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RESEARCH ARTICLE

Residual contents of the toxic metals (lead and cadmium), and the trace elements (copper and zinc) in the bovine meat and dairy products: residues, dietary intakes, and their health risk assessment

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ABSTRACT

Residual content of lead (Pb), cadmium (Cd), copper (Cu), and zinc (Zn) in the retailed meat and dairy products in Egypt was estimated. Calculation of estimated weekly intake (EWI) and health risks was followed. Results revealed detection of tested metals in all samples at variable concentrations with higher Pb and Cd than maximum permissible limits. However, the calculated EWI did not exceed the provisional tolerable weekly intake established by the World Health Organization. Furthermore, calculation of the hazard ratio and index for mixed metal contaminants did not exceed one, indication that products consumption would not pose adverse effects on consumers.

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Introduction

Bovine meat products such as beef mince, sausage, luncheon, and basterma, and dairy products as milk, cheese, dried milk, and yoghurt are considered as primary sources for the animal derived protein with high bioactive peptides, vitamins and provide humans with part of their needs from energy (Stadnik and Kęska 2015). In addition, such products represent essential sources for the trace elements such as copper (Cu), and zinc (Zn) that needed for normal physiological functions of the body systems (Pogorzelska-Nowicka *et al.* 2018). At the same time, these meat and dairy products might pose a health hazard because of the possible transfer of some food poisoning organisms, and chemical residues such as heavy metals to the consumers.

Heavy metals are known for their bioaccumulation and biomagnification nature. Metals such as lead (Pb), and cadmium (Cd) are of toxic nature and of no-known physiological values (Thompson and Darwish 2019). Pb toxicity is responsible for many deaths among children around the world (Darwish *et al.* 2016). Pb can also act as a neurotoxin and adversely affect the renal functions and the gastrointestinal tract (Cunningham and Saigo 1997).

Cadmium is another toxic metal that can be transferred to the human *via* the food chain. The United States environmental protection agency classified Cd as a group B1 carcinogen (IARC 2016). Furthermore, Cd is the primary cause of the *itai-itai* disease that is characterized by renal failure and osteomalacia (Nishijo *et al.* 2017). In addition, repeated exposure to small concentrations of Cd is linked to multiple organ damage such as liver, kidney, testes, breast, and nervous system (Darwish *et al.* 2019).

Zinc is an essential trace element that plays key roles in the catalytic functions of more than 100 enzymes in the body. Zinc is also needed for the regulation of the gene expression of many cell components. In addition, Zn is important for the cell wall maintenance. Zinc deficiency is a major problem in many developing countries and can lead to several health challenges including anemia, reduced immunity, hypogonadism, and dwarfism. Meat and dairy

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products are considered as a major supply for the human needs of Zn (Roohani *et al.* 2013, Pogorzelska-Nowicka *et al.* 2018).

Copper is one essential element that plays important roles in the biochemistry and physiology of the living organisms as it acts as a co-factor for several enzymes. Furthermore, Cu is an important element for the cellular respiration. However, excess exposure to Cu might lead to induction of the oxidative damage to the cell organelles (Darwish *et al.* 2014).

Investigation of the residual contents of toxic metals and trace elements was done in Egypt in variable food matrices such as fish (Morshdy *et al.* 2013, 2019), meat, edible offal (Darwish *et al.* 2015). However, estimation of the content of Pb, Cd, Zn, and Cu in the meat products including beef mince, sausage, luncheon, and basterma and dairy products as milk, cheese, dried milk, and yoghurt had received less attention. In addition, calculation of the dietary intakes and the potential health risks associated with ingestion of contaminated meat and dairy products among the Egyptian population had been less investigated.

One major task for the food safety sector is to ensure the safety and the wholesomeness of the retailed meat and dairy products, therefore, this study was designed to estimate the residual content of Pb, Cd, Zn, and Cu in the retailed meat and dairy products in Egypt. In addition, calculation of the dietary intakes and the associated health risks with detected metals was followed. The public health significance of the studied metals was further discussed.

