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Residual Levels of Toxic Metals and Estimation of their Dietary Intakes, and Non-Carcinogenic Risks Associated with the Consumption of Meat and Edible Offal of Camel in Egypt and Saudi Arabia

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SIA, SME, SA collected samples from Saudi Arabia and processed them. MHEK, ASA, MMA, TMG and HAK collected samples from Egypt and performed samples' digestions and metal measurements, and wrote the manuscript. SIA, SME, SA arrnaged funds for the research. All authors approved the final version of the manuscript.

Key words
Camel meat, Offal, Lead, Cadmium, Arsenic, Mercury, Egypt, Saudi Arabia

ABSTRACT

Camel meat and edible offal are regarded as exotic meats around the world. However, such meat kinds are regarded as emerging meat sources rich in animal-derived protein in particular countries such as Egypt and Saudi Arabia. Camel meat and offal supplies humans with part of their needs from essential amino acids, minerals, vitamins, and polyunsaturated fatty acids. Toxic metals such as lead (Pb), cadmium (Cd), arsenic (As), and mercury (Hg) are of no-known physiological importance. The objectives of the present study were to quantitatively estimate the residual levels of Pb, Cd, As, and Hg in camel meat and edible offal including round, liver, kidney, and tongue in samples collected from Zagazig slaughterhouse, Egypt and Al-Ahsa slaughterhouse, Saudi Arabia. Dietary intakes and potential health risks associated with the consumption of camel meat and edible offal among Saudi and Egyptian populations were additionally calculated in a comparative way. The obtained results indicated that edible offal including liver, kidney and tongue had higher levels of the tested metals compared with the muscle. Samples collected from Egypt had significantly ($p < 0.05$) higher metal residues than that collected from Saudi Arabia. Cadmium content exceeded the established maximum permissible limits (MPL) in 65%, 20%, and 70% of liver, kidney, and tongue samples collected from Egypt, while only 35%, and 20% of the Saudi liver and kidney samples exceeded MPL. Arsenic residue levels exceeded MPL in 50%, 50%, and 25% of the Egyptian liver, kidney, and tongue samples. None of the examined samples exceeded MPL for Pb, and Hg. Calculation of the hazard ratio (HR), and hazard index (HI) for Egyptian and Saudi adults and children indicated that HI was higher than one for Egyptian children consuming liver, kidney, and tongues of the camel. Therefore, it is highly recommended to reduce the daily consumption of such offal samples, particularly among children in Egypt.

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INTRODUCTION

Arabian camel (*Camelus dromedarius*) is regarded as an important animal particularly in the Middle Eastern countries and in the Arabian Peninsula. Camel is also a historical animal that plays key roles in the culture of countries like Egypt and Saudi Arabia. Camel is also known as the ship of the dessert because of its survivability to the extremely hard environmental conditions (Darwish *et al.*, 2010; Kadim *et al*., 2008). This animal plays significant roles in the economy of several countries as it is reared for milk, meat, wool, and leather production (Kurtu, 2004). Camel's meat is a major source of animal-derived protein rich in essential amino acids, and minerals in many parts of Africa such as Egypt, Sudan, Somalia, and Ethiopia, in Asia such as Saudi Arabia, other Gulf countries, India and in Australia. The camel meat production increased to reach about 0.7% of the world meat production (FAO, 2001). Camel meat is believed to play medicinal roles as it is regarded as healthier red meat source than that of beef and mutton, because the scanty amounts of fat, and cholesterol (El-Ghareeb *et al*., 2019). Edible offal of camel such as liver, kidney, and tongue is very popular in the Middle East, particularly in Egypt because of its unique aroma and flavor (Tang *et al*., 2020).

Egypt and Saudi Arabia are considered the biggest two countries in the Middle East, in terms of their space area, population and resources. There is a significant advancement in the economy and industrial activities in the two countries, beside the common agricultural activities in Egypt (Darwish *et al*., 2015). Similar activities were associated with the release of environmental pollutants such as heavy metals in different Asian and African countries (Yasotha *et al*., 2021; Shezi *et al*., 2022).

