, mean levels of Cd and Fe in water sample was higher than their permissible limits. Concentrations in poultry feeds were generally higher than their levels in broiler meat. Multivariate analyses indentified varied factors responsible for the accumulation of these metals in feeds, water samples, and broiler meat examined. The estimated daily intake (EDI) of Cd, Fe, and Zn were higher than their oral reference doses (Rfds) for both the children and adults' populations. The hazard quotients (HQ) of Cd, Fe, Pb, and Zn for both populations were also higher than one (1). The hazard index of the metals for populations were higher than one but, the children class were more susceptible to non-carcinogenic risks. The incremental lifetime cancer risk (ILCR) for the carcinogens varied between medium to very high cancer risk classes. The total cancer risk (TCR) for the children and adults' populations were 1.54E-01 and 1.43E-01, respectively higher than the standard limits. The relative risk (RR) analysis identified Cd as metal with the highest potential to cause cancer and non-cancer risks in the consumers of broiler meat from the studied poultry farms. This study has indicated the significance of assessing poultry feeds and related products to establish to the quality.*

KEYWORDS: poultry-related foods, inorganic toxicants, poultry-related foods, broiler meat, cancer and non-cancer risks, Uyo, Nigeria.

INTRODUCTION

Poultry farming is one of the major sources of income for a greater proportion of people in Akwa Ibom State and Nigeria at large. In developing countries such as Nigeria, people live in the cities more than in the rural areas (Anríquez and Stloukal, 2008). Meat and eggs derived from poultry farming are widely consumed in Nigeria because there is no tribal, sex, age, or religious barriers (Adedokun *et al*., 2019). In Akwa Ibom State, the recent killings and kidnappings along the water ways has increased the prices of sea foods hence, most homes now depend on poultry meats and eggs as their sources of protein. During the processing of poultry feeds, mineral elements such as Cu, Fe, Mn, Se, and Zn are added as supplements (Suleiman *et al*., 2015; Lv *et al*., 2023). These supplements are applied for proper growth and development of the birds (M'Sadeq *et al*., 2018; Igwemmar and Kakulu 2022). They improve the development and quality of eggs produced (Chowdhury *et al.*, 2021). However, highly toxic metals are also introduced as contaminants to these feeds during processing (Hossain *et al*., 2014; Adekanmi, 2021).

Drinking water is another source of essential and non-essential elements in poultry farms (Fairchild *et al*., 2006; Abbas *et al*., 2008). Reports have shown that, contaminants in water systems have adverse negative effects on the aquatic and human lives over time (Abah *et al*., 2019; Lin *et al*., 2022). According to Barnhart *et al*. (2021), aquatic and human lives are dependent on the quality of water available.

The quality foods available for birds in poultry farms can affect the quality of meat and eggs harvested (Gerber *et al*., 2020; Kodani *et al*., 2022). Consumption of contaminated foods is one of the major routes through which metal loads in human beings are elevated. The consumption of contaminated foods can cause serious human health problems and even death (Rather *et al.*, 2017; Sadiku *et al*., 2020; Attiq, 2023). In spite of this, high levels of toxic metals in poultry meats and eggs have been reported (Korish and Attia, 2020; Aljohani, 2023). It has also been reported that, the consumption of contaminated poultry-related foods over time can expose the consumers to cancer and non-cancer risks (Bari *et al*., 2015; Kamaly and Sharkawy, 2023).

Waste products from poultry farms are generally used as organic fertilizers for the improvement of plants growth (Tronina and Bubel, 2008; Rahman *et al*., 2022). The application of contaminated manures in farms can increase the metal loads in both the soil and crops culivated (Adekiya *et al*., 2019; Zhen *et al*., 2020; Ebong *et al*., 2022). Plants have potential of bio-accumulating metals from contaminated soil environment and transfer them into the food chain (Benson and Ebong, 2005; Priya *et al*., 2023). Contaminated poultry waste products have the tendency of contaminating both the surface and groundwater (Oyewale *et al*., 2019; Muhammad *et al*., 2020; Abioye *et al*., 2022). The quality of air in an environment can also be impacted negatively by contaminated wastes from poultry farms and other industries (Israel *et al*., 2008; Seidavi *et al*., 2019; Gržinić *et al*., 2023).

The negative impact of elevated levels of metals in soil and edible plant most times manifest in human body via food chain (Nyiramigisha and Komariah, 2021; Tindwa and Singh, 2023). Consequently, health problems related to poultry farming are ultimately transferred to the consumers (Tsiodras *et al*., 2008; Hermans *et al*., 2012). Based on widespread utilization of chicken meats, eggs, and organic wastes from poultry farms, the quality of these products should be properly assessed to ascertain their suitability or otherwise for human consumption. Thus, periodic evaluation of trace metals in foods and their sources including poultry products could be a panacea for saving lives globally (Koch *et al*., 2022; Mitra *et al*., 2022; Hassan *et al*., 2023). Assessing human health risks in poultry-related food is a necessary tool to evaluate the potential negative risks associated with the exposure to these products. It also helps in the planning and formulating of environmental policies regarding poultry farming and the related foods (Kampa and Castanas, 2008; Zhang *et al*., 2023).

Previous works on poultry and the related products in the study area concentrated on the economic aspect of poultry farming (Ajah *et al*., 2023; Inyang *et al*., 2023; Ebong and Awatt, 2023). A few scientific researchers were done separately on the metal loads in poultry feeds, meat, and water (Jonah and Essien, 2020; Udom, 2021; Ubong *et al*., 2022). These studies never concentrated on specific poultry farms to identify the source or sources of trace metals and their health implications on the consumers. However, this work aimed at identifying the actual source of trace metals in the broiler meat from poultry farms investigated by assessing the feeds and water in these farms. It also aimed at establishing the cancer and non-cancer health risks associated with the exposure to these metals through the consumption of broilers harvested from the studied poultry farms. Hence, the outcome of this study will provide comprehensive information on the inorganic contaminants and their associated health risks via the consumption of broilers from the poultry farms examined.