Material and methods

All reagents used were of analytical grade and the standard solutions of Pb, Cd, Zn, and Cu were purchased from Merck, Darmstadt, Germany.

Collection of samples

A total of 160 meat and dairy products were sampled according to the simple random sampling method. The number of samples was determined based on a confidence level of 90%, and a margin of error of 10%. Samples included beef mince, sausage, luncheon, basterma, raw milk, kariesh cheese, dried milk powder, and yoghurt (20 of each) were collected from retail markets and grocery stores in Zagazig city, Egypt. Samples (50 g of each) were transferred cooled in plastic falcon tubes to the laboratory for extraction and heavy metal analysis.

Sample preparation and extraction

For sample preparation and extraction, one gram from each sample was mixed with 10 ml of the digestion mixture (3 parts of HNO_3 and 2 parts of $HClO_4$). The mixture was kept overnight at room temperature for digestion, and then placed at heated water bath (70 °C) with swirling at 30 min intervals for 3 h. Metals concentrations were directly measured using an atomic absorption spectrophotometer (PerkinElmer 2380).

The accuracy of the analysis was confirmed by using blanks, measuring the samples in duplicate, and using certified reference material IAEA-142/TM (muscle homogenate), Vienna, Austria. Mean recoveries ranged from 95% to 105% to the tested metals. Recovered concentrations of the certified samples were within 3–5% of the certified values. All materials and equipment used in the analysis were washed with diluted nitric acid to avoid external contamination with heavy metals.

Dietary intakes of heavy metals

The estimated daily intake (EDI) and estimated weekly intake (EWI) (μg/kg/day) values for the tested heavy metals were calculated using the equations described by the Human Health Evaluation Manual (US EPA 2010):

$$EDI = C * F_{IR}/BW$$
$$EWI = EDI * 7$$

Where C is the concentration of the tested metal in the sample (ppm wet weight); F_{IR} is the food ingestion rate in Egypt, which was estimated at 85.7 g/day for meat products, 120 ml/day for milk, 10 g/day for dried milk powder, 65 g/day for kariesh cheese, and yoghurt (FAO 2003); BW is the body weight of Egyptian adults, which was set at 70 kg.

Health risk assessment

The non-cancer risks associated with the consumption of the metal-contaminated meat and dairy products among the Egyptian populations were calculated using the guidelines recommended by the US EPA (2010). Therefore, the EDI was compared with the recommended reference doses (RfD) (0.001 mg/kg/day for Cd, 0.004 mg/kg/day for Pb, and 0.3 mg/kg/day for Zn) (US EPA 2010), to generate the hazard ratio (HR) as following:

$$HR = EDI/RfD * 10^{-3}$$

The hazard ratios can be summed to calculate a hazard index (HI) for estimation of the health risks associated with mixed contaminants.

$$HI = \sum HRi$$

where *i* represents each metal.

If the value of HR and/or HI exceeded one, this indicates a potential risk to human health, whereas a result less than or equal one indicates no risk.

Statistical analysis

The Tukey-Kramer HSD difference test (JMP) (SAS Institute, Cary, NC, USA) was used for statistical comparisons ($p < 0.05$).

Results and discussion

Residual contents of heavy metals in meat and dairy products

Lead

The obtained results in the present study revealed detection of Pb in all examined meat and dairy product samples as displayed in Figure 1. Among the examined meat products, basterma had significantly ($p < 0.05$) the highest residual Pb content (ppm) (0.15 ± 0.02), followed by sausage (0.11 ± 0.01), beef mince (0.08 ± 0.01), and luncheon (0.08 ± 0.01), respectively (Figure 1(A)). Lower Pb content was detected in the retailed beef meat products in Spain at 0.007 ppm (González-Weller *et al.* 2006). In agreement with the obtained results of the present study, Pb was detected at 0.16 ppm in German sausage retailed in Iran (Abedi *et al.* 2011). Higher Pb contents (14.84 and 16.69 ppm) were recorded in poultry Frankfurter and burger retailed in Saudi Arabia (Korish and Attia 2020). Similarly, higher Pb contents were reported in pork meat products (mortadella, baked ham, raw ham, cured sausage, salami, and würstel) from South Italy with a range of 0.22–0.38 ppm (Barone *et al.* 2021).

