



Trace element composition of *Gallus gallus domesticus* eggs and health risks associated with their consumption in Port Harcourt, Nigeria

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ABSTRACT

Using a multivariate approach, this study investigated the trace element composition and concentration in *Gallus gallus domesticus* eggs. A total of 51 freshly laid *Gallus gallus domesticus* egg samples were randomly obtained and analyzed for trace elements using a flame atomic absorption spectrophotometer. The results showed that the levels of zinc, lead, nickel, cobalt, manganese, iron and copper were in the range of $2.16 \pm 0.03 - 7.43 \pm 0.98$ mg/L, $<0.001 \pm 0.00 - 9.86 \pm 1.01$ mg/L, $<0.001 \pm 0.00 - 1.44 \pm 0.30$ mg/L, $<0.001 \pm 0.00 - 0.35 \pm 0.01$ mg/L, $<0.001 \pm 0.00 - 0.76 \pm 0.04$ mg/L, $2.93 \pm 1.01 - 11.52 \pm 1.54$ mg/L and $0.03 \pm 0.05 - 0.967 \pm 0.06$ mg/L, respectively. However, mercury, chromium, and cadmium were not detected in all the samples. This suggests the absence of toxicity risk due to these non-essential elements in the eggs, especially when they are consumed. Moreover, the target hazard quotient and total target hazard quotient of the eggs were less than 1, thereby suggesting no probable harm, as well as non-carcinogenic hazards respectively.

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1. Introduction

Avian eggs are an important source of nutrients. This food source contains most of the proteins, lipids, vitamins, and minerals required for good health and human well-being.

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Avian products (meat and eggs) are consumed in different parts of the world (1, 2).

Some important birds such as ducks, chickens, turkeys, etc. are reared on the homestead. Globally, poultry products are considered essential components of daily diets, whether in developed or developing countries (3 - 5). Poultry birds are fed with feeds produced from



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agricultural activities, as well as water from different sources such as surface, ground, and rain waters (6 - 10).

The risks associated with trace elemental composition of food and food products continue to attract the attention of environmentalists, environmental health officers, and food scientists among others. This is because trace elements have been detected in several food sources, including animals (11), mushrooms (12), fishes (14), and soil and vegetation (15 - 22). Furthermore, trace elements have been detected in water sources consumed by humans (23 - 24). As such, food safety from a public health perspective is now a major concern worldwide, especially in recent past decades. This is due to the increasing demand to monitor food and allied products that may be potentially contaminated with toxicants such as trace elements. Overall, this approach is aimed at understanding the full spectrum of health-related problems, as well as the establishment of a concentration threshold and safe limits.

Poultry birds can be used to provide evidence for environmental pollution since they can bioaccumulate trace elements contained in their diet and surroundings. Eggs of poultry birds have been reported to contain trace elements (25 - 27), this being an indication that adult birds and hens laying eggs possess the tendency to bioaccumulate toxic elements ingested through feeds and water. Trace elements are mostly grouped as vital (copper, zinc, iron, chromium, etc.) and non-vital (mercury, cadmium, and lead). The vital elements play an essential role in biological processes,

while the non-vital metals have no known biological functions accredited to them. As such, reported some health effects associated with these non-vital elements may be harmful to life forms even at low concentrations. The human and animal health hazards associated with trace elements have been comprehensively documented by Turdi and Yang (28); Izah et al. (23; 29); Dashtizadeh et al. (30); Xie et al. (31). Most importantly, health risks of trace elements are known to exhibit source, time, and concentration-dependent effects.

Exposure to trace elements through the intake of *Gallus gallus domesticus* eggs may constitute a health risk, particularly in regions (urban and suburban centers across Nigeria, and the globe) where they are consumed in high amounts as a key and cheap source of protein for young adults, infants and pregnant women (32 - 34). Eggs are available for sale in most grocery shops and open markets. Studies on the health effects of trace elements in chicken and its eggs have been carried out in different parts of the world, including China (35), the Kingdom of Saudi Arabia (36), Poland (37), Korea Republic (38), Iran (39), Togo (40), Bangladesh (32), Palestine (41), United States of America (42), amongst others. A common characteristic of the different studies is the reported presence of trace elements in poultry products, which were observed at varying concentrations, including at toxic levels. Meanwhile, there is a scarcity of information concerning the health risk of trace elements in urban areas of Nigeria. The earlier work of Fakayode and Olu-Owolabi (43) on trace metal contents of chicken eggs in Ibadan is among the most prominent literature on this study.

Whereas, similar studies are yet to be conducted for Port Harcourt metropolis. This is despite the comparable importance of dietary egg protein to the teeming population of both cities.

The level of trace metals in environmental and food samples is often compared with regulatory threshold limits stipulated by the World Health Organization, and other national agencies of different countries. However, the threshold limits do not provide sufficient information concerning the toxicity and cumulative hazard attributed to each element. As a result, the health risk assessment approach is utilized for estimating the potential hazards associated with the consumption of food containing trace elements. The health risk is often evaluated based on carcinogenic and non-carcinogenic hazards. In recent times, the adoption of a multivariate approach to study the effects of public-environmental health variables is gaining popularity. For instance, it has recently been applied in the study of trace metals in the air (44), plants (45 - 49) as well as in soil and other habitats (50 - 55). Some multivariate analyses such as cluster analysis have been applied for the study of trace metals in fish (56), water (24), and some other food products, but information about the application of multivariate analysis such as principal component analysis, cluster analysis, etc., in the study of trace elements in chicken eggs, is scarce in the literature.

The goal of this study is to determine (1) the level of trace elements in eggs of poultry (2) the public health implications of trace metal loads in poultry eggs, and (3) the relationships and sources of metals in poultry eggs sold in public areas of Port Harcourt, Rivers State, Southern Nigeria.

