Mercury exposure Assessment in fish and humans from Sundarban Mangrove Wetland of India

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Present study had documented total mercury levels in six commonly consumed fish species, and performed a cross-sectional study on local residents to gauge their intake of fish (via dietary survey) and mercury exposure (via hair biomarker analyses). Mean total mercury content in edible composites of locally-caught fishes (topse, hilsa, mackerel, topse, sardinella, khoira) was low and ranged from 0.01 to 0.11 ug g® mercury, dry weight. In a cross-sectional study of 58 area residents, the mercury content in hair ranged from 0.25 to 1.23 ug g® with a mean of 0.65 ± 0.23 ug g®. Hair mercury level was not influenced by gender, age, or occupation. Mean number of meals consumed per week was 3.1 ± 1.1, and all participants consumed at least one fish meal per week. When related to fish consumption, a significant positive association was found between number of fish meals consumed per week and hair mercury levels.

[Keywords: Exposure assessment, Methylmercury, Fish consumption, Biomarkers, Sundarban]

Introduction

Mercury is a heavy metal of global public health concern. Approximately 6,000 tons of mercury is released into the environment annually and concentrations continue to rise in many regions of the world. A majority of this mercury is released from coal-fired power plants, and largely from point sources in India and China. Epidemiological studies in China have started to document elevated exposures to mercury, though in India much less is known about human exposures and associated health risks.

The public is primarily exposed to mercury (as methylmercury) through fish consumption. Though released from most industries as an inorganic compound, upon deposition into aquatic ecosystems microorganisms can methylate this form of mercury into methylmercury. As a methylated chemical, mercury can effectively cross biological membranes and accumulate in organisms, biomagnify through aquatic food chains, and build up in the tissues of fish-consumers. The concentrations of mercury in tissues of fish-consumers may be 10-million times greater than ambient levels in the environment. In India, there exist several studies showing the presence of mercury in fish, and in many cases the measured values exceed consumption guidelines set by the U.S. EPA (0.3 ug g®) or WHO (1.0 ug g®).

Despite the ubiquity of mercury in Indian fish, little is known about human exposures via fish consumption. More than half India’s population is estimated to eat fish and seafood on a regular basis and over 30% of its population relies upon it from
livelihood). Fish and seafood are a major source of dietary proteins, essential elements, and omega-3 fatty acids. In addition, harvesting these items is of immense recreational, economical, and cultural importance to several groups. Clearly, mercury contamination of fish may have a range of deleterious societal impacts in India.

Within India, the Sundarban region of the state of West Bengal is an area worth studying in terms of mercury exposure. The Sundarban wetland is a vast mega-delta in West Bengal, India, comprising over 100 islands and approximately 6.5 million people. Sagar Island is the most significant fishing center of Sundarban, particularly during the winter months. The potential source of mercury in this region are industrial sources (Paper factories, electronic industries, etc), agricultural run-offs (mercury-containing fungicides) and sewage sludge from the upper stretch of Ganga river. With rapid development of electronic industries in West Bengal, a large number of outdated electronic products in the form of ‘e-waste’ contribute to the mercury sources in the study area. The estuary receives raw sewage from the megalcity of Calcutta located 85 km upstream. Nearby and bordering states (Bihar, Orissa, West Bengal) contain more than 50% of India’s coal resources. Studies from the Sundarban coastal regions have documented mercury in sediment. In addition to potential health impacts, the presence of mercury may degrade fish and seafood which are critical to the sustenance, livelihood, and economy of local residents. The goal of this project was to increase understanding of mercury exposure in the region (and by extension, India given the dearth of information on this matter) by addressing the following objectives: A) to document total mercury levels in six commonly consumed fish species; and B) to perform a cross-sectional study on local residents to gauge their intake of fish (via dietary survey) and mercury exposure (via hair biomarker analyses).

Materials and Methods

The study was performed within the Sundarban coastal region of the state of West Bengal, India. The sampling locations occur at the southernmost front of Sagar Island (Figure 1), which is the largest island formed at the mouth of the Hugli estuary. The island is approximately 300 km² in area with a population of over 160,000 individuals. Six species common to the region were sampled: pomphret (Pampus pampus), hilsa (Tenualosa ilisha), mackerel (Rastrelliger kanagurta), sardin (Sardinella sp), topse (Polynemus paradises) and khoira (Setipinna phasa). After returning to the laboratory the weights and lengths of the fish species were noted. The fishes were cut open and the edible muscles were sliced and washed with Milli-Q water. The samples were then dried at 40°C until dryness (3-4 days) and pulverized with a mortar and pestle. A composite of 5-6 individuals from each species was analyzed for total mercury.