MATERIALS AND METHODS

Study Area

Uyo is the capital of Akwa Ibom State that locates in the south-south region of Nigeria. The metropolitan area lies between latitude 04° 59' N and Longitude 07° 57' E. Akwa Ibom State is one of the major oil producing areas in Niger Delta Area of Nigeria. Due to the oil and oil-related activities in the State, the population is high (NPC, 2006). The area has a land mass of about 28.48km^2 in the equatorial rain forest zone (Akpan *et al*., 2014; Udoh and Igbokwe, 2014). The town has two outstanding seasons (wet and dry) during April to November and March to December, respectively. Uyo has an estimated annual rainfall of 1000 mm with a slight variation in temperature (Essien and Cyrus, 2019).

Sample collection and treatment

The feeds, water samples, and broilers were randomly sampled from poultry farms along Abak, Aka, Nwanaiba, and Oron Roads within Uyo Metropolis following the methods of Okolo *et al*. (2022). At each of the poultry farms, five hundred grams each of starter and finisher feeds were collected in polyethylene bags. These samples were conveyed to a laboratory in the Department of Chemistry, University of Uyo, Nigeria and air-dried for three (3) weeks. After drying, the samples were homogenized with a ceramic pestle and mortar. These samples were later filtered through a 2 mm sieve and stored in well-labeled containers before digestion. The starter and finisher feeds from each farm were mixed together properly to obtain composite sample for each location. One gram of the blended feed from each farm was digested with 20 mL of Aqua Regia in a conical flask. The flask was placed on a hot plate until a clear solution was obtained. The flask was allowed to cool, on cooling, the mixture was filtered with Whatman No. 1 Filter paper into a 50mL volumetric flask and filled to mark with deionized water. The filtrates were transferred into clean sample bottles for metal analysis (Ande *et al*., 2020).

Water samples were collected from plastic tanks at the studied farms in 2L polyethylene bottles previously washed with detergent diluted with HNO_3 and deionized water. Prior to sample collection, the polyethylene bottles were cleaned twice with the water to be sampled. Water samples collected were acidified with 1ml conc. HNO_3 on site before transporting them to the laboratory. These samples were stored at a temperature of 4°C before metal analysis (APHA/AWWA/WPCF, 2017; APHA, 2017).

Matured old broilers (*Gallus gallus domesticus)* (Six (6) weeks and above) were obtained from of the farms investigated, slaughtered with stainless steel knife, then the muscle, liver, heart, and gizzard were harvested. The parts collected were washed using deionized water and cut into pieces with stainless steel knife a mixed together based on the location. Two grams (2g) was weighed and placed in an oven at 105°C for two hours. The dried samples were put in a digestion flask and digested with a mixture of 1ml conc. $HClO_4$ and 5ml conc. HNO_3 on a hotplate until the mixture was colourless. The volume of the digest in the digestion flask was raised to 50ml with deionized water. The mixture obtained was filtered using Whatman No. 42 Filter Paper into polyethylene bottles and preserved at a temperature of 4°C before analysis (Hossain *et al*., 2023). Concentrations of Cd, Cr, Cu, Fe, Mn, Ni, Pb, and Zn were obtained in these poultry-related products using atomic absorption spectrophotometer (UNICAM 969 Model) following the methods of AOAC. (2004).

Cancer and non-cancer health risk assessment

The carcinogenic and non-carcinogenic risks associated with the exposure to trace metals determined via the consumption of broiler meat from the poultry farms were evaluated by the estimated daily intake (EDI), hazard quotients (HQ), and hazard index (HI). The incremental lifetime cancer risk (ILCR), total cancer risk (TCR), and relative risk (RR) were also calculated (USEPA, 2018; Salihu *et al*., 2019).

Estimated daily intake (EDI)
The EDI of trace metals for the children and adults populations via the consumption of broiler meat was computed with Equation (1) below.

$$EDI = \frac{Cm \times Df}{BW} \text{------------------------------} (1)$$

Where Cm denotes the concentration ($mgkg^{-1}$) of trace metals determined in the broiler meat; Df indicates the daily intake rate of the broiler meat which is 46.59 kg for children and 60.95 kg for the adults (Ekhator *et al*., 2017; Patrick-Iwuanyanwu and Chioma, 2017); BW is the body weight which is 24 kg for children and 70 kg for the adults' population (Ekhator *et al*., 2017).

Hazard quotient (HQ)
Hazard quotient (HQ) of the metals for the children and adults' populations via the consumption of broiler meat from the poultry farms investigated was calculated using Equation (2) according to Chien *et al*. (2002) and Oyekunle *et al*. (2020).

$$HQ = \frac{EDI}{RfD} \text{--------------------------------} (2)$$

Where EDI is the values obtained for estimated daily intake (EDI) in Equation (1); Rfd represents the recommended oral reference dose for the metals which according to USEPA, (2000) are Cd (0.001), Cr (1.500), Cu (0.040); Fe (0.700), Mn (0.140), Ni (0.020), Pb (0.004), and Zn (0.300) mg kg^{-1} day^{-1}.

Hazard index (HI)
Hazard index (HI) of the trace metals through the consumption of broiler meat from poultry farms investigated for the children and adults populations was computed by means of Equation (3).

$$HI = \Sigma HQ = HQCd + HQCr + HQCu + HQFe + HQMn + HQNi + HQPb + HQZn - (3)$$

Where ΣHQ represents the sum of all the hazard quotients (HQ) of the trace metals in broiler meat from poultry farms examined.

Incremental lifetime cancer risk (ILCR)
The incremental lifetime cancer risk (ILCR) associated with cancer-causing metals in both the children and adults populations through the consumption of broiler meat were calculated with Equation (4) below.

$$ILCR = CSF \; x \; EDI \text{------------------------------} (4)$$

Where CSF indicates the cancer slope factors for the cancer-causing metals shown in Table 3 based on the USEPA IRIS, (2011) standard; EDI is the estimated daily intake rate of cancer-causing metals in Table 3.