Regarding dairy products, raw milk had the highest residual content (ppm) of Pb (0.14 ± 0.01), followed by yoghurt (0.12 ± 0.01), kariesh cheese (0.09 ± 0.02), and dried milk powder (0.05 ± 0.005), respectively (Figure 1(B)). In line with the obtained results in the current investigation, Castro-González *et al.* (2018) reported comparable levels of Pb in raw milk (0.03 ppm) and ranchero cheese (0.11 ppm) in Mexico. In addition, Sujka *et al.* (2019) detected Pb in milk samples from various regions in Poland at 0.01–0.23 ppm. Besides, Pšenková *et al.* (2020) detected Pb at levels <0.1 ppm in raw cow's milk in Slovakia. However, lower residual

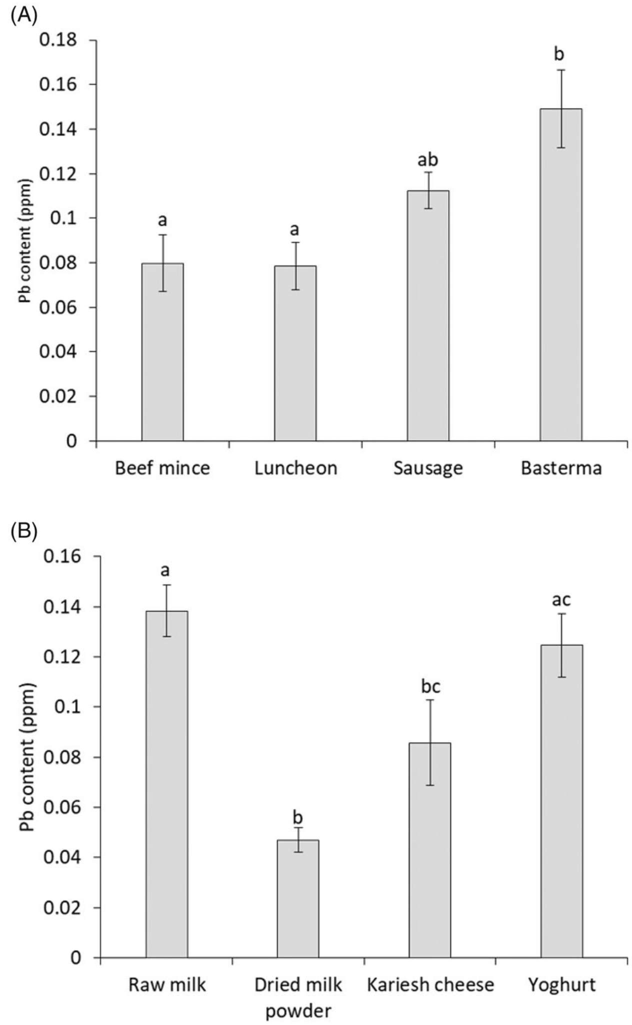

Figure 1. Lead (Pb) residual contents (ppm) in (A) Meat products and (B) Dairy products marketed in Egypt. Data represent means ± SE ($n = 20$ each product). Columns carrying different letter are statistically significant at $p < 0.05$.

content of Pb (0.006–0.01 ppm) was reported in milk samples from Tabriz, Iran (Beikzadeh *et al.* 2019).

