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Heavy metal contents in salted fish retailed in Egypt: Dietary intakes and health risk assessment

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Abstract

Background: In Egypt, salted fish is considered a typically processed fish, including salted sardine, salted mullet (feseikh), keeled mullet (sahlia), and herrings. High-quality protein, polyunsaturated fatty acids, vital amino acids, and trace minerals such as magnesium and calcium are all abundant in fish. However, eating salted fish can expose people to toxins found in the environment, such as heavy metals.

Aim: In Zagazig, Egypt, four types of locally produced salted fish—salted sardine, feseikh, sahlia, and herrings—were tested for heavy metals, specifically lead (Pb), cadmium (Cd), arsenic (As), and mercury (Hg). Second, the assessed heavy metals linked to the Egyptian population's consumption of salted fish were used to calculate estimated daily intakes (EDIs) and potential health hazards, such as hazard quotient (HQ) and hazard index (HI).

Methods: Samples of salted herrings, feseikh, sahlia, and sardines were gathered from the markets in Zagazig. Samples of salted fish were subjected to acid digestion and then heavy metal extraction. Atomic absorption spectrometers (AAS) were used to measure heavy metals. HI, HQ, and EDI were computed computationally.

Results: With the exception of mercury, which was not found in the salted herrings, the recorded results showed that all of the tested metals were present in the samples that were evaluated. The herrings contained residual Pb and Cd contents that were highest, followed by sardine, feseikh, and sahlia, in that order. After sardine, herrings, and sahlia, feseikh has the greatest concentration. Sardine, feseikh, and sahlia had the highest quantities of mercury, in that order. A number of samples were found to be above the maximum allowable levels. There were no apparent hazards associated with consuming such conventional fish products, according to the computed HQ and HI values for the heavy metals under investigation based on the daily intakes.

Conclusion: Samples of salted fish sold in Zagazig, Egypt, had high quantities of the hazardous elements Pb, Cd, As, and Hg. Due to the bioaccumulation and biomagnification characteristics of these studied metals, such data should be taken carefully even though the computed health hazards revealed no potential problems.

Keywords: Heavy metals, Salted fish, Estimated daily intakes, Health risk assessment, Egypt.

Introduction

Fish is considered an essential source of protein because of its high biological value, polyunsaturated fatty acids, vitamins, and minerals including calcium and phosphorus. Egypt is becoming a more fish-consuming nation as a result of the scarcity of red meat and the cheaper cost of fish than chicken and meat (Morshdy *et al.*, 2019).

Fish preservation and the creation of novel fish products have long been accomplished through the

use of salting. Fish salting is associated with the creation of traditional fish products in Egypt, which are utilized as a significant source of protein in addition to being consumed on special occasions. Salted fish products that are locally produced and stink are salted mullet (fesiekh), salted sardine, salted keeled mullet (sahlia), and salted herrings that are smoked after being salted (Abbas *et al.*, 2021). Fish preserved for an extended period of time with salting has a positive

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microbiological impact on the fish. The chemical residues in the used fish, it has minimal effect.

Lead (Pb), cadmium (Cd), mercury (Hg), and arsenic (As) are examples of heavy metals that are distinguished by their capacity for bioaccumulation and biomagnification. When fish products are produced, metals from the surrounding aquatic environment as well as from the salting and smoking procedures can seep into the fish flesh (Morshdy *et al.*, 2021). Consequently, eating tainted fish can allow these toxicants to enter the human body (Morshdy *et al.*, 2013). However, not much is known regarding the presence of heavy metals in salted fish sold for retail in Egypt.

Heavy metals have several detrimental effects on human health. Lead, for instance, is one of the heavy metals that have been linked to many child fatality incidents, according to reports from China (Xu et al., 2014) and Zambia (Yabe et al., 2015). Furthermore, Pb has detrimental effects on IQ and mental health (Thompson and Darwish, 2019). Cadmium is another heavy metal that has no known physiological function. Cadmium is the primary cause of itai-itai illness, which has been connected to significant fish intake in Japan. Kidney failure and osteomalacia are the hallmarks of this illness (Nishijo et al., 2017). Furthermore, according to the IARC (2016), Cd is categorized as a group B1 carcinogen. Arsenic is another dangerous heavy metal that has been linked to a number of health problems, including skin irritation, cancer, and damage to multiple organs (Reichard and Puga, 2010). Japan saw the first cases of Minamata sickness in the 1950s as a result of eating mercury-contaminated fish. Furthermore, autism and abnormalities in neurological behavior are linked to mercury exposure (Clifton, 2007).