Total cancer risk (TCR)

The total cancer risk (TCR) for the exposure to cancer-related metals by the children and adults populations via the consumption of broiler meat from the studied poultry farms was approximated with Equation (5).

$$TCR = \Sigma ILCR = ILCRCd + ILCRCr + ILCRCu + ILCRNi + ILCRPb - - - - - - - - - (5)$$

Where $\Sigma ILCR$ specifies the sum of individual incremental lifetime cancer risk of the metals. The total cancer risk is categorizes into the following groups: 1.0E-01- 1.0E-03 denotes the very high cancer risk class, 1.0E-04 signifies the high cancer risk class, 1.0E-05 depicts the medium cancer risk class, 1.0E-06 belong to the low cancer risk class, and TCR less than 1.0E-06 belong to the negligible cancer risk class (USEPA, 1999).

Relative risk (RR)

The most damaging of all the metals causing cancer and non-cancer threats was identified with relative risk (RR) as indicated in Equation (6) following the procedures of Yu *et al*. (2014) and Adebiyi *et al.* (2020).

$$RR\ (\%) = \frac{Cm}{Rfd} X\ 100 - (6)$$

Where Cm is the concentration of trace metals determined; Rfd represents the oral reference dose.

Data Analysis

Analysis of results obtained was done using IBM SPSS Statistics 20 (IBM USA). Multivariate analyses (Principal component analysis and Hierarchical cluster analysis) were carried out with Duncan's multiple range tests at 90% confidence limit. Factor analysis was executed by means of Varimax Rotation procedures on eight (8) properties and values from 0.549 and above were classified as significant. Cluster analysis was achieved by Dendrograms to segregate the properties based on their similar sources and properties.

RESULTS AND DISCUSSION

Trace Metals in the Studied Poultry-Related Products

Results for the concentrations of trace metals in feeds, water and broiler meat from the poultry farms investigated are shown in Table 1.

Table 1: Trace metals in poultry-related samples within Uyo Metropolis

	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
				Poultry Feeds				
MIN	0.175	0.036	1.045	41.382	0.005	0.003	0.002	1.934
MAX	0.192	0.040	1.183	44.260	0.007	0.007	0.006	2.140
SD	0.009	0.002	0.071	1.295	0.001	0.002	0.002	0.091
MEAN	0.185	0.038	1.109	42.819	0.007	0.005	0.005	2.026
RL	1.0	0.01	8.0	80.0	60.0	0.5	5.00	40.00
Water Samples								
MIN	0.008	0.007	0.976	3.410	0.002	0.002	0.001	0.701
MAX	0.014	0.010	1.018	3.580	0.004	0.003	0.002	0.814
SD	0.003	0.001	0.019	0.084	0.001	0.001	0.001	0.060
MEAN	0.011	0.009	1.002	3.499	0.003	0.003	0.002	0.757
RL	0.003	0.05	2.00	2.00	0.05	0.07	0.01	3.00
Broiler Meat								
MIN	0.097	0.015	0.017	11.752	0.002	0.001	0.002	1.724
MAX	0.113	0.019	0.020	12.205	0.003	0.002	0.003	1.803
SD	0.007	0.002	0.001	0.197	0.001	0.001	0.001	0.040
MEAN	0.106	0.017	0.019	12.030	0.003	0.001	0.002	1.763
RL	0.5	1.0	0.4	180.0	0.5	0.5	0.1	150.0

Results obtained showed that Cd in the feeds varied from 0.175 to 0.19 2 $mgkg^{-1}$. This range is higher than 0.028 - 0.094 $mgkg^{-1}$ reported by Amitaye and Okwagi, (2019). Concentrations of Cr in the feeds varied between 0.036 and 0.040 $mgkg^{-1}$. This is lower than 0.001 - 0.62 $mgkg^{-1}$ obtained by Ifie *et al*. (2022) in chicken feeds investigated. Levels of Cu in feeds examined varied from 1.045 to 1.183$mgkg^{-1}$, this is higher than 0.04 - 1.21 µg/ml reported by Suleiman *et al*. (2015). Concentrations of Fe ranged from 41.382 to 44.260 $mgkg^{-1}$ and are lower than 164.81 – 283.09 µg/g reported by Igwemmar *et al*. (2022). Mn in the feeds investigated ranged from 0.005 to 0.007 $mgkg^{-1}$. The obtained range is lower than 0.94 - 3.12 µg/ml obtained by Suleiman *et al*. (2015). Ni concentrations in the feeds varied between 0.003 and 0.007 $mgkg^{-1}$. This is lower than 2.25 - 4.87µg/ml reported by Okoye *et al*. (2011). Pb in the studied poultry feeds ranged from 0.002 to 0.006 $mgkg^{-1}$. The obtained range is below 3.8–33.6 $mgkg^{-1}$ reported by Mahesar *et al*. (2010) however; higher than below detectable limit (BDL) recorded by Ande *et al*. (2020). Concentrations of Zn in the studied poultry feeds varied from 1.934 to 2.140$mgkg^{-1}$. The reported range is lower than 49.75 – 85.82 $mgkg^{-1}$obtained in poultry feeds by Igwemmar *et al*. (2022). Generally, the mean concentrations of all the metals examined in poultry feeds in Table 1 are below their recommended limits by FAO/WHO (2000). Hence, these feeds may not have adverse effects on the birds and the consumers.