2. Materials and Methods

2.1. Study area

The study area is Port Harcourt, Rivers State, Southern Nigeria, which is located within latitudes 6°58'N to 7°06'N and Longitude 4°40'E to 4°55'E. The area is situated within the lowland swamp forest ecological zone, bordered by mangrove swamp forests to the East, West, and South (57, 58). Port Harcourt is the capital of Rivers State. It is an important industrial and commercial hub in Nigeria. This is because of its seaport, vast oil reserve, mineral mining activities, and population density (59). The atmospheric condition of the area is characterized by two distinct seasons (wet and dry), with temperature and relative humidity of 28 ± 10 °C and 50 - 95 % respectively throughout the year. Port Harcourt experiences heavy rainfall for about eight months (March - October), averaging 2500 mm/annum (60). Geologically, it has a flat topography that is underlined by superficial silty clay and silty sand soil, with a water table fewer than 10 m below the ground surface (59).

2.2. Sample collection

Due to the prevalence of open-market systems along both minor and major highways in Nigeria, food and food product vending points are directly exposed to vehicular emissions. Seventeen study areas were randomly selected for sampling within Port Harcourt metropolis, Rivers State, Nigeria. These include: YKC-Woji (PE1), Rumurolo (PE2), Eneka (PE3), New Odani (PE4), Igbo-Etche (PE5), Rumukwurushi Pipeline (PE6), Eliozi (PE7), Old Odani (PE8), Elelewon (PE9), Rumukwurushi Town (PE10), Tank/East-West Road (PE11), Atali (PE12), Rumuodara (PE13), Slaughter/Trans-Woji (PE14), Eliowhani (PE15),

Akpajo (PE16), Oroigwe/Elimgbu (PE17). Freshly laid poultry eggs were purchased from retailers in each of the study areas [PE1 – PE17]. At each location, samples were bought from three different retailers. Hence, a total of fifty-one poultry egg samples were obtained. Samples were stored in pre-sterilized, clean polyethylene vials and transported to the laboratory immediately. The samples were instantly pre-digested and analyzed at the laboratory.

2.3. Trace elements determination of *Gallus gallus domesticus* eggs

The eggshells were cracked with a sterile spatula, and the content was dispensed into a sterile 100 mL glass beaker. Samples were vortexed with a spatula, then approximately 20 mL of egg albumen and yolk mixture was transferred into a 100 mL glass beaker, followed by the addition of 1 ml of nitric acid (HNO₃) 69 % (v/v) and 10 mL hydrochloric acid (HCl) 37 % (v/v). The egg mixture was digested at medium heat to near-dryness before being left to cool for 30 min. The clear solution (acid-digest) was filtered into a 20 mL measuring cylinder using 125 mm diameter Whatmann's filter paper. The filtrate was made up to the 20 mL mark in the measuring cylinder using distilled water. The instrument was first calibrated using standard test solutions of the respective metals. Thereafter, filtrates were individually aspirated into the GBC 908PBMT Flame Atomic Absorption Spectrometer (FAAS). The concentration of the test elements (zinc, lead, nickel, cadmium, cobalt, manganese, iron, copper, chromium, and mercury) was concurrently extrapolated from an absorbance-concentration plot.

The concentration was expressed as mg/L. Operational conditions of the FAAS (including slit width, noise, wavelength, lamp current, and flame composition) and quality assurance considerations (including the limit of detection, the limit of quantification, quantity of standard, and percentage recovery) were carried out in line with the step previously employed by Izah et al. (44), Aigberua et al. (56, 61); Izah and Aigberua (15). After every 10 samples, the respective standard solutions were rerun to ensure deviation was within the pre-set acceptable range of 90 – 110%.

2.4. Health Hazard Assessment of *Gallus gallus domesticus* eggs

The potential of toxic elements causing harmful effects on humans is referred to as a health risk. The consumption of toxic substances from food (such as poultry eggs) and drinks often leads to health problems, which are mostly assessed using non-carcinogenic indices such as Estimated Daily Intake (EDI), also known as chronic daily intake or average daily intake, target quotient, hazard index, and carcinogenic hazard.

2.4.1. Non-carcinogenic hazards

The estimated daily intake of the egg (EDIE)

The estimated daily intake of the egg (EDIE) was determined following the method described by Hashemi et al. (2019); Aliu et al. (2021); Shaheen et al. (2016).

$$EDIE (\mu\text{g}/\text{kg}/\text{bw}/\text{day}) = (\text{LTE} \times \text{EiR})/(\text{Bw})$$

Where LTE is the level of trace elements in mg/kg; EIR is the ingestion rate of egg in g/day, and BW in kg is the body weight. Adult and children's EIR are 11.30 and

6.50 respectively, with BW of 60 and 27 respectively (62).

Target hazard quotients (THQe) and total target hazard quotients of *Gallus gallus domesticus* eggs (TTHQe): The THQe and TTHQe were determined following the method described in Hashemi et al. (2019); Shaheen et al. (2016).

$$\text{THQe} = (\text{LTE} \cdot \text{EF} \cdot \text{ED} \cdot \text{EIR}) / (\text{RFD} \cdot \text{BW} \cdot \text{AT}) \times 0.001$$

$$\text{TTHQe} = \sum \text{THQe}$$

Where EF is the exposure frequency (365 days/year) for both groups; ED is the exposure duration in years; RFD is the oral reference dose in mg/kg/bw/day; AT is the average time (365 × ED for both groups). Adult and children ED are 70 and 14, respectively, with BW of 60 and 27 respectively; RFD for lead = 0.004, nickel = 0.02, copper = 0.04, zinc = 0.3, cobalt = 0.02, manganese = 0.140, iron = 0.70 (62). When THQe and TTHQe are >1, it portends adverse health effects, while THQe <1 depicts the absence of health effects (24; 62).

2.4.2. Carcinogenic Hazard risk *Gallus gallus domesticus* eggs

The carcinogenic hazard of trace elements in eggs (CHe) reflects the probability of a person developing cancer over a lifetime of exposure. The CH was determined following the method described by Hashemi et al. (63), Shaheen et al. (62).

$$\text{CHe} = (\text{LTE} \cdot \text{EF} \cdot \text{ED} \cdot \text{EIR} \cdot \text{CSF}) / (\text{BW} \cdot \text{AT}) \times 0.001$$

The CSF for lead is 0.0085 mg/kg/day (62; 63). The allowable estimated lifetime risks for carcinogens range from 10⁻⁶ (the likelihood of 1 in 1,000,000 people acquiring cancer) to 10⁻⁴ (the probability of 1 in 1,000,000 people developing cancer) (24; 62).

