

Residues of some Toxic Heavy Metals and Trace Elements in Chicken Eggs

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Abstract

Presence of residues of heavy metals and trace elements in chicken eggs represent chemical hazards from the point of view of both Food safety sector due to consumption of contaminated eggs and poultry medicine sector due to the possible harmful effects of metals on chicken embryos, embryonic deaths, in particular. The objective of this study was to estimate the residual concentrations of the toxic metals (lead, cadmium, arsenic and nickel) and trace elements (zinc and copper) in three kinds of chicken eggs. The examined chicken eggs included balady (produced from free-range house fed chickens), commercial eggs, produced from laying hens fed on commercial ration and organic eggs produced from laying hens fed on organic based ration. A total of 60 fresh egg samples were collected randomly from (20, each) during the period from June to October 2016 Sharkia Governorate, Egypt. The achieved results declared that balady chicken eggs had the highest load of all metals examined followed by commercial and finally organic eggs. The residual concentrations of all examined metals exceeded the maximum permissible limits set by Food and Agricultural Organization (FAO)/ World Health Organization (WHO), especially in the balady {Pb (0.34 ± 0.03); Cd (0.18 ± 0.02); As (0.14 ± 0.017); Zn (23.17 ± 0.88); Ni (1.31 ± 0.08), and Cu (11.47 ± 0.66)}, followed by commercial then organic eggs. High metal concentrations in chicken eggs represent major hazards on both human and chicken embryos that might lead to sever toxicological implications. The adverse outcomes of the examined metals on both human and chicken embryos were discussed.

Keywords: Heavy Metals, Trace Elements, Chicken Eggs.

Introduction

Eggs as a food matrix represent a major source of animal-derived protein, minerals and vitamins. Eggs, especially produced by free-range chicken are considered ideal samples for monitoring environmental pollutants because chickens live under the same environment as humans [1]. Heavy metals and trace elements are found in the environment as a result of geochemical changes or due to anthropogenic activities. Some of the trace elements are needed to certain limit by living organisms to maintain normal body functions such as zinc (Zn) and copper (Cu). Non-essential elements such as lead (Pb), cadmium (Cd), arsenic (As) and nickel (Ni) have well-documented toxic effects. Monitoring of such contaminants is a matter of importance for risk assessment of both animals and humans [2].

Chickens, either free-range or under intensive rearing systems, are exposed to vast array of heavy metals and trace elements via inhalation of polluted air or ingestion of contaminated feed. Poultry feed contains high

concentrations of minerals which is directly added to the ration to fulfill the bird body requirements and in many cases, exceed the bird needs. Additionally, poultry ration in many cases contain bone or fish meals, which are considered major sources of feed contamination with metals. Some of these metals can cross the egg shell and even represent a risk factor for the chicken embryos [3]. Estimation of metal concentrations in chicken eggs is an important subject, which is related to food safety, poultry medicine and an environmental alarming issue. This study was undertaken to estimate the concentrations of four toxic metals (lead, cadmium, arsenic and nickel) and two trace elements (zinc and copper) in balady, commercial and organic chicken eggs and compare these concentrations with the standard national permissible limits.

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Materials and Methods

Collection of samples

A total of 60 fresh egg samples were collected randomly from balady, commercial and organic eggs (20, each). Balady egg samples were collected from free-range chickens (house-fed) at villages of Sharkia Governorate, Egypt, during the period from June to October 2016. Commercial eggs were collected from laying hens' farms (depend on commercial layers' ration), while organic eggs were produced from laying hens fed on organic based rations.

Sample preparation and extraction

Heavy-metal levels in the examined eggs were measured according to Wet digestion method [4]. In summary, egg white and yolk were mixed and homogenized together. Samples were firstly dried at 70°C to get constant weight. Then, one gram of each sample was mixed with 10 mL 3:2 HNO₃ (65%v/v): HC₁O₄ (70%v/v). The mixture was digested for 5 h in water bath at 70°C with swirling at 30 min intervals to ensure complete digestion. After cooling, the digest was transferred into 15 mL falcon tubes and stored at room temperature until metal analyses.