Cadmium

Cadmium was detected in all examined samples in the current study. Among the tested meat products, basterma had the highest Cd residual content (ppm) (0.08 ± 0.009), followed by sausage (0.05 ± 0.005), luncheon (0.04 ± 0.006), and beef mince (0.006 ± 0.001), respectively (Figure 2(A)). In agreement with the recorded Cd concentrations in the meat product examined in the present study, Abd-Elghany *et al.* (2020) recorded levels of Cd ranged between 0.30 and 0.59 ppm in the sheep meat and edible offal in Kuwait. Besides, Barone *et al.* (2021) recorded Cd levels of 0.01–0.03 ppm in the pork meat products in South Italy. However, lower Cd concentrations (2.2–13.5 ppb)

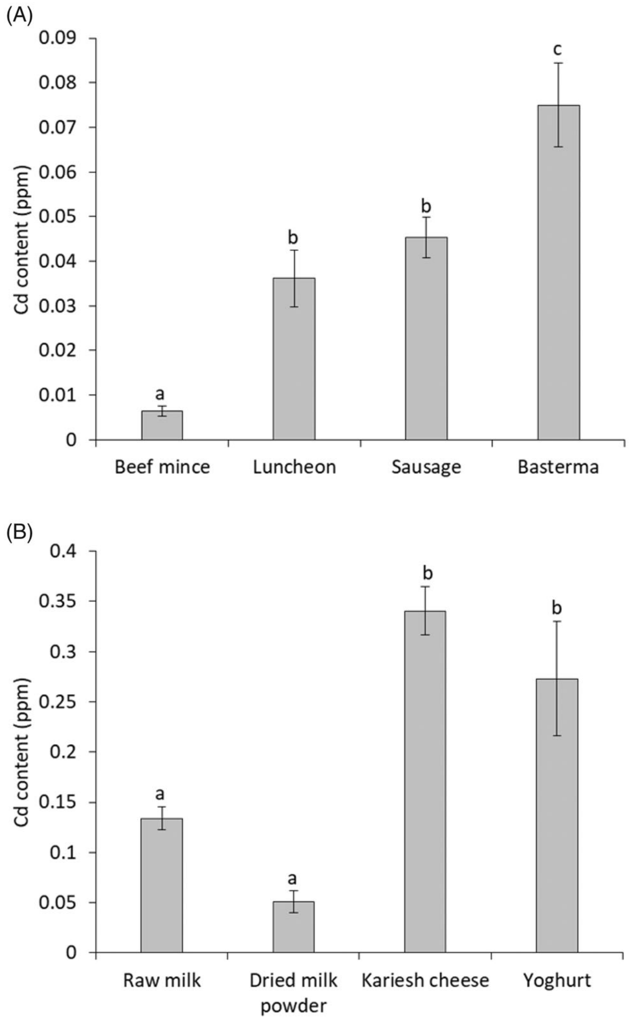

Figure 2. Cadmium (Cd) residual contents (ppm) in (A) Meat products and (B) Dairy products marketed in Egypt. Data represent means ± SE ($n = 20$ each product). Columns carrying different letter are statistically significant at $p < 0.05$.

