Environmental Science & Technology

pubs.acs.org/est

Article

# Compound-Specific Stable Isotope Analysis Provides New Insights for Tracking Human Monomethylmercury Exposure Sources

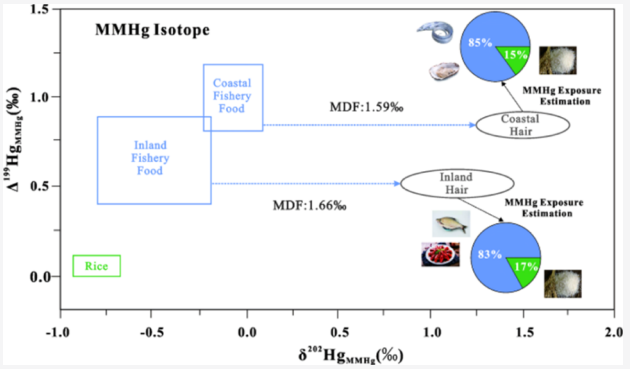
Shaochen Yang, Bo Wang, Chongyang Qin, Runsheng Yin, Ping Li,* Jinling Liu, David Point, Laurence Maurice, Jeroen E. Sonke, Leiming Zhang, and Xinbin Feng

Cite This: *Environ. Sci. Technol.* 2021, 55, 12493−12503

Read Online

ACCESS | Metrics & More | Article Recommendations | Supporting Information

**ABSTRACT:** Monomethylmercury (MMHg) exposure can induce adverse neurodevelopmental effects in humans and is a global environmental health concern. Human exposure to MMHg occurs predominately through the consumption of fishery foods and rice in Asia, but it is challenging to quantify these two exposure sources. Here, we innovatively utilized MMHg compound-specific stable isotope analyses (MMHg-CSIA) of the hair to quantify the human MMHg sources in coastal and inland areas, where fishery foods and rice are routinely consumed. Our data showed that the fishery foods and rice end members had distinct $\Delta^{199}Hg_{MMHg}$ values in both coastal and inland areas. The $\Delta^{199}Hg_{MMHg}$ values of the human hair were comparable to those of fishery foods but not those of rice. Positive shifts in the $\delta^{202}Hg_{MMHg}$ values of the hair from diet were observed in the study areas. Additionally, significant differences in $\delta^{202}Hg$ versus $\Delta^{199}Hg$ were detected between MMHg and inorganic Hg (IHg) in the human hair but not in fishery foods and rice. A binary mixing model was developed to estimate the human MMHg exposures from fishery foods and rice using $\Delta^{199}Hg_{MMHg}$ data. The model results suggested that human MMHg exposures were dominated (>80%) by fishery food consumption and were less affected by rice consumption in both the coastal and inland areas. This study demonstrated that the MMHg-CSIA method can provide unique information for tracking human MMHg exposure sources by excluding the deviations from dietary surveys, individual MMHg absorption/demethylation efficiencies, and the confounding effects of IHg.




**KEYWORDS:** *monomethylmercury, human hair, fishery food, rice, isotope*



## 1. INTRODUCTION

Mercury (Hg) is a toxic trace metal of global concern and has been listed as one of the top 10 chemicals of public health concern by the World Health Organization.[1,2] The toxic mechanisms and biogeochemical behaviors of Hg largely depend on its chemical forms.[3,4] Monomethylmercury (MMHg) is one of the predominant forms of organic Hg, which results from methylation of inorganic Hg (IHg) by bacteria or due to abiotic reactions in the environment, and is more neurotoxic and bioaccumulative than IHg in organisms.[3−5] Human exposure to MMHg can lead to long-term neurocognitive deficits in children, which result in global socioeconomic costs of over US$20 billion.[6] Underlying the persistence and biomagnification of MMHg in aquatic systems, fishery food intake has been identified as the primary route of human exposure to MMHg.[7] In addition, rice consumption could also pose great risks of MMHg exposure to individuals using rice as a staple food due to the more active Hg methylation and higher MMHg levels in rice paddies than in other kinds of farmlands.[8−10] To quantify the human MMHg intake levels from fishery foods and rice consumption, numerous studies have adopted the dietary calculation method.[7,10,11] However, the uncertainties that result from statistical biases in questionnaires, MMHg bioaccessibility, and personal demethylation efficiencies are poorly understood,[11−13] which lead to large deviations between the MMHg daily intakes and MMHg levels in biomarkers (such as the human hair). As a result, increasing efforts have been made to develop new approaches to more accurately estimate the contributions of fishery foods and rice to human MMHg burdens, especially in areas where both rice and fish serve as staple foods.[11]

The application of stable Hg isotopes is based on its two-dimensional isotope system of mass-dependent fractionation (MDF) and mass-independent fractionation (MIF) and is an

Received: March 18, 2021
Revised: August 6, 2021
Accepted: August 10, 2021
Published: September 1, 2021

effective approach to trace the sources and biogeochemical processes of Hg in the environment.[14−16] MDF occurs in most physical, chemical, or biological processes,[17−19] whereas MIF is predominately affected by specific photochemical processes (e.g., photochemical $Hg^{2+}$ reduction and photochemical MMHg degradation) for both odd and even Hg isotopes.[14] Prior studies have demonstrated that $\delta^{202}$Hg could indicate the human internal demethylation process because the hair of fish consumers is enriched by nearly +2.0‰ in $\delta^{202}$Hg relative to the consumed fish.[3] The hair $\Delta^{199}$Hg (or $\Delta^{201}$Hg) values could identify dietary MMHg exposure sources considering the absence of Hg photochemical processes during the metabolism.[20−22] Thus, the $\delta^{202}$Hg and $\Delta^{199}$Hg values of the total Hg (THg) appear to have the potential to trace the dietary MMHg sources of human exposure. However, recent studies have reported that the human hair displays significant correlations between the MMHg fraction and $\delta^{202}Hg_{THg}$ or $\Delta^{199}Hg_{THg}$,[23] which suggests distinct isotope properties between the IHg and MMHg fractions. Therefore, using the isotope values of THg to quantify the MMHg exposure sources contains large uncertainties since the human hair, fishery foods, and rice occasionally present low MMHg/THg values,[22,24−28] and the mechanisms that are behind the biogeochemical behaviors of different Hg species are poorly understood.[29] Although gas chromatography (GC) has been used for the separation and identification of chemical Hg compounds,[24] this online method may cause variations of up to 0.5‰ in $\delta^{202}$Hg, which result in low precision when analyzing the MMHg isotope values.[30,31] Recently, a selective extraction method (SEM) has been developed for compound-specific stable isotope analysis (CSIA), which can extract MMHg from aqueous biological samples without GC separation and offers high precision for the MMHg isotope values.[29] However, no research has focused on the MMHg isotope values in the human hair and their direct link to dietary MMHg.[20] It is urgent to distinguish the MMHg exposure pathways and to calculate the relative contributions from each source, and this knowledge may be used to reduce the health risks of MMHg exposure.

Here, we selected one inland and two coastal fish-rich cities in South China, where the inhabitants may have a high probability of MMHg exposure via the consumption of fishery foods and rice,[7,32−34] to investigate MMHg-CSIA in fishery food, rice, and the human hair. This study aims to study and differentiate the isotope properties between MMHg and IHg in these biological matrices and quantify the contributions from the two pathways of human MMHg exposure. This is pioneering work that uses the CSIA approach, which can provide new insights into the risk assessment and risk control of human MMHg exposure.

## 2. MATERIALS AND METHODS

**2.1. Sample Collection and Preparation.** The human hair, fishery food, and rice samples were collected in three cities in South China, namely, Zhoushan, Xiamen, and Wuhan, in April 2016, December 2017, and September 2017, respectively (Figure S1). The first two cities are located in coastal areas, and the third is located in an inland area. A total of 26 coastal hair, 19 inland hair, 27 coastal commercial fishery food, 19 inland commercial fishery food, 4 coastal rice, and 5 inland rice samples were collected for use in this study. The hair samples were collected from individuals who had lived in the study area for at least 1 year and used rice as their staple food. All hair samples had weights of approximately 200−300 mg, and a questionnaire, which included "age", "height", "weight", "staple food", and "specific Hg exposure sources" information, was also used. Individuals in inland areas without any seafood intake were selected and were identified by the questionnaires. The questionnaire information is shown in Table S1. Every participant signed a consent agreement before the sampling took place. This study obtained an ethics approval from the Institute of Geochemistry, Chinese Academy of Sciences. The selected fishery food samples were obtained from local supermarkets and included fish, crustaceans, and mollusks, which represented the main aquatic products in the sampling areas.[35] After collection, the muscle tissues of the fishery food samples were preserved in an ice box and were transported to the laboratory as quickly as possible. The sampled species and relative yields are shown in Table S2. The rice samples were collected in supermarkets that were located in the districts where the human hair samples were obtained and represent the rice that is consumed by the local inhabitants. Due to the fact that this study aims to focus on fishery food MMHg exposure since we selected the aquatic products cities, only small quantities of rice samples were collected in this study.

In the laboratory, the hair samples were washed with a nonionic detergent, distilled water, and acetone to remove surface dust and were then air-dried. The washed hair samples were ground with a ball mill to ensure homogenization and were finally stored in sealed bags for further analysis.[33] The seafood and freshwater muscle samples were separated, lyophilized, and homogenized with a pulverizer and were stored at 4 °C.[36] The rice samples (approximately 100 g each) were air-dried, crushed, and passed through a 150-mesh sieve. No isotope fractionation of Hg occurred during these treatments according to previous studies.[20,21]

**2.2. Hg Concentration Analysis.** All human hair (approximately 0.05−0.1 g), fishery food (approximately 0.1−0.2 g), and rice (approximately 0.5−1.0 g) samples were digested with 5 mL of $HNO_3$ (65%, v/v) at 95 °C for 3 h, and their THg concentrations were then determined by the cold vapor atomic fluorescence spectroscopy (CVAFS) method.[37] For the analyses of the MMHg concentrations, the human hair and fishery food samples were digested with 25% $HNO_3$, while the rice samples were digested by the KOH-methanol and solvent extraction techniques.[38] The MMHg concentrations were measured by aqueous ethylation, purge, trap, and GC-CVAFS (Brooks Rand Model III, Brooks Rand Lab, America) detection and followed Method 1630.[38] In this study, the THg and MMHg concentrations are expressed as ng/g for the wet weights for fishery food and ng/g for the dry weights for the human hair and rice samples.