With consideration for the preceding information, this investigation was conducted to determine the residual Pb, Cd, As, and Hg concentrations in four varieties of retail salted fish products sold in Egypt: salted sardine, herrings, fesiekh, and sahlia. In addition, we computed the Egyptian consumers' anticipated daily intakes and the possible health concerns related to eating salted fish.

Materials and Methods

The present investigation was carried out in compliance with the criteria issued by Zagazig University in Egypt. The best quality chemicals were bought from Merck in Darmstadt, Germany, and utilized in this investigation. *Sample collection*

In Zagazig City, Sharkia Governorate, Egypt, 80 haphazard salted fish samples, comprising 20 pieces of salted herring, salted sardine, fesiekh, and sahlia, were gathered from salted fish businesses and hypermarkets. Samples were refrigerated until measurements and heavy metal extraction were completed. The Central Laboratory, Faculty of Veterinary Medicine, Zagazig University, Egypt, was where metal measurements were conducted, and the Meat Hygiene Laboratory was where samples were prepared.

Sample preparation

Sample preparation and metal measurements were finished in accordance with earlier protocols (Darwish *et al.*, 2015, 2018). One gram of muscle tissue from each salted fish sample was essentially digested using 5 ml of a digestion solution that contained 3 ml of nitric acid (65%) and 2 ml of perchloric acid (70%). The homogenate was allowed to stand at room temperature for 12 hours. After that, the mixture was incubated for 3 hours at 70°C in a water bath, stirring every 30 minutes. After bringing the digested mixture to room temperature and diluting it with 20 ml double distilled water (DDW), filter paper was used to filter it. The filtrate was kept at room temperature until the heavy metals were measured. *Analytical method*

The Perkin Elmer® PinAAcleTM 900T atomic absorption spectrophotometer (Shelton, CT) was utilized for the measurement of lead and cadmium using graphite furnace atomic absorption spectroscopy. In contrast, hydride generation/cold vapor atomic absorption spectroscopy (Shelton, CT) was used to measure As and Hg. The central laboratory of Zagazig University's Faculty of Veterinary Medicine in Egypt conducted heavy metal assessments.

Quality assurance

The National Research Council of Canada's DORM-3 fish protein reference material was used to guarantee the correctness of the analytical procedures. The limits of detection (μ g/g) for the metals that were analyzed were 0.001 for Hg, 0.005 for Cd, 0.02 for As, and 0.01 for Pb. The studied metals had recovery rates ranging from 90% to 105%. The determination coefficient (R^2) varied from 0.980 to 0.999 for every metal that was analyzed.

Estimated daily intake (EDI)

The following equation was used to compute the values of the EDI for the observed heavy metals obtained from the Egyptian population's intake of salted fish:

EDI $(\mu g/kg/dav) = Cm*FIR/BW$ (US EPA, 2010)

FIR is the rate of fish intake by the Egyptian population, and Cm is the concentration of the analyzed metal (μ g/g ww). FIR was established at 48.57 g/day (FAO, 2010); adult Egyptian body weight (BW) is 70 kg.

Evaluation of health risks

The noncarcinogenic danger of the tested heavy metals was expressed by its hazard quotient (HQ), which was computed using the following formula:

 $HQ = EDI/RFD*10^{-3}$ (US EPA, 2017)

where RFD stands for recommended reference dosage (mg/kg/day), and the values for Pb, Cd, As, and Hg were established as follows: 0.004, 0.001, 0.0003, and 0.0005 (US EPA, 2017).