Heavy metals such as lead (Pb), cadmium (Cd), arsenic (As), and mercury (Hg) are of no-known physiological functions. However, such metals were implicated in several adverse health effects (Thompson and Darwish, 2019). For instances, Pb was responsible for several cases of deaths among children, and impairment of the intellectual functions among chiclren (Darwish *et al*., 2016). Pb and Hg were regarded as neurotoxins and adversely affect the mental health (Cunningham and Saigo, 1997). Cd is classified by the US Environmental Protection Agency as a group B1 carcinogen (IARC, 2016). Additionally, Cd was identified as the main contributor to the itai-itai disease, which exhibits symptoms of renal failure and osteomalacia (Nishijo *et al*., 2017). Multiple organ damage has been related to ingesting high dosages of as (Thompson and Darwish, 2019). Thus, it is crucial for both the food safety and public health sectors to monitor the residual amounts of these harmful components in human food.

As camel lives under the same environment as humans, therefore it can act as a bioindicator for the human exposure to the environmental pollutants. Estimation of the toxic metal residues in the camel meat and edible offal had received little attention in Egypt and Saudi Arabia. Therefore, the present study was taken to first estimate the residual levels of the toxic metals (Pb, Cd, As, and Hg) in the meat, liver, kidney, and tongue of camels slaughtered at Zagazig slaughterhouse, Egypt, and Al-Ahsa slaughterhouse, Saudi Arabia. In addition, the dietary intakes and potential health risk assessment were done.

MATERIALS AND METHODS

All experiments were done following the guidelines of both Zagazig and King Faisal Universities. All chemicals were of the highest quality and purchased from Merck, Germany.

Samples collection

Two hundred samples were randomly collected from camel muscle (round), liver, kidney, and tongue. Samples were collected equally from camels slaughtered at Zagazig (Egypt) and Al-Ahsa (Saudi Arabia) slaughterhouses (n = 100, 25 samples from each tissue). Samples were collected during the period of March to June 2022. Samples were collected from adult, healthy male camels. Tissue samples were collected and kept in plastic falcon tubes at −20 °C until extraction and measurement of heavy metals.

Sample preparation and extraction

Finerty *et al*. (1990) method was used for the extraction of heavy metals. In a nutshell, one g of each tissue was digested overnight at room temperature in 10 mL of the digestion solution, which is composed of a mixture of nitric acid at 65% (3 volumes) and perchloric acid at 70% (2 volumes). The mixture was then heated for three hours in a water bath (70°C). Following digestion, the produced extracts were stored at room temperature in firmly covered bottles until metal analysis. After obtaining the necessary authorizations, digested samples that were obtained in Saudi Arabia were sent to Egypt for metal analysis.

Metal measurement

Using hollow cathode lamps with an air-acetylene flame and an atomic absorption spectrophotometer (Shimadzu AAS 6800, Shimadzu, Japan), heavy metal residues, including Pb and Cd, were detected. Using hydride generation/cold vapor atomic absorption spectroscopy, As, and Hg were detected (Shelton, CT, USA). The average recovery rate varied between 94 and 105%. Based on standard curves created for each of the analysed metals, the concentrations of the detected heavy metals were calculated. Wet weight (ww) basis was used to record the results, which were documented as µg/g.

Quality assurance and control

As a quality assurance parameter for this analysis, metal concentrations were measured in the certified reference muscle homogenate, IAEA-142/TM (Vienna, Austria). The heavy metal concentrations in the certified samples were within 5% of the certified levels in the reported concentrations. Every measurement was carried out three times.

Estimated daily intake (EDI)

Based on the US Environmental Protection Agency's (USEPA, 2010) proposed formula, the estimated daily intake (EDI) of the studied heavy metals was determined:

$EDI = C_m X F_{IR} / BW.$

As EDI is based on µg/kg/day, BW is the body weight of adults (70 kg) and children (30 kg) (Bortey-Sam *et al*., 2015), C_m is the concentration of the metal in the sample (µg/g ww), F_{IR} is the meat ingestion rate, which was estimated at 146 g/day (Adam *et al*., 2014; FAO, 2003).

Health risk assessment

Based on an algorithm proposed by the US Environmental Protection Agency (USEPA, 2010), the non-cancer risk related to the ingestion of camel meat and offal among Egyptian and Saudi populations was determined.

Hazard ratio = EDI/HR X 10^{-3}

Where EDI stands for estimated daily intake and RfD for recommended reference doses (0.001 for Cd, 0.004 for Pb, 0.0003 for As, and 0.0005 for Hg) is the recommended doses in mg/kg/day (USEPA, 2010).