Results for the distribution of trace metals in water samples from poultry farms investigated are shown in Table 1. Concentrations of Cd in water samples from the studied farms ranged from 0.008 to 0.014 mg/L. This is lower than 0.00 – 0.019 mgL^{-1} obtained in water samples from poultry farm at Maritime Region of Togo by Idrissa *et al*. (2020). Cr ranged between 0.007 and 0.010 mgL^{-1}in the water samples examined which is below 0.00–0.31 mgL^{-1}reported by Chen *et al*. (2022) in a related study. Cu varied from 0.976 to 1.018mgL^{-1} in the studied water samples. The obtained range is higher than 0.11 - 0.27 reported in Mecca Almokaramah, Saudi Arabia by Hussein *et al*. (2013). Concentrations of Fe varied from 3.410 to 3.580mgL^{-1}, and are lower than 17.0563-59.4264 mgL^{-1} reported in Osun State, Nigeria by Ogunwale *et al*. (2021). Mn in the studied water samples ranged from 0.002 to 0.004mgL^{-1}. The obtained range is lower than 0.02 – 0.44 mgL^{-1} reported in Assiut Governorate, Egypt by Sayed and Omar, (2013). The range of 0.002 – 0.003 $mgL^{-1}$was recorded for Ni in the studied water samples. The range is below 0.00–0.65 mgL-1 obtained in a similar study by Chen *et al*. (2022). Levels of Pb in water samples from farms investigated ranged from 0.001 to 0.002mgL^{-1} which is lower than 0.021-0.2283 mgL^{-1} obtained by Ogunwale *et al*. (2021). Zn concentrations varied between 0.701 and 0.814 mgL^{-1}, this is higher than 0.09 - 0.23mgL^{-1} but lower than 3.0420–21.1450 obtained by Hussein *et al*. (2013) and Ogunwale *et al*. (2021), respectively.

Results recorded for trace metals water samples from the studied poultry farms indicated that Cr, Cu, Mn, Ni, Pb, and Zn are below their recommended limits of 0.05, 2.0, 0.05, 0.07, 0.01, and 3.00mgL^{-1}, respectively by WHO, (2011). However, the mean concentrations of Cd and Fe were higher than 0.003 and 2.00 mgL^{-1}, respectively recommended by WHO (2011). This is consistent with the findings by Idrissa *et al*. (2020) in water samples from poultry farms in Togo. High level of Cd in water can accumulate in the organs such as kidney, lung, liver and reproductive organs of the chicken (Adekanmi, 2021). This may result in the reduction of growth, oxidative stress, hormonal imbalance, and poor conversion of feeds by birds. It could also reduced both the egg laying capacity and the quality of egg in a poultry (Yang *et al*., 2012; Kim *et al.*, 2019; Zhu *et al.*, 2019; Wu *et al*., 2022). According to Kubier *et al*. (2019), the major natural source of Cd is weathering of Cd-contaning rocks although; the anthropogenic sources are numerous. The primary source of Fe in groundwater is the natural rock weathering processes (Wang *et al*., 2023). Elevated levels of Fe reported can change the turbidity and colour of the water due the formation of iron (III) oxide (Fe_2O_3) (Fairchild *et al*., 2006). When the level of Fe in water is higher than 0.3mgL^{-1} as reported in this study, iron bacteria that utilize water and other water-holding containers as their source of nutrients will accumulate (Oyeku and Eludoyin, 2010).

Results for the concentrations of trace metals in broiler meat from the studied farms are shown in Table 1. Concentrations of trace metals in the broiler meat examined varied as follows: Cd ranged from 0.097 to 0.113 $mgkg^{-1}$. The obtained range is below the 0.01–4.60 $mgkg^{-1}$reported for Cd by Iwegbue *et al*. (2008). Cr varied between 0.015 and 0.019 $mgkg^{-1}$ lower than 0.0–0.08 $mgkg^{-1}$ reported by Hossain *et al*. (2023). Cu in the broilers investigated recorded a range of 0.017 – 0.020

mgkg^{-1}. This is lower than 3.117 – 5.066 obtained in a similar study by Mottalib *et al*. (2018). Fe ranged between 11.752 and 12.205 mgkg^{-1}, this is lower than 10-310 mgkg^{-1} obtained by Ogbomida *et al*. (2018). Mn in the studied meat ranged from 0.002 to 0.003mgkg^{-1}. This is lower than 0.01 – 0.09 mgkg^{-1}reported in a similar work by Asegbeloyin *et al*. (2015). A range of 0.001 – 0.002 mgkg^{-1} was recorded for Ni in this work. The obtained range for Ni is lower than 0.577 - 0.852 mgkg^{-1}reported by Okolo *et al*. (2022) in a related study. Pb and Zn concentrations in meats investigated ranged as follows: 0.002 – 0.003 mgkg^{-1} and 1.724 – 1.803 mgkg^{-1}, respectively. Concentrations of Pb in the studied meat are lower than 0.0138 – 0.0191 mgkg^{-1} reported in a related work by Ersoy *et al*. (2015). Levels of Zn reported are also lower than 0.089 - 2.094 mgkg^{-1} obtained in edible parts of chicken by Ogu and Akinnibosun (2020).

The mean concentrations of Cd, Cr, Cu, Fe, and Mn obtained in the studied meats (0.106±0.007, 0.017±0.002, 0.019±0.001, 12.030±0.197, and 0.003±0.001mgkg^{-1}, respectively) are all below their recommended limits of 0.5, 1.0, 0.4, 180.0, and 0.5mgkg^{-1}, respectively by FAO/WHO, (2011). The mean values of Ni (0.001±0.001 mgkg^{-1}) and Zn (1.763±0.040 mgkg^{-1}) are also within their limits in Table 1. The mean value of Pb (0.002±0.001 mgkg^{-1}) is lower than 0.1 mgkg^{-1} recommended by FAO/WHO, (2011). Consequently, broilers from the farms investigated were deficient of Cu, Fe, Mn, and Zn which are essential for the normal growth (Yang *et al*., 2011). This can result in severe problems for the broilers in the poultry farms investigated (Olgun, 2017; Olukosi *et al*., 2018; Lin *et al*., 2020). Notwithstanding the low levels of Pb reported, the weight of the broilers could be reduced due to the high toxicity of the metal even at very low concentrations (Adekanmi, 2021; Ebrahimi *et al*., 2023). The low levels of Cd, and Ni may not have adverse effect on the broilers since the elements are not essential for their normal enzymatic and growth of animals (Naseri *et al*., 2020; Al-Waeli *et al.*, 2013; Wu *et al*., 2013; Li *et al*., 2014; Wu *et al.*, 2015). Studies have also shown that, the concentrations of Cr obtained in the studied broilers can affect their growth negatively (Dalólio *et al*., 2021; Youssef *et al*., 2022).

