



Mercury contamination in fishermen's hair in Zabol Chahnimeh Reservoirs, Iran

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Article Info	Abstract
Article type: Research Article	Aquatic ecosystems contaminated by heavy metals are a worldwide concern. These metals are toxic and persistent. This research aimed to measure mercury in the hair of fishermen, working at Chahnimeh Reservoirs, Sistan and Baluchistan Province, south-east of Iran. Samples were collected from April 2012 to October 2012 from 40 local fishermen. Those fishermen with amalgam-filled teeth (a source of mercury) were excluded. The average amount of mercury in the samples was 1.56 µg/g (ranging from 0.1 to 3.65 µg/g). In 29 fishermen (72%) hair mercury levels exceeded the reference dose of the USEPA. The results in this study showed that height and age were not significantly related to mercury levels, but body weight and BMI were. Fish consumption ($p=0.001$), smoking ($p=0.018$), weight ($r=0.35$, $p=0.02$) and BMI ($r=0.35$, $p=0.02$) were significantly related to hair mercury levels. Fruit ($p=0.017$) and dairy consumption ($p<0.001$) were inversely related to the concentration of mercury in hair.
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Introduction

Aquatic ecosystems contaminated by heavy metals are a worldwide concern. These metals are toxic and persistent. They can accumulate in the food chain, enter drinking water sources and cause serious health hazards for humans (Baramaki Yazdi et al., 2012).

Mercury is now a major environmental pollutant. It is released from both natural and anthropogenic sources. Mercury has significant adverse effects on fetal neurodevelopment and damages liver and

kidney tissues in adults (Khan et al., 2019; Yang et al., 2020). The most common forms of mercury in aquatic ecosystems are elemental mercury (Hg^{2+}) bonded with inorganic or organic chemicals; and organic mercury in the form of methyl or di-methyl-mercury (Djermanovic et al., 2020).

The levels of mercury have been studied in many human populations in Iran (Barghi et al., 2012; Dahmardeh Behrooz et al., 2012; Golpayegani et al., 2015; Okati and Esmaili-sari 2018), but little research has been done on mercury concentrations in

people living in Sistan and Baluchistan Province, south-east of Iran.

The Chahnimeh water reservoirs are located in the Sistan and Baluchistan Province. Their water has been supplied from the Hirmand River since 1983. These reservoirs hold around 1.5 billion m³ of water, are located in the south of the Sistan Plain, and cover an area of 30,000 hectares (Figure 1). The water stored in these reservoirs is used to irrigate the Sistan Plain and provides potable water for Zabol and Zahedan cities. The total capacity of Chahnimeh reservoirs at its best, is one-seventh of the Hamoun International Wetland. It is also an important wintering site for migratory birds and the most important and the only place for catching fish in the Sistan region, at times when the Hamoun wetland is dry. Chahnimeh reservoirs have served as a water storage, especially during the droughts that have happened over the last three decades. The Hirmand (Helmand) River is a transboundary river, that has faced a decline in streamflow in recent decades. The river is the most important water source shared between Iran and Afghanistan and plays an important role in the livability and economy of this region. However, fast agricultural developments (UNODC, 2016) in the basin of Hirmand river threaten the region's ecological stability.

Mercury contamination in the general population with no occupational exposure, happens mainly through consuming fish or other seafood contaminated with methylmercury (MeHg) (Agah et al., 2007; Giangrosso et al., 2016) or through the release of mercury from dental fillings (Bates 2006; Clarkson et al., 2003). Mercury contamination in humans increases with increased fish consumption; and increased mercury contamination in the fish consumed (Clarkson, 2002).

Mercury has been measured in human breast milk, hair, nail, blood, and adipose tissue (Barghi et al., 2012; Dahmardeh Behrooz et al., 2012; Park et al., 2017; Zolfaghari et al., 2007). Assessment of hair samples is a non-invasive method with advantages such as easy collection, low cost, easy transfer and storage used widely

for investigating mercury exposure in humans (Xiaojie et al., 2008) (Esteban and Castaño 2009). Hair samples provide information about short- and long-term mercury exposures (Esteban and Castaño 2009).

Fishermen are a population that have been reported to have hair mercury concentrations significantly higher than others, which is probably because of high fish consumption or their closeness to polluted sites such as ports or industrial wastewater (Giangrosso et al., 2016).