**2.3. Hg Isotope Analysis.** Approximately 0.03 g of the hair sample, 0.3 g of the fishery food sample, and 1.0 g of the rice sample were digested in 5 mL of $HNO_3$ at 95 °C for 3 h, and 0.5 mL of BrCl (30%) was subsequently added into the digested solution to maintain the Hg stability. The digested solution was diluted with Milli-Q water to approximately 20% acid and 1 ng/g of THg prior to measuring the Hg isotope compositions.[23] The Hg isotopes were measured by Nu-Plasma II multicollector−inductively coupled plasma mass spectrometry (MC−ICP−MS) and followed the procedures of a previous study.[39] The $\delta^{202}$Hg and $\Delta^{199}$Hg values were expressed relative to NIST-SRM-3133 (National Institute of Standards and Technology Standard Reference Materials


https://doi.org/10.1021/acs.est.1c01771
Environ. Sci. Technol. 2021, 55, 12493−12503

3133) by following the protocol recommended by Blum and Bergquist.[40]

The MMHg isotope composition measurements were improved by an SEM that was established by Masbou et al.[29] The SEM details are shown in the Supporting Information, Section S1. The efficiencies needed to be verified after the MMHg extraction procedures were completed, which included verifying the purity and recovery of each sample, and the procedure is also shown in the Supporting Information, Section S1. The purity (%) of MMHg in the extracts was used to determine the relative values of the IHg impurities, and the recoveries (%) of MMHg in each sample were used to determine the extraction efficiencies of the SEM. The purified MMHg extracts were diluted with 10% $HNO_3$ (v/v) to produce a final THg concentration of 1 ng/g, and the Hg isotopes were then measured by MC−ICP−MS.[39]

The isotope ratios of the IHg compounds in the samples were calculated from the THg and MMHg fractions, as described in eqs 1−3 below[28]

$$\delta^{202}Hg_{THg} = \delta^{202}Hg_{MMHg} \times R + \delta^{202}Hg_{IHg} \times (1 - R) \quad (1)$$

$$\Delta^{199}Hg_{THg} = \Delta^{199}Hg_{MMHg} \times R + \Delta^{199}Hg_{IHg} \times (1 - R) \quad (2)$$

$$\Delta^{201}Hg_{THg} = \Delta^{201}Hg_{MMHg} \times R + \Delta^{201}Hg_{IHg} \times (1 - R) \quad (3)$$

where $R$ is the percentage of MMHg that is contained in the THg concentration (MMHg %) in each sample. The uncertainties in the IHg isotope ratios were simulated by a Monte Carlo simulation approach ($n$ = 10,000 times) and used the pseudorandom number generation function in Python software. In this approach, we assumed that the uncertainty in the MMHg % value was 5% and that the uncertainties in the $\delta^{202}$Hg, $\Delta^{199}$Hg, and $\Delta^{201}$Hg values were 0.08, 0.06, and 0.06‰, respectively (i.e., the same as that of UM-Almaden solution detected in this study).

**2.4. Binary Mixing Model.** To quantitatively estimate the contribution of Hg exposure from each source, both a statistical method and isotope mixing model were used in this study. The daily absorption amount (DAA) method was used to estimate the THg or MMHg exposures through the use of statistical data, and the details are shown in the Supporting Information, Section S2.[23] The uncertainties in the DAA were also calculated by the Monte Carlo simulation approach ($n$ = 50,000 times) by using the pseudorandom number generation function of Python software. A binary mixing model was developed in this study to evaluate the quantitative contributions to the MMHg exposures from the diet, and the equations are shown as follows:[20,23]

$$\Delta^{199}Hg_{hairMMHg} = F_{rice} \times \Delta^{199}Hg_{riceMMHg} + F_{fish}\Delta^{199}Hg_{fishMMHg} \quad (4)$$

$$F_{rice} = (\Delta^{199}Hg_{hairMMHg} - \Delta^{199}Hg_{fishMMHg}) / (\Delta^{199}Hg_{riceMMHg} - \Delta^{199}Hg_{fishMMHg}) \quad (5)$$

$$F_{fish} = 1 - F_{rice} \quad (6)$$

where $\Delta^{199}Hg_{hairMMHg}$, $\Delta^{199}Hg_{fishMMHg}$, and $\Delta^{199}Hg_{riceMMHg}$ represent the $\Delta^{199}$Hg values in the MMHg of the hair, fishery food, and rice, respectively, and $F_{fish}$ and $F_{rice}$ represent the MMHg fractions in the fishery food and rice sources, respectively. The mean $\Delta^{199}Hg_{fishMMHg}$ value in the local fishery food was used as the end member for the fishery food source, and the mean $\Delta^{199}Hg_{riceMMHg}$ value in the local rice was used as the end member for the rice dietary source. The uncertainties in the binary mixing model were also estimated by the Monte Carlo simulation approach. The above-described method is based on the binary method shown in the Supporting Information, Section S3, and assumes that fishery food and rice are the dominant MMHg sources of human MMHg exposure.

**2.5. Quality Control and Data Analysis.** The quality control approach consisted of a method blank, spiked blank, duplicate samples, and certified reference materials (CRMs). The limits of detection for THg and MMHg were 0.02 and 0.003 ng/g, respectively. The recoveries (%) of the THg and MMHg concentrations in CRMs are shown in Table S3. The relative percentage differences from the duplicate analysis in all fishery food, rice, and hair samples were <10% for both the THg and MMHg analyses (Table S4). The recoveries from THg isotope digestion were in the range of 80−120% (Table S5). The results of the purities and recoveries for the SEM are shown in Table S6. The mean SEM purities (%) in the fishery food, rice, and hair were 96.3 ± 11.8%, 90.8 ± 10.2%, and 92.7 ± 9.43%, respectively, and the mean SEM recoveries were 99.9 ± 11.9%, 91.2 ± 10.4%, and 100 ± 18.8%, respectively, which indicated that the SEM could extract the MMHg levels from these biological samples completely and accurately. In this study, the NIST-3133 standard solution was used as a bracketing standard, and NIST-3177 was used as a secondary standard. The average $\delta^{202}$Hg and $\Delta^{199}$Hg values were −0.55 ± 0.08‰ and 0.00 ± 0.08‰ ($n$ = 25, Mean ± 2SD), respectively, in NIST-3177, which were consistent with previously published data.[39,40] BCR-482 and NIES-13 were used as the THg isotope standards, and NIES-13 and TORT-2 were selected as the MMHg isotope standards. The isotope compositions of these CRMs are shown in Table S7.

The correlations between these measured variables were characterized by using linear regression analysis, and the statistical significance of the correlations was evaluated by SPSS 19.0 software for Windows. The THg and MMHg concentrations and Hg isotope data in all samples were compared using the $t$-test with a 5% significance level.

## 3. RESULTS

**3.1. THg and MMHg Levels.** The THg and MMHg concentrations and MMHg/THg ratios (%) for all samples that were collected in the coastal and inland areas are shown in Table 1. Wide ranges for the THg (e.g., 5.04−87.0 ng/g) and MMHg concentrations (e.g., 3.74−78.2 ng/g) were observed in the fishery food across the study areas (Table S8), and comparable levels of THg and MMHg were observed between the coastal and inland areas (Table 1). The MMHg/THg ratios of all fishery foods varied from 31 to 93% (Table S8), and the average ratio was slightly higher in inland areas than in coastal areas (Table 1). Compared with the fishery foods, we found much lower THg (3.14−5.10 ng/g) and MMHg concentrations (0.860−3.05 ng/g) as well as MMHg/THg ratios (24.4−77.4%) in the rice samples across the study areas (Table S9). Again, the THg and MMHg levels in the rice



https://doi.org/10.1021/acs.est.1c01771
Environ. Sci. Technol. 2021, 55, 12493−12503

Table 1. THg, MMHg, MMHg/THg, the MMHg Isotope, and the IHg Isotope (Mean ± 1 SD) in the Human Hair, Fishery Food, and Rice of Coastal and Inland Areas

| sample | $n$ | THg (ng/g) | MMHg (ng/g) | MMHg/THg (%) | $\delta^{202}Hg_{MMHg}$ (‰) | $\Delta^{199}Hg_{MMHg}$ (‰) | $\Delta^{201}Hg_{MMHg}$ (‰) | $\delta^{202}Hg_{IHg}$ (‰) | $\Delta^{199}Hg_{IHg}$ (‰) | $\Delta^{201}Hg_{IHg}$ (‰) |
|---|---|---|---|---|---|---|---|---|---|---|
| Coastal Area | | | | | | | | | | |
| human hair | 26 | 1930 ± 1280 | 1110 ± 565 | 60.9 ± 14.3 | 1.51 ± 0.63 | 0.83 ± 0.17 | 0.65 ± 0.17 | 0.24 ± 0.95 | 0.45 ± 0.27 | 0.36 ± 0.27 |
| fishery food | 27 | 30.2 ± 18.8 | 21.2 ± 14.9 | 67.8 ± 15.6 | −0.08 ± 0.39 | 0.99 ± 0.54 | 0.81 ± 0.44 | 0.08 ± 1.20 | 0.85 ± 0.63 | 0.73 ± 0.49 |
| rice | 4 | 4.06 ± 0.561 | 1.61 ± 0.560 | 38.8 ± 9.73 | −0.68 ± 0.06 | 0.03 ± 0.09 | 0.03 ± 0.06 | −1.49 ± 0.39 | 0.02 ± 0.18 | 0.07 ± 0.09 |
| Inland Area | | | | | | | | | | |
| human hair | 19 | 795 ± 517 | 582 ± 364 | 73.5 ± 9.90 | 1.13 ± 0.55 | 0.51 ± 0.16 | 0.37 ± 0.12 | 0.33 ± 0.92 | 0.17 ± 0.23 | 0.08 ± 0.22 |
| fishery food | 19 | 29.8 ± 23.7 | 22.4 ± 18.6 | 75.7 ± 11.3 | −0.53 ± 0.52 | 0.63 ± 0.48 | 0.48 ± 0.44 | −0.52 ± 1.05 | 0.51 ± 0.66 | 0.56 ± 0.53 |
| rice | 5 | 3.84 ± 0.779 | 2.28 ± 0.646 | 60.3 ± 16.6 | −0.89 ± 0.16 | 0.07 ± 0.05 | 0.06 ± 0.06 | −1.19 ± 0.82 | 0.00 ± 0.06 | 0.05 ± 0.15 |