Generally, levels of all the metals were relatively higher in the studied poultry feeds than in the meat as observed by Korish and Attia (2020). This could be attributed to (i) Use of essential elements by the broilers for normal enzymatic activities (Lv *et al*., 2023). (ii) Unnecessary mineral secretion owing to low availability, egestion, and formation of insoluble complexes (Aksu *et al*., 2011; Stafford *et al*., 2016).

Multivariate Analyses

Results for the principal component analysis and Cluster Analysis are shown in Table 2 and Figure 1 – 3, respectively.

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Table 2: Principal component analysis of relative loading of trace metals in the feeds, water samples, and meat from poultry farms investigated

	Poultry Feed		**Water**			**Meat**		
Parameter	**F1**	**F2**	**F1**	**F2**	**F3**	**F1**	**F2**	**F3**
Cd	0.718	-0.644	-0.545	0.783	0.300	-0.585	-0.736	0.341
Cr	0.641	-0.097	0.304	0.950	0.071	-0.184	0.805	0.564
Cu	0.995	-0.093	-0.688	0.726	-0.013	0.676	-0.607	0.417
Fe	0.923	-0.366	0.921	0.357	-0.157	0.405	0.212	0.889
Mn	0.886	0.421	0.874	0.018	0.487	-0.890	0.455	-0.040
Ni	0.809	0.465	-0.948	-0.187	0.256	0.882	0.441	-0.169
Pb	0.356	0.922	0.948	0.187	-0.256	0.882	0.441	-0.169
Zn	0.956	-0.128	0.370	-0.164	0.914	0.931	-0.355	-0.082
% Total Variance	65.6	22.8	55.1	28.3	16.6	52.6	29.1	18.3
Cumulative %	65.6	88.4	55.1	78.2	100.0	52.6	81.7	100.0
Eigenvalue	5.25	1.83	4.41	2.27	1.32	4.21	2.32	1.47

Trace metals determined in the poultry feeds, water, and meat were analyzed with principal component analysis (PCA) to identify the underlining factors responsible for the accumulation of trace metals in the studied poultry-related samples (Ebong and Mkpenie, 2016; Ebong *et al*., 2022a; 2023a). The PCA results in Table 2 indicate two (2) underlining factors responsible for the accumulation of trace metals in poultry meat. These two factors have Eigenvalue higher than one (1) with 88.4% of the total variance. Factor one (F1) donated 65.6% of the total variance with significant influence by all the metals determined except Pb (Table 2). This could be attributed to contamination (anthropogenic factors) (Adamse *et al*., 2017; Aljohani, 2023; Kamaly and Sharkawy, 2023). Factor two (F2) contributed 22.8% to the entire variance with strong impact by Pb only (Table 2). This indicates the negative impact of contamination during the processing of poultry feeds on the quality of poultry products (Adekanmi, 2021). In water, the PCA results in Table 2 show three (3) major factors with Eigenvalues higher than one (1). Factor one (1) contributed 55.1% of the total variance with considerable influence by Fe, Mn, and Pb. This shows the influence of both human and natural factors on the quality of water in the studied poultry farms (Ozoko *et al*., 2022; Ullah *et al*., 2022). Factor two donated 28.3% to the total variance with substantial influence by Cd, Cr, Cu, and Zn. This represents the influence of industrial wastes and geological enrichments on the water quality within the studied farms (Khatri and Tyagi, 2015; Baba and Gündüz, 2017; Akhtar *et al*., 2021). Factor three contributed 16.6% of the total variance with significant influence by Zn only. This represents impact of both the natural and anthropogenic factors on the quality of water in the studied poultry farms (Noulas *et al*., 2018; Tu *et al*., 2020). The PCA of poultry meat revealed three principal factors with Eigenvalue higher than one and a total variance of 100%. Factor one added 52.6% to the total variance with strong influence by Cu, Ni, Pb, and Zn. This represents the influence of poultry feeds and water on the quality of poultry-related food products (Iwegbue *et al*., 2008; Adekanmi, 2021). Factor two donated 29.1% to the entire variance with significant influence by Cr only. This signifies the influence of animal diets on the metal load of the birds in poultry farms examined (Farag *et al*., 2017; Mottalib *et al*., 2018). Factor three (F3) added 18.3% of the total variance with strong influence through Cr and Fe. This also signifies the impact of feeds and water on the quality of the poultry products (Iwegbue *et al*., 2008; Ebbing *et al*., 2019; Lin *et al*., 2020).