The aim of this study was to determine the level of mercury in fishermen in Sistan region and the factors affecting the level of mercury among these people who comprise a large part of the population in the region. This study can also provide indirect estimates about mercury contamination in this region.

Materials and Methods

The Chahnimeh water reservoirs in the east of Iran located between 61° 37' to 61° 46' longitudes, and 30 ° 43' to 30° 52' latitudes have been formed in four geological depressions. This cross-sectional study was conducted from April 2012 to October 2012. A number of posters were installed in locations close to fishing areas in the south of Zabol, a large city in the region and the Chahnimeh and fishermen were asked to voluntarily participate in this research. Eventually 40 fishermen volunteered to participate.

Hair samples were taken from forty fishermen working in the Sistan area (Figure 1). In order to evaluate the impact of fish consumption on mercury intake, fishermen with amalgam-filled teeth were excluded. This was important because amalgam used for dental filling is a source of mercury contamination in humans (Ritchie, 2002).

A questionnaire about demographic information and factors probably affecting the amount of mercury intake was completed for each fisherman. This questionnaire included questions about their weight, height, number of filled-teeth, the number of fish meals they ate, fruits, vegetables and dairy products per week.

The hair sample of each participant was taken from the occipital region using clean scissors. Hair bunch samples were at least three centimeters long. These samples were stored in plastic bags. In order to measure mercury, hair samples were washed with a nonionic detergent and then rinsed two times with deionized water. After this, samples were dried at 60 °C in an electric oven (Mortada et al., 2002); and were then cut into small pieces.

About 0.2 grams of dry hair was taken from each sample and was placed in a 25 ml Teflon digestion tank including 4 ml concentrated ultrapure 65% nitric acid (Suprapure, Merck, Darmstadt, Germany) and 4 ml 30% hydrogen peroxide (30% Merck, Suprapur). The tank was closed tight and placed in an electric oven at 180°C for 45 minutes. After this, the tank was taken out of the oven and allowed to cool in room temperature. Then the solution was transferred into a colorimetric tube, and 5% hydrochloride (Suprapure, Merck, Darmstadt, Germany) was added to it, until it reached 10 milliliters; and was then thoroughly mixed.

Mercury was measured by cold vapour atomic absorption spectrophotometry (AAS), Perkin Elmer Analyst 700, equipped with a MHS 15 CVAAS system (Perkin–Elmer Instruments, Waltham, MA,

USA). Results were reported as milligrams per kilogram dry weight. Procedural blanks and certified reference material (CRMs, e.g. DOLT-2, DORM-2 and IAEA086) were included in each sample batch. In order to determine the detection limit of mercury in the hair samples, blank samples were injected three times for analysis and the result with 3-times the standard deviation of the procedural blanks was 0.08 µg/g dw. The precision and accuracy of the applied analytical method was estimated based on CRMs, e.g. DOLT-2 and DORM-2 and IAEA086 mercury in human scalp hair. The results of our CRMs measurements (Table 1) were a good estimate of the real values. In each sample batch, procedural blanks and certified reference material (CRMs, e.g. DOLT-2 and DORM-2) were included. For each matrix, analyses of three blank samples and analysis of reagent blanks was performed. In order to estimate the accuracy and precision of the chemical analysis; sample blanks, standard blanks and three analytical duplicates with the concentration of 1.2 µg/g were injected and their mean and its 95% confidence interval was calculated. Quantification was based on multi-level calibration on the concentrations of 0.1, 0.5, 1, 3 and 5 µg/g; and then the standard calibration curve was drawn with 99% accuracy.