The risk associated with mixed metals was calculated using a hazard index (HI). To calculate a hazard index and determine the risk of mixed metals, the hazard ratios (HRs) can be added together. This equation can be used to produce a danger index:

HI = HRi, with i standing for each metal.

A value less than 1 means there are no health hazards, while an HR and/or HI >1 indicates a potential harm to human health.

Statistical analysis

ANOVA was used for statistical analysis, followed by the Tukey-Kramer HSD difference test (JMP) (SAS Institute, Cary, NC, USA). where a significance threshold of p < 0.05 was used. Means ± SD were used to express metal concentrations.

RESULTS AND DISCUSSION

The recorded results in Figure 1A showed that the liver, kidney, and tongue tissues of camel samples collected from Egypt had significantly the highest mean concentrations of Pb (µg/g ww) followed by the same tissues that collected from Saudi Arabia, liver (0.09±0.01); kidney (0.08±0.01), and tongue (0.08±0.01); while the muscle samples collected from both Egypt and Saudi Arabia had the lowest Pb residues (0.04±0.004, and 0.03±0.003, respectively). It is evident from the obtained results that liver (0.14±0.02), and tongue (0.13±0.01) had significantly ($p < 0.05$) the highest Cd residues, followed by kidney (0.07±0.01), and finally muscle (0.03±0.003) in camel samples collected from Egypt. While samples collected from Saudi Arabia showed the following trend liver (0.09±0.01) > kidney (0.06±0.01) > tongue (0.04±0.004) > muscle (0.006±0.001). In general samples collected from Egypt had higher Cd content compared with that collected from Saudi Arabia (Fig. 1B). Similar Pb, and Cd concentrations were recorded in the camel's muscle and edible offal retailed in Saudi Arabia (El-Ghareeb *et al*., 2019). However, higher concentrations of Pb, and Cd were recorded in the edible tissues collected from other farm animals such as cattle, sheep, and goat in Ghana (Bortey-Sam *et al*., 2015), and Egypt (Darwish *et al*., 2015). Detection of Pb, and Cd in the different tissues of camel in both Egypt and Saudi Arabia indicates exposure of camel during their lifetime to Cd either via inhalation or ingestion of contaminated feed and water (Thompson and Darwish, 2019).

Same to Pb, and Cd, As content in the samples collected from Egypt was significantly higher than the corresponding samples collected from Saudi Arabia. As the recorded mean concentrations of As (µg/g ww) in the liver, kidney, tongue, and muscle were 0.05±0.005, 0.05±0.004, 0.04±0.01, and 0.006±0.001, respectively. In the case of the same tissues that collected from Saudi Arabia, As average concentrations were 0.02±0.003, 0.03±0.003, 0.03±0.002, and 0.003±0.001, respectively (Fig. 1C). The obtained results of the present study were comparable to the recorded in camel tissues from Saudi Arabia (El-Ghareeb *et al*., 2019), and in sheep and goats collected from Ghana (Bortey-Sam *et al*., 2015). However, lower concentrations were recorded in meat retailed in Taiwan (Chen *et al*., 2013). Arsenic is commonly used in livestock farms as a feed additive to increase the feed conversion ratio, this could explain the detected concentrations of As in the present study (Hu *et al*., 2017). Mercury was detected in all examined samples in the present study, Egyptian samples showed higher Hg residues than that recorded in the Saudi samples. Where liver (0.008±0.001), and kidney (0.007±0.001) had significantly ($p < 0.05$) higher Hg residues than tongue (0.003±0.001), and muscle (0.003±0.001), respectively. However, no significant differences were observed between liver (0.003±0.0003), kidney (0.003±0.0003), tongue (0.003±0.0002), and muscles (0.002±0.0002) samples collected from Saudi Arabia (Fig. 1D). Relatively similar concentrations were recorded in the different tissues of wild living animals such deer and boar in Hungary, and Slovakia (Lehel *et al.*, 2017; Malova *et al.*, 2019). Detection of Hg in the camel tissues even at low concentrations indicates exposure of camel to environmental pollution via soil, water, or feed (Thompson and Darwish, 2019).