2.5. Statistical Analysis

Descriptive and inferential statistics were calculated using the Statistical Package for Social Science, while Minitab 17 was utilized for statistical analysis. The information was presented as mean ± standard deviation. To show statistical differences, a one-way analysis of variance was performed at p = 0.050. Waller-Duncan test statistics were used to identify the source of differences in egg quality from the different locations. Pearson's correlation was utilized to show a significant association among the test elements. Cluster analysis using the complete linkage method was used to measure the Euclidean distance between the different locations and elements, thereby revealing dependent and independent groupings or relationships. Furthermore, principal component analysis (PCA) was used to identify sources of significant variations or outlier values among test elements.

3. Results

3.1. Composition and Concentration of trace elements in *Gallus gallus domesticus* eggs

The trace element composition and concentration in *Gallus gallus domesticus* eggs in Port Harcourt, Nigeria were assessed. Varying amounts of zinc, lead, nickel, cobalt, manganese, iron, and copper were found in the samples collected from seventeen locations across the city (Table 1). However, mercury, chromium, and cadmium were not found in the poultry eggs from the different sample locations within the Port Harcourt metropolis. Concentration ranged from 2.16 ± 0.03 to 7.43 ± 0.98 mg/L for zinc, undetected to 9.86 ± 1.01 mg/L for lead, undetected to 1.44 ± 0.30 mg/L for

nickel, undetected to 0.35 ± 0.01 mg/L for cobalt, undetected to 0.76 ± 0.04 for manganese, $2.93 \pm 1.01 - 11.38 \pm 0.53$ for iron and 0.03 ± 0.05 to 0.97 ± 0.06 for copper (Table 1). The highest concentration of zinc was recorded for PE11, closely followed by PE10 and PE4, whilst the three locations did not depict reasonable statistical deviation according to Waller Duncan's Post-hoc analysis. The highest concentration of lead, nickel, cobalt, manganese, iron, and copper was recorded for locations PE2, PE5, PE9, PE6, PE12, and PE6 respectively (Table 1). Correlative relationship analysis of the different metals revealed zinc had a significant relationship with iron and copper in all the sample sites while copper had a similar close association with manganese and iron (Supplementary Table 1). On the other hand, the characteristic concentration of lead, nickel, manganese, iron, and copper was not influenced by other trace elements recorded for the study (Supplementary Table 1).

3.2. Relationships and sources of trace metals in *Gallus gallus domesticus* eggs

Cluster analysis using the complete linkage method measured by Euclidean distance of trace elements found in *Gallus gallus domesticus* eggs based on sample location revealed three major clades with eleven compound clusters for seventeen sample locations (Fig. 1). The different clades are; clade 1 – cluster one (PE2 and PE12), clade 2 clusters two (PE16), and three (PE7 and PE17), and clade 3 clusters four (PE9), five (PE10), six (PE3 and PE11), seven (PE4 and PE6), eight (PE8), nine (PE5 and PE13), ten (PE14) to eleven (PE1 and PE5). The different distances suggest clades 1 and 2 may be closely related.

A similar analysis conducted based on the trace elements recorded from eggs in the different locations also revealed three clades (Fig. 2). However, closer relationships were observed between zinc and iron, nickel and copper, cobalt and manganese, but lead depicted no relationship with other trace elements.

Principal component analysis revealed three components (PC1, PC2, and PC3), with PC1 and PC2 accounting for more than 50 % of the observed variation (Supplementary Table 2). Together the three components accounted for 69.30 % of observed variations in trace metal composition across the different locations. Not much difference was observed in the percentage variance when the principal components were rotated but the loadings of each metal varied slightly (Supplementary Table 3).

3.3. Public health implications of trace metals in

Gallus gallus domesticus eggs

For adults and children, the result of the estimated daily intake of trace elements from *Gallus gallus domesticus* eggs in Port Harcourt suggest variable toxicity levels (Table 2). Considering the recommended dietary intake and allowance of 2000, 18, and 15 $\mu\text{g.kg/BW/day}$ for Cu, Fe, and Co respectively, it may be safe to imply that the levels observed in the study are within acceptable limits. In the study, zinc ranged from 2.16 – 7.43 mg/L. In the same vein, the estimated daily intake (EDIe) ($\mu\text{g.kg/BW/day}$) of trace elements contained in *Gallus gallus domesticus* eggs consumed by children in Port Harcourt, Nigeria is presented in Table 3. Similarly, low to no health risk observations are deductible for results obtained from total target hazard quotients (TTHQe), and carcinogenic hazard of lead, particularly for adults and children (Tables 4 and 5).

Table 1. Composition and concentration of trace elements in *Gallus gallus domesticus* eggs in Port Harcourt, Nigeria