samples were comparable between the coastal and inland areas (Table 1). The hair samples displayed obviously higher THg (1930 ± 1280 ng/g) and MMHg (1110 ± 565 ng/g) levels but lower MMHg/THg ratios (60.9 ± 14.3%) in the coastal areas than those in the inland areas (THg: 795 ± 517 ng/g; MMHg: 582 ± 364 ng/g; MMHg/THg ratio: 73.5 ± 9.9%), as shown in Tables 1 and S10. In addition, we present the obtained questionnaire information in Table S1 and found no significant correlation between the personal data (e.g., age, height, or weight) and hair Hg levels (e.g., $lgC_{THg}$ or $lgC_{MMHg}$) in the study areas (Figure S2).

**3.2. $\delta^{202}$Hg of MMHg and IHg.** The details of $\delta^{202}Hg_{MMHg}$ and $\delta^{202}Hg_{IHg}$ values are summarized in Tables S11 and S12, respectively, and the $\delta^{202}Hg_{THg}$ values are listed in Table S10 and Section S4, respectively. The fishery food samples exhibited substantial variations in the $\delta^{202}Hg_{MMHg}$ (−1.03−0.59‰) values, whereas the rice samples presented a much smaller range (−1.05 to −0.60‰) (Table S11). The coastal fishery foods were enriched with heavier MMHg isotopes (−0.08 ± 0.39‰) than the inland fishery foods (−0.53 ± 0.52‰). This result is consistent with the rice $\delta^{202}Hg_{MMHg}$ characteristics in the coastal (−0.68 ± 0.06‰) and inland areas (−0.89 ± 0.16‰, Tables 1 and S13). All hair samples exhibited positive $\delta^{202}Hg_{MMHg}$ values (0.05−2.98‰), while significantly higher $\delta^{202}Hg_{MMHg}$ values were observed in the coastal hair samples (1.51 ± 0.63‰) than in the inland hair samples (1.13 ± 0.55‰, Table S13). Positive offsets for the $\delta^{202}Hg_{MMHg}$ levels were observed between the human hair samples and diets (Table S14). In the coastal areas, the offset values from fishery food and rice to the hair were 1.59 and 2.19‰, respectively. Similarly, these values in the inland areas were 1.66 and 2.02‰, respectively (Figure 1).

In contrast, no significant differences in $\delta^{202}Hg_{IHg}$ values were found in the fishery food samples between the coastal (0.08 ± 1.20‰) and inland areas (−0.52 ± 1.05‰). The $\delta^{202}Hg_{IHg}$ values of the rice samples in the coastal (−1.49 ± 0.39‰) and inland areas (−1.19 ± 0.82‰) were also roughly consistent, as shown in Table S13. Correspondingly, the hair $\delta^{202}Hg_{IHg}$ values between the coastal (0.24 ± 0.95‰) and inland areas (0.33 ± 0.92‰) were comparable (Table S13). As a result, the $\delta^{202}Hg_{IHg}$ values of fishery food, rice, and the hair displayed similar patterns between the coastal and inland areas.

A comparison of the $\delta^{202}Hg_{MMHg}$ and $\delta^{202}Hg_{IHg}$ values in the fishery food, rice, and hair samples is shown in Table S15. We observed comparable $\delta^{202}$Hg values between MMHg and IHg in the fishery food and rice samples but significantly higher $\delta^{202}$Hg values for MMHg than for IHg in the hair samples (Table S15).

**3.3. $\Delta^{199}$Hg of MMHg and IHg.** The $\Delta^{199}Hg_{MMHg}$ and $\Delta^{199}Hg_{IHg}$ values are shown in Tables S11 and S12, respectively. The fishery food samples exhibited a 2‰ variation in $\Delta^{199}Hg_{MMHg}$ (0.09−2.03‰), whereas the rice samples exhibited a smaller range (−0.09−0.14‰) in the study areas (Table S11). Significant differences in the $\Delta^{199}Hg_{MMHg}$ values were observed between the coastal fishery food (0.99 ± 0.54‰) and inland fishery food samples (0.63 ± 0.48‰, Tables 1 and S13). However, the $\Delta^{199}Hg_{MMHg}$ values in the rice samples were remarkably comparable between the coastal (0.03 ± 0.09‰) and inland areas (0.07 ± 0.05‰, Tables 1 and S13). The $\Delta^{199}Hg_{MMHg}$ values of the hair samples varied from 0.18 to 1.24‰ in the study areas (Table S11). Similar to fishery food, the coastal hair samples exhibited



https://doi.org/10.1021/acs.est.1c01771
Environ. Sci. Technol. 2021, 55, 12493−12503

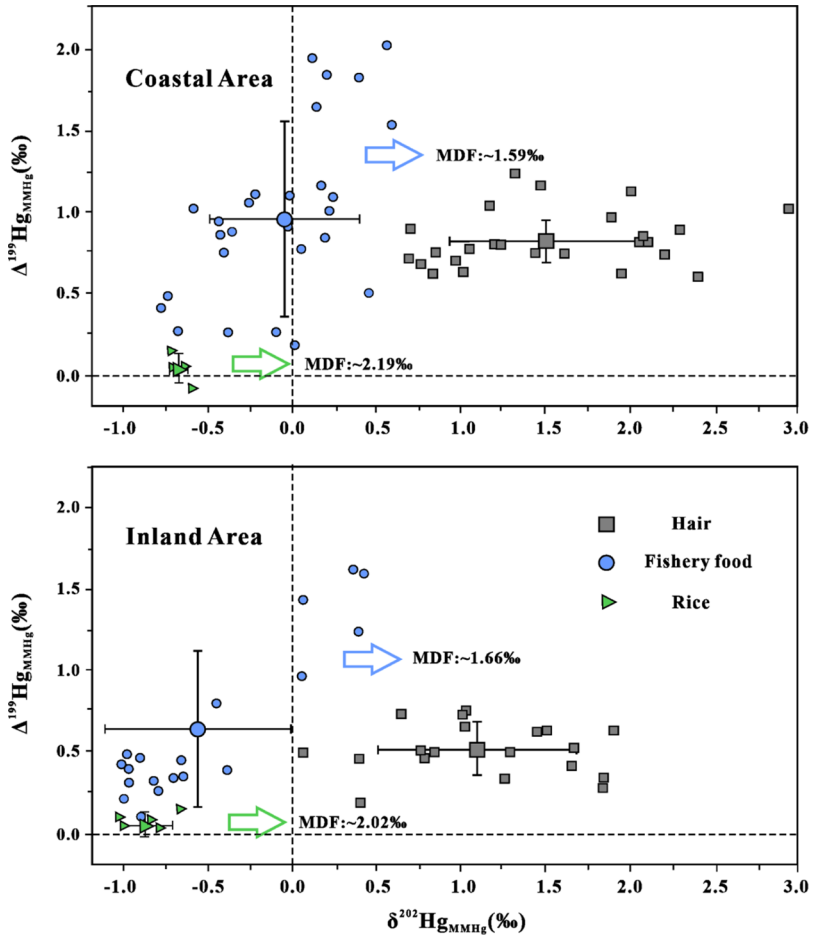



**Figure 1.** MMHg isotope properties ($\delta^{202}Hg_{MMHg}$ and $\Delta^{199}Hg_{MMHg}$) in fishery food, rice, and hair samples in coastal and inland areas. The small shape patterns indicate the $\delta^{202}Hg_{MMHg}$ and $\Delta^{199}Hg_{MMHg}$ values of each sample, and the large shape patterns with ±1 SD represent the mean values of $\delta^{202}Hg_{MMHg}$ and $\Delta^{199}Hg_{MMHg}$ for each sample type (e.g., fishery food, rice, or the hair). The blue arrow indicates the offset of $\delta^{202}Hg_{MMHg}$ between fishery food and the hair, whereas the green arrow indicates the offset of $\delta^{202}Hg_{MMHg}$ between rice and the hair.

higher $\Delta^{199}Hg_{MMHg}$ values (0.83 ± 0.17‰) than the inland hair samples (0.51 ± 0.16‰), as shown in Tables 1 and S12. The average $\Delta^{199}Hg_{MMHg}$ value for the hair samples was slightly lower than that for fishery food but was significantly higher than that for rice in the study areas (Table S14), which suggested that the MMHg in the hair samples was dominated by fishery food consumption rather than by rice consumption.

For the IHg, the fishery food and rice samples showed wide ranges of $\Delta^{199}Hg$ (−0.31−2.24‰ and −0.15−0.28‰, respectively). No significant differences in the $\Delta^{199}Hg_{IHg}$ values between the coastal and inland areas (Table S13) were found in the fishery food or rice samples, but the coastal hair samples (0.45 ± 0.27‰) exhibited significantly higher $\Delta^{199}Hg_{IHg}$ values than the inland hair samples (0.17 ± 0.23‰).