Hair is used to gauge exposure to methylmercury. Hair samples of 58 area residents were collected using methods outlined previously after obtaining consent. Approximately 20-30 strands of hair were cut from the occipital region of the scalp. In addition to sampling hair, a brief survey was administered to gather information on gender, age, occupation, and fish consumption habits.

Concentration of total mercury in each fish and hair sample was measured in a Direct Mercury Analyzer 80 (DMA-80, Milestone Inc, CT) according to U.S. EPA Method 7473 as previously described by...
About 10–30 mg of dried sample was weighted in a nickel sampling boat and placed into the DMA-80. Following decomposition of sample at 800°C, liberated mercury was next trapped using gold and then subsequently desorbed, carried to an absorbance cell and quantified spectrophotometrically. Analytical accuracy and precision were determined through the use of certified Standard Reference Materials (SRM) and intermittent analysis of duplicate samples. Recoveries of mercury in the DOLT-4 SRM (National Research Council of Canada) and the Japanese NIES hair SRM#13 were 105.3% (range: 104.7 – 106.0%) and 80.2% (range: 77.1 – 83.2%), respectively. Variability (measured by %RSD) of the two SRMs was < 6.0%. None of the results were adjusted based upon the reported SRM recoveries. The detection limit (0.037 ng mercury) was calculated as 3 times the standard deviation of the mean blank value, and none of the samples fell below this value. All mercury values are reported on a dry weight basis.

The research protocol was approved by the Ethics Committee of India. All the subjects were taken prior consent for analysis of the scalp hair for mercury with the help of short demonstration of the procedure before them.

Data Analysis

All statistical operations were performed using SPSS (v11.5, Chicago IL). Preliminary data analysis included tabulation of descriptive statistics for all measurements. The primary relationships of interest were associations between hair mercury levels and fish consumption, gender, age and occupation, and were evaluated using parametric statistical procedures. All data are indicated as mean ± standard deviation.

Results and Discussion

The length and weight of the sampled fish and their feeding habits are summarized in Table 1. The mean total mercury content in the fish composites ranged from 0.01 to 0.11 µg g⁻¹ (Table 1). The largest mercury concentration was found in topse followed by hilsa, mackerel, sardinella, and khaira. Topse (*Polynemus paradiseus*) being the predator fish mainly feeding on crustaceans (mainly shrimps), small fishes and benthic organisms whereas khaira (*Setipinna phasa*) mainly feeds on mysids and copepods. None of the total mercury values in these fishes exceed fish consumption guideline values used by the USEPA (0.3 µg g⁻¹) or WHO (1.0 µg g⁻¹).

The concentrations reported in this study are generally lower than a recent NGO study focused on mercury in fish from several markets in Calcutta. For example, values for total mercury in topse (0.41 µg g⁻¹), khoira (0.21 µg g⁻¹) and hilsa (0.69 µg g⁻¹) in the report by Chacraverti and Kumar were about 10-times greater than what we report here. In addition, the values we report in this study (generally less than 0.1 µg g⁻¹) are also lower than values measured in a range of other fish species sampled from the Ganges river in West Bengal and from the East Calcutta Wetlands. This comparatively low values of mercury may be related with different sample sizes, ages and