Sample locations	Zn, mg/L	Pb, mg/L	Ni, mg/L	Co, mg/L	Mn, mg/L	Fe, mg/L	Cu, mg/L
PE1	6.35±0.36f	0.04±0.05a	0.13±0.02ab	0.00±0.00a	0.01±0.01a	6.28±0.51cde	0.85±0.00hi
PE2	4.74±0.13cde	9.86±1.01f	1.04±0.02f	0.01±0.01a	0.01±0.01a	8.02±0.49cdef	0.85±0.038hi
PE3	6.67±0.15fg	0.00±0.00a	0.00±0.00a	0.00±0.00a	0.00±0.00a	9.29±0.06fgh	0.83±0.06hi
PE4	7.33±0.09gh	4.28±0.54d	0.43±0.00c	0.13±0.06b	0.00±0.00a	8.30±1.54def	0.84±0.03hi
PE5	4.76±0.17cde	0.00±0.00a	1.44±0.30g	0.03±0.01a	0.18±0.01b	8.13±1.75cdef	0.33±0.04bcd
PE6	6.44±0.03f	4.05±0.72d	0.00±0.00a	0.01±0.02a	0.76±0.04g	6.05±0.04bcd	0.97±0.06i
PE7	3.15 ±0.01b	2.69±0.25c	0.77±0.08e	0.00±0.00a	0.00±0.00a	2.93±1.01a	0.47±0.02def
PE8	4.37±0.17cd	2.39±0.56c	0.05±0.05ab	0.00±0.00a	0.01±0.01a	8.31±0.58ef	0.33±0.02bcd
PE9	5.43±0.07e	3.88±0.11d	0.01±0.01ab	0.35±0.01c	0.68±0.01f	11.38±0.53h	0.25±0.02bc
PE10	7.34±0.02gh	0.00±0.00a	0.02±0.03ab	0.00±0.00a	0.01±0.02a	11.21±0.58gh	0.71±0.14gh
PE11	7.43±0.98h	0.10±0.17a	0.17±0.04b	0.15±0.02b	0.59±0.01e	8.30±2.09def	0.36±0.33cde
PE12	6.35±0.01f	8.67±0.10e	0.00±0.00ab	0.01±0.01a	0.00±0.00a	11.52±1.54h	0.16±0.01ab
PE13	5.10±0.57de	0.10±0.18a	0.53±0.03cd	0.00±0.00a	0.47±0.02d	9.04±1.04fg	0.03±0.05a
PE14	4.66±0.57cd	0.08±0.13a	0.04±0.04ab	0.00±0.00a	0.25±0.05c	5.88±1.35bc	0.56±0.02fg
PE15	6.69±0.54fg	0.49±0.23ab	0.61±0.01de	0.00±0.00a	0.02±0.03a	6.15±1.20cde	0.55±0.04rfg
PE16	4.05±0.02c	0.01±0.01a	0.64±0.03de	0.00±0.00a	0.16±0.02b	3.87±1.08ab	0.15±0.05ab
PE17	2.16±0.03a	1.26±0.02b	0.01±0.02ab	0.00±0.00a	0.60±0.08e	3.81±0.66ab	0.255±0.01bc

Keys:

Data are presented as mean ± standard deviation (n=3). The same letters along the column indicate no statistical deviation according to Waller Duncan's statistics

Zn = Zinc, Pb = Lead, Ni = Nickel, Co = Cobalt, Mn = Manganese, Fe = Iron and Cu = Copper

Woji (PE1), Rumurolo (PE2), Eneka (PE3), New Odani (PE4), Igbo - Etche (PE5), Rumukwurushi pipeline (PE6), Eliozu (PE7), Old Odani (PE8), Elelenwon (PE9), Rumukwurushi town (PE10), Tank/East-West road (PE11), Atali (PE12), Rumuodara (PE13), Slaughter (PE14), Eliowhani (PE15), Akpajo (PE16), Oroigwe/Elingbu (PE17)

Supplementary Table 1. Pearson correlation of trace element in *Gallus gallus domesticus* eggs in Port Harcourt, Nigeria

Parameters	Zn	Pb	Ni	Co	Mn	Fe	Cu
Zn	1						
Pb	-0.00	1					
Ni	-0.25	0.07	1				
Co	0.22	0.11	-0.15	1			
Mn	-0.11	-0.12	-0.27	0.45**	1		
Fe	0.54**	0.26	-0.24	0.36**	-0.05	1	
Cu	0.45**	0.14	-0.09	-0.14	-0.24	-0.05	1

Keys:

Data are presented as mean ± standard deviation (n=3). The same letters along the column indicate no statistical deviation according to Waller Duncan statistics

Zn = Zinc, Pb = Lead, Ni = Nickel, Co = Cobalt, Mn = Manganese, Fe = Iron and Cu = Copper

** . Statistical association at $p=0.01$; n=3, N=51

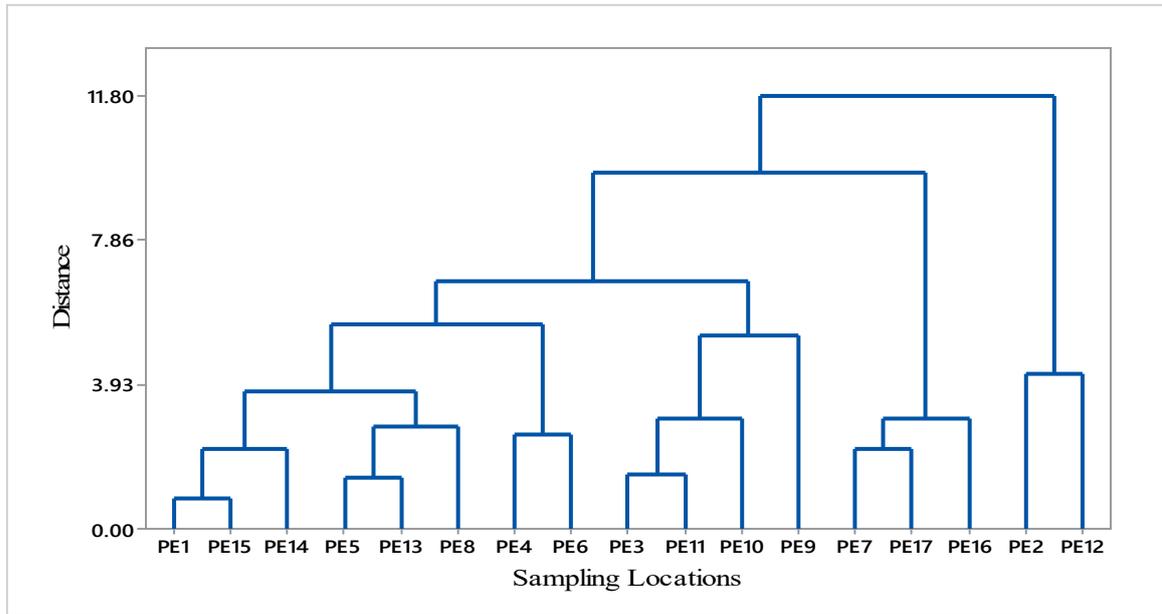


Figure 1. Cluster analysis using complete linkage method measured by Euclidean distance of trace elements found in the *Gallus gallus domesticus* eggs across vendor locations in Port Harcourt, Nigeria