Although the fishery food and rice samples showed comparable $\Delta^{199}Hg_{MMHg}$ and $\Delta^{199}Hg_{IHg}$ values, the hair samples displayed significantly higher $\Delta^{199}Hg_{MMHg}$ values when compared with the $\Delta^{199}Hg_{IHg}$ values (Table S15). Interestingly, we observed significant positive correlations between the $\Delta^{199}Hg_{MMHg}$ and $\Delta^{199}Hg_{IHg}$ values in all fishery food samples ($R^2$ = 0.36, $p$ < 0.01, Figure S3) and hair samples ($R^2$ = 0.13, $p$ < 0.01, Figure S4) in this study.

## 4. DISCUSSION

**4.1. Comparison of MMHg Isotopes between Coastal and Inland Areas.** Earlier studies have characterized the $\delta^{202}Hg_{THg}$ and $\Delta^{199}Hg_{THg}$ values in aquatic organisms, such as in inland rivers ($\delta^{202}Hg_{THg}$: −0.92−−0.40‰; $\Delta^{199}Hg_{THg}$: −0.09−0.55‰),[3] inland lakes ($\delta^{202}Hg_{THg}$: −0.96−0.08‰; $\Delta^{199}Hg_{THg}$: 0.49−2.52‰),[41] the Great Lakes ($\delta^{202}Hg_{THg}$: −0.61−2.06‰; $\Delta^{199}Hg_{THg}$: 0.58−7.16‰),[42,43] coastal areas ($\delta^{202}Hg_{THg}$: −2.09−0.98‰; $\Delta^{199}Hg_{THg}$: −0.04−2.56‰),[36,44−46] and open oceans ($\delta^{202}Hg_{THg}$: 0.00−1.84‰; $\Delta^{199}Hg_{THg}$: 1.00−5.50‰).[47] The present study is the first to report on the MMHg isotopes in fish samples. The sources and formation mechanisms of MMHg in water prior to its bioaccumulation have been shown to directly affect the $\delta^{202}Hg_{MMHg}$ and $\Delta^{199}Hg_{MMHg}$ levels in aquatic organisms.[42,43,47] For instance, the MMHg in the open oceans and Great Lakes is primarily methylated from Hg that is supplied by precipitation with positive $\Delta^{199}Hg$ and negative $\delta^{202}Hg$ values,[47−49] whereas the MMHg in inland rivers or inland lakes is methylated from Hg that originates from sediments or terrestrial runoff with a nearly zero $\Delta^{199}Hg$ and very negative $\delta^{202}Hg$ values.[50−52] In the open oceans and Great Lakes, the water column is the dominant site for in situ MMHg generation, where extensive photochemical degradation occurs.[47] The above factors can, to a large extent, cause the


https://doi.org/10.1021/acs.est.1c01771
Environ. Sci. Technol. 2021, 55, 12493−12503

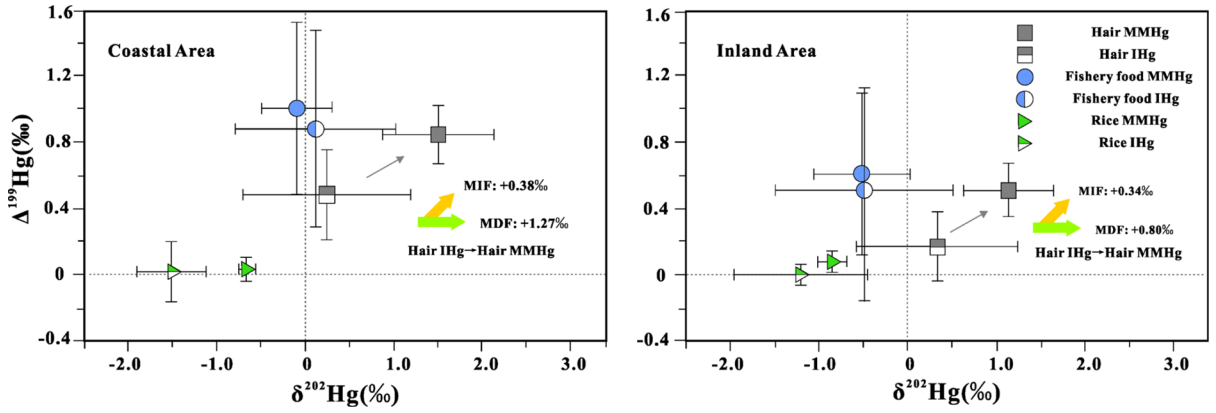



**Figure 2.** $\delta^{202}$Hg and $\Delta^{199}$Hg values (mean ± 1 SD) of MMHg and IHg in fishery food, rice, and hair samples in coastal and inland areas. The green arrow indicates an increase from hair $\delta^{202}Hg_{IHg}$ to $\delta^{202}Hg_{MMHg}$, whereas the yellow arrow indicates an increase from hair $\Delta^{199}Hg_{IHg}$ to $\Delta^{199}Hg_{MMHg}$. However, the $\delta^{202}$Hg and $\Delta^{199}$Hg values of MMHg and IHg in fishery food and rice are comparable.

MMHg in the open oceans or Great Lakes to have relatively high $\delta^{202}Hg_{MMHg}$ and $\Delta^{199}Hg_{MMHg}$ values. In this study, the coastal fishery food samples were captured from nearshore and estuarine areas; therefore, the MMHg isotopes may be characterized as a mixture of open ocean isotopes (with high $\delta^{202}Hg_{MMHg}$ and $\Delta^{199}Hg_{MMHg}$ values) and terrestrial runoff isotopes (with low $\delta^{202}Hg_{MMHg}$ and $\Delta^{199}Hg_{MMHg}$ values). The inland fishery food samples were generally captured in the Yangtze River in Wuhan, which had relatively lower $\delta^{202}Hg_{MMHg}$ and $\Delta^{199}Hg_{MMHg}$ values (Figure 1). Therefore, we proposed that the distinct sources and behaviors of Hg in the coastal and inland aquatic systems resulted in clear differences in the MMHg isotopes in fishery foods. In contrast, the coastal rice samples exhibited higher $\delta^{202}Hg_{MMHg}$ values than and comparable $\Delta^{199}Hg_{MMHg}$ values to inland rice samples (Figure 1), which reflected the MMHg isotope compositions in the paddy soils of these two areas since the MMHg in the rice grains generally originated from paddy soils.[27,28,53] The above-presented MMHg isotope profiles can be useful indicators for the environmental origin of MMHg prior to its entry into the food web.

The hair $\Delta^{199}Hg_{MMHg}$ values (0.83 ± 0.17‰, $n$ = 26) from the coastal area in this study were lower than the hair $\Delta^{199}Hg_{THg}$ values that were reported for seafood consumers in southern Italy (1.20 ± 0.19‰, $n$ = 21), the Faroe Islands (1.27 ± 0.03‰, $n$ = 6), Northern Europe (1.17 ± 0.24‰, $n$ = 11), and North America (1.86 ± 0.12‰, $n$ = 11)[20,22,44,54] but were much higher than the hair $\Delta^{199}Hg_{THg}$ values of inland fish consumers in the Bolivian Amazon (0.18 ± 0.03‰, $n$ = 6)[3] and the hair $\Delta^{199}Hg_{MMHg}$ values in inland areas reported in this study (0.51 ± 0.16‰, $n$ = 19). Due to the absence of MIF in the food web,[20] the $\Delta^{199}Hg_{MMHg}$ values in hair samples could provide direct information on the dominant MMHg exposure pathways in humans.[3,22] As shown in Figure 1, the $\Delta^{199}Hg_{MMHg}$ values in the coastal and inland areas represented the mixing effects from the two end members, which included fishery food and rice. The consistency in the $\Delta^{199}Hg_{MMHg}$ values between the hair and fishery food samples indicated that the human MMHg exposure in the coastal and inland areas was mainly due to fishery food consumption but was marginally due to rice consumption. The significant difference in the hair $\Delta^{199}Hg_{MMHg}$ values between the coastal and inland areas may result from the higher $\Delta^{199}Hg_{MMHg}$ values in seafood than in freshwater fishery food.[22] Furthermore, we found that the $\delta^{202}Hg_{MMHg}$ offsets between fishery food and the hair in the coastal (+1.59‰) and inland areas (+1.66‰) were slightly lower than the offsets that were reported by Laffont et al. (+2.00‰) and Bonsignore et al. (+2.20‰) and were comparable with the results reported by Li et al. (+1.75‰).[20,22,44] These offsets could reflect the process of human internal demethylation, with the elimination of MMHg with the lighter Hg isotope. However, the demethylation ability of each person is different, which could be affected by the daily ingested diet.[55] Consequently, individual variations could be the main factor that impacts the $\delta^{202}Hg_{MMHg}$ offsets between the fishery food and human hair samples.

