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Distribution and speciation of mercury affected by humic acid in mariculture sites at the Pearl River estuary[☆]

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A B S T R A C T

At the Pearl River Estuary of southern China, mercury and its environmental problems have long been a great concern. This study investigated the distribution and speciation of mercury compounds that are significantly influenced by the increasing content of humic acid (HA, a model natural organic matter) in this region. The inorganic mercury and methyl mercury, being adsorbed and converted at different HA levels, were studied in sediments and surface water at both mariculture and their reference sites. In mariculture sediments with higher HA content (up to 4.5%), more mercury were adsorbed at different compound levels, promoting the methylation and accumulation of mercury ($P < 0.05$) at the sediment-water interface. Seasonal shift in environmental temperature might control the HA content, subsequently favouring mercury methylation (maximum $1.75 \pm 0.08\ \mathrm{mg\ L^{-1}\ d^{-1}}$) under warm weather conditions. In reference sites received less HA wastes, lower adsorption capacity and methylation rate were observed for mercury in sediments and surface water. Our work points to the significant roles of HA on mercury distribution and speciation both spatially and seasonally, thus addressing the impacts of mariculture activities on estuary eco-system.

1. Introduction

Mercury (Hg) exists in elemental, inorganic and organic forms. Elemental and inorganic mercury can cause severe damage to nervous and immune systems (Li and Tse, 2015). They can be converted to organic mercury with methyl mercury as one of the most toxic forms under favourable conditions. Methylation of mercury is prone to occur in tropical and subtropical climates (governing the Pearl River estuary) with enhanced state entropy of reactants at higher temperature (Fleck et al., 2016). Methyl mercury, a carcinogenic substance with good water solubility, can be bio-accumulated in living organisms and bio-magnified through aquatic food chains (Beauvais-Fluck et al., 2017). It is frequently detected to exceed the maximum limit regulated by the World Health Organization in marine fish species which constitutes an important fraction of seafood intake by humans (Chen et al., 2014). The environmental problems and detrimental impacts of methyl mercury on human health therefore have long been great concerns for decades. This is especially important in southern China which serves as an important aquaculture base but lacks of detailed research data related to mercury speciation in different ecological compartments (Zhang and Wong, 2007). To meet the urgent requirements for pollution control and water safety, more comprehensive investigation and evaluation of the ecological impacts of mercury and in particular its environmental behaviours should be initiated.

Humus is the main organic component in natural sediments. It is categorized as humic acid (HA), fulvic acid and humin according to the solubility of each individual substance. Among which HA can be dissolved in alkaline but not in acidic solutions, while fulvic acid in both acidic and alkaline, and humin in neither conditions (Bonomaluwa and Palutnicowa, 1987). These three humus compounds share similar molecular structures with only differences between molecular weight, elemental ratio and functional groups

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etc. The deposited HA have been widely found to form stable complexes with mercury, thus preventing soluble species such as Hg^{2+} from being leached into surface water layers (Chiasson-Gould et al., 2014). The released Hg^{2+} ions interact actively with dissolved HA in aqueous phases, largely owing to their higher mobility with better mass transfer. More importantly, the free Hg^{2+} ions are likely to interconvert between redox and inorganic/organic states, e.g. Hg^{2+} being reduced to Hg^0 and methylated to methyl mercury, whose transport, fate and eco-toxicity shall be influenced significantly in the presence of HA (Gu et al., 2011; Zheng et al., 2012). The mercury compounds can be methylated in both biotic and abiotic pathways (Cesário et al., 2017). It has been noted that the increase in the methylating efficacy of microorganisms is promoted by dissolved organic matters and nutrients (Mazrui et al., 2016). However, insufficient information is available concerning the specific influences of natural HA and related factors on mercury methylation in sediments and surface water.

Estuary, a typical transitional and highly dynamic environment, is of great scientific research interest and well aware of mercury contamination as compared to freshwater systems (Yin et al., 2016). For a long period, the fishery industry has dominated the local economy at the estuary of Pearl River, southern China. In this area, organic carbon-rich food, usually small trash fish or artificially minced/powered marine meat/plant containing or producing humic substances, is intensively applied to culture commercial fish (Zhou et al., 2004). Among which, only a minority of the HA dosed as carbon sources can be digested by cultured fish, while the major parts are not consumed or excreted in their original forms through urine or feces (Liang et al., 2011). The HA may account for up to 90% of the dissolved organic matter in the culture water (Leonard et al., 2002). It has been proven by previous studies that mariculture activities would affect the physical, chemical and biological parameters and thereby determining the speciation, bioavailability and toxicity of mercury in the aquatic environment (Jiang et al., 2017; Liang et al., 2017). For example, mariculture might lead to variations between the ratios of total mercury to methyl mercury subjected to the methylating activity of sulfate-reducing bacteria and sulfide concentration affected by food supply and fish excretion. Therefore, it is crucial to examine the fate and transformation of mercury affected by the elevated content of HA discharged into the mariculture zones.