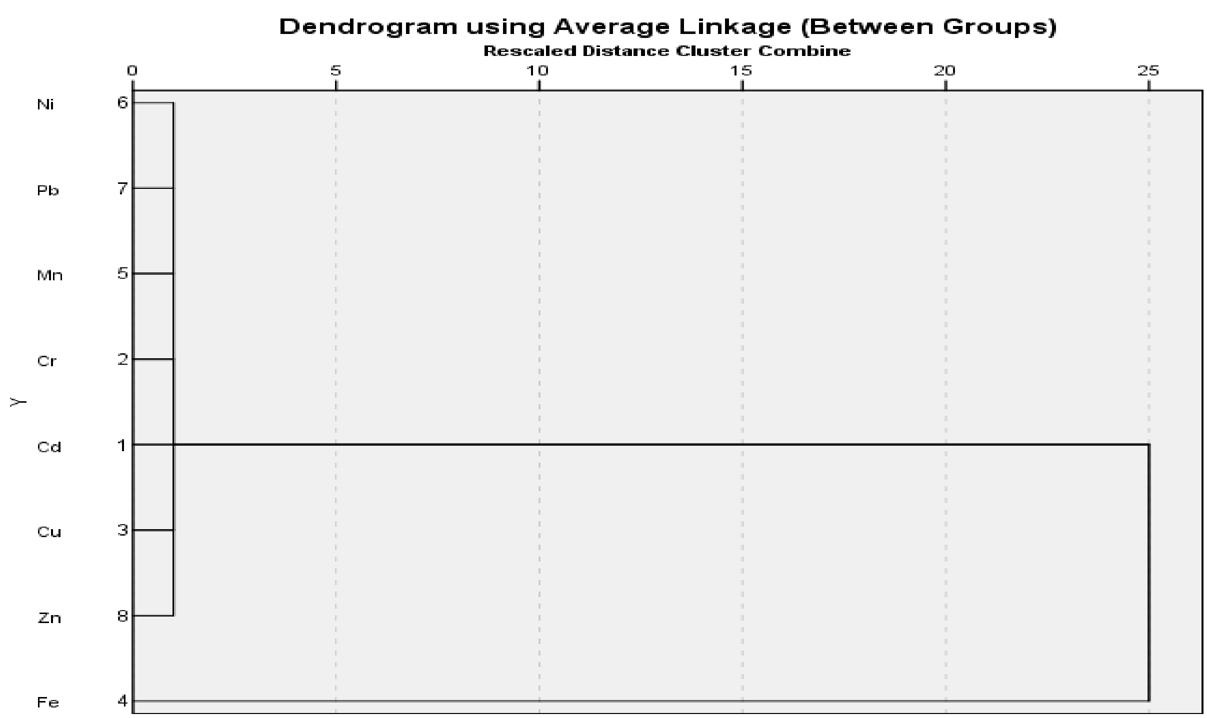


Figure 1: Hierarchical clusters of trace in poultry feeds

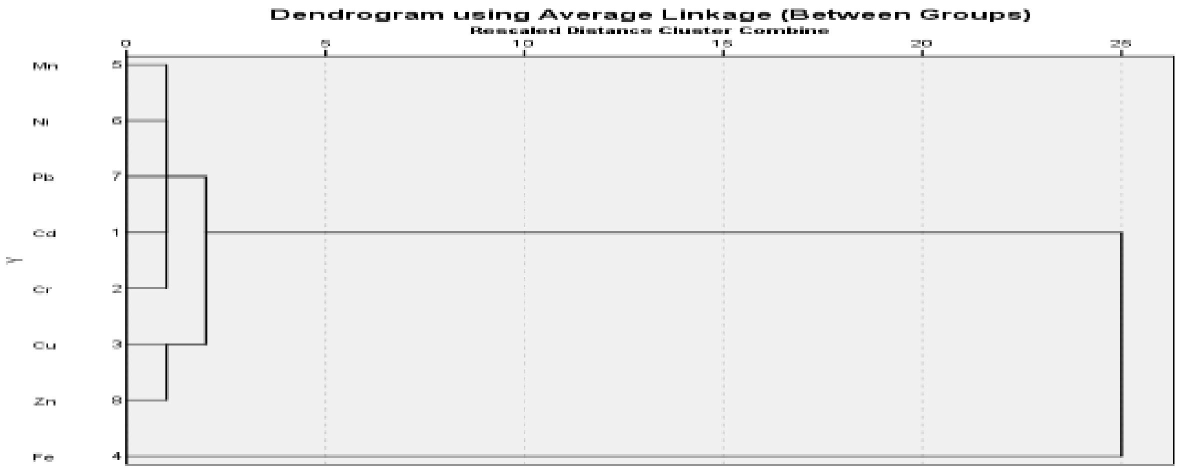


Figure 2: Hierarchical clusters of trace in water samples from the studied poultry farms.

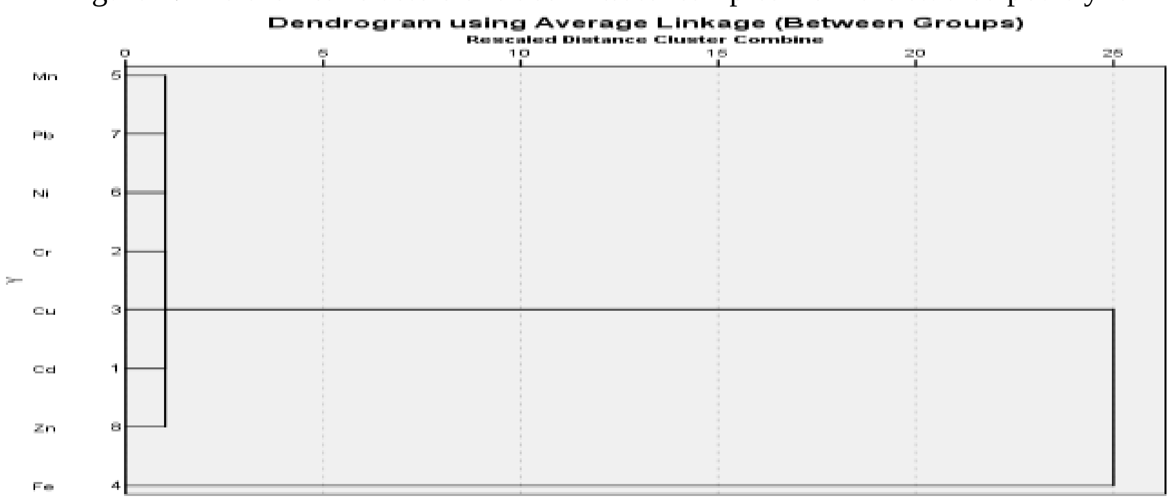


Figure 3: Hierarchical clusters for trace in broiler meat

The Hierarchical clusters of trace metals in the feeds, water, and broiler meats from the poultry farms investigated are shown in Figures 1, 2, and 3, respectively. Cluster analysis indicates the source and common properties among members of similar cluster (Ebong *et al*., 2023b). Hierarchical cluster analysis (HCA) for the poultry feeds assessed revealed two principal clusters (Figure 1). The first cluster connects all the trace metals determined together except Fe. This is consistent with the findings by PCA for poultry feeds in Table 1 and may depict a familiar source for these metals. The second cluster links Fe only which is similar to the findings at Lemna Dumpsite soil by Ebong *et al*. (2019). This show that majority of Fe found in the poultry feeds examined could be from a source different from other metals.