Measurement of heavy metal concentrations

All reagents were analytical grade and standard solutions of Zn, Cu, Pb, Cd, As and

Ni were purchased from Merck, Darmstadt, Germany. Metal concentrations were directly measured by using an atomic absorption spectrophotometer (PerkinElmer 2380), using hollow cathode lamps, equipped with air-acetylene flame. Mean recoveries for all metals ranged from 95 to 105 %. The detection limits (ppm) were 0.05 (Pb), 0.005 (Cd), 0.02(As), 0.01 (Ni) 0.1 (Zn), and 0.05 (Cu), respectively. Samples and blank solutions were measured in triplicates.

Statistical analysis

Statistical significance was evaluated using Tukey–Kramer honestly significant difference tests, with $p < 0.05$ considered as significant (JMP program, SAS Institute, Cary, NC, USA) [5].

Results and Discussion

Estimation of heavy metals in eggs gives an indication about chicken exposure to metals during their lifetime. Chicken exposure to heavy metals and trace elements is due to ingestion of repeated small doses of such chemicals via feed and water. These metals can pass to the egg representing a risk factor for both chicken embryos in hatching eggs and humans who consume contaminated table eggs. In this study, we estimated the concentration of toxic metals (Pb, Cd, As and Ni) and trace elements (Zn and Cu) in whole egg.

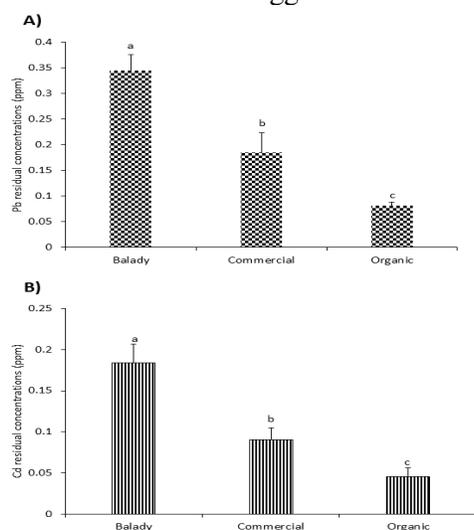


Figure 1: Lead and cadmium residual concentrations in chicken egg; A) Lead (Pb); B) Cadmium (Cd). The results were recorded as mean \pm SE (ppm/wet weight) of Pb and Cd concentrations in chicken egg (n=20 each of balady, commercial and organic eggs). Means carrying different small letter are statistically significant among the examined chicken egg type. (P < 0.05).

Lead (Pb)

Balady eggs had significantly ($P < 0.05$) the highest Pb concentrations followed by commercial and organic eggs. The mean \pm SE values of Pb concentrations in the examined egg samples were 0.34 ± 0.03 , 0.18 ± 0.04 and 0.08 ± 0.007 ppm in balady, commercial and organic eggs, respectively (Figure 1A). The recorded concentrations in this study were comparable with Pb concentrations found in chicken eggs in Egypt (Giza) and Bangladesh [6,7]. However, higher Pb concentration (0.42 ppm) was reported in Malaysia [8].

Lead concentrations recorded in this study exceeded FAO/WHO [9] recommended concentration (0.1 ppm) in 85%, 30% and 10% of the examined balady, commercial and organic eggs, respectively (Figure 4). Excess lead in food can cause serious damage to the brain, kidneys, nervous system and red blood cells in humans [10]. Food poisoning due to lead intoxication is recently reported in

children in Nigeria, Zambia and China [11-13]. When lead finds the way to the chicken body, it accumulates in the bone, egg (shell, yolk and albumen), soft tissues such as liver and kidneys and leads to elevated blood lead levels [14]. The recorded concentrations of Pb residues in the egg in this study is very close to the toxic concentrations of Pb (1 ppm) that lead to Pb intoxication in chicken. When chicken receive such concentrations of Pb in their diet, signs of depression, poor growth rate and food conversion ratio occur. Acute lead poisoning in chickens may develop muscle weakness, ataxia, and loss of appetite, followed by marked weight loss, eventual cessation of egg production and severe anemia may develop. Young chickens are more susceptible than adult chickens. Chronic Pb toxicity in chicken leads to severe muscle atrophy, degeneration of neurons and lower immune response [15].