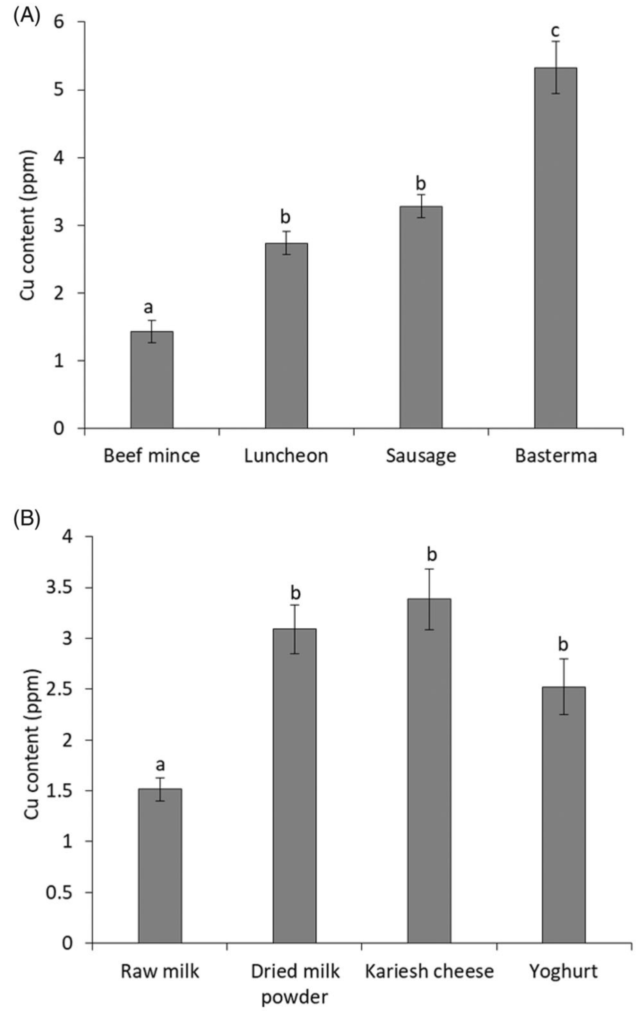

Figure 3. Copper (Cu) residual contents (ppm) in (A) Meat products and (B) Dairy products marketed in Egypt. Data represent means ± SE ($n = 20$ each product). Columns carrying different letter are statistically significant at $p < 0.05$.

were demonstrated in marketed sausage in Iran (Abedi *et al.* 2011).

Kariesh cheese had significantly the highest Cd residues (ppm) (0.34 ± 0.02), followed by yoghurt (0.27 ± 0.06), raw milk (0.13 ± 0.01), and dried milk powder (0.05 ± 0.01), respectively (Figure 2(B)). Similar levels (0.28 ± 0.16 ppm) were reported in cow's milk marketed in Benha, Egypt (Malhat *et al.* 2012). Higher Cd residues (~5.57 ppm) were reported in the pasteurized milk samples retailed in Iran (Sobhanardakani 2018). Lower Cd residues (0.0001–0.007 ppb) were detected in milk, butter milk, cream, cheese, and yoghurt samples from different regions in Poland (Sujka *et al.* 2019).

Copper

Copper was detected in all examined meat product samples with a range of 0.46–2.55 ppm in beef mince, 1.24–4.13 in luncheon, 2.59–8.33 ppm in basterma, and 2.24–4.36 ppm in sausage. Basterma had the highest Cu content followed by sausage, luncheon, and beef mince (Figure 3(A)). Comparing the recorded Cu residues in the present work with other reports, lower Cu levels (0.80 ± 0.01) were recorded in the rabbit meat and viscera in Chile (Valenzuela *et al.* 2011). Barone *et al.* (2021) reported relatively similar Cu concentrations (1.08–1.21 ppm) in the pork meat products retailed in Italy.

Copper was also detected in all examined dairy product samples. Copper content ranged between 1.25–5.65 ppm in kariesh cheese, 1.15–4.58 ppm in dried milk powder, 0.98–5.12 ppm in yoghurt, and 0.75–2.37 ppm in raw milk samples (Figure 3(B)). In agreement with the obtained results in the current investigation, Malhat *et al.* (2012) recorded that Cu residues in the cow's milk sold in Benha city, Egypt