Moreover, the noncancer danger of mixed metals was calculated using a hazard index (HI). HI was computed using the subsequent equation:

Since each metal is represented by an i, HI = \sum HRi

A possible risk to human health was suggested when either HQ or HI value was more than 1.

Statistical analysis

The statistical analysis was performed using the Tukey–Kramer honestly significant difference (HSD) test (JMP) and a one-way analysis of variance (SAS Institute, Cary, NC). p < 0.05 was regarded as statistically significant. The data were presented in the form of means \pm standard deviation.

Ethical approval

Not needed for this study.

Results

The current study's documented findings showed that Pb was found in all of the samples that were analyzed. The Pb concentration (μ g/g ww) of herrings was 0.72 \pm 0.03, significantly (p < 0.05) higher than that of sahlia (0.32 ± 0.06), feseikh (0.32 ± 0.01), and salted sardine (0.19 ± 0.01), in that order (Fig. 1). In the salted sardine, feseikh, sahlia, and herrings samples under examination, the percentages of samples that exceeded the maximum permitted limits (MPLs) of Pb were 0%, 15%, 20%, and 100%, respectively. When consuming such salted fish products, the EDI values (μ g/kg/day) for Pb varied from 0.132 to 0.501. In every product, the HQ values were less than 1 (Table 1).

In addition, cadmium was found in every salted fish sample that was tested. Like Pb, herrings had the

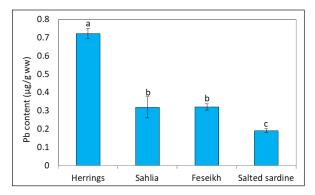


Fig. 1. Shows the residual lead (Pb) content ($\mu g/g$ ww) in the salted fish that was tested. Means \pm standard error are shown for each variety of salted fish (n = 20). At p < 0.05, there is a substantial difference between columns that carry different letters.

greatest Cd level ($\mu g/g \text{ ww}$) (0.09 ± 0.006), with sahlia (0.05 ± 0.003), salted sardine (0.03 ± 0.003), and feseikh (0.06 ± 0.003) following (Fig. 2). The data that was acquired showed that the amounts of Cd in the salted sardine, feseikh, sahlia, and herrings that were investigated were higher than the MPL by 5%, 55%, 40%, and 95%, respectively. For Cd, the EDI values varied from 0.021 to 0.065. Similar to Pb, no product's HQ values were more than 1 (Table 1).

In addition, arsenic was found in every salted fish sample that was tested; the range of concentrations was $0.86-1.53 \ \mu g/g$ ww in feseikh, $0.55-1.23 \ \mu g/g$ ww in sahlia, $0.42-0.84 \ \mu g/g$ ww in herrings, and $0.16-0.39 \ \mu g/g$ ww in salted sardine. In terms of its As residues, feseikh was ranked highest, followed by salted sardin, herrings, and sahlia, in that order (Fig. 3). 90% of the salted sardine samples that were analyzed, as well as the samples of sahlia, feseikh, and herrings, had arsenic contents that were higher than the MPL for total arsenic. In salted sardine, the EDI values for As varied from 0.651 to 2.565 in feseikh. In every product, the HQ values were less than 1 (Table 1).

Mercury was most likely found in all of the samples of salted sardine, feseikh, and sahlia that were tested, although it was not found in any of the tests of herrings. The studied sahlia, feseikh, and salted sardine had average Hg concentrations (μ g/g ww) of 0.006 ± 0.0002, 0.003 ± 0.0002, and 0.002 ± 0.0002, respectively (Fig. 4). The determined HQ values for Hg did not exceed 1, and none of the tested samples went over the predetermined MPL limits (Table 1). In no salted fish product did the computed HI values for total pollutants surpass 1.

Discussion

Heavy metals are characterized by their bioaccumulative and biomagnification nature, and cannot be eliminated via various fermentation and salting processes performed on salted fish (Thompson and Darwish, 2019). The latter is very popular in Egypt and consumed on special occasions and festivals since ancient times. Heavy metals can reach the fish during their lifetime and accumulate in the fish's flesh and various tissues. Subsequently, such pollutants can find their way into the human body via ingestion of such contaminated products. Several studies were conducted before to estimate toxic metal residues in raw and

Table 1. Daily intake and health risk assessment of the tested heavy metals due to ingestion of salted fish.