### 4.2. Comparison of MMHg and IHg Isotope Values.

The $\delta^{202}$Hg and $\Delta^{199}$Hg values in MMHg found in this study were similar to those of IHg in both the fishery food and rice samples, as shown in Figure 2 and Table S15. This finding is not consistent with the results reported in previous studies that suggested higher $\delta^{202}$Hg and $\Delta^{199}$Hg values for MMHg than for IHg in fish and rice samples.[26,28,53] MMHg in the environment is mainly derived from microbial methylation.[14,56,57] Previous studies have demonstrated that bacterial cellular uptake activities influence IHg fractionation prior to methylation, which can impart distinct $\delta^{202}$Hg values to the IHg and MMHg pools.[56,57] However, once MMHg is present in the water column or sediment, photochemical processes and microbial demethylation activities could also impact the pools and $\delta^{202}$Hg values of MMHg and IHg.[47,58] Therefore, multiple processes and transformations of Hg in the aquatic environment are responsible for a widespread range of $\delta^{202}$Hg values for IHg and MMHg. It is unclear whether the MMHg that accumulated in the aquatic food web would display higher $\delta^{202}$Hg values than IHg.[58,59] Given the similarity of the $\Delta^{199}Hg_{MMHg}$ and $\Delta^{199}Hg_{IHg}$ values in the fishery food between the coastal and inland areas, we hypothesized that a portion of the IHg in fishery food tissues was derived from in vivo demethylation of MMHg.[60,61] The slope of $\Delta^{199}$Hg/$\Delta^{201}$Hg can provide valuable information on the photochemical reaction types, with slopes of 1.36 ± 0.04 (1SE) for the photodegradation of MMHg and 1.00 ± 0.02 for the photoreduction of IHg.[40] Our data showed slopes of 1.22 ± 0.02 ($n$ = 46, 1SE) for $\Delta^{199}Hg_{MMHg}/\Delta^{201}Hg_{MMHg}$ and 1.08 ± 0.06 for $\Delta^{199}Hg_{IHg}/\Delta^{201}Hg_{IHg}$ in fishery foods. The former slope is in accordance with previous values (~1.20) that were reported for fish from coastal and open oceans (Figure


https://doi.org/10.1021/acs.est.1c01771
Environ. Sci. Technol. 2021, 55, 12493−12503

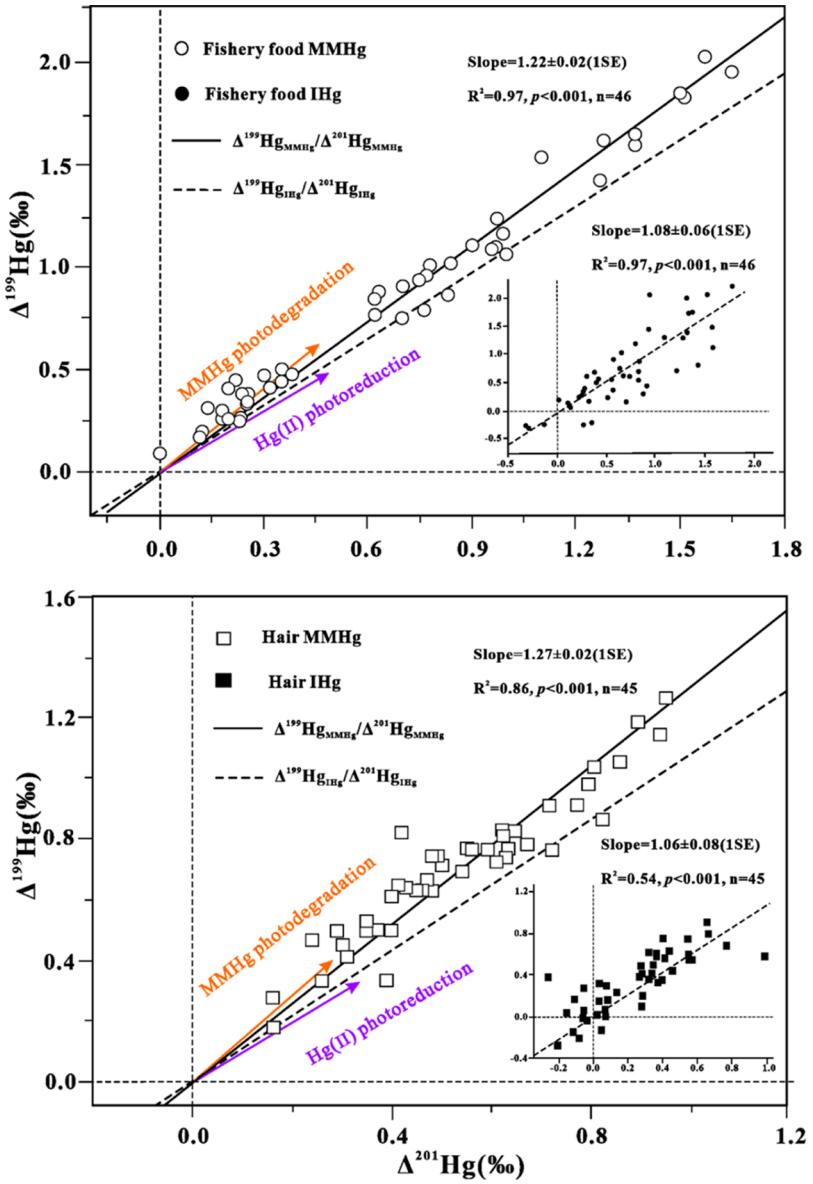



**Figure 3.** Slopes of $\Delta^{199}Hg/\Delta^{201}Hg$ of MMHg and IHg in fishery food and hair samples. The orange arrow indicates the slope of MMHg photodegradation (slope = 1.36 ± 0.04), whereas the purple arrow indicates the slope of Hg(II) reduction (slope = 1.00 ± 0.02). The SDs of the $\delta^{202}Hg_{MMHg}$ and $\Delta^{199}Hg_{MMHg}$ values are 0.10 and 0.06‰ for hair samples and 0.07 and 0.04‰ for fishery food, respectively. The SD of each IHg that was calculated by the Monte Carlo model is shown in the Supporting Information.

3).[45,47,62] Interestingly, the latter value is slightly higher than that for aqueous oxidized ligand-bounded Hg[Hg(II)] photoreduction (1.00 ± 0.02), which suggests that the IHg in fish muscle tissues may be partially derived from in vivo demethylation of MMHg. In addition, the significant positive correlation between $\Delta^{199}Hg_{MMHg}$ and $\Delta^{199}Hg_{IHg}$ in fishery foods (Figure S3, $R^2 = 0.36$, $p < 0.01$) may further support this hypothesis. Further studies should investigate the MMHg and IHg isotope properties in aquatic organisms in various systems (e.g., rivers, lakes, estuaries, and oceans) to clarify the fate of Hg in aquatic food webs.

Although previous studies indicated that the main form of Hg in the hair is MMHg,[20,63] IHg exposure through various pathways (such as Hg vapor exposure and IHg intake) may result in a non-negligible fraction of IHg in the hair.[22,64] Hence, it is necessary to study the isotopic properties of MMHg and IHg in the human hair separately. Differences of 1.27 and 0.80‰ between the hair $\delta^{202}Hg_{MMHg}$ and $\delta^{202}Hg_{IHg}$ values were found in the coastal and inland areas, respectively, which are likely related to the different metabolic processes of MMHg and IHg in the human body.[3,54,65] Significantly higher $\Delta^{199}Hg$ values were also detected in MMHg than in IHg (Figure 2), and the slope of $\Delta^{199}Hg_{MMHg}/\Delta^{201}Hg_{MMHg}$ in the hair (1.27 ± 0.02, 1SE, $n$ = 45, Figure 3) was different from that of $\Delta^{199}Hg_{IHg}/\Delta^{201}Hg_{IHg}$ (1.06 ± 0.08, 1SE, $n$ = 45), which suggested that the MMHg and IHg in the hair retained different mechanisms [e.g., MMHg photodegradation or Hg(II) reduction].[37] The MMHg in the hair is primarily due to fishery food consumption, as discussed above. However, the IHg in the hair was reported to mainly come from rice and vegetable intake and inhalation.[20,22,64] Generally, the IHg in rice, vegetables, air, and other nonfish intake sources should have $\Delta^{199}Hg_{IHg}$ values near 0 because the IHg in these matrices lacks photochemical reactions.[23,28,53] Absorbing IHg from


https://doi.org/10.1021/acs.est.1c01771
Environ. Sci. Technol. 2021, 55, 12493−12503

these sources is supposed to cause the hair $\Delta^{199}Hg_{IHg}$ values to be near 0. Interestingly, we found positive $\Delta^{199}Hg_{IHg}$ values in the hair samples of individuals from both the coastal and inland areas (coastal: 0.45 ± 0.27‰; inland: 0.17 ± 0.23‰), which suggested other possible IHg exposure sources. The positive correlation between $\Delta^{199}Hg_{IHg}$ and $\Delta^{199}Hg_{MMHg}$ in the hair samples suggested that IHg may demethylate from MMHg with positive $\Delta^{199}Hg$ values, which is consistent with the results obtained by Sherman et al.[54] Furthermore, we suspected that the positive $\Delta^{199}Hg_{IHg}$ values in the hair can also result from absorbing IHg from fishery food due to the high positive $\Delta^{199}Hg_{IHg}$ values in fishery food. This observation indicated that the IHg in the hair could be a mixture of complex sources present in the study areas, which is different from MMHg (nearly all from fishery food intake). We suggest that the IHg exposure model that is estimated by the Hg isotope approach should consider a new end member with positive $\Delta^{199}Hg_{IHg}$ values that is derived from fishery food.

**4.3. MMHg Exposure Sources.** This study developed a new binary mixing model based on $\Delta^{199}Hg_{MMHg}$ ($BIM_{MMHg}$) to estimate the relative contributions of human MMHg exposure from fishery food. The THg and MMHg exposure assessment methods, such as the THg DAA method ($DAA_{THg}$), MMHg DAA method ($DAA_{MMHg}$), and THg binary isotope mixing method ($BIM_{THg}$), were also adopted for comparison.[9,10,20,23] The details and statistical data for the $DAA_{THg}$ and $DAA_{MMHg}$ methods are shown in the Supporting Information, Section S2 and Table S16.