<table>
<thead>
<tr>
<th>Common Name</th>
<th>Species Name</th>
<th>Length (mm)</th>
<th>Weight (gm)</th>
<th>Total Mercury (µg/g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Topse</td>
<td><em>Polynemus paradiseus</em></td>
<td>17.8 ± 1.8</td>
<td>19.6 ± 10.9</td>
<td>0.033</td>
</tr>
<tr>
<td>Khoira</td>
<td><em>Setipinna phasa</em></td>
<td>16.1 ± 1.3</td>
<td>55.5 ± 18.9</td>
<td>0.010</td>
</tr>
<tr>
<td>Pomphret</td>
<td><em>Pampus pampus</em></td>
<td>22.7 ± 1.6</td>
<td>145.2 ± 46.6</td>
<td>0.105</td>
</tr>
<tr>
<td>Mackerel</td>
<td><em>Rastrelliger kanagurta</em></td>
<td>17.9 ± 1.3</td>
<td>60.9 ± 13.4</td>
<td>0.048</td>
</tr>
<tr>
<td>Hilsa</td>
<td><em>Tenualosa ilisha</em></td>
<td>25.0 ±0.2</td>
<td>166.8 ± 10.1</td>
<td>0.058</td>
</tr>
<tr>
<td>Sardine</td>
<td>Sardinella sp</td>
<td>28.5 ± 1.3</td>
<td>165.8 ± 22.7</td>
<td>0.027</td>
</tr>
</tbody>
</table>
characteristics of the captured environment\textsuperscript{19,20}. In contrast similar low values of mercury (0.002 – 0.198 µg g\textsuperscript{-1}) in fishes were obtained in commonly consumed fish species from Taiwan\textsuperscript{21}. According to Wang\textsuperscript{22} the assimilation of metals mainly depend the food conditions such as the food density and food type\textsuperscript{23,24,25}. These external conditions may significantly affect the ingestion, digestion, solubilization\textsuperscript{26,27}, membrane transport\textsuperscript{28}, and gut passage time\textsuperscript{29} and subsequently affect the dietary Assimilation Efficiency.

The present work represents a case study with limited sample size from Sundarban wetland. In order to evaluate the fish advisory level further studies are required with adequate number of collected fish species from this region. For the epidemiological portion of this study, 58 participants were recruited and equally distributed between genders. The mean age was 27.3 ± 14.0, and ranged from 4 to 70 years. In terms of occupation, 31% were involved in fishing, 24.1% were students, 32.8% were housewives, 6.9% were children, and the rest were involved in other activities. Nearly all (84.5%) participants were literate. None of the women (n=29) smoke or drank alcohol, whereas 23/29 men smoked and 8/29 men drank.

The mercury content in hair ranged from 0.25 to 1.23 µg g\textsuperscript{-1}, with a mean of 0.65 ± 0.23 µg g\textsuperscript{-1} (Table 2). There were no statistically significant differences in hair mercury values when stratified according to gender, age, or occupation (Table 2). Despite concerns of mercury pollution in India there exist few mercury human biomonitoring studies from the country for us to compare our work to. In a study of individuals associated with the Bhabha Atomic Research Center (BARC) in Bombay, mean values in blood (5.2 µg/L), urine (6.2 µg/L) and hair (1.2 µg g\textsuperscript{-1}) were reported though sample size and quality control values were not reported\textsuperscript{30}. As part of an international study comparing trace element exposures across five countries, in a sample of 255 from Bombay and New Delhi the mean hair mercury value was 1.3 ppm\textsuperscript{31}. In a study of 354 residents of Agra, the mean hair mercury values in males (0.73 µg g\textsuperscript{-1}, range: 0-21) and females (0.77 µg g\textsuperscript{-1}, range: 0-19.5) were similar\textsuperscript{32}.

Table 2–Hair total mercury values (µg/g) in residents from the Sundarban Wetlands of West Bengal, India. Data are stratified according to gender, age, and occupation with ANOVA p-values reported in column 1.