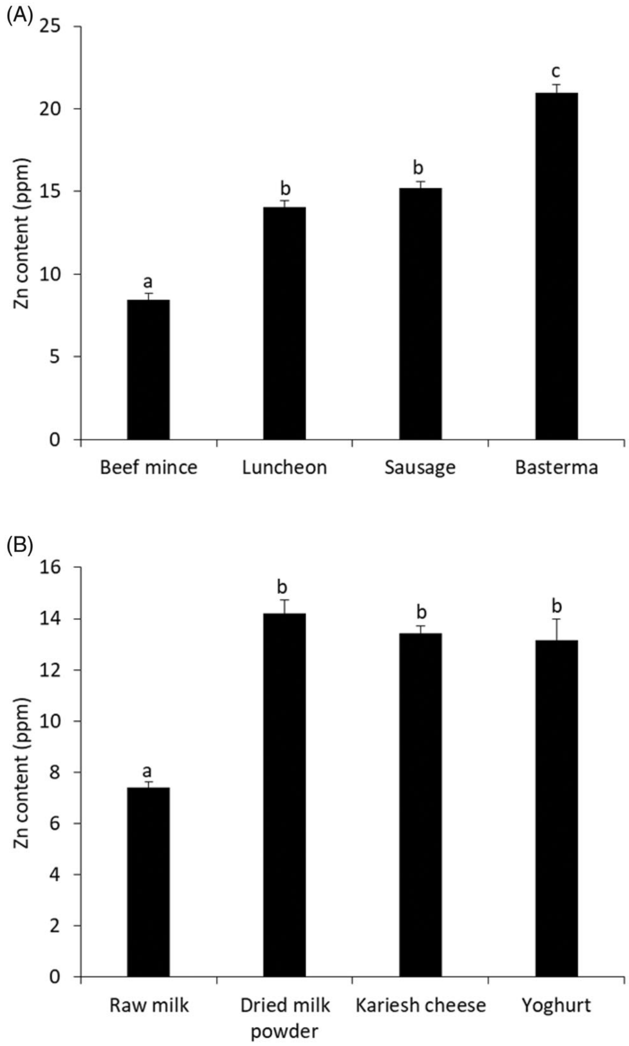

Figure 4. Zinc (Zn) residual contents (ppm) in (A) Meat products and (B) Dairy products marketed in Egypt. Data represent means ± SE ($n = 20$ each product). Columns carrying different letter are statistically significant at $p < 0.05$.

was 2.94 ± 1.09 ppm. Furthermore, comparable Cu levels (0.01–4.94 ppm) to that reported in the present study were recorded in the dairy products including raw milk, yoghurt, and cheese retailed in Poland (Sujka *et al.* 2019). However, Meshref *et al.* (2014) recorded lower Cu levels (0.09–0.60 ppm) in dairy products including milk, kariesh cheese, butter, and rice pudding retailed in Beni-Suef, Egypt. Higher Cu residues (8.41 and 9.77 ppm) were reported in the raw milk and pasteurized milk samples retailed in Iran, respectively (Sobhanardakani 2018).

Zinc

Zinc was detected in all examined meat product samples. Basterma had the highest Zn content (ppm) (20.94 ± 0.53), followed by sausage (15.21 ± 0.37), luncheon (14.06 ± 0.41), and beef mince (8.44 ± 0.42), respectively (Figure 4(A)). Similarly, Zn levels of 9.5 ± 0.35 in the rabbit meat and viscera in Chile (Valenzuela *et al.* 2011). Furthermore, Zn residues of 5.71–7.32 ppm were recorded in the pork meat products marketed in Italy (Barone *et al.* 2021). Higher Zn content (121.27 ± 7.45 ppm) was reported in the retailed meat in Nigeria (Ihedioha *et al.* 2014).

In the same manner, Zn was detected in all examined dairy product samples at variable concentrations. Zn contents (ppm) were 14.22 ± 0.49, 13.44 ± 0.26, 13.15 ± 0.83, and 7.41 ± 0.21 in the examined dried milk powder, kariesh cheese, yoghurt, and raw milk, respectively (Figure 4(B)). Likely, Malhat *et al.* (2012) recorded Zn levels of 4.77–10.75 ppm in cow's milk retailed in El-Qaliubiya governorate, Egypt. Besides, relatively similar Zn levels (3.01–8.59 ppm) were reported in dairy products including milk, kariesh cheese, butter, and rice pudding retailed in Beni-Suef, Egypt (Meshref *et al.* 2014). In addition, Zn levels ranged between 0.10 and 56.44 ppm were reported in the marketed dairy products in Poland (Sujka *et al.* 2019). Higher Zn residues (90.12–253.70 ppm) were reported in the raw milk and pasteurized milk samples retailed in Iran (Sobhanardakani 2018).