In the coastal areas, 85 ± 15%, 86 ± 3%, and 73 ± 11% of the MMHg in the human hair and 80 ± 3% of the THg in the human hair were due to fishery food consumption, as estimated by the $BIM_{MMHg}$, $DAA_{MMHg}$, $BIM_{THg}$, and $DAA_{THg}$ methods, respectively (Figure 4). All the methods produced similar results in the coastal areas, except for a somewhat lower estimation from the $BIM_{THg}$ method. Due to the massive production and widespread consumption habits of fishery food

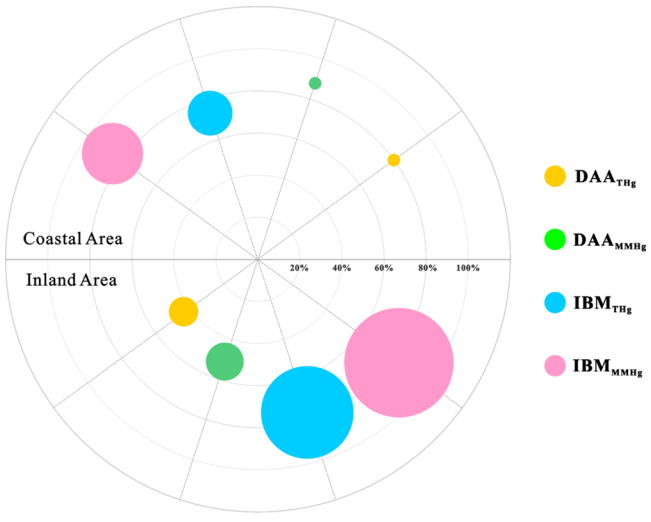



**Figure 4.** Estimated contributions of fishery food intake to human Hg exposure. Circles filled with different colors represent the relative contributions of human THg/MMHg exposure through fishery food consumption that are estimated by DAATHg (yellow), DAAMMHg (green), IBMTHg (blue), and IBMMMHg (pink). The radii of the circles represent the 1 SD values of the estimated methods, which were calculated by the Monte Carlo model.

in coastal areas,[35] the dietary intake of MMHg may be less affected by individual variations and living conditions,[9] which would lead to similar outputs from the $DAA_{MMHg}$ and $BIM_{MMHg}$ methods. By considering the different exposure sources of MMHg and IHg to the human body, the $\Delta^{199}Hg$ values of IHg were significantly lower than those of MMHg in the hair samples. In addition, IHg accounted for 17−81% of the THg in the human hair in the coastal areas (Table S10). Such a non-negligible fraction of IHg would obviously impact the THg isotope values in hair samples, which may explain the discrepancy in the MMHg exposure estimations between the $BIM_{THg}$ and $BIM_{MMHg}$ methods. The MMHg exposures that were estimated by the $BIM_{THg}$ method in previous studies focused on individuals whose hair Hg was mostly in the form of MMHg.[3,20,22] In situations where individuals are exposed to complicated IHg sources, the THg isotopes in the hair may not accurately indicate the MMHg exposure sources, and using the MMHg isotope values to identify the MMHg exposure sources in the hair would be a good approach.

In the inland areas, 83 ± 26%, 49 ± 11%, 75 ± 22%, and 44 ± 7% of the human MMHg (or THg) exposures were estimated to be from fishery food consumption by the $BIM_{MMHg}$, $DAA_{MMHg}$, $BIM_{THg}$, and $DAA_{THg}$ methods, respectively (Figure 4). These numbers were quite different from those in the coastal areas. The $BIM_{MMHg}$ and $BIM_{THg}$ methods yielded similar results, which were much higher than those that were shown by the $DAA_{MMHg}$ and $DAA_{THg}$ methods. The similar results from the $BIM_{MMHg}$ and $BIM_{THg}$ methods could be explained by the relatively high MMHg % values in the hair samples in inland areas, which minimized the disturbance of IHg. The large discrepancies in the exposure estimations between the DAA and isotope methods should largely be caused by the uncertainties in the dietary statistics.[20] People living in inland areas may have different fish-consumption habits depending on their income levels and living areas, which are completely different from coastal areas. For instance, higher-income families may consume substantially more fishery food than lower-income families, and this factor was not explicitly considered in our data sampling, which could lead to statistical uncertainties in the daily intake of fishery food. Other uncertainties in our data statistics can result from the individual variations in the demethylation ability of MMHg and the distinct absorption efficiencies of fish and rice MMHg in the human gastrointestinal phase.[11,55] When compared with the MMHg isotope methods, the DAA methods cannot avoid or estimate these uncertainties due to the lack of direct evidence and indicators.

This study demonstrated that inaccuracies can be caused by the IHg fraction in hair samples when using the THg isotopes to identify and quantify the MMHg exposure sources. Similarly, the variations in individual daily food intake and MMHg absorption efficiency induced large uncertainties when using the DAA model.[20] MMHg isotope analysis is an efficient method for tracking human MMHg exposure sources by linking dietary MMHg intakes directly to human exposures. Although rice is the staple food in the study areas, fishery food intake is still the principal pathway for human MMHg exposure, which implies that the health risk of MMHg exposure via rice consumption should not be overestimated in fish-rich areas.[66] However, in typical Hg-polluted areas, rice may accumulate significant amounts of MMHg from polluted paddy soils.[53] In these areas, the relative contributions of the rice and fishery food intakes to human MMHg exposures that


https://doi.org/10.1021/acs.est.1c01771
Environ. Sci. Technol. 2021, 55, 12493−12503

are solely determined by the MMHg isotopes are still unclear. The MMHg-CSIA method can be adopted to track human MMHg sources in typical Hg-polluted areas.

## ■ ASSOCIATED CONTENT

### Supporting Information

The Supporting Information is available free of charge at https://pubs.acs.org/doi/10.1021/acs.est.1c01771.

SEM method; DAA model; THg isotope binary method; THg isotope values; questionnaire information about residents in ZS, XM, and WH; fishery food species and yields sampled in coastal and inland areas; THg and MMHg results ofCRMs; duplicate samples for the SEM method and THg digest; THg-digested recovery of fishery food, rice, the hair, and CRM; purity and recovery of the SEM method of fishery food, rice, the hair, and CRM; quality control (mean ± 1 SD) of THg and the MMHg isotope of CRM; concentrations (wet weight) and isotope compositions of THg and related information about fishery samples; concentrations (dry weight) and isotope compositions of THg and related information about rice samples; concentrations (dry weight) and isotope compositions of THg and related information about hair samples; MMHg isotopes and recovery and purity in fishery food, rice, and the human hair; calculated IHg isotopes (mean and 1 SD) of fishery food, rice, and the human hair; statistical significance values ($p$) among the variables of of coastal and inland areas; statistical significance values ($p$) among the MMHg and IHg isotope values in all samples; statistical significance among the MMHg and IHg isotope values in all samples; parameters used in the diet calculation model; sampling sites of fishery food, rice, and the human hair; correlations between hair LgC and ages, heights, and weights; $\Delta^{199}Hg_{IHg}$ and $\Delta^{199}Hg_{MMHg}$ in fishery food in coastal and inland areas; and $\Delta^{199}Hg_{IHg}$ and $\Delta^{199}Hg_{MMHg}$ in the hair in coastal and inland areas (PDF)

## ■ AUTHOR INFORMATION


### Corresponding Author

**Ping Li** − *State Key Laboratory of Environmental Geochemistry, Institute of Geochemistry, Chinese Academy of Sciences, Guiyang 550081, China; CAS Center for Excellence in Quaternary Science and Global Change, Xi'an 710061, China;* orcid.org/0000-0002-0145-4122; Email: liping@mail.gyig.ac.cn

### Authors

**Shaochen Yang** − *Hubei Key Laboratory of Critical Zone Evolution, School of Earth Sciences, China University of Geosciences, Wuhan 430074, China*

**Bo Wang** − *State Key Laboratory of Environmental Geochemistry, Institute of Geochemistry, Chinese Academy of Sciences, Guiyang 550081, China*

**Chongyang Qin** − *State Key Laboratory of Environmental Geochemistry, Institute of Geochemistry, Chinese Academy of Sciences, Guiyang 550081, China*

**Runsheng Yin** − *State Key Laboratory of Ore Deposit Geochemistry, Institute of Geochemistry, Chinese Academy of Sciences, Guiyang 550081, China*

**Jinling Liu** − *Hubei Key Laboratory of Critical Zone Evolution, School of Earth Sciences, China University of Geosciences, Wuhan 430074, China;* orcid.org/0000-0002-9989-6550

**David Point** − *Observatory Midi-Pyrénées, Geosciences Environment Toulouse Laboratory, Research Institute for the Development (IRD), University of Toulouse and CNRS, Toulouse 31400, France*

**Laurence Maurice** − *Observatory Midi-Pyrénées, Geosciences Environment Toulouse Laboratory, Research Institute for the Development (IRD), University of Toulouse and CNRS, Toulouse 31400, France*

**Jeroen E. Sonke** − *Observatory Midi-Pyrénées, Geosciences Environment Toulouse Laboratory, Research Institute for the Development (IRD), University of Toulouse and CNRS, Toulouse 31400, France;* orcid.org/0000-0001-7146-3035

**Leiming Zhang** − *Air Quality Research Division, Science and Technology Branch, Environment and Climate Change Canada, Toronto M3H 5T4, Canada;* orcid.org/0000-0001-5437-5412

**Xinbin Feng** − *State Key Laboratory of Environmental Geochemistry, Institute of Geochemistry, Chinese Academy of Sciences, Guiyang 550081, China; CAS Center for Excellence in Quaternary Science and Global Change, Xi'an 710061, China;* orcid.org/0000-0002-7462-8998

Complete contact information is available at: https://pubs.acs.org/10.1021/acs.est.1c01771


### Notes

The authors declare no competing financial interest.

## ■ ACKNOWLEDGMENTS

This study was funded by the CAS Interdisciplinary Innovation Team (JCTD-2020-20); the National Natural Science Foundation of China (41573132 and 41921004); the Bureau of Frontier Sciences and Education, Chinese Academy of Sciences (QYZDJ-SSW-DQC005-03); the Youth Innovation Promotion Association, Chinese Academy of Sciences (2017442); and the CAS "Light of West China" Program.

## ■ REFERENCES


(1) Selin, N. E. Global biogeochemical cycling of mercury: a review. *Annu. Rev. Environ. Resour.* **2009**, *34*, 43−63.

(2) World Health Organization. *Ten Chemicals of Major Public Health Concern*. (accessed 15 August 2019), 2018.

(3) Laffont, L.; Sonke, J. E.; Maurice, L.; Hintelmann, H.; Pouilly, M.; Sánchez Bacarreza, Y.; Perez, T.; Behra, P. Anomalous mercury isotopic compositions of fish and human hair in the Bolivian amazon. *Environ. Sci. Technol.* **2009**, *43*, 8985−8990.