HI	Hg			As			Cd			Pb			
	HQ	EDI	%	HQ	EDI	%	HQ	EDI	%	HQ	EDI	%	
0.251	0.002	0.003	0	0.195	0.651	90	0.021	0.021	5	0.033	0.132	0	Salted sardine
0.868	0.002	0.005	0	0.769	2.565	100	0.042	0.042	55	0.055	0.223	15	Feseikh
0.673	0.004	0.008	0	0.579	1.932	100	0.035	0.035	40	0.055	0.221	20	Sahlia
0.581	NA	NA	0	0.391	1.301	100	0.065	0.065	95	0.125	0.501	100	Herrings

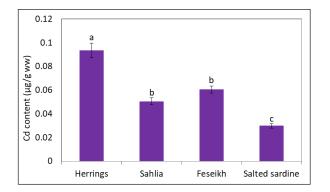


Fig. 2. Shows the residual cadmium (Cd) content (μ g/g ww) in the salted fish that was tested. Means \pm standard error are shown for each variety of salted fish (n = 20). At p < 0.05, there is a substantial difference between columns that carry different letters.

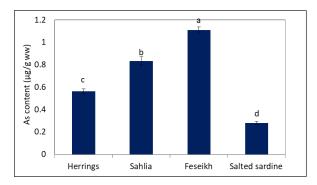


Fig. 3. Shows the residual arsenic (As) content ($\mu g/g$ ww) in the salted fish that was tested. Means \pm standard error are shown for each variety of salted fish (n = 20). At p < 0.05, there is a substantial difference between columns that carry different letters.

canned fish (Ashraf et al., 2006; Morshdy et al., 2013; Leung et al., 2014; Morshdy et al., 2019). However, few reports investigated the heavy metal load in the salted fish. In the current study, the tested metals have no biological values but might adversely affect human health. All tested metals were detected in all examined salted fish products except for the mercury which was not detected in the salted and smoked herrings. This can be attributed to the fact that during the manufacturing process of the smoked herrings, the fish is exposed to elevated temperature which leads to evaporation of the mercury even if present in the raw fish. Studying the contamination of fermented fish with environmental pollutants has received less attention. However, in the studies performed heavy metals were detected at variable rates. For instance, Pb and Cd were detected at comparable levels to what was recorded in the current study in the fermented salted shrimp sampled in Korea, Cd was detected at concentrations up to 0.5 ppm, while lead was detected up to levels reaching 0.8 ppm (Heo

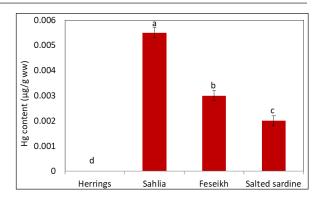


Fig. 4. Shows the residual mercury (Hg) content ($\mu g/g$ ww) in the salted fish that was tested. Means \pm standard error are shown for each variety of salted fish (n = 20). At p < 0.05, there is a substantial difference between columns that carry different letters.

et al., 2005). Besides, Storelli et al. (2011) detected Hg, Pb, and Cd in the salted anchovies collected from Italian supermarkets. The anchovies were harvested primarily from the Mediterranean Sea and the Atlantic Ocean. The recorded concentrations were in agreement with the recorded values in the current investigation. Higher levels of Pb were recorded in the salted sardine and smoked herrings sampled from Damanhour city, Egypt reaching values of 2.097 ± 0.224 (salted sardine), and 1.929 ± 0.211 (herrings) mg/kg wet weight, respectively, with an incidence of 100% of each. All samples exceeded the established MPL for Pb (El-Kewaiev et al., 2011). In addition, higher concentrations for Hg and Pb were recorded in salted sardine and feseikh samples collected from Menoufia governorate, Egypt, with 65%, and 80% of sardine and feseikh samples regarded as unacceptable for Hg residues, and 45%, and 65% of the two salted fish types are unacceptable for Pb residues (Elrais et al., 2018). Arsenic content was recorded at higher levels in salted fish retailed in Malaysia (Jeevanaraj et al., 2020). The salting process had minor effects on the accumulated metals as reported before (Abbas et al., 2021).