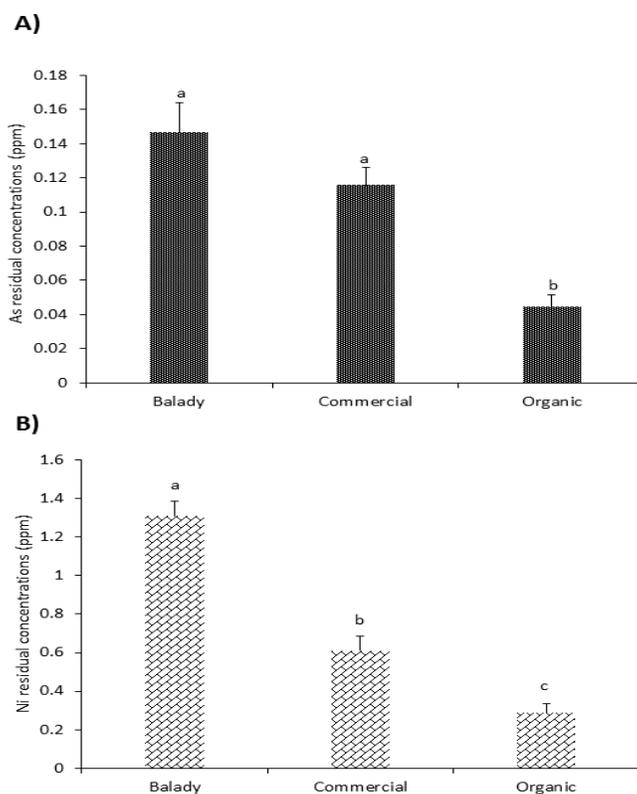


Figure 2: Arsenic and nickel residual concentrations in chicken egg; A) Arsenic (As); B) Nickel (Ni). The results were recorded as mean \pm SE (ppm/wet weight) of As and Ni concentrations in chicken egg (n=20 each of balady, commercial and organic eggs). Means carrying different small letter are statistically significant among the examined chicken egg type. ($P < 0.05$).

Cadmium (Cd)

Cadmium contamination of environment and food chain is usually due to the abuse of Cd in industry of batteries, pigments and stabilizers. In this study, the mean Cd concentrations recorded in the examined egg samples were 0.18 ± 0.02 , 0.09 ± 0.01 and 0.04 ± 0.01 ppm in balady, commercial and organic eggs, respectively. Similar to Pb, Cd concentrations were significantly higher in balady eggs followed by commercial and organic eggs (Figure 1B). The recorded concentrations in this study are in agreement with the previous reports in Malaysia and Egypt [6,8]. Unlikely, higher Cd concentration (0.3 ppm) was recorded in Bangladesh [7]. Nisianakis et al. [16] recorded lower Cd concentrations (1.44 ± 0.138 ppb) in chicken eggs marketed in Greece.

Comparing the obtained concentrations in this study with the allowed Maximum Permissible Limits (MPL) of Cd in eggs decided by FAO/WHO (0.1 ppm), it is clear that 40% and 20% of the examined balady and commercial eggs exceeded that limit (Figure 4). Cadmium is known by its severe toxicity and long-term exposure to Cd is associated with kidney malfunction, mutations, cancers and embryonic deaths [17]. The outbreak of Itai-Itai disease in Cd-polluted areas in Japan is an evidence for the toxic effects of Cd on the bones. The major symptoms of Itai-Itai disease patients include severe osteomalacia accompanied with multiple bone fractures and renal dysfunction [18]. In chicken, Cadmium is reported to cause thyroid dysfunction with concomitant increase of lipid peroxidation in chicken after oral exposure to 2.5 mg/kg daily for 15 days [19].