(4) Clarkson, T. W. The three modern faces of mercury. *Environ. Health Perspect.* **2002**, *110*, 11−23.

(5) Clarkson, T. W.; Magos, L. The toxicology of mercury and its chemical compounds. *Crit. Rev. Toxicol.* **2006**, *36*, 609−662.

(6) Bellanger, M.; Pichery, C.; Aerts, D.; Berglund, M.; Castaño, A.; Cejchanová, M.; Crettaz, P.; Davidson, F.; Esteban, M.; Fischer, M. E.; Gurzau, A. E.; Halzlova, K.; Katsonouri, A.; Knudsen, L. E.; Kolossa-Gehring, M.; Koppen, G.; Ligocka, D.; Miklavčič, A.; Reis, M. F.; Rudnai, P.; Tratnik, J. S.; Weihe, P.; Budtz-Jørgensen, E.; Grandjean, P.; DEMO/COPHES. Economic benefits of methylmercury exposure control in Europe: monetary value of neurotoxicity prevention. *Environ. Health* **2013**, *12*, 3.

(7) Cheng, J.; Gao, L.; Zhao, W.; Liu, X.; Sakamoto, M.; Wang, W. Mercury levels in fisherman and their household members in

Zhoushan, China: Impact of public health. *Sci. Total Environ.* **2009**, *407*, 2625−2630.
(8) Feng, X.; Li, P.; Qiu, G.; Wang, S.; Li, G.; Shang, L.; Meng, B.; Jiang, H.; Bai, W.; Li, Z.; Fu, X. Human exposure to methylmercury through rice intake in mercury mining areas, Guizhou Province, China. *Environ. Sci. Technol.* **2008**, *42*, 326−332.
(9) Zhang, H.; Feng, X.; Larssen, T.; Qiu, G.; Vogt, R. D. In Inland China, Rice, Rather than Fish, Is the Major Pathway for Methylmercury Exposure. *Environ. Health Perspect.* **2010**, *118*, 1183−1188.
(10) Li, P.; Feng, X.; Yuan, X.; Chan, H. M.; Qiu, G.; Sun, G.-X.; Zhu, Y.-G. Rice consumption contributes to low level methylmercury exposure in southern China. *Environ. Int.* **2012**, *49*, 18−23.
(11) Gong, Y.; Nunes, L. M.; Greenfield, B. K.; Qin, Z.; Yang, Q.; Huang, L.; Bu, W.; Zhong, H. Bioaccessibility-corrected risk assessment of urban dietary methylmercury exposure via fish and rice consumption in China. *Sci. Total Environ.* **2018**, *630*, 222−230.
(12) Li, P.; Feng, X.; Chan, H.-M.; Zhang, X.; Du, B. Human Body Burden and Dietary Methylmercury Intake: The Relationship in a Rice-Consuming Population. *Environ. Sci. Technol.* **2015**, *49*, 9682−9689.
(13) Karimi, R.; Fitzgerald, T. P.; Fisher, N. S. A Quantitative synthesis of mercury in commercial seafood and implications for exposure in the United States. *Environ. Health Perspect.* **2012**, *120*, 1512.
(14) Blum, J. D.; Sherman, L. S.; Johnson, M. W. Mercury isotopes in earth and environmental sciences. *Annu. Rev. Earth Planet. Sci.* **2014**, *42*, 249−269.
(15) Tsui, M. T.-K.; Blum, J. D.; Kwon, S. Y. Review of stable mercury isotopes in ecology and biogeochemistry. *Sci. Total Environ.* **2020**, *716*, 135386.
(16) Yin, R.; Feng, X.; Shi, W. Application of the stable-isotope system to the study of sources and fate of Hg in the environment: A review. *Appl. Geochem.* **2010**, *25*, 1467−1477.
(17) Kritee, K.; Barkay, T.; Blum, J. D. Mass dependent stable isotope fractionation of mercury during mer mediated microbial degradation of monomethylmercury. *Geochim. Cosmochim. Acta* **2009**, *73*, 1285−1296.
(18) Jimenez-Moreno, M.; Perrot, V.; Epov, V. N.; Monperrus, M.; Amouroux, D. Chemical kinetic isotope fractionation of mercury during abiotic methylation of Hg(II) by methylcobalamin in aqueous chloride media. *Chem. Geol.* **2013**, *336*, 26−36.
(19) Zheng, W.; Hintelmann, H. Mercury isotope fractionation during photoreduction in natural water is controlled by its Hg/DOC ratio. *Geochim. Cosmochim. Acta* **2009**, *73*, 6704−6715.
(20) Li, M.; Sherman, L. S.; Blum, J. D.; Grandjean, P.; Mikkelsen, B.; Weihe, P.; Sunderland, E. M.; Shine, J. P. Assessing sources of human methylmercury exposure using stable mercury isotopes. *Environ. Sci. Technol.* **2014**, *48*, 8800−8806.
(21) Rothenberg, S. E.; Yin, R.; Hurley, J. P.; Krabbenhoft, D. P.; Ismawati, Y.; Hong, C.; Donohue, A. Stable mercury isotopes in polished rice (Oryza Sativa L.) and hair from rice consumers. *Environ. Sci. Technol.* **2017**, *51*, 6480−6488.
(22) Laffont, L.; Sonke, J. E.; Maurice, L.; Monrroy, S. L.; Chincheros, J.; Amouroux, D.; Behra, P. Hg speciation and stable isotope signatures in human hair as a tracer for dietary and occupational exposure to mercury. *Environ. Sci. Technol.* **2011**, *45*, 9910−9916.
(23) Du, B.; Feng, X.; Li, P.; Yin, R.; Yu, B.; Sonke, J. E.; Guinot, B.; Anderson, C. W. N.; Maurice, L. Use of Mercury isotopes to quantify mercury exposure sources in inland populations, China. *Environ. Sci. Technol.* **2018**, *52*, 5407−5416.
(24) Laffont, L.; Maurice, L.; Amouroux, D.; Navarro, P.; Monperrus, M.; Sonke, J. E.; Behra, P. Mercury speciation analysis in human hair by species-specific isotope-dilution using GC-ICP-MS. *Anal. Bioanal. Chem.* **2012**, *405*, 3001−3010.
(25) Queipo-Abad, S.; Gonzalez, P. R.; Martínez-Morillo, E.; Davis, W. C.; Alonso, J. I. G. Concentration of mercury species in hair, blood and urine of individuals occupationally exposed to gaseous elemental mercury in Asturias (Spain) and its comparison with individuals from a control group formed by close relatives. *Sci. Total Environ.* **2019**, *672*, 314−323.
(26) Meng, M.; Sun, R.-y.; Liu, H.-w.; Yu, B.; Yin, Y.-g.; Hu, L.-g.; Chen, J.-b.; Shi, J.-b.; Jiang, G.-b. Mercury isotope variations within the marine food web of Chinese Bohai Sea: Implications for mercury sources and biogeochemical cycling. *J. Hazard. Mater.* **2020**, *384*, 121379.
(27) Yin, R.; Feng, X.; Meng, B. Stable mercury isotope variation in rice plants (Oryza sativa L.) from the Wanshan mercury mining district, SW China. *Environ. Sci. Technol.* **2013**, *47*, 2238−2245.
(28) Li, P.; Du, B.; Maurice, L.; Laffont, L.; Lagane, C.; Point, D.; Sonke, J. E.; Yin, R.; Lin, C.-J.; Feng, X. Mercury isotope signatures of methylmercury in rice samples from the Wanshan mercury mining area, china: environmental implications. *Environ. Sci. Technol.* **2017**, *51*, 12131.
(29) Masbou, J.; Point, D.; Sonke, J. E. Application of a selective extraction method for methylmercury compound specific stable isotope analysis (MeHg-CSIA) in biological materials. *J. Anal. At. Spectrom.* **2013**, *28*, 1620−1628.
(30) Dzurko, M.; Foucher, D.; Hintelmann, H. Determination of compound-specific Hg isotope ratios from transient signals using gas chromatography coupled to multicollector inductively coupled plasma mass spectrometry (MC-ICP/MS). *Anal. Bioanal. Chem.* **2009**, *393*, 345−355.
(31) Rodríguez-González, P.; Epov, V. N.; Bridou, R.; Tessier, E.; Guyoneaud, R.; Monperrus, M.; Amouroux, D. Species-specific stable isotope fractionation of mercury during Hg(II) methylation by an anaerobic bacteria (Desulfobulbus propionicus) under dark conditions. *Environ. Sci. Technol.* **2009**, *43*, 9183−9188.
(32) Liu, X.; Cheng, J.; Yuling, S.; Honda, S. i.; Wang, L.; Liu, Z.; Sakamoto, M.; Liu, Y. Mercury concentration in hair samples from Chinese people in coastal cities. *J. Environ. Sci.* **2008**, *20*, 1258−1262.
(33) Liu, J.-L.; Xu, X.-R.; Yu, S.; Cheng, H.; Peng, J.-X.; Hong, Y.-G.; Feng, X.-B. Mercury contamination in fish and human hair from Hainan Island, South China Sea: Implication for human exposure. *Environ. Res.* **2014**, *135*, 42−47.
(34) Zhang, W.; Zhang, X.; Tian, Y.; Zhu, Y.; Tong, Y.; Li, Y.; Wang, X. Risk assessment of total mercury and methylmercury in aquatic products from offshore farms in China. *J. Hazard. Mater.* **2018**, *354*, 198−205.
(35) China Fishery Press. *China Fishery Statistical Yearbook*; China Fishery Press: Beijing, China, 2016.
(36) Liu, C.-B.; Hua, X.-B.; Liu, H.-W.; Yu, B.; Mao, Y.-X.; Wang, D.-Y.; Yin, Y.-G.; Hu, L.-G.; Shi, J.-B.; Jiang, G.-B. Tracing aquatic bioavailable Hg in three different regions of China using fish Hg isotopes. *Ecotoxicol. Environ. Safe.* **2018**, *150*, 327−334.
(37) USEPA. *Method 1631: Revision E, Mercury in Water by Oxidation, Purge and Trap, and Cold Vapor Atomic Fluorescence Spectrometry*; USEPA: Washington, 2002.
(38) Liang, L.; Horvat, M.; Cernichiari, E.; Gelein, B.; Balogh, S. Simple solvent extraction technique for elimination of matrix interferences in the determination of methylmercury in environmental and biological samples by ethylation-gas chromatography-cold vapor atomic fluorescence spectrometry. *Talanta* **1996**, *43*, 1883−1888.
(39) Yin, R.-S.; Feng, X.-B.; Foucher, D.; Shi, W.-F.; Zhao, Z.-Q.; Wang, J. High Precision Determination of Mercury Isotope Ratios Using Online Mercury Vapor Generation System Coupled with Multicollector Inductively Coupled Plasma-Mass Spectrometer. *Chin. J. Anal. Chem.* **2010**, *38*, 929−934.
(40) Bergquist, B. A.; Blum, J. D. Mass-Dependent and -Independent Fractionation of Hg Isotopes by Photoreduction in Aquatic Systems. *Science* **2007**, *318*, 417−420.
(41) Kwon, S. Y.; Blum, J. D.; Nadelhoffer, K. J.; Timothy Dvonch, J.; Tsui, M. T.-K. Isotopic study of mercury sources and transfer between a freshwater lake and adjacent forest food web. *Sci. Total Environ.* **2015**, *532*, 220−229.
(42) Lepak, R. F.; Janssen, S. E.; Yin, R.; Krabbenhoft, D. P.; Ogorek, J. M.; DeWild, J. F.; Tate, M. T.; Holsen, T. M.; Hurley, J. P.