Figure 2 illustrates three (3) main clusters with the first one linking Mn, Ni, Pb, and Cr together. Cluster two joins Cu and Zn as one while, the third cluster links Fe only. This displays the different sources and properties of metals in water samples investigated in the separate clusters (Ebong *et al.*, 2018; Ayeni and Soneye, 2013).

The HCA for the broiler meat from the studied poultry farms revealed two major clusters. The first cluster put all the trace metals in one cluster except Fe while, the second cluster links Fe only. This is similar to the PCA for the broilers shown in Table 1.

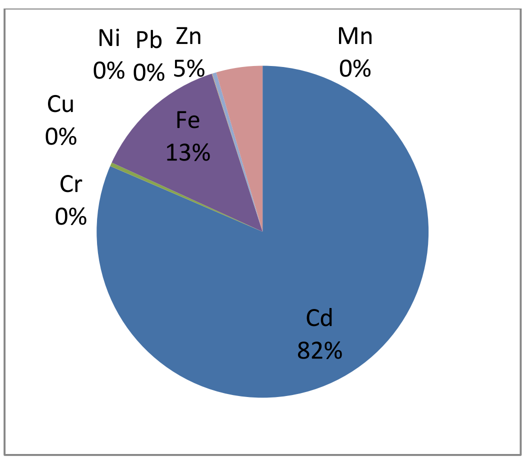
Health Risk Assessment

Table 3: Results of chronic daily intake (CDI) rate, cancer, and non-carcinogenic risks of trace metal in the studied poultry-related product,

	Children				Adults				
	EDI	HQ	ILCR	EDI	HQ	ILCR	RR	%RR	CSF_o
Cd	0.206	206.0	7.80E-02	0.092	92.0	3.50E-2	106.0	81.40	0.38
Cr	0.033	0.022	1.70E-02	0.015	0.01	8.00E-02	1.10E-02	0.01	0.5
Cu	0.037	0.925	5.60E-02	0.017	0.425	2.60E-02	4.75E-01	0.37	1.5
Fe	23.35	33.36	-	10.48	14.96	-	17.19	13.20	NA
Mn	0.006	0.043	-	0.003	0.021	-	2.10E-02	0.02	NA
Ni	0.002	0.10	3.40E-03	0.001	0.05	2.00E-03	5.00E-02	0.04	1.7
Pb	0.004	1.00	3.40E-05	0.002	0.50	2.00E-05	5.00E-01	0.38	0.0085
Zn	3.422	11.41	-	1.535	5.12	-	5.88	4.52	NA
HI		252.86			113.09		-		
TCR			1.54E-01			1.43E-01	-		

Results for the estimated daily intake rate of trace metals via the consumption of broiler meat from the studied poultry farms indicated that, Cr, Cu, Mn, Ni, and Pb for both populations were below their recommended oral reference doses (RfDs) by USEPA, (2000). However, the mean EDI values of Cd, Fe, and Zn were higher than their RfD limits for both children while Zn was within the limit in the adults‛ population. The mean EDI values for Cu, Mn, Ni, and Pb are also lower than their Provisional tolerable daily intake (PTDI) values of 0.50, 0.14, and 0.005 $mgkg^{-1}day^{-1}$, respectively for children and adults populations by EPAU, (2016). Conversely, EDI values for Cd, Cr, Fe, and Zn in children were above their PTDI values (0.001, 0.003, 0.80, and 1.00 $mgkg^{-1}day^{-1}$, respectively) by EPAU, (2016). However, the mean EDI value for Cr in the adults' population is lower than a PTDI value of 0.003 $mgkg^{-1}day^{-1}$. This is an indication that Cd, Cr, Fe, and Zn may have adverse health problems on the consumers of broiler meat from the studied farms. Results obtained also revealed that, children were at higher risk than the adults' population (Table 3). The reported higher risk for the children is consistent with the findings by Samad *et al*. (2023).

Results for the mean hazard quotient (HQ) of metals for both the children and adults populations revealed values higher than one (1) (Table 3). This is similar to the results obtained by Kamunda *et al*. (2016) and Apau *et al*. (2022) in their studies. Thus, the consumers of broiler meat from the studied poultry farms may be exposed to non-cancer risks over time (Tschinkel *et al*., 2020; Udiba *et al*., 2022). Levels of Cd, Fe, Pb, and Zn were the major contributors to the high HQ values obtained in this study.