Heavy metals occur naturally in the environment and find their way into the animal body *via* contaminated feed and water. Such metals accumulate in the animal tissues such as muscles and mammary glands and might pass into the milk. Meat and dairy products might be contaminated with heavy metals due to the use of contaminated raw materials or other industrial ingredients (Morshdy *et al.* 2019, Thompson and Darwish 2019). Therefore, future approaches are still needed to screen heavy metal residues in soil, water, animal feed, and animal offal such as livers and kidneys. Besides, monitoring of the residual contents of other toxic elements such as arsenic, and mercury, and trace elements such as iron, and calcium in meat and dairy products are also needed for comprehensive understanding of the Egyptian scenario of heavy metal residues in food.

Dietary intakes and human health risk assessment

Lead

By comparing the detected residual concentrations of Pb in the examined samples with the established maximum permissible limits (MPL), it was clear that 70%, 50%, 35%, and 25% of the examined basterma, sausage, beef mince, and luncheon, respectively exceeded MPL of Pb in meat and meat products (0.1 ppm) (EC

2006). Whereas, 100%, 100%, 90%, and 80% of the examined raw milk, yoghurt, kariesh cheese, and dried milk samples, respectively exceeded MPL of Pb in raw milk and dairy products (0.02 ppm) (EC 2006). Excessive consumption of foods contaminated with Pb might lead to severe health hazards such as toxicity, mutagenicity, carcinogenicity, and disturbances in the haeme-synthesis and central nervous system (EFSA 2010, Darwish *et al.* 2016). However, calculation of the EWI of Pb in the examined samples revealed that raw milk (EWI = 1.659 µg/kg/week) and basterma (EWI = 1.281 µg/kg/week) could cause the highest intake of Pb. These values were within the recommended provisional tolerable weekly intake (PTWI) for Pb (25 µg/kg/week) established by WHO (2010). Calculation of the HR values for Pb *via* ingestion of meat and dairy products revealed values far below one, indicating that ingestion of such products will not bear potential risks on human health for Pb exposure. In agreement with this assumption, Darwish *et al.* (2015) concluded that ingestion of cattle and sheep meat in Egypt will not pose potential risks for Pb exposure. Similarly, Sujka *et al.* (2019) reported that despite presence of higher levels of Pb in dairy products than the recommended MPL, calculation of the dietary intakes of Pb revealed no potential risks among the consumers in Poland.

Cadmium

Cadmium residue levels in the examined samples were higher than the established MPL (0.05 ppm) (EC 2006) in 100%, 95%, 90%, 55%, 30%, 20%, 20%, and 0% of the examined kariesh cheese, yoghurt, raw milk, basterma, sausage, luncheon, dried milk powder, and beef mince, respectively (Table 1). Ingestion of foods contaminated with Cd is associated with the occurrence of the nephropathy, renal failure, softening of bones, and carcinogenesis (Morshdy *et al.* 2013). The highest EWI of Cd were recorded for kariesh cheese (2.212 µg/kg/week), followed by yoghurt (1.778 µg/kg/week), raw milk (1.603 µg/kg/week); while the lowest EWI values were recorded for dried milk powder (0.049 µg/kg/week), and beef mince (0.056 µg/kg/week), respectively. The recorded EWI of Cd for all examined products were within the established PTWI for Cd (7 µg/kg/week) (WHO 2010). Besides, the calculated HR values for Cd in all examined samples did not exceed one (Table 1), suggesting that ingestion of such meat and dairy products would not pose a public health risk. In agreement with the reported results in the current work, Sujka *et al.* (2019) reported that Cd residues in the dairy products in Poland would not have potential risks among the Polish consumers. Unlikely, Darwish *et al.* (2015) reported that ingestion of cattle and sheep offal in Egypt might pose potential risks associated with Cd.