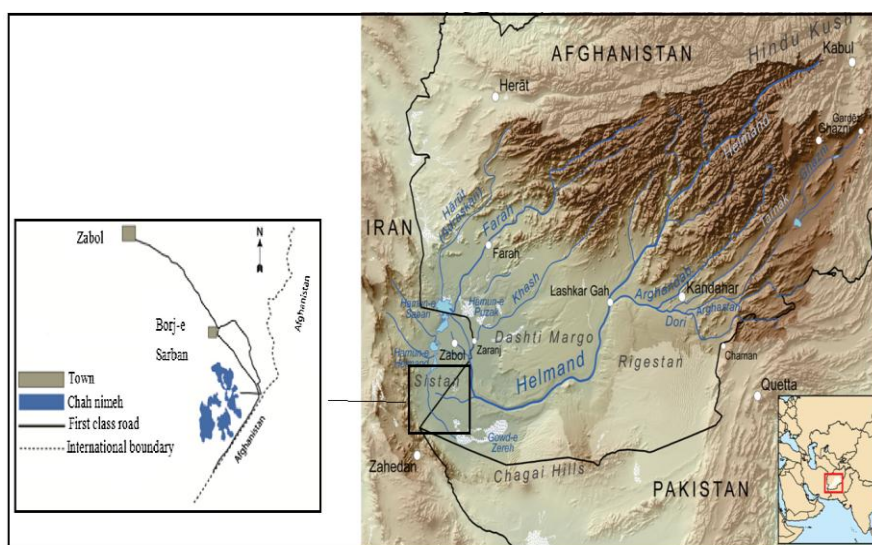


Figure 1. Map showing sampling locations

Table 1. The results of the certified samples in $\mu\text{g/g}$.

	Certified value	Our results	Recovery
DOLT-3	3.37±0.14	3.39±0.07	99±4
DORM-2	4.64±0.26	4.41±0.10	105±2
IAEA 086	0.534-0.612	0.556-0.611	96±3

Statistical analysis was performed using SPSS16 software. The normality of variables was assessed using Kolmogorov–Smirnov test which showed that mercury concentrations were normally distributed ($P>0.05$). The differences in Hg concentrations in subgroups were determined by independent *t*-tests. Spearman's rank correlation coefficients were used to test the correlations between mercury levels with weight, height, and age of the fisherman. A *p*-value less than 0.05 indicated statistical significance.

Results and Discussion

The average amount of mercury in the samples was $1.56 \mu\text{g/g}$ (ranging from 0.1 to $3.65 \mu\text{g/g}$). The reference intake dose (RfD) of mercury according to USEPA is $0.1 \mu\text{g}$ mercury per kilogram of body weight per day, and this corresponds to about $1.0 \mu\text{g/g}$ in hair samples (USEPA 2005). In this study, 29 fishermen (72%) had mercury levels over the RfD, with the range being from 1.11 to $3.65 \mu\text{g/g}$ (Figure 2). None of the volunteers had filled teeth and none of them had orthodontic gear.

The mean mercury concentration was significantly higher in hair samples obtained from fishermen eating fish ≥ 2 times a week (number=27) ($1.90 \pm 0.90 \mu\text{g/g}$) compared to those who consumed fish <2 meals per week (number=13) ($0.85 \pm 0.65 \mu\text{g/g}$, $p=0.001$) ($p < 0.05$) (Figure 2). Also, mercury levels in 81.4% of fishermen who ate fish more than 2 times per week were higher than the RfD (USEPA) (Figure 2). The source of this contamination is more likely to be fish consumption, because mercury levels in the fish samples from this region in the year

2012 were higher than WHO, FDA and US-EPA permissible limits (Dahmardeh Behrooz et al., 2013). Mercury in the fish of this area appears to originate from the use of fertilizers and pesticides in the agricultural lands around the Hirmand River in Iran and Afghanistan.

The results also indicated significant differences between mercury concentration in smoking fisherman (number=9) ($2.21 \pm 0.95 \mu\text{g/g}$) and non-smoking ones (number=31) ($1.37 \pm 0.81 \mu\text{g/g}$, $p=0.018$). Mercury levels in 100% of smoking fishermen were higher than RfD (USEPA) while in only 18% of non-smoking fishermen mercury levels were higher than RfD (USEPA) (Figure 2). This might be due to mercury in tobacco entering the body of smoking persons (Kowalski and Wiercinski, 2009). Higher mercury contamination among smokers has also been seen in other studies (Hong et al., 2013; Pirard et al., 2018; Rumiantseva et al., 2021).

The mean mercury concentration in fishermen who consumed dairy products once a week was (number = 24) ($2.40 \pm 0.73 \mu\text{g/g}$) and for those who consumed dairy more than once a week was (number = 16) ($0.99 \pm 0.50 \mu\text{g/g}$) ($p < 0.001$), (Figure 2). Also, mercury levels in 100% of fishermen who were eating dairy once a week were higher than RfD (USEPA) while it was higher in only 18% of fishermen eating dairy more than once a week (Figure 2).

Some studies suggest consuming milk products may decrease the absorption of mercury (Abdel-Salam et al., 2010; Reid, 2015). Future research is needed to clarify the link between dairy consumption and mercury levels.