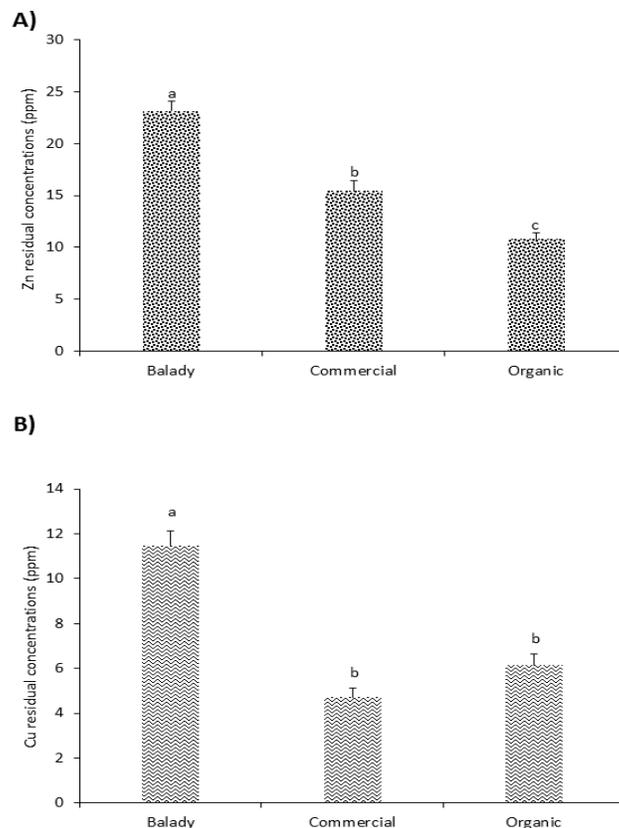


Figure 3: Zinc and Copper residual concentrations in chicken egg; A) Zinc (Zn); B) Copper (Cu). The results were recorded as mean \pm SE (ppm/wet weight) of Zn and Cu concentrations in chicken egg (n=20 each of balady, commercial and organic eggs). Means carrying different small letter are statistically significant among the examined chicken egg type. (P < 0.05).

Arsenic (As)

Chicken receives several arsenical compounds supplied normally to the ration for the control of diseases and improvement of the body gain and feed conversion ratio. These arsenical compounds are mostly excreted unchanged in chicken litter and subsequently may contaminate the produced eggs [20]. The results recorded in Figure 2A showed the mean residual concentrations of As (ppm) in the examined chicken egg samples. Balady and commercial eggs had significantly higher mean concentrations of As (0.14 ± 0.017 and 0.12 ± 0.01) compared with organic eggs (0.04 ± 0.006). The recorded concentrations of As in commercial eggs in this study are in agreement with that reported in Greece [16]. Higher concentrations of As were recorded in chicken eggs marketed in Malaysia (0.3 ppm) and Bangladesh (0.3 ppm) [7,8]. Lower values were reported in commercial eggs marketed in Giza, Egypt (0.033 ppm) [6]. Twenty-five

percentage of balady eggs (Figure 4) exceeded MPL of As (0.1 ppm) established by FAO/WHO [9].

Acute or subacute arsenic exposure might induce gastrointestinal disturbances ranging from mild abdominal cramping and diarrhea to severe life-threatening hemorrhagic gastroenteritis associated with shock. Chronic exposure to arsenic is related to skin lesions, cancers, cardiovascular effects, diabetes, pulmonary malfunction, neurological symptoms, and developmental and reproductive toxicity [21]. Arsenic effects in chicken were investigated after dietary exposure of chicken to elevated concentrations of arsenic trioxide (7.5-30 mg/kg). Arsenic triggered chicken defense and produced nervous disorders represented by induction of the expression of inflammatory-related genes in the cerebrum, cerebellum, thalamus, brain stem and myelencephalon [22].