The recorded results in Table I showed that none of the examined samples in both Egypt and Saudi Arabia exceeded the established maximum permissible limits (MPL) of Pb (0.1 µg/g ww in muscles, and 0.5 µg/g ww in offal, EC, 2006). However, Cd concentrations exceeded MPL of Cd (0.05 µg/g ww in muscles, and 0.1 µg/g ww in offal, EC, 2006) in muscles, liver, kidney, and tongue collected from Egypt at 0%, 65%, 20%, and 70%, respectively, and 0%, 35%, 20%, and 0% in the same tissues collected from Saudi Arabis, respectively. Arsenic residual concentrations exceeded the established MPL in muscle and tissues (0.05 µg/g ww in muscles and offal, EC, 2006) at 0%, 50%, 50%, and 25% in muscles, liver, kidney, and tongue collected from Egypt, respectively; while all samples collected from Saudi Arabia did not exceed MPL of As. Similarly, Hg did not exceed the set MPL of Hg (0.05 µg/g ww in muscles and offal, EC, 2006). Comparing results of the present study with other reports showed that 87.5% of samples collected from livestock in Alcudia Valley, Spain exceeded MPL of Pb (Pareja-Carrera *et al.*, 2014). Cd concentrations were higher than EU limits in hunted game animals in Slovakia (Gašparík *et al.*, 2017). El-Ghareeb *et al.* (2019) stated that 20% and 10% of liver and kidney samples of Camel collected from Saudi Arabia exceeded MPL of As. Malova *et al.* (2019) reported that 29% of the examined samples of wild living animals in Slovakia exceeded MPL of Hg set by European Union.