The data obtained demonstrate the inter-species differences in the accumulation of heavy metals; for example, sardines had the lowest concentrations of various metals, while herrings and feseikh had the greatest residual amounts. This could rely on where the fish is in the food chain. The high concentrations of Pb and As in the salted fish products under examination indicate that the fish were either heavily exposed to these metals during their lives or that the contamination occurred during post-processing or in the salting tanks (Morshdy *et al.*, 2019; Abbas *et al.*, 2021).

The present study's measured quantities of the heavy metals under examination frequently surpassed the permissible maximum permitted level (MPL) (European Commission, 2007). As a result, the EDI, HQ, and HI calculations in this study were expanded to assess the possible health hazards related to the consumption of such fish products. The measured HQ and HI values for the metals under examination showed values much below one for every metal tested, suggesting that there were no health hazards connected to these metals from salted fish. According to Jeevanaraj *et al.* (2020), salted fish sold in Malaysia had higher HI values. According to Morshdy *et al.* (2021), heavy metals are linked to a number of toxicological and harmful health outcomes, including various organ damage, carcinogenesis, and pediatric fatalities.

Conclusion

The current study's results showed that Pb, Cd, As, and Hg were detected in the salted fish under examination at varying concentrations. Samples had residual levels higher than the required MPL on multiple occasions, especially for Pb, and As. There were no possible hazards identified for the Egyptian population by the computed EDI, HQ, or HI. The bioaccumulation and biomagnification of heavy metals necessitate cautious handling of such assumptions.

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Authors' contributions

MM and NM collected samples and performed extraction and metal analysis. Validation of data AM, AT, WD, WE, AFA, conceptualization AM, AT, WD, WE, AFA, resources, WD, WE, AFA, supervising the work AM, AT, WD, and HM. MM and WD drafted the manuscript.

Conflict of interest

The authors declare that there is no conflict of interest. *Funding*

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Data availability

Data will be made available upon a reasonable request.

References

- Abbas, M.M.M., Shehata, S.M., Talab, A.S. and Mohamed, M.H. 2021. Effect of traditional processing methods on the cultivated fish species, Egypt. Part I. Mineral and heavy metal concentrations. Biol. Trace Elem. Res. 200, 2391– 2405.
- Ashraf, W., Seddigi, Z., Abulkibash, A. and Khalid, M. 2006. Levels of selected metals in canned fish consumed in Kingdom of Saudi Arabia. Environ. Monit. Assess. 117(1–3), 271–279.
- Clifton, J.C. 2nd. 2007. Mercury exposure and public health. Pediatr. Clin. N. Am. 54(2), 237–269.
- Darwish, W.S., Atia, A.S., Khedr, M.H. and Eldin, W.F.S. 2018. Metal contamination in quail meat: residues, sources, molecular biomarkers, and human health risk assessment. Environ. Sci. Pollut. Res. 25, 20106–20115.