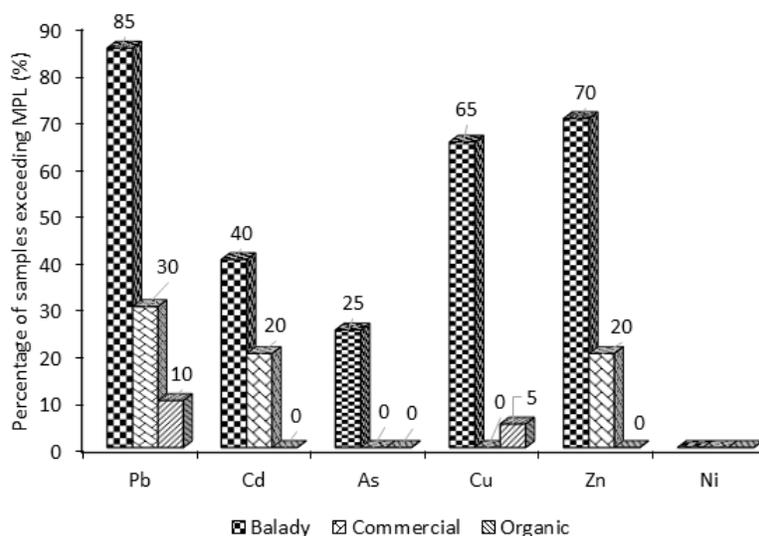


Figure 4: Percentage of chicken egg samples exceeding maximum permissible limits of heavy metals and trace elements under study.

Nickel (Ni)

Nickel concentrations analysis in this study was recorded in Figure 2B. Balady eggs were significantly the highest in the Ni load (ppm) (1.31 ± 0.08), followed by commercial eggs (0.61 ± 0.74) and organic eggs (0.29 ± 0.05).

Results of Nickel concentrations in this study were corresponding to that recorded in Malaysia (1.11 ppm) and Bangladesh (1.35 ppm) [7,8].

There is no report about MPL of nickel (Figure 4). Nickel is a metal of no known

biological function in humans and animals. However, dietary exposure to nickel compounds is associated with dermatitis and cancer risk in humans [23]. The activity of liver enzymes was increased by dietary supplementation of 500 mg Ni/kg, indicating damage of the liver parenchyma. The results of serum biochemistry were confirmed by a mild or moderate form of pathological focal fatty infiltration of the liver in broilers [24].

Zinc (Zn)

The obtained results in Figure 3A showed the mean concentrations of Zn in the examined samples.

It is clear from this figure that balady eggs had significantly the highest Zn concentrations followed by commercial eggs and finally eggs produced on organic base. The mean \pm SE residual concentrations of Zn (ppm) in the examined samples were 23.17 ± 0.88 , 15.42 ± 1.03 and 10.77 ± 0.59 in the examined balady, commercial and organic eggs, respectively. The reported Zn concentrations in this study were corresponding to that reported in chicken eggs (15.75 ± 4.05 ppm) collected from Campania region in Southern Italy [25]. Unlikely, higher Zn concentrations were reported in upper Egypt (56.8 ± 2.89 ppm) [6]; lower Zn concentrations were reported in chicken eggs produced in Peshawar, Pakistan [3].

Comparing the obtained Zn concentrations in chicken eggs from this study with MPLs established by FAO/WHO [9] revealed that all the examined organic eggs were within the allowed Zn MPL (20 ppm), however 70% and 20% of the examined balady and commercial eggs exceeded this limit (Figure 4). Zinc is one of the essential trace elements that is needed for the normal body functions. It is essential for the function of many enzymes and receptors in the body. Furthermore, Zn has a documented antioxidant and anti-inflammatory effects and promote the immune system in animals and humans [26]. However, the role of dietary Zn supplementation in increasing the body gain or the feed conversion efficiency is not clear. For instance, chickens fed on diets supplemented with Zn (600 mg/kg) had a slightly lower body weight, compared with those given 0 or 300

mg/kg of Zn. Nevertheless, this body weight decrease did not affect meat yield [27].

Copper (Cu)

Copper residual concentrations were also estimated in this study. Similar to Zn, balady eggs had significantly ($p < 0.05$) the highest Cu residual concentration followed by both of organic and commercial eggs. The recorded Cu residues (ppm) in the examined samples were 11.47 ± 0.66 , 4.17 ± 0.42 and 6.15 ± 0.51 in balady, commercial and organic eggs, respectively (Figure 3B). The obtained concentrations of Cu in this study were in agreement with the previous published reports from Egypt, Pakistan and Bangladesh [3,6,7]. Unlikely, lower residual Cu concentrations were recorded in chicken eggs from Greece and Italy [16,25].

