HUMAN EXPOSURE ASSESSMENT TO MERCURY THROUGH HAIR ANALYSIS IN COASTAL VILLAGES OF THE VALPARAISO REGION (CHILE)

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ABSTRACT

In the present study, we assessed mercury exposure of residents of different sites of Valparaíso Region (Chile) due to fish consumption and atmospheric contamination from coal-burning power plant and copper smelter. Total mercury concentration was determined in 199 hair samples and in 14 species of marine fish, purchased in the supermarket or collected in the studied fishing villages. The total mercury (THg) concentrations ranged from 0.04 to 1.78 $\mu\text{g g}^{-1}$ in human hair samples and from 0.04 to 3.87 $\mu\text{g g}^{-1}$ in fish samples. Importantly, hair mercury concentration was significantly higher in the case of population exposed to mercury exclusively by fish consumption (e.g. Quintay and Zapallar), in comparison to the residents exposed to industrial emissions and fish consumption (e.g. Puchuncaví, Las Ventanas, Quintero). Finally, high mercury concentrations were observed in fish types commonly consumed by the residents of the fishing villages, evidencing human exposure to mercury through fish consumption.

Keywords: Mercury concentration; Human hair; Fish consumption

1. INTRODUCTION

The mercury (Hg) is recognized as one of the most hazardous environmental pollutants (Baeyens et al., 2003; Dryżalowska & Falandysz 2014) due to its high degree of persistence in the environment and its toxicity to different biological species (Li and Wang 2007). Mercury contamination is a worldwide problem because of its long-range transport and its ubiquity in global marine ecosystems; thus meaning that the entire global population is potentially exposed (Esteban et al., 2015).

In the environment, this element proceeds from natural sources, such as rock weathering or volcanic activity; and anthropogenic sources, principally incineration and burning of fossil fuels (e.g. coal-fired power plants). Once released, Hg persists in the environment, where it can be transferred to the air, water, sediments, soil and it can be incorporated to biota (Li et al., 2006, NRC (National Research Council), 2000; Chen et al., 2016; Yusa et al., 2017; Mailman et al., 2006).

In the atmosphere, mercury exists primarily as the inorganic forms that can be divided into vapor phase mercury (Hg) and particulate Hg (PHg). Hg⁰ is a stable form of mercury and has a long atmospheric residence time (Xu et al., 2013). Contrarily, once emitted from sources, Hg²⁺ is easily transferred to PHg during the transport of air masses, and it is removed from the atmosphere (e.g. wet or dry deposition) (Fu et al., 2008). Although, Hg can be deposited in terrestrial and aquatic ecosystems being converted, by microbial activity, into more toxic methylmercury and subsequently be bioaccumulated and biomagnified in aquatic food webs (Watras et al., 1998; Black et al., 2011; Shao et al., 2013).

For human beings, the exposition to toxic levels of mercury can cause neurological, nephrological, cardiac and reproductive disorders as well as genetic damage (Zahir et al., 2005; (ATSDR), 1999). The exposition sources to Hg is primarily through the consumption of contaminated fish, especially large predatory fish species such as tuna, swordfish, shark and whale (NRC (National Research Council), 2000, Boening 2000; UNEP (United Nations Environment Programme), Brodzka 2009, Marin, et al., 2017), besides the respiration of vapor Hg and PHg, coming from industrial activities (Fu et al., 2011). In this sense, high mercury concentration can be emitted from coal fired power plants, producing enrichment of the atmospheric mercury contents and increasing the exposition risk for contaminated air respiration (Beckers and Rinklebe, 2017).

In this way, mercury concentrations in blood and hair have been used as biomarkers to assess the human exposition to this element (Chien et al., 2010). In this way, hair samples appear as interesting alternative because it is a non-invasive matrix and it is easy to sample. Besides, hair Hg concentrations are higher than in other human samples (e.g. blood, urine, nails, among others) and it correlates to concentrations in blood, since mercury is incorporated into hair follicles by the formation of a MeHg-cysteine complex (Esteban et al., 2015; McDowell et al., 2004).