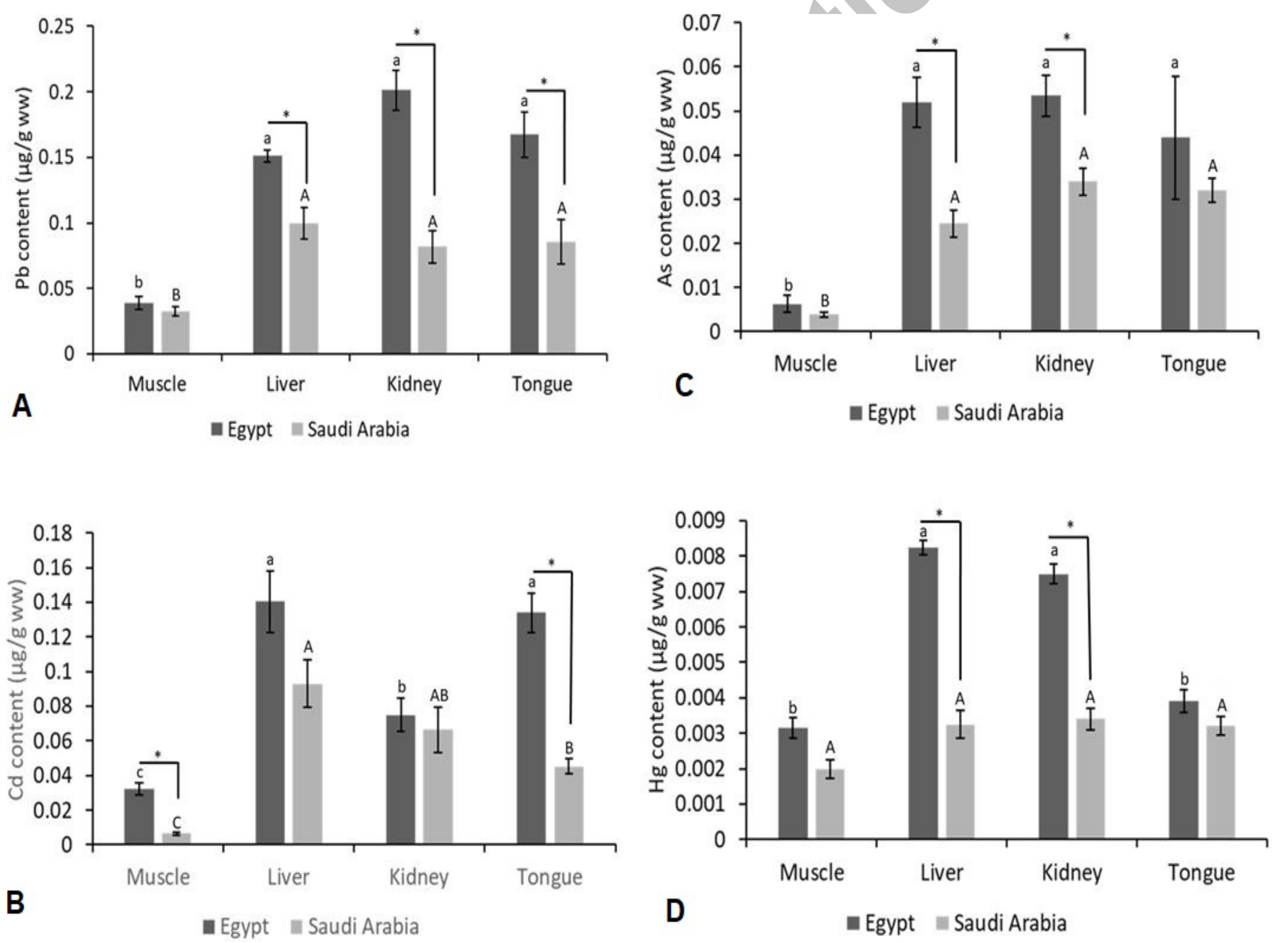

Fig. 1. A) lead (Pb), B) cadmium (Cd), C) arsenic (As), D) mercury (Hg) residual contents (µg/g ww) in camels' liver, kidney, tongue, and muscle retailed in Egypt and Saudi Arabia. Columns carrying different letters (a, b, and c) are significantly different among Egyptian samples; while columns carrying different letters (A, B, and C) are significantly different among Saudi samples. Star marks refers to significant difference between Egyptian and Saudi samples, $P < 0.05$.

Table I. Samples (%) exceeding maximum permissible limits of the estimated heavy metals in the camel's meat in Egypt and Saudi Arabia.

	Pb (µg/g)		Cd (µg/g)		As (µg/g)		Hg (µg/g)	
	Egypt	Saudi Arabia	Egypt	Saudi Arabia	Egypt	Saudi Arabia	Egypt	Saudi Arabia
Muscle	0	0	0	0	0	0	0	0
Liver	0	0	65	35	50	0	0	0
Kidney	0	0	20	20	50	0	0	0
Tongue	0	0	70	0	25	0	0	0

Mean±SEM; n, test of significance/ ANOVA
Values represent percentages (%) of samples exceeding the established maximum permissible limits of the tested heavy metals according to EU (2006).

Table II. Estimated daily intakes (EDI) and health risks (HR) associated with toxic metal residues in camel's meat consumed by adults in Egypt and Saudi Arabia.

Location	Sample	Pb (µg/g)		Cd (µg/g)		As (µg/g)		Hg (µg/g)		HI
		EDI	HR	EDI	HR	EDI	HR	EDI	HR	
Egypt	Muscle	0.081	0.020	0.067	0.067	0.013	0.043	0.006	0.013	0.144
	Liver	0.314	0.078	0.292	0.292	0.108	0.361	0.017	0.034	0.767
	Kidney	0.419	0.104	0.156	0.156	0.111	0.371	0.015	0.031	0.664
	Tongue	0.349	0.087	0.279	0.279	0.091	0.305	0.008	0.016	0.688
Saudi Arabia	Muscle	0.067	0.016	0.013	0.013	0.008	0.026	0.004	0.008	0.065
	Liver	0.207	0.051	0.194	0.194	0.051	0.170	0.006	0.013	0.429
	Kidney	0.170	0.042	0.138	0.138	0.070	0.236	0.007	0.014	0.432
	Tongue	0.178	0.044	0.094	0.094	0.066	0.222	0.006	0.013	0.375

Table III. Estimated daily intakes (EDI) and health risks (HR) associated with toxic metal residues in camel's meat consumed by children in Egypt and Saudi Arabia.