The recorded Cu concentrations in this study exceeded MPL (10 ppm) of FAO/WHO (JECFA, 2005) [9] in 65% and 5% of the examined balady and organic eggs, respectively (Figure 4).

Copper is one of the essential trace elements that is needed as a cofactor for various enzymes such as cytochrome oxidase and superoxide dismutase, Cu is also an essential element for cellular respiration and free radical defense. Exposure to excess concentrations of Cu lead to oxidative damage to single macromolecules such as lipoproteins, DNA, or thiol containing enzymes [28]. In the poultry industry, copper sulfate is added in a concentration of 125 to 250 ppm to poultry rations in the United States as a growth promoter. However, the exact mechanisms behind this growth promotion activity are not clear. One of the possible mechanisms could be attributed to the bactericidal, bacteriostatic, or both, effects of Cu on the gastrointestinal tract (GIT) microbiota [29].

From all obtained results in this study, it notes worthy that different kinds of chicken eggs had different metal accumulation pattern, which might be attributed to bird-strain differences that might subsequently affect metal detoxification pathways.

Conclusion

It is clear that chicken eggs represent a major source of both human and chicken

embryo exposure to various metals and trace elements, which might represent a potential hazard leading to various biological consequences. Thus, we highly recommend continuous screening of metal load in eggs produced by different avian species as well as ration introduced to birds, which represent the major source of chicken exposure to these metals.

Conflict of interest

The authors declare that there is no conflict of interest.

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الملخص العربي

بقايا بعض المعادن الثقيلة السامة والعناصر النادرة في بيض الدجاج

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يمثل تواجد بقايا المعادن الثقيلة والعناصر النادرة ببيض الدجاج أحد المخاطر الكيميائية من وجهتي نظر القائمين على حبة الغذاء وذلك نظرا للاستهلاك الأدمي لبيض المائدة وكذلك للقائمين علي طب الطيور لما لهذه السموم من مخاطر صحية وبصفة خاصة على أجنة الدواجن في بيض التفريخ. استهدفت هذه الدراسة قياس تركيزات متبقيات بعض المعادن السامة مثل الرصاص، الكاديوم، الزرنيخ، النيكل وكذلك بعض العناصر النادرة مثل الزنك والنحاس في ثلاثة أنواع من بيض الدجاج. تمثلت هذه الأنواع في البيض البلدي (وهو المنتج من الدجاج المربي تربية منزلية) والبيض التجاري (المنتج من مزارع انتاج البيض والتي تعتمد على علائق البياض التجارية) وكذلك البيض الأبيض العضوي الناتج عن تغذية الدجاج البياض علي أعلاف عضوية خالية من الإضافات. وقد تم تجميع عينات البيض (٦٠ عينة) بطريقة عشوائية من بعض قرى محافظة الشرقية خلال الفترة من شهر يونيو إلي شهر أكتوبر ٢٠١٦ م. وأوضحت النتائج احتواء البيض البلدي علي أعلى معدل من متبقيات المعادن تحت الدراسة; Zn(23.17 ± 0.88); As (0.14 ± 0.017); Ni(1.31 ± 0.08), and Cu(11.47 ± 0.66), { Pb (0.34± 0.03);Cd (0.18 ± 0.02); }، وقد وجد ارتفاع في متبقيات المعادن المسجلة في هذه الدراسة عن الحدود القصوى والمسموح بها من الهيئات العالمية خاصة في البيض البلدي والبيض التجاري. والجدير بالذكر أن تواجد تركيزات مرتفعة من تلك المعادن يشكل خطورة كبيرة على كل من المستهلك و أجنة الطيور وذلك لما لها من تأثيرات سامة عليهما. هذا وقد تمت مناقشة الأهمية الصحية للمعادن تحت الدراسة.