Some studies suggest that concentrations in human hair samples is positively correlated to fish consumption, especially in coastal regions where fish is consumed as primary source of protein (USEPA 2010, Gibb et al., 2016; Esteban

et al., 2015; Oken et al., 2005; Domanico et al., 2017; Cheng et al., 2009). In this way, Díez et al., 2008 found a strong correlation between hair Hg concentration and fish consumption in the urban population from southern Italy. Equally, Yusa et al., 2017 (Spain) analyzed different fish (swordfish, canned tuna, salmon, trout, sole, hake, whiting), finding positive correlation with Hg levels in hair. Likewise, Shao et al., 2013 (China) evaluated hair mercury levels and different species of fish, in order to determine potential health risks associated with dietary consumption of mercury, demonstrating that Hg intake via fish consumption is significantly correlated with Hg accumulated in human hair.

Generally, the consumption of contaminated fish is the primary route of exposure to mercury, although some studies show other sources. In this way, (Díez et al., 2011) found that mean mercury levels were greater in people living close to industrial mining activity sites compared with a site placed hundreds of kilometers away.

In Chile, the impact of mercury contamination has not been deeply studied and the reports have been focused on sediments (Yáñez et al., 2013) or food contamination (Muñoz et al., 2017). However, the evaluation of human exposition is very limited; with only two reports available. In one of the reports (Bruhn et al., 1995), the hair samples from pregnant and breastfeeding women living in south of Chile have been analyzed. In this study, the mean hair mercury concentration reached 1.81 mg kg⁻¹ (corresponding to mothers who live in the coastal region) which was significantly associated with frequency of fish consumption. In other report (Bruhn et al., 1994), total mercury content in scalp hair was analyzed in nursing women (South of Chile) with normal to high fish and seafood consumption as well as in women with negligible or no fish and seafood consumption. The results showed a higher concentration hair mercury of woman's with high fish consumption.

In the present study, we assessed mercury exposure of residents of different sites of Valparaíso Region (Chile) due to fish consumption and atmospheric contamination from coal-burning power plant and copper smelter. Likewise, we assessed total mercury concentrations in fish samples purchased in the supermarket or collected in the studied fishing villages. To the best of our knowledge, no similar studies have been reported for Chilean population.

2. MATERIALS AND METHODS

2.1 Sample Collection

During the year 2016, hair samples were collected from 199 volunteers, residents of different towns and villages of the Valparaíso region (Las Ventanas, La Greda, Valle Alegre, Puchuncaví, Los Maitenes, Maitencillo, Campiche, Zapallar and Quintay). The purpose was to assess human exposure to mercury due to fish consumption and atmospheric contamination from coal-burning power plant and copper smelter. Specifically, the sites Puchuncaví, La Greda and Los Maitenes are located close to copper smelter and coal-burning power plant and these places are strongly impacted by these activities (Parra et al., 2014). In contrary, Quintay is a typical fishing cove, without reported contribution of industrial contamination.

For hair sample analysis, approximately 1.0 g of human hair was collected from the occipital region with a stainless steel scissors. The samples were placed and sealed in polyethylene bags, properly identified, and transported to the laboratory for treatment and analysis.

In order to assess the mercury level in fish samples, different fish samples were purchased from the local fisheries and some supermarkets (See Figure 1). Fresh and frozen samples were placed in plastic bags and transported to the laboratory and then immediately stored at -20°C .

Figure 1. Concentrations of total mercury ($\mu\text{g g}^{-1}$, dry weight) in fish samples collected in studied sites and purchased at different supermarkets.

2.2 Chemical analysis

Before the Hg determination, hair samples were cut into short segments (about 5 mm) and washed successively with acetone and Milli-Q water. The fish samples were homogenized, freeze-dried, crushed and ground into fine powder and stored to 4°C until analysis.

The digestion of hair and fish samples was carried out in a closed-vessel microwave oven. Specifically, 200 mg of the sample were placed inside a closed Teflon vessel and heated at 180°C during 20 minutes, applying 1000 watts of power with concentrated nitric acid and hydrogen peroxide (4:1 v/v). Finally, the samples were diluted to 25 ml with MilliQ water. All samples were analyzed in triplicate and concentrations are reported in dry weight basis.