Keys:

Woji (PE1), Rumurolo (PE2), Eneka (PE3), New Odani (PE4), Igbo-Etche (PE5), Rumukwurushi pipeline (PE6), Elioizu (PE7), Old Odani (PE8), Elelenwon (PE9), Rumukwurushi Twn (PE10), Tank/East-West Road (PE11), Atali (PE12), Rumuodara (PE13), Slaughter (PE14), Eliowhani (PE15), Akpajo (PE16), Oroigwe/Elimgbu (PE17)

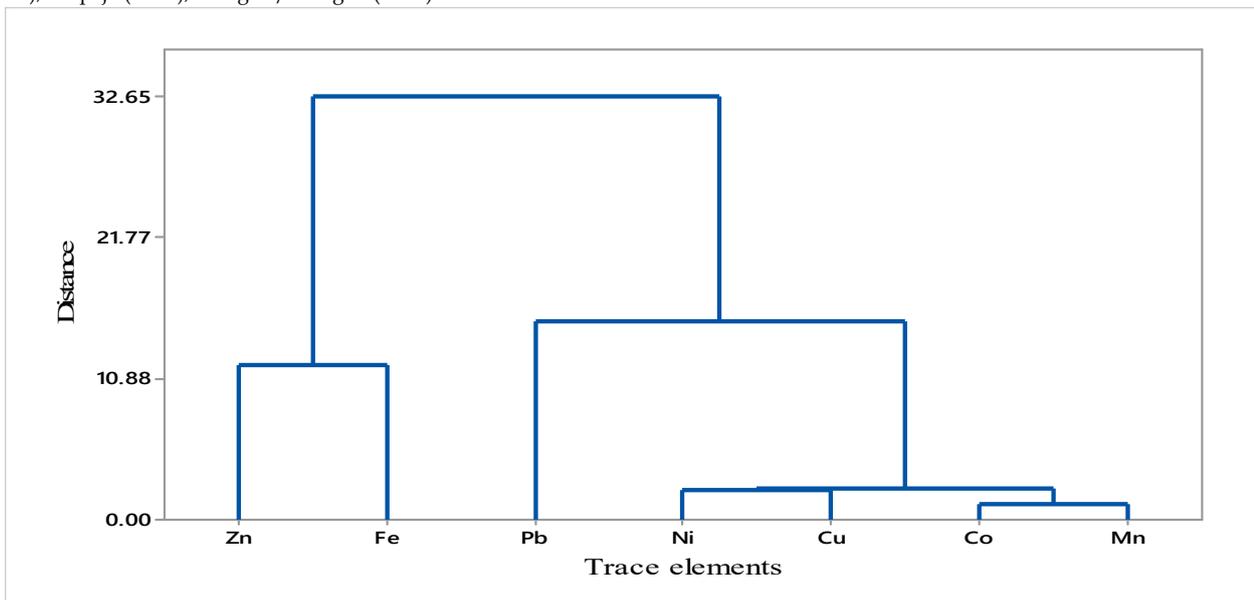


Figure 2. Cluster analysis using complete linkage method measured by Euclidean distance of trace elements in poultry eggs sold in Port Harcourt, Nigeria

Keys:

Zn = Zinc, Pb = Lead, Ni = Nickel, Co = Cobalt, Mn = Manganese, Fe = Iron and Cu = Copper

Supplementary Table 2. Principal component analysis of trace element in *Gallus gallus domesticus* eggs sold in Port Harcourt, Nigeria

Parameters	Principal Component		
	1	2	3
Zn	0.77	0.40	-0.24
Pb	0.24	0.24	0.74
Ni	-0.53	0.21	0.50
Co	0.61	-0.53	0.23
Mn	0.20	-0.81	-0.18
Fe	0.79	0.065	0.32
Cu	0.25	0.69	-0.34
Total	2.02	1.67	1.16
% of Variance	28.79	23.90	16.62
Cumulative %	28.78	52.68	69.30

Keys:

Zn = Zinc, Pb = Lead, Ni = Nickel, Co = Cobalt, Mn = Manganese, Fe = Iron and Cu = Copper

Supplementary Table 3. Rotated Principal component analysis of trace element in *Gallus gallus domesticus* eggs sold in Port Harcourt, Nigeria

Parameters	Principal Component		
	1	2	3
Zn	0.88	0.02	0.20
Pb	-0.07	-0.12	0.81
Ni	-0.55	-0.44	0.26
Co	0.13	0.75	0.35
Mn	-0.11	0.81	-0.24
Fe	0.48	0.31	0.63
Cu	0.65	-0.48	-0.05
Total	1.76	1.75	1.34
% of Variance	25.16	24.99	19.15
Cumulative %	25.16	50.15	69.30

Keys:

Zn = Zinc, Pb = Lead, Ni = Nickel, Co = Cobalt, Mn = Manganese, Fe = Iron and Cu = Copper

Table 2. Estimated daily intake (EDIE) ($\mu\text{g.kg/BW/ day}$) of trace elements contained in *Gallus gallus domesticus* eggs consumed by adults in Port Harcourt, Nigeria