- Darwish, W.S., Hussein, M.A., El-Desoky, K.I., Ikenaka, Y., Nakayama, S., Mizukawa, H. and Ishizuka, M. 2015. Incidence and public health risk assessment of toxic metal residues (cadmium and lead) in Egyptian cattle and sheep meats. Int. Food. Res. J. 22(4), 1719–1726.
- El-Kewaiey, I.A., Ali, O.M.A. and Saleh, O.M.A. 2011. Incidence of heavy metals residues in salted and smoked fish products. Assiut Vet. Med. J. 57(131), 1–17.
- Elrais, A., Hassanien, F., Hassan, M.A., Latif, H.M. and El-Zahaby, D. 2018. Detection of some heavy metals in fresh and salted fish. Benha Vet. Med. J. 35(2), 169–177.
- European Commission. 2007. Commission Regulation (EC) 2007. No 333/ 2007 of 28 March 2007 laying down the methods of sampling and analysis for the official control of the levels of lead, cadmium, mercury, inorganic tin, 3-MCPD and benzo(a) pyrene in foodstuffs. Off J Eur Union. L88, 29–38.
- FAO. 2010. Nutrition country profiles—Egypt. Rome, Italy: FAO. 2010.
- Heo, O.S., Oh, S.H., Shin, H.S. and Kim, M.R. 2005. Mineral and heavy metal contents of salt and saltedfermented shrimp. Korean J. Food Sci. Technol. 37(4), 519–524.
- IARC. 2016. monographs on the identification of carcinogenic hazards to humans. Available via *https://monographs.iarc.fr/agents-classified-by-the-iarc/*
- Jeevanaraj, P., Ahmad Foat, A., Tholib, H. and Ahmad, N.I. 2020. Heavy metal contamination in processed seafood and the associated health risk for Malaysian women. Br. Food J. 122(10), 3099–3114.
- Leung, H.M., Leung, A.O., Wang, H.S., Ma, K.K., Liang, Y., Ho, K.C., Cheung, K.C., Tohidi, F. and Yung, K.K. 2014. Assessment of heavy metals/ metalloid (As, Pb, Cd, Ni, Zn, Cr, Cu, Mn) concentrations in edible fish species tissue in the Pearl River Delta (PRD), China. Mar. Pollut. Bull. 78(1–2), 235–45.
- Morshdy, A.E., Hafez, A.E., Darwish, W.S., Hussein, M.A. and Tharwat, A.E. 2013. Heavy metal residues in canned fishes in Egypt. Jpn. J. Vet. Res. 61, S54–S57.
- Morshdy, A.E.M.A., Darwish, W.S., Daoud, J.R.M. and Sebak, M.A.M. 2019. Estimation of metal residues in *Oreochromis niloticus* and *Mugil cephalus* intended for human consumption in Egypt: a health risk assessment study with some reduction trials. J. Consum. Protect. Food Saf. 14, 81–91.
- Morshdy, A.E.M., Darwish, W.S., Hussein, M.A., Mohamed, M.A.A. and Hussein, M.M. 2021. Lead and cadmium content in Nile tilapia (*Oreochromis niloticus*) from Egypt: a study for their molecular biomarkers. Sci. Afr. 12, e00794.
- Nishijo, M., Nambunmee, K., Suvagandha, D., Swaddiwudhipong, W., Ruangyuttikarn, W. and

Nishino, Y. 2017. Gender-specific impact of cadmium exposure on bone metabolism in older people living in a cadmium-polluted area in Thailand. Int. J. Environ. Res. Public Health 14(4), 401.

- Reichard, J.F. and Puga, A. 2010. Effects of arsenic exposure on DNA methylation and epigenetic gene regulation. Epigenomics 2(1), 87–104.
- Storelli, M.M., Giachi, L., Giungato, D. and Storelli, A.
 2011. Occurrence of heavy metals (Hg, Cd, and Pb) and polychlorinated biphenyls in salted anchovies.
 J. Food Prot. 74(5), 796–800.
- Thompson, L.A. and Darwish, W.S. 2019. Environmental chemical contaminants in food: review of a global problem. J. Toxicol. 2019, 2345283.
- US EPA. 2010. Integrated risk information system (IRIS). Cadmium (CASRN-7440-43-9). 2010.

Available via http://www.epa.gov/iris/subst/0141. htm

- US EPA. 2017. Human health risk assessment. 2017. Available via https://www.epa.gov/risk/hu-manhealth-risk-assessment
- Xu, J., Sheng, L., Yan, Z. and Hong, L. 2014. Blood lead and cadmium levels of children: a case study in Changchun, Jilin Province, China. West Ind. Med. J. 63, 29–33.
- Yabe, J., Nakayama, S.M., Ikenaka, Y., Yohannes, Y.B., Bortey-Sam, N., Oroszlany, B., Muzandu, K., Choongo, K., Kabalo, A.N., Ntapisha, J., Mweene, A., Umemura, T. and Ishizuka, M. 2015. Lead poisoning in children from townships in the vicinity of a lead-zinc mine in Kabwe, Zambia. Chemosphere 119C, 941–947.