In order to evaluate the accuracy of mercury determination method, certified reference material was analyzed. Specifically, the IAEA-085 (human hair, certified in total mercury) and Tuna Fish ERM- CE 464 (tuna fish, certified in total mercury) were analyzed and the recovery was higher than 97% of certified mercury content, considered satisfactory.

2.3 Instrumental

An atomic fluorescence spectrometer (Millennium Merlin, Model 10.025, PS Analytical, Orpington, Kent, England) was used to determine the Hg concentrations by cold vapor-atomic fluorescence spectrometry (CV-AFS). The mercury vapor was generated using a mixer with three channel system: Sample (flow: 9.0 ml min^{-1}); HCl solution (1.5 M, flow: 9 ml min^{-1}); and, NaBH_4 (7.5 % w/v, flow: 4.5 ml min^{-1}) prepared in NaOH 2% w/v.

2.4 Statistical methods

All statistical analyses were performed using Statgraphics Centurion XV (Statpoint Technologies, Inc.). Mercury concentrations in the hair samples were tested for normality and were found to be distributed not normally. For this, the Mann-Whitney test was used to evaluate statistical differences between medians (95 % confidence) between the typical fishery (Quintay) and the other cities and villages considered in this study. Additionally, a log-transformation was carried out with mercury concentrations to approximate the normal distribution and the differences between the groups, age and sex were tested using one way ANOVA and Tukey test (95% confidence).

3. RESULTS AND DISCUSSION

3.1 Mercury concentrations in fish

Considering that fish is the most consumed seafood in Chilean coastal territory, the mercury concentration was evaluated different in fish samples purchased from the local fisheries and some supermarkets. The group of fish collected included big predators (canned tuna, shark, albacore), fat fish (salmon) and small fish (hake, Pacific pomfret, Tilapia, Alaska Pollock and Pangasius).

The Hg contents of the collected fish samples are showed in Figure 2. As expected, the fresh and frozen albacore presented the highest concentration of mercury (3.87 and $1.70\text{ }\mu\text{g g}^{-1}$, respectively), as well as the canned tuna ($1.44\text{ }\mu\text{g g}^{-1}$); coincidentally with similar studies carried out by (Yusa et al., 2017).

Figure 2. Box plot of the distribution of total mercury concentrations (THg) in collected hair samples.

These samples presented levels higher than limits established for Chilean Ministry of Health, in according with actual regulation established for maximum mercury contents in fish samples (0.5 mg kg^{-1} and 1.0 mg kg^{-1} for non-predators and predators, respectively) (Minsal, 2018). Additionally, based on methylmercury concentration in fish and consumption frequency, the human exposition to mercury in artisanal fisheries, such as Quintay, can be evaluated. In fish tissues, a great proportion of mercury is present in Methylmercury form and then the THg can be used for this purpose. Based on self-declared consumption frequency (2 times per week) and considering a typical fish-portion (200 g wet weight), the mercury exposure for fish consumption ranged to $55.8\text{--}970\text{ }\mu\text{g}$ per week, depending of fish-type consumed. For a 70 kg individual, it would represent a mercury dose between $0.12\text{--}2.04\text{ }\mu\text{g kg}^{-1}$ (body weight) day^{-1} . Considering that in fish tissues methylmercury form is the predominant form of Hg (> 90%) Díez et al., 2008, these doses are above the US EPA reference dose for methylmercury of $0.1\text{ }\mu\text{g kg}^{-1}$ (body weight) day^{-1} (U.S. EPA, 2001) and, for some fish, the World Health Organization tolerable intake for methylmercury of $0.2\text{ }\mu\text{g kg}^{-1}$ (body weight) day^{-1} (WHO, 2004). Evidently, the exposition risk is evidenced and it can be considerable highest for female (mean weight: 60 kg) and children (For 11–15 year: 50 kg ; < 11 year, less to 50 kg) as described previously (Black, 2011).

3.2 Hair mercury concentrations

The Table 1 shows the average, maximum and minimum of hair mercury concentrations of analyzed samples. These concentrations ranged from 0.04 to $1.78\text{ }\mu\text{g g}^{-1}$. Importantly, the average concentrations found for all samples did not exceed the recommended level for mercury in hair of $1.0\text{ }\mu\text{g g}^{-1}$ (National Research Council, 2000).