Sampling locations	Zn	Pb	Ni	Co	Mn	Fe	Cu
PE1	1.20E+00	6.53E-03	2.50E-02	0.00E+00	1.88E-03	1.18E+00	1.60E-01
PE2	8.93E-01	1.86E+00	1.96E-01	1.32E-03	1.44E-03	1.51E+00	1.59E-01
PE3	1.26E+00	0.00E+00	0.00E+00	0.00E+00	0.00E+00	1.75E+00	1.56E-01
PE4	1.38E+00	8.05E-01	8.12E-02	2.44E-02	0.00E+00	1.56E+00	1.57E-01
PE5	8.97E-01	0.00E+00	2.72E-01	6.34E-03	3.44E-02	1.53E+00	6.12E-02
PE6	1.21E+00	7.62E-01	0.00E+00	2.01E-03	1.42E-01	1.14E+00	1.82E-01
PE7	5.93E-01	5.07E-01	1.44E-01	0.00E+00	0.00E+00	5.52E-01	8.85E-02
PE8	8.23E-01	4.50E-01	9.04E-03	0.00E+00	1.32E-03	1.56E+00	6.27E-02
PE9	1.02E+00	7.30E-01	1.32E-03	6.64E-02	1.28E-01	2.14E+00	4.63E-02
PE10	1.38E+00	0.00E+00	2.83E-03	0.00E+00	2.01E-03	2.11E+00	1.34E-01
PE11	1.40E+00	1.92E-02	3.13E-02	2.91E-02	1.11E-01	1.56E+00	6.79E-02
PE12	1.20E+00	1.63E+00	0.00E+00	8.80E-04	0.00E+00	2.17E+00	3.06E-02
PE13	9.60E-01	1.95E-02	9.97E-02	0.00E+00	8.93E-02	1.70E+00	6.40E-03
PE14	8.78E-01	1.44E-02	7.66E-03	0.00E+00	4.77E-02	1.11E+00	1.05E-01
PE15	1.26E+00	9.31E-02	1.15E-01	4.39E-04	4.46E-03	1.16E+00	1.03E-01
PE16	7.63E-01	2.45E-03	1.21E-01	0.00E+00	2.98E-02	7.30E-01	2.73E-02
PE17	4.07E-01	2.36E-01	1.88E-03	0.00E+00	1.13E-01	7.17E-01	4.61E-02

Keys:

Zn = Zinc, Pb = lead, Ni = nickel, Co = cobalt, Mn = manganese, Fe = iron and Cu = copper

Woji (PE1), Rumurolo (PE2), Eneka (PE3), New Odani (PE4), Igbo-Etche (PE5), Rumukwurushi pipeline (PE6), Eliozu (PE7), Old Odani (PE8), Elelenwon (PE9), Rumukwurushi town (PE10), Tank/East-West Road (PE11), Atali (PE12), Rumuodara (PE13), Slaughter (PE14), Eliowhani (PE15), Akpajo (PE16), Oroigwe/Elimgbu (PE17)

Table 3. Estimated daily intake (EDIe) ($\mu\text{g.kg/BW/ day}$) of trace elements contained in *Gallus gallus domesticus* eggs consumed by children in Port Harcourt, Nigeria

	Zn	Pb	Ni	Co	Mn	Fe	Cu
PE1	1.53E+00	8.35E-03	3.19E-02	0.00E+00	2.41E-03	1.51E+00	2.05E-01
PE2	1.14E+00	2.37E+00	2.51E-01	1.69E-03	1.85E-03	1.93E+00	2.04E-01
PE3	1.61E+00	0.00E+00	0.00E+00	0.00E+00	0.00E+00	2.24E+00	2.00E-01
PE4	1.76E+00	1.03E+00	1.04E-01	3.12E-02	0.00E+00	2.00E+00	2.01E-01
PE5	1.15E+00	0.00E+00	3.48E-01	8.11E-03	4.40E-02	1.96E+00	7.82E-02
PE6	1.55E+00	9.74E-01	0.00E+00	2.57E-03	1.82E-01	1.46E+00	2.33E-01
PE7	7.58E-01	6.48E-01	1.84E-01	0.00E+00	0.00E+00	7.05E-01	1.13E-01
PE8	1.05E+00	5.75E-01	1.16E-02	0.00E+00	1.69E-03	2.00E+00	8.01E-02
PE9	1.31E+00	9.34E-01	1.69E-03	8.49E-02	1.63E-01	2.74E+00	5.91E-02
PE10	1.77E+00	0.00E+00	3.61E-03	0.00E+00	2.57E-03	2.70E+00	1.72E-01
PE11	1.79E+00	2.46E-02	4.00E-02	3.72E-02	1.42E-01	2.00E+00	8.67E-02
PE12	1.53E+00	2.09E+00	0.00E+00	1.12E-03	0.00E+00	2.77E+00	3.91E-02
PE13	1.23E+00	2.49E-02	1.27E-01	0.00E+00	1.14E-01	2.18E+00	8.19E-03
PE14	1.12E+00	1.85E-02	9.79E-03	0.00E+00	6.10E-02	1.42E+00	1.34E-01
PE15	1.61E+00	1.19E-01	1.47E-01	5.61E-04	5.70E-03	1.48E+00	1.32E-01
PE16	9.75E-01	3.13E-03	1.54E-01	0.00E+00	3.81E-02	9.33E-01	3.49E-02
PE17	5.20E-01	3.02E-01	2.41E-03	0.00E+00	1.45E-01	9.16E-01	5.90E-02

Keys:

Zn = Zinc, Pb = lead, Ni = Nickel, Co = Cobalt, Mn = Manganese, Fe = Iron and Cu = Copper

Woji (PE1), Rumurolu (PE2), Eneka (PE3), New Odani (PE4), Igbo-Etche (PE5), Rumukwurushi pipeline (PE6), Elioizu (PE7), Old Odani (PE8), Elelenwon (PE9), Rumukwurushi town (PE10), Tank/East-West Road (PE11), Atali (PE12), Rumuodara (PE13), Slaughter (PE14), Eliowhani (PE15), Akpajo (PE16), Oroigwe/Elimgbu (PE17)

Table 4. Carcinogenic hazard of lead and total target hazard quotients (TTHQe) of trace elements in *Gallus gallus domesticus* eggs consumed by adults in Port Harcourt, Nigeria