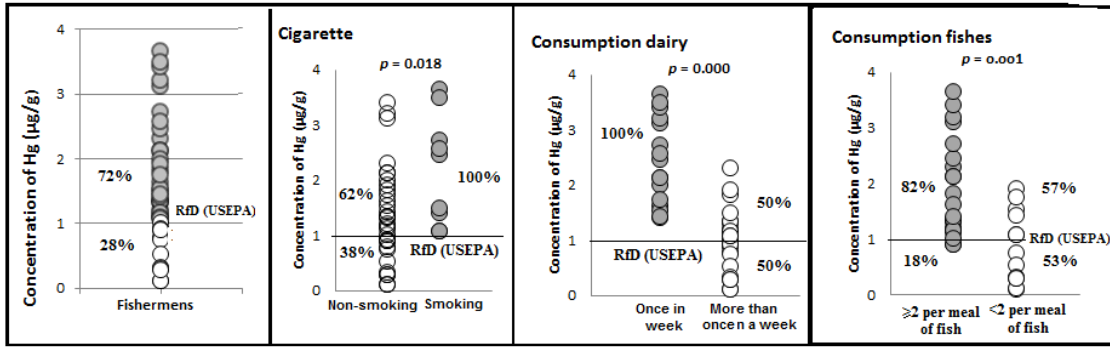


Figure 2. Difference between mercury level and exposure source

From left to right; mercury level in all fishermen, mercury level in smoking and non-smoking fisherman, mercury level in fishermen consuming dairy products once a week compared with those who consumed dairy more than once a week and mercury level in fishermen that ate fish ≥ 2 per week compared with fishermen that ate fish < 2 per week, respectively.

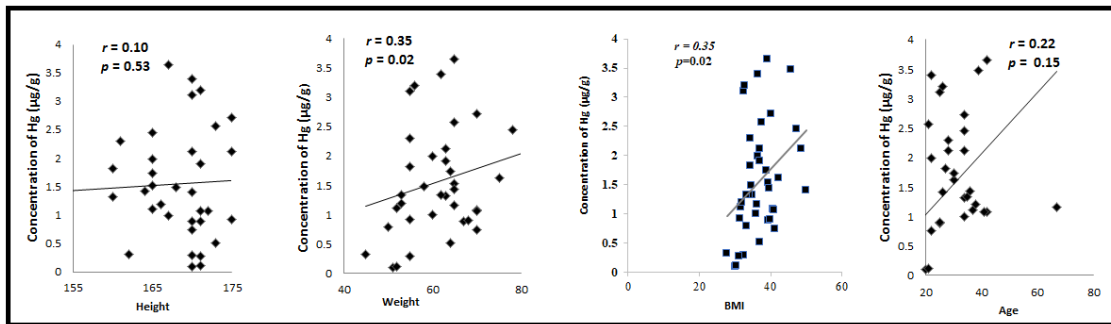


Figure 3. Correlation between hair mercury and age, weight, height and BMI among the participants.

The results of our study showed that height and age were not significantly related to mercury levels, but body weight and Body Mass Index (BMI) were (Figure 3). Results from studies conducted in other countries also indicated that as weight increases, so does hair mercury concentrations (Babi et al., 2000; Díez et al., 2008).

Lim et al. also reported a significant positive correlation ($p < 0.0001$) between hair mercury concentration and BMI in Korea (Lim et al., 2010), although other studies such as the one from Naples, Italy, in which total mercury concentrations in human hair ranged from 0.221 to 3.402 $\mu\text{g/g}$, did not show a statistically significant correlation between total mercury concentrations in human hair and BMI ($r=0.028$; $p=0.663$) (Díez et al., 2008). Also no statistically significant correlation was seen between total mercury in human hair and BMI ($r=-0.15$, $p=0.29$) or methylmercury in human hair and BMI ($r = 0.05$; $p = 0.72$) in Brazil (Barbosa et al., 2001); and no significant correlation was

reported between hair levels of Hg and BMI ($r=0.095$, $p=0.313$) in Eritrea (Astolfi et al., 2020).

Conclusion

The levels of mercury in hair of fishermen from the study area suggest that more extensive and intensive mercury monitoring of items consumed by humans combined with health directives are necessary to protect humans from poisoning by mercury.

Author contributions

All the authors contributed to this work.

Availability of data and materials

The authors declare that the data and materials for this work are available.

Conflict of interest

The authors declare no conflict of interest. All the authors have read and approved the manuscript.

Ethical statement

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee of the University of Zabol.

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