<table>
<thead>
<tr>
<th></th>
<th>N</th>
<th>Mean (±SD)</th>
<th>Median</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>All participants</td>
<td>58</td>
<td>0.65 (0.23)</td>
<td>0.60</td>
<td>0.25-1.23</td>
</tr>
<tr>
<td>Gender (p=0.92)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>29</td>
<td>0.66 (0.26)</td>
<td>0.60</td>
<td>0.25-1.23</td>
</tr>
<tr>
<td>Female</td>
<td>29</td>
<td>0.65 (0.20)</td>
<td>0.59</td>
<td>0.35-1.09</td>
</tr>
<tr>
<td>Age (p = 0.15)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;18 years</td>
<td>20</td>
<td>0.73 (0.22)</td>
<td>0.71</td>
<td>0.36-1.12</td>
</tr>
<tr>
<td>19-35 years</td>
<td>18</td>
<td>0.62 (0.26)</td>
<td>0.59</td>
<td>0.25-1.23</td>
</tr>
<tr>
<td>&gt;35 years</td>
<td>20</td>
<td>0.60 (0.19)</td>
<td>0.60</td>
<td>0.31-1.04</td>
</tr>
<tr>
<td>Occupation (p=0.35)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fisherman</td>
<td>18</td>
<td>0.64 (0.28)</td>
<td>0.61</td>
<td>0.26-1.23</td>
</tr>
<tr>
<td>Homemaker</td>
<td>19</td>
<td>0.60 (0.15)</td>
<td>0.58</td>
<td>0.35-0.87</td>
</tr>
<tr>
<td>Student</td>
<td>14</td>
<td>0.67 (0.21)</td>
<td>0.60</td>
<td>0.36-1.09</td>
</tr>
<tr>
<td>Child</td>
<td>4</td>
<td>0.86 (0.18)</td>
<td>0.94</td>
<td>0.59-0.96</td>
</tr>
<tr>
<td>Other</td>
<td>3</td>
<td>0.72 (0.44)</td>
<td>0.78</td>
<td>0.25-1.12</td>
</tr>
</tbody>
</table>
In a study of autistic children from Chennai, the mean hair mercury value in controls (0.37 µg g⁻¹, n=50) was significantly lower than values measured in cases (0.65 – 3.1 µg g⁻¹, n=15 per group, 3 groups according to Childhood Autism Rating Scale values). The mercury biomarker values reported in these aforementioned studies are similar to our findings. In addition, the mean hair mercury values are similar to values reported from other countries such as Pakistan, Iran, USA, and Germany. The mercury levels are also lower than values observed among populations who depend on fish as a principal component of their diet, such as mothers from the Faroe Islands (median: 4.5 µg g⁻¹) and Seychelle Islands (5.8 µg g⁻¹).

Moving beyond biomarker measures, our study also asked about fish consumption and reported a significant association (p<0.001) between fish consumption (queried as number of meals consumed per week) and hair mercury levels (Figure 2). Individuals with the highest mean hair mercury values also indicating to consuming 5 or more fish meals per week. To our knowledge, this is the first study in an Indian population to make such an association, and thus can be added to a large database of studies from across the world. We do acknowledge key limitations of our survey methods (i.e., specific fish not identified, portion sizes lacking), and these should be addressed in the future to help increase understanding of what specific fish (and possibly other food items) that may contribute to mercury burdens in India. For example, carefully linked fish consumption surveys and biomarker studies in the U.S. have enabled researchers to show that consumption of tuna contribute a majority of the mercury to the average citizen (while in certain areas of China this mercury is largely derived from rice consumption).

In terms of fish consumption, the mean number of meals consumed per week was 3.1 (1.1), ranged from 1 to >5, and was normally distributed. All participants consumed at least one fish meal per week. As indicated earlier, those that consumed five or more fish servings per week (1.1 ppm, n=9) had significantly more hair mercury that those consuming one meal per week (0.3 ppm, n=2). To perform a robust analysis, fish consumption was stratified into individuals consuming two or fewer meals per week, three meals, and four or more meals. In doing so, a significant difference was observed when these groupings were compared against mean hair mercury values (F=72.9, p<0.001, Figure 2). Such an association has been documented in several other populations worldwide and we believe our data is the first from an Indian population.

Fish consumption did not vary according to gender or occupation. Males consumed an average of 3.1 ± 1.0 fish meals per week while females consumed an average of 3.1 ± 1.1 fish meals per week. Fisherman (n=18) consumed an average of 3.0 ± 1.1 fish meals per week and this was not significantly different from the other occupational groupings. There was a significant age-related difference in consumption. The mean age of individuals that consumed more than 3 fish meals per week was 18.1 ± 11.1 years and this was significantly greater than mean age of individuals that consumed 3 fish meals per week (29.0 ± 15.5 years) and those that ate less than 3 fish meals per week (34.7 ± 8.9 years). In the literature, there is conflicting information concerning age-related differences in mercury biomarker levels.

Fig. 2—Hair total mercury values (µg/g) in residents from the Sundarban Wetlands of West Bengal (India) in relation to number of self-reported meals of fish consumed per week. Letters denote significant (p<0.001) differences between the bars based on a one-way ANOVA.
Conclusions

Present study provide information into the THg concentration in fish samples from Sundarban and the probable ecotoxicological health hazards from fish consumption. Data pertaining to the THg levels in scalp hair samples of the residents of Sundarban demand further research as the concentration supersedes the WHO prescribed level.

Acknowledgements

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