Sampling locations	Zn	Pb	Ni	Co	Mn	Fe	Cu	TTHQe	Carcinogenic hazard
PE1	3.99E-03	1.63E-03	1.25E-03	0.00E+00	1.35E-05	1.69E-03	4.01E-03	1.26E-02	5.55E-08
PE2	2.98E-03	4.64E-01	9.80E-03	6.59E-05	1.03E-05	2.16E-03	3.98E-03	4.83E-01	1.58E-05
PE3	4.19E-03	0.00E+00	0.00E+00	0.00E+00	0.00E+00	2.50E-03	3.91E-03	1.06E-02	0.00E+00
PE4	4.60E-03	2.01E-01	4.06E-03	1.22E-03	0.00E+00	2.23E-03	3.93E-03	2.17E-01	6.85E-06
PE5	2.99E-03	0.00E+00	1.36E-02	3.17E-04	2.46E-04	2.19E-03	1.53E-03	2.09E-02	0.00E+00
PE6	4.05E-03	1.90E-01	0.00E+00	1.00E-04	1.02E-03	1.63E-03	4.55E-03	2.02E-01	6.47E-06
PE7	1.98E-03	1.27E-01	7.20E-03	0.00E+00	0.00E+00	7.88E-04	2.21E-03	1.39E-01	4.31E-06
PE8	2.74E-03	1.13E-01	4.52E-04	0.00E+00	9.42E-06	2.24E-03	1.57E-03	1.20E-01	3.83E-06
PE9	3.41E-03	1.83E-01	6.59E-05	3.32E-03	9.13E-04	3.06E-03	1.16E-03	1.94E-01	6.21E-06
PE10	4.61E-03	0.00E+00	1.41E-04	0.00E+00	1.44E-05	3.02E-03	3.36E-03	1.11E-02	0.00E+00
PE11	4.67E-03	4.80E-03	1.57E-03	1.45E-03	7.93E-04	2.23E-03	1.70E-03	1.72E-02	1.63E-07
PE12	3.99E-03	4.08E-01	0.00E+00	4.40E-05	0.00E+00	3.10E-03	7.64E-04	4.16E-01	1.39E-05
PE13	3.20E-03	4.87E-03	4.98E-03	0.00E+00	6.38E-04	2.43E-03	1.60E-04	1.63E-02	1.65E-07
PE14	2.93E-03	3.61E-03	3.83E-04	0.00E+00	3.41E-04	1.58E-03	2.61E-03	1.15E-02	1.23E-07
PE15	4.20E-03	2.33E-02	5.73E-03	2.19E-05	3.18E-05	1.65E-03	2.58E-03	3.75E-02	7.91E-07
PE16	2.54E-03	6.12E-04	6.04E-03	0.00E+00	2.13E-04	1.04E-03	6.83E-04	1.11E-02	2.08E-08
PE17	1.36E-03	5.91E-02	9.42E-05	0.00E+00	8.09E-04	1.02E-03	1.15E-03	6.35E-02	2.01E-06

Keys:

Zn = Zinc, Pb = Lead, Ni = Nickel, Co = Cobalt, Mn = Manganese, Fe = Iron and Cu = Copper; Woji (PE1), Rumurolo (PE2), Eneka (PE3), New Odani (PE4), Igbo- Etche (PE5), Rumukwurushi pipeline (PE6), Elioizu (PE7), Old Odani (PE8), Elenenwon (PE9), Rumukwurushi town (PE10), Tank/East-West road (PE11), Atali (PE12), Rumuodara (PE13), Slaughter (PE14), Eliowhani (PE15), Akpajo (PE16), Oroigwe/Elimgbu (PE17).

Table 5. Carcinogenic hazard of lead and total target hazard quotients (TTHQe) of trace elements in *Gallus gallus domesticus* eggs consumed by children in Port Harcourt, Nigeria.

Sampling locations	Zn	Pb	Ni	Co	Mn	Fe	Cu	TTHQe	Carcinogenic hazard of Pb
PE1	4.59E-04	1.88E-04	1.44E-04	0.00E+00	1.55E-06	1.94E-04	4.62E-04	1.45E-03	6.39E-09
PE2	3.43E-04	5.34E-02	1.13E-03	7.58E-06	1.19E-06	2.48E-04	4.58E-04	5.56E-02	1.82E-06
PE3	4.82E-04	0.00E+00	0.00E+00	0.00E+00	0.00E+00	2.88E-04	4.50E-04	1.22E-03	0.00E+00
PE4	5.29E-04	2.32E-02	4.67E-04	1.40E-04	0.00E+00	2.57E-04	4.52E-04	2.50E-02	7.87E-07
PE5	3.44E-04	0.00E+00	1.56E-03	3.65E-05	2.83E-05	2.52E-04	1.76E-04	2.40E-03	0.00E+00
PE6	4.65E-04	2.19E-02	0.00E+00	1.16E-05	1.17E-04	1.87E-04	5.24E-04	2.32E-02	7.45E-07
PE7	2.27E-04	1.46E-02	8.28E-04	0.00E+00	0.00E+00	9.07E-05	2.54E-04	1.60E-02	4.96E-07
PE8	3.15E-04	1.29E-02	5.20E-05	0.00E+00	1.08E-06	2.57E-04	1.80E-04	1.37E-02	4.40E-07
PE9	3.92E-04	2.10E-02	7.58E-06	3.82E-04	1.05E-04	3.52E-04	1.33E-04	2.24E-02	7.14E-07
PE10	5.30E-04	0.00E+00	1.63E-05	0.00E+00	1.65E-06	3.47E-04	3.87E-04	1.28E-03	0.00E+00
PE11	5.37E-04	5.53E-04	1.80E-04	1.67E-04	9.12E-05	2.57E-04	1.95E-04	1.98E-03	1.88E-08
PE12	4.59E-04	4.70E-02	0.00E+00	5.06E-06	0.00E+00	3.57E-04	8.79E-05	4.79E-02	1.60E-06
PE13	3.68E-04	5.60E-04	5.73E-04	0.00E+00	7.34E-05	2.80E-04	1.84E-05	1.87E-03	1.90E-08
PE14	3.37E-04	4.15E-04	4.41E-05	0.00E+00	3.92E-05	1.82E-04	3.01E-04	1.32E-03	1.41E-08
PE15	4.83E-04	2.68E-03	6.59E-04	2.52E-06	3.66E-06	1.90E-04	2.97E-04	4.31E-03	9.10E-08
PE16	2.93E-04	7.04E-05	6.95E-04	0.00E+00	2.45E-05	1.20E-04	7.85E-05	1.28E-03	2.39E-09
PE17	1.56E-04	6.80E-03	1.08E-05	0.00E+00	9.31E-05	1.18E-04	1.33E-04	7.31E-03	2.31E-07