The concentrations found were similar to other studies from people living in coastal sites. For example, Black et al., 2011 found a mean mercury concentrations of $0.21\pm 0.22\text{ }\mu\text{g g}^{-1}$ in human hair collected in Batswana where the higher levels were associated with the fish consumption. Equally, Li et al., 2006 found values of mercury in hair samples similar with this study ($0.42\text{ }\mu\text{g g}^{-1}$), attributed to the greater consumption of fish among

the residents of coastal sites from China. The study of Díez et al., 2008 was also consistent with our results. Herein, the authors found an average mercury concentration of $0.64\text{ }\mu\text{g g}^{-1}$ in hair samples collected in Italy, where these values were strongly correlated with fish consumption. Marcinek et al., 2017 equally found that the mean value of mercury in hair samples was found to be $0.174\pm 0.137\text{ }\mu\text{g g}^{-1}$ with a statistically significant correlation ($p<0.05$) between the content of Hg in hair of the studied population and the fish consumption. In the same way, other studies have found higher average concentrations of mercury in the hair ($>1.0\text{ }\mu\text{g g}^{-1}$) compared to this study, where the levels found have been correlated with fish consumption (Esteban et al., 2015, Yusa et al., 2017, Shao et al., 2013).

Table 1. Hair mercury concentrations ($\mu\text{g g}^{-1}$) and population characteristics at the studied sampling sites.

Sites	Sex	N	Age range	Maximum	Minimum	Average	Mann-Whitney test ^A	Anova-Tukey (log[THg]) ^B
Las Ventanas	Male	9	15-81	1.78	0.06	0.49	abc	bc
	Female	4	6-39	0.72	0.11	0.32		
La Greda	Male	6	5-53	0.98	0.05	0.32	ab	a
	Female	23	2-86	0.37	0.04	0.07		
Valle Alegre	Female	11	8-59	0.26	0.06	0.07	d	a
Puchuncaví	Male	15	2-48	1.53	0.04	0.17	d	a
	Female	24	2-71	0.54	0.06	0.12		
Los Maitenes	Male	6	6-73	0.59	0.08	0.19	d	ab
	Female	12	4-70	0.62	0.07	0.12		
Maitencillo	Male	5	37-67	1.06	0.08	0.41	bc	bc
	Female	14	7-83	0.44	0.05	0.24		
Campiche	Male	10	14-83	0.09	0.05	0.03	ab	a
	Female	19	4-70	0.71	0.06	0.13		
Zapallar	Male	6	6-8	0.84	0.08	0.44	a	c
	Female	4	3-41	0.45	0.09	0.27		
Quintay	Male	4	22-81	0.76	0.23	0.59	a	c
	Female	8	23-79	0.74	0.06	0.37		

^A: Grouping established based on differences respect to Quintay. B: Grouping established based on Tukey test ($p < 0.05$)

The descriptive statistics on the Hg concentrations in hair in the different sampling sites are showed in Table 1. The data presents are no normal distributed and Mann-Whitney test was applied. From this analysis, the people from Quintay (typical fisheries) presents higher mercury levels in hair respect to Puchuncaví, Los Maitenes and Valle Alegre. Interestingly, the last places present highest mercury concentration in soil samples, attributed to atmospheric deposition of PHg from industrial emissions (Bernalte et al., 2015). Additionally, a long term study conducted in this sector evidenced moderate to high pollution risk in the area, derived from atmospheric deposition in soil samples (Rueda et al., 2016). These results suggest that industrial emissions do not contribute significantly to mercury exposure in the studied population, confirming that the ingestion of food containing mercury is the principal source for human exposition.

CONCLUSIONS

In our study, Hg levels were determined in the hair of people of different sites of Valparaíso Region, exposed to mercury by fish consumption and atmospheric contamination coming from different industries. According to the results of the present study, the people who frequently eat fish present an elevated level of exposure to mercury, as is the case of the residents of Quintay (fishing village). Finally, these results suggest that industrial emissions do not contribute significantly to mercury exposure in the studied population.

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