Copper

Copper content exceeded the established MPL (5 ppm) (EC 2006) in 55% of basterma samples, 15% of kariesh cheese samples, and 5% of yoghurt samples (Table 1). The recorded EWI of Cu exceeded the PTWI of Cu (3.5 µg/kg/week) (WHO, 2010), in all examined meat and dairy products except for dried milk powder (EWI = 3.087 µg/kg/week); while beef mince represented the major source for Cu exposure among the examined products (EWI = 85.603 µg/kg/week). There is no report on RfD values for Cu, therefore, HR of Cu was not applicable in this study. High intake of Cu was similarly reported in Poland (Sujka *et al.* 2019). Copper is needed for the normal functions of several enzymes in the body; however, excessive intake of Cu might lead to hepatic cirrhosis, hyperthyroidism, and allergic reactions (Darwish *et al.* 2014).

Zinc

All examined meat and dairy product samples had Zn content within the established MPL (50 ppm) (EC 2006). However, EWI of Zn for all samples exceeded

Table 1. Dietary intake and health risk assessment of Pb, Cd, Cu, and Zn in the examined meat and dairy products.

	Pb			Cd			Cu			Zn			
	%	EWI	HR	%	EWI	HR	%	EWI	HR	%	EWI	HR	HI
Beef mince	35	0.686	0.024	0	0.056	0.008	0	85.603	NA	0	72.352	0.034	0.066
Luncheon	25	0.672	0.024	20	0.308	0.044	0	23.436	NA	0	120.526	0.057	0.125
Sausage	50	0.966	0.034	30	0.392	0.056	0	28.098	NA	0	130.361	0.062	0.152
Basterma	70	1.281	0.046	55	0.644	0.092	55	45.654	NA	0	179.487	0.085	0.223
Raw milk	100	1.659	0.059	90	1.603	0.229	0	18.165	NA	0	88.914	0.042	0.330
Dried milk powder	80	0.049	0.002	20	0.049	0.007	0	3.087	NA	0	14.224	0.007	0.016
Kariesh cheese	90	0.553	0.019	100	2.212	0.316	15	21.987	NA	0	87.388	0.042	0.377
Yoghurt	100	0.812	0.029	95	1.778	0.254	5	16.373	NA	0	85.463	0.041	0.324

%: refers to percentage of samples exceeding the established maximum permissible limits.
EWI: Estimated weekly intakes.
HR: Hazard ratio.
HI: Hazard index.

the recommended PTWI (7 µg/kg/week). Among the examined samples, basterma would provide the highest Zn intake followed by sausage, luncheon, raw milk, kariesh cheese, yoghurt, beef mince, and dried milk powder (Table 1). The calculated HR values for Zn were far below one, indicating that such levels of Zn would not pose potential health risks on the consumer. Similar values for Zn intake were reported in Italy (Licata *et al.* 2004), and South Korea (Khan *et al.* 2014). Zn is an essential trace element for the normal function of the body systems; however, excessive Zn intake might lead to some adverse effects such as vomiting, insomnia, and neurodegenerative diseases (Faa *et al.* 2008).

Collectively, calculation of the HI for mixed contaminations among the examined meat and dairy products revealed values below one, indicating that consumption of such products would not pose health risks among the Egyptian consumers.

Conclusions

The obtained results in the present study revealed detection of Pb, Cd, Cu, and Zn in all examined samples at variable concentrations. Several samples had residual levels higher than the recommended MPL. However, the calculated EWI did not exceed the PTWI for the metal contaminants for all examined samples indicating that consumption of such products would not pose any hazards on the public health.

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Disclosure statement

No potential conflict of interest was reported by the author(s).

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