Keys:

Zn = Zinc, Pb = Lead, Ni = Nickel, Co = Cobalt, Mn = Manganese, Fe = Iron and Cu = Copper

Woji (PE1), Rumurolu (PE2), Eneka (PE3), New Odani (PE4), Igbo- Etche (PE5), Rumukwurushi pipeline (PE6), Eliozu (PE7), Old Odani (PE8), Elemenwon (PE9), Rumukwurushi town (PE10), Tank/East-West Road (PE11), Atali (PE12), Rumuodara (PE13), Slaughter (PE14), Eliowhani (PE15), Akpajo (PE16), Oroigwe/Elimgbu (PE17)

4. Discussion

The evaluation of trace element composition and concentration in *Gallus gallus domesticus* eggs and the health risks associated with their consumption in Port Harcourt have been assessed. Generally, results suggest little to no toxic levels of trace elements in the eggs. However, it remains to be seen if bioaccumulation from consumption over time may portend health risks. Trace element composition may change over time, especially concerning environmental, economic, and social indices. This was presented in a study by Marelli et al. (64), where the physicochemical properties of eggs were reported to be dynamic; being influenced by external factors. This includes production systems and consumer demand (64). It is known that poultry eggs are highly susceptible to trace metal toxicity which may potentially have diverse impacts (38). However, some trace elements may remain undetectable in eggs. The range and significance level of trace elements found in the present study (Table 1) is indicative of in-feed trace metal levels in diets, especially for the egg-laying *Gallus gallus domesticus*. Trace metals at high concentrations are known to cause effects such as hepatic dysfunction, liver inflammation, and increased metabolic activities in both humans and animals (13; 23). One way to reduce the load and effects of these in-feed trace elements is by quercetin supplementation. The work of Zoidis et al. (65) on dietary supplementation for *Gallus gallus domesticus* revealed promising results as quercetin supplementation led to an altered elemental profile in eggs. The author further opined that the presence of quercetin led to differentiation in the deposition of trace elements in *Gallus gallus domesticus* eggs.

Apart from the correlative relationship between some trace elements assessed in the study, our results suggest the metals depict divergent sources, indicating mutual independence and similarity in behavior during transport. From Fig. 1 and 2, two major clusters were formed. Closer clusters between variables indicate direct similarity among variables being compared. The near-distance cluster depicts a statistical connection, while distant clusters indicate strong disassociation of the variables. The dissimilarity clusters may be connected to the high concentration of trace metals. From Tables 3 and 4, the statistical deviation in the concentration of trace metals contained in eggs from different locations is responsible for the variance and slight spreading observed. The trace metals (zinc, lead, nickel, cobalt, manganese, iron, and copper) content of eggs could be influenced by the level of metallic ions in feeds, water, and antibiotics used for rearing chickens. In addition, ions adhere to feeding trays and atmospheric deposition. Thus, metals could emanate from natural and human activities.

Results obtained in this study agree with that of Hashemi et al. (63) from Iran wherein they reported non-carcinogenic risk from the consumption of eggs. This is due to the low estimated and tolerable daily intake loads of trace elements found in egg varieties from the present study. By extension, this study suggests that Port Harcourt residents are likely not to experience adverse health effects or cancer risks from egg consumption. Despite this, low-concentration exposure over a long period may portend toxic effects. However, our findings disagree with those of Shaheen et al. (62) that found toxic levels of nickel, copper, lead, etc., in eggs from Bangladesh. Contrary to the results from the present study, Shaheen et al. (62) reported that

the toxic levels of trace elements from the consumption of eggs are likely to portend carcinogenic and non-carcinogenic health effects. Also, eggs of other animals like blue mussels, leatherback turtles, and wild ducks have been reported to contain toxic levels of diverse trace metals (66 - 68). Therefore, the eggs of domesticated and non-domesticated animals may be potentially dangerous for human consumption. Furthermore, this implies that there are multiple sources of heavy metal assimilation into *Gallus gallus domesticus* eggs other than their feed. Apart from the toxic levels of trace elements in eggs, eggs are known to contain cholesterol. Hence, if over-consumed, may lead to the development or aggravation of cardiovascular diseases. Kuang et al. (69) reported that egg consumption can potentially lead to elevated cholesterol absorption and the disruption of cholesterol homeostasis that is hitherto regulated by endogenous biosynthesis, dietary intakes, and breakdown through utilization and excretion.

Long-term monitoring and the institution of relevant policies to regulate chicken feed composition are necessary. Analysis of the physicochemical properties of chicken feed (starter, grower, and layer) have revealed significant levels of trace elements, including chromium, cobalt, lead, manganese, and nickel. Consequently, these elements are biomagnified into edible meat and eggs of poultry birds (36). Due to the short lifespan of chickens, the full physiological, growth, development, and metabolic effects might not show up until the birds are sacrificed.

The modern trend of adding metal additive to bird feeds also contributes to the potential composition and concentration effects (70). As revealed through this study, and the work of Lopez-Alonso (71), presently, the most important issue is with finding the right balance between identifying trace minerals needed by livestock, and determination of their toxic or lethal doses. This assertion is supported by Perello et al. (72, 73) as shown by the importance that was attached to the idea of determining acceptable trace elements exposure and dietary limits.

5. Conclusion

In conclusion, the health risks posed by the contamination of agricultural products, whether by inorganic, organic, or microbiological micro-macro pollutants, continue to be a cause of concern around the world. In this study, the projected daily metal intake from egg-eating in Nigeria was found to be within the regulatory requirements, assuaging concerns about metal toxicity in persons who consume it. Additionally, the results of TTHQ suggest there may be no risk to one's health, including no carcinogenic risk as this was within acceptable limits. However, the metals in these eggs must be examined regularly to avoid any potential health risks.

Conflict of interest

The author declares that there is no conflict of interest.

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