Location	Sample	Pb (µg/g)		Cd (µg/g)		As (µg/g)		Hg (µg/g)		HI
		EDI	HR	EDI	HR	EDI	HR	EDI	HR	
Egypt	Muscle	0.189	0.047	0.157	0.157	0.030	0.102	0.015	0.031	0.338
	Liver	0.734	0.183	0.682	0.682	0.253	0.843	0.040	0.080	1.790
	Kidney	0.979	0.244	0.365	0.365	0.260	0.867	0.036	0.073	1.550
	Tongue	0.814	0.203	0.652	0.652	0.214	0.713	0.018	0.037	1.606
Saudi Arabia	Muscle	0.158	0.039	0.031	0.031	0.018	0.062	0.009	0.019	0.152
	Liver	0.484	0.121	0.452	0.452	0.119	0.397	0.015	0.031	1.003
	Kidney	0.398	0.099	0.323	0.323	0.165	0.551	0.016	0.033	1.008
	Tongue	0.417	0.104	0.221	0.221	0.155	0.519	0.015	0.031	0.875

Discrepancies in environmental contamination scenarios and raising techniques are the main causes of the differences in the metal-accumulation patterns between this study and studies from other regions of the world. Additionally, inter-species variations in metal accumulation may be explained by variations in xenobiotic metabolizing enzymes and metal-detoxification capacities (Komsta-Szumska and Chmielnicka, 1983). Due to their substantial function in the metabolism and detoxification of xenobiotics, the liver and kidney of the camel in this study had higher levels of hazardous metals and trace elements than the muscle did (Jarup and Akesson, 2009).

Dietary intake and health risk assessment

The assessment of the health risks linked to Saudi Arabia's population's exposure to metals through the ingestion of camel offal is little known. Therefore, the current study calculated the daily intakes (EDI), hazard ratios (HR), and hazard indices (HI) of the examined elements to estimate the health risks among adults and children in Egypt and Saudi Arabia. The EDI (µg/kg/day) values for Pb among Egypt adults ranged between 0.081 (muscle) and 0.491 (kidney); for Cd ranged between 0.067 (muscle) and 0.0292 (liver); for As ranged between 0.013 (muscle) and 0.111 (kidney); and for Hg ranged between 0.006 (muscle) and 0.017 (liver). Such EDI values for Pb among Saudi Adults ranged between 0.067 (muscle) and 0.207 (liver); for Cd ranged between 0.013 (muscle) and 0.194 (liver); for As ranged between 0.008 (muscle) and 0.070 (kidney); and for Hg ranged between 0.004 (muscle) and 0.007 (kidney) (Table II). The calculated non-carcinogenic hazard ratios and hazard indices were far below one as shown in Table II, indicating no potential risks associated with the consumption of camel meat and edible offal. Further calculation of EDI, and HR values among Egyptian and Saudi children indicated the same trend as in the adults for each single tested metal. However, the calculated HI values for mixed contaminants among Egyptian children were higher than one for liver (1.790), kidney (1.550), and tongue (1.606) (Table III) indicating potential risk associated with the consumption of such edible offal among children in Egypt. The EDI of the studied heavy metals that consumed by the Egyptian and Saudi populations were equivalent to those that had been reported in Egypt (Darwish *et al*., 2015) and China (Jiang *et al*., 2016). The reported HR and HI values were lower than those from Ghana (Bortey-Sam *et al*., 2015) and Egypt (Darwish *et al*., 2015). The analyzed metals have a considerable impact on human health. Pb, for example, was the cause of numerous child fatalities in China (Xu *et al*., 2014) and Zambia (Yabe *et al*., 2015). Cadmium was linked to kidney dysfunction, and lung cancers (Nishijo *et al*., 2017). Arsenic was associated with skin lesions, metastatic carcinomas, and developmental and reproductive disorders (Thompson and Darwish, 2019). Mercury was also associated with neurological disorders (Cunningham and Saigo, 1997). Therefore, even minute concentrations of the tested toxic metals will accumulate by time and lead to several toxicological outcomes.

CONCLUSION

All tested samples included metals that have been proven to pose health hazards. This may be an indication of potential health risks for consumers, particularly youngsters, given the high meat intake in the research area. To analyze the amounts of heavy metals in various farm animals that accurately reflect the situations of environmental pollution in the study area, ongoing monitoring studies are still required.

NOVELTY STATEMENT

To the best of our knowledge this is the first study to compare the current levels of toxic metals in camel meat and offal in Saudi Arabia, and Egypt, and to calculate their dietary intakes, and their health risk assessment among Saudi and Egyptian populations.

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IRB approval

All the proceedure were approved by the institutional review board (IBR number DSR1811007).

Ethical statement

All experiments were done following the guidelines of both Zagazig and King Faisal Universities. Where no living animals nor human subjects were used in the present study.

Statement of conflict of interest

The authors have declared no conflict of interest.

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