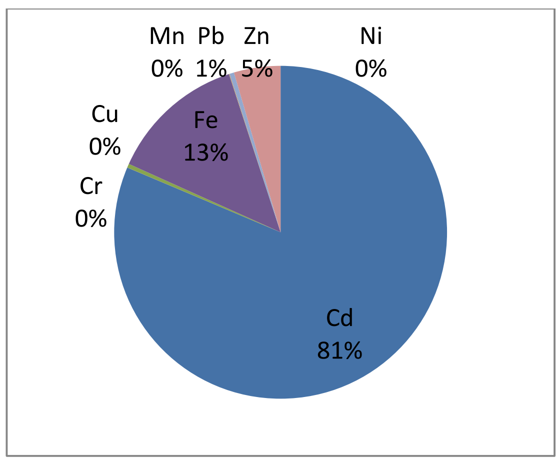


Figure 4: Average hazard quotient (HQ) of trace metals for children (A) and adults (B) populations

The mean hazard index (HI) values of metals for the children and adults' populations are 252.86 and 113.09, respectively (Table 3). The high HI values obtained in this study are in agreement with the results reported by Loh *et al*. (2023) and Omali *et al*. (2023). The reported values are higher than one (1) hence; the consumers of broiler meat from the studied farms could be exposed to serious non-carcinogenic health problems (Chao-yang *et al*., 2018; Meseret *et al*., 2020). The children population showed higher mean HI value than the adults. Consequently, the children population could be more exposed to the associated non-carcinogenic risks than the adults (Krupnova *et al*., 2019; Focus *et al*., 2021). The mean HQ values of Cd contributed 82 and 81%

of the total HI in the children and adults' populations, respectively (Figures 4A and B). The study revealed that Cd could be a potential health problem to the consumers of broiler meat from the studied farms.

Prolonged human exposure to Cd, Cr, Cu, Ni, and Pb may result in cancer-related health problems (Cao *et al*., 2014). In this study, the exposure rate to these carcinogens by the consumers of the studied meat was assessed using the incremental lifetime cancer risk (ILCR) (Goudarzi *et al*., 2021; Zhao and You, 2021). As shown in Table 3, the mean ILCR values for Cd, Cr, Cu, Ni, and Pb are 7.80E-02, 1.70E-02, 5.60E-02, 3.40E-03, and 3.40E-05, respectively in the children population. Results in Table 3 also reveal mean ILCR values of 3.50E-2, 8.00E-02, 2.60E-02, 2.00E-03, and 2.00E-05 for Cd, Cr, Cu, Ni, and Pb, respectively for the adults' population. Hence, the cancer risk associated with the exposure to Cd, Cr, and Ni were in the very high cancer risk class for both populations (Mohammadi *et al*., 2019). However, the cancer risk related to the exposure to Pb was in the medium class for both populations (USEPA, 2012; Ramadan and Haruna, 2019). Higher cancer risks related with exposure to the carcinogens examined via broiler meat were observed for the younger than the adults' population as reported by Kasozi *et al.* (2021) and Samad *et al*. (2023).

Results for the total cancer risks (TCR) of the carcinogens (Cd, Cr, Cu, Ni, and Pb) due to the consumption of the studied meat are indicated in Table 3. TCR values of 1.54E-01 and 1.43E-01were recorded for the children and adults populations, respectively. The reported TCR values for both populations are beyond the acceptable range of 10^{-6} - 10^{-4} by USEPA, (2011). The high TCR values obtained calls for concern since a higher proportion of human beings in the study area irrespective of age and sex depend on poultry-related products as their major protein source. Consequently, the consumers of broilers from the studied farms may be exposed to cancer and cancer-related risks (Ogidi *et al*., 2021). Cd contributed the highest proportion (51%) to the total TCR for children while, Cr donated the highest TCR value for the adults' population. The orders for the contributions of the carcinogens to the total TCR in are Children and adults are Cd > Cu > Cr > Ni > Pb and Cr > Cd > Cu > N I > Pb, respectively. This indicates that Pb contributed the lowest value of the TCR in both the children and adults' populations.

Results of relative risk (RR) for both the carcinogenic and non-carcinogenic problems are shown in Table 3. The results revealed the potentials of each of the metals in affecting the consumers through the consumption of studied meat. Results obtained indicated the following order for the metals: Cd > Fe > Zn > Pb > Cu > Ni > Mn > Cr for both populations. Cd contributed the highest proportion (81.4 %) while; Cr donated 0.01 % of the entire RR obtained. Consequently, Cd was the metal with the most devastating tendency to cause both the cancer and non-cancer risks to the consumers of meat from the studied farms (Rahimzadeh *et al*., 2017; Furtak *et al*., 2022). The high RR value reported is in agreement with the ones by Kumar and Kumar, (2021) and Adebiyi *et al*.

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(2020). Hence, the consumption of broilers from the farms investigated may result in high accumulation of Cd and the attendants' human health problems.

CONCLUSION

The research was to assess the quality of poultry feeds and water sources in some poultry farms in Uyo. Nigeria. It also evaluated the concentrations of metal toxicants in matured broilers from poultry farms investigated to establish their suitability for human consumption. Results obtained revealed that, the water source and feeds utilized in poultry farms can elevate the level of metals in poultry-related products. Contaminated poultry feeds and water can affect negatively the quality of meat and eggs harvested from poultry farms. The results also indicated higher levels of Cd and Fe in the water sources used in the studied poultry farms. The study indicated that, broiler from poultry farms examined were deficient of essential elements such as Cu, Fe, Mn, and Zn which can affect the quality of meat. The principal component analysis identified two, three, and three factors responsible for metal toxicants in feeds, water, and broilers, respectively. The estimated daily intake rate of metals intake via the consumption of broiler meat from the studied farms indicated higher levels of Cd, Fe, and Zn. The provisional tolerable daily intake rate of Cd, Cr, Fe, and Zn were also higher than their limits. The higher hazard quotient and hazard index values obtained revealed the high potential of the metals to cause non-cancer problems in the consumers, and the children population was more vulnerable. The incremental lifetime cancer risk and total cancer risk were higher than their recommended limits indicating the tendency of broiler meat from the studied poultry farms at causing cancer problems in the consumers. Results obtained revealed that, Cd levels in the studied broiler meat can cause serious cancer and non-cancer health problems in the consumers. The study has indicated that, the assessment of poultry-related foods to establish their levels of metal contaminants is a life-saving tool for the consumers. The outcome of this study is useful to poultry farmers to know the feeds suitable for optimal farm outputs. The study will assist the institutions concern in their future policy making and implementations.

Conflict of interest

There is no conflict of interest associated with this manuscript.

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