https://doi.org/10.1021/acs.est.1c01771
*Environ. Sci. Technol.* 2021, 55, 12493−12503

Factors affecting mercury stable isotopic distribution in piscivorous fish of the Laurentian Great Lakes. *Environ. Sci. Technol.* **2018**, *52*, 2768−2776.
(43) Perrot, V.; Pastukhov, M. V.; Epov, V. N.; Husted, S.; Donard, O. F. X.; Amouroux, D. Higher Mass-Independent Isotope Fractionation of Methylmercury in the Pelagic Food Web of Lake Baikal (Russia). *Environ. Sci. Technol.* **2012**, *46*, 5902−5911.
(44) Bonsignore, M.; Tamburrino, S.; Oliveri, E.; Marchetti, A.; Durante, C.; Berni, A.; Quinci, E.; Sprovieri, M. Tracing mercury pathways in Augusta Bay (southern Italy) by total concentration and isotope determination. *Environ. Pollut.* **2015**, *205*, 178−185.
(45) Senn, D. B.; Chesney, E. J.; Blum, J. D.; Bank, M. S.; Maage, A.; Shine, J. P. Stable isotope (N, C, Hg) study of methylmercury sources and trophic transfer in the Northern Gulf of Mexico. *Environ. Sci. Technol.* **2010**, *44*, 1630−1637.
(46) Balogh, S. J.; Tsui, M. T.-K.; Blum, J. D.; Matsuyama, A.; Woerndle, G. E.; Yano, S.; Tada, A. Tracking the fate of mercury in the fish and bottom sediments of Minamata Bay, Japan, using stable mercury isotopes. *Environ. Sci. Technol.* **2015**, *49*, 5399−5406.
(47) Blum, J. D.; Popp, B. N.; Drazen, J. C.; Choy, C. A.; Johnson, M. W. Methylmercury production below the mixed layer in the North Pacific Ocean. *Nat. Geosci.* **2013**, *6*, 879−884.
(48) Kwon, S. Y.; Blum, J. D.; Yin, R.; Tsui, M. T.-K.; Yang, Y. H.; Choi, J. W. Mercury stable isotopes for monitoring the effectiveness of the Minamata Convention on Mercury. *Earth−Sci. Rev.* **2020**, *203*, 103111.
(49) Motta, L. C.; Blum, J. D.; Johnson, M. W.; Umhau, B. P.; Popp, B. N.; Washburn, S. J.; Drazen, J. C.; Benitez-Nelson, C. R.; Hannides, C. C. S.; Close, H. G.; Lamborg, C. H. Mercury cycling in the North Pacific Subtropical Gyre as revealed by mercury stable isotope ratios. *Glob. Biogeochem. Cycles* **2019**, *33*, 777−794.
(50) Gehrke, G. E.; Blum, J. D.; Slotton, D. G.; Greenfield, B. K. Mercury isotopes link mercury in San Francisco Bay Forage fish to surface sediments. *Environ. Sci. Technol.* **2011**, *45*, 1264−1270.
(51) Kwon, S. Y.; Blum, J. D.; Chen, C. Y.; Meattey, D. E.; Mason, R. P. Mercury Isotope Study of Sources and Exposure Pathways of Methylmercury in Estuarine Food Webs in the Northeastern U.S. *Environ. Sci. Technol.* **2014**, *48*, 10089−10097.
(52) Li, M.; Schartup, A. T.; Valberg, A. P.; Ewald, J. D.; Krabbenhoft, D. P.; Yin, R.; Balcom, P. H.; Sunderland, E. M. Environmental origins of methylmercury accumulated in subarctic estuarine fish indicated by mercury stable isotopes. *Environ. Sci. Technol.* **2016**, *50*, 11559−11568.
(53) Qin, C.; Du, B.; Yin, R.; Meng, B.; Fu, X.; Li, P.; Zhang, L.; Feng, X. Isotopic fractionation and source appointment of methylmercury and inorganic mercury in a paddy ecosystem. *Environ. Sci. Technol.* **2020**, *54*, 14334−14342.
(54) Sherman, L. S.; Blum, J. D.; Franzblau, A.; Basu, N. New insight into biomarkers of human mercury exposure using naturally occurring mercury stable isotopes. *Environ. Sci. Technol.* **2013**, *47*, 3403−3409.
(55) Guo, G.; Yumvihoze, E.; Poulain, A. J.; Man Chan, H. Monomethylmercury degradation by the human gut microbiota is stimulated by protein amendments. *J. Toxicol. Sci.* **2018**, *43*, 717−725.
(56) Wang, Y.; Janssen, S. E.; Schaefer, J. K.; Yee, N.; Reinfelder, J. R. Tracing the Uptake of Hg(II) in an Iron-Reducing Bacterium Using Mercury Stable Isotopes. *Environ. Sci. Technol. Lett.* **2020**, *7*, 573−578.
(57) Janssen, S. E.; Schaefer, J. K.; Barkay, T.; Reinfelder, J. R. Fractionation of Mercury Stable Isotopes during Microbial Methylmercury Production by Iron- and Sulfate-Reducing Bacteria. *Environ. Sci. Technol.* **2016**, *50*, 8077−8083.
(58) Yin, R.; Feng, X.; Zhang, J.; Pan, K.; Wang, W.; Li, X. Using mercury isotopes to understand the bioaccumulation of Hg in the subtropical Pearl River Estuary, South China. *Chemosphere* **2016**, *147*, 173−179.
(59) Jackson, T. A.; Whittle, D. M.; Evans, M. S.; Muir, D. C. G. Evidence for mass-independent and mass-dependent fractionation of the stable isotopes of mercury by natural processes in aquatic ecosystems. *Appl. Geochem.* **2008**, *23*, 547−571.
(60) Wang, X.; Wang, W.-X. Selenium induces the demethylation of mercury in marine fish. *Environ. Pollut.* **2017**, *231*, 1543−1551.
(61) Sun, R.; Yuan, J.; Sonke, J. E.; Zhang, Y.; Zhang, T.; Zheng, W.; Chen, S.; Meng, M.; Chen, J.; Liu, Y.; Peng, X.; Liu, C. Methylmercury produced in upper oceans accumulates in deep Mariana Trench fauna. *Nat. Commun.* **2020**, *11*, 3389.
(62) Madigan, D. J.; Li, M.; Yin, R.; Baumann, H.; Snodgrass, O. E.; Dewar, H.; Krabbenhoft, D. P.; Baumann, Z.; Fisher, N. S.; Balcom, P.; Sunderland, E. M. Mercury stable isotopes reveal influence of foraging depth on mercury concentrations and growth in Pacific Bluefin tuna. *Environ. Sci. Technol.* **2018**, *52*, 6256−6264.
(63) McDowell, M. A.; Dillon, C. F.; Osterloh, J.; Bolger, P. M.; Pellizzari, E.; Fernando, R.; de Oca, R. M.; Schober, S. E.; Sinks, T.; Jones, R. L.; Mahaffey, K. R. Hair Mercury Levels in U.S. Children and Women of Childbearing Age: Reference Range Data from NHANES 1999-2000. *Environ. Health Perspect.* **2004**, *112*, 1165−1171.
(64) Sherman, L. S.; Blum, J. D.; Basu, N.; Rajaee, M.; Evers, D. C.; Buck, D. G.; Petrlik, J.; DiGangi, J. Assessment of mercury exposure among small-scale gold miners using mercury stable isotopes. *Environ. Res.* **2015**, *137*, 226−234.
(65) Du, B.; Yin, R.; Fu, X.; Li, P.; Feng, X.; Maurice, L. Use of mercury isotopes to quantify sources of human inorganic mercury exposure and metabolic processes in the human body. *Environ. Int.* **2021**, *147*, 106336.
(66) Xu, X.; Han, J.; Pang, J.; Wang, X.; Lin, Y.; Wang, Y.; Qiu, G. Methylmercury and inorganic mercury in Chinese commercial rice: Implications for overestimated human exposure and health risk. *Environ. Pollut.* **2020**, *258*, 113706.


https://doi.org/10.1021/acs.est.1c01771
*Environ. Sci. Technol.* 2021, 55, 12493−12503