The major objective of this study was to evaluate the roles of HA governing the behaviours of mercury in the aquatic environment. More specifically, the followings were investigated in mariculture sites compared to their reference sites at the Pearl River estuary: (1) Mercury adsorption which determined its distribution between aqueous and solid phases at different compound levels affected by natural HA; and (2) Mercury methylation influenced by increased contents of HA and varied temperature at different seasons in both sediments and surface water. The HA-mercury interactions were interpreted by isothermal models, mercury speciation and statistical analyses to provide real insights into the biogeochemical behaviours of complex contaminants in the estuary eco-system.

2. Materials and methods

2.1. Sample collection and pretreatment

Two mariculture sites and their reference sites (200–1000 m away from each other), i.e. Guishan Island (between Mainland and Hong Kong) and Daya Bay (completely isolated from the Pearl River), were investigated at the Pearl River estuary in southern China. Briefly, surface seawater was sampled using polyethylene bottles that were pre-treated with 0.5% (v/v) HCl. In addition, sediment core samples were collected from the 0–25 cm surface layers by using a customized columnar sampler. All samples were collected in duplicates and immediately transported from each site to the laboratory in ice bath. Water samples were stored at −20 °C in darkness until analyses. Sediment samples were freeze-dried, ground into powder, homogenized by a 0.154-mm sieve made of stainless steel and kept in sealed storage containers. The detailed geographic and sampling information can be further referred to supplementary information (Fig. S1 and Table S1) and our previous study (Tao et al., 2016).

2.2. Mercury quantification and QA/QC

Mercury quantification was based on total and methylated concentrations. Total mercury in sediments was digested using ~0.2 g dry samples with 5.0 mL aqua regia at 95 °C for 5 h. All mercury were oxidized to divalent mercury by 0.5% (v/v) BrCl and then reduced to elemental mercury by $SnCl_2$. The total concentrations of mercury were analyzed by Total Mercury Manual System (Brooks Rand Labs, USA) equipped with a cold vapor atomic fluorescence spectrometer, according to Method 1631 (USEPA, 2002). Methyl mercury in sediments was extracted by $HNO_3/CuSO_4/$dichloromethane prior to stripping to water (Liang et al., 2004). The concentrations of methyl mercury were detected after distillation and ethylation followed by gas chromatography coupled to cold vapor atomic fluorescence spectrometry, according to Method 1630 (USEPA, 2001).

Analytical quality assurance and/or quality control (QA/QC) were conducted with standard curves, method blanks (solvent) and reagent blanks as well as 10% replicate samples in each sample batch. Identical or similar reference materials were used to verify the analytical methods. Four certified reference materials of NIST 1641e (National Institute of Standards & Technology, USA), GBW08675 and GBW(E)070009 (National Institute of Metrology, China) and IAEA 433 (International Atomic Energy Agency, Austria) were employed to evaluate the accuracy of total and methyl mercury measurements respectively. The detailed QA/QC information is shown in Table S2.

2.3. Humic acid extraction and purification

Extraction and purification of natural HA were carried out following the standard method set by international humic substance society (Swift, 1996). Firstly, the sampled sediments were mixed with 0.5 M HCl at a solid-liquid ratio of 1:10 (g mL^{-1}). After vigorous stirring for 5 h, the mixture was kept at room temperature (25 ± 5 °C) for another 5 h. All settled solids were centrifuged and washed with deionized (DI) water for 3 times upon removal of supernatant. Secondly, all solids separated in step 1 were dissolved in 0.4 M NaOH solution also at a solid-liquid ratio of 1:10. Residual solids were removed by centrifugation after 5-h stirring and 5-h settlement. Thirdly, the brown solution obtained in step 2 was adjusted to pH = 2.0 for precipitation of HA with 6.0 M HCl, kept overnight and subsequently the supernatant was removed by centrifugation. Fourthly, the centrifuged solids from step 3 were re-dissolved and re-precipitated with 6.0 M HCl and 0.4 M NaOH for 3 times. The final solids were washed with 1% HF-99% HCl mixture to remove silicate minerals, washed with DI water for several times and adjusted to pH = 7.0 with 6.0 M HCl. After the complete removal of Cl^- ions (tested with $AgNO_3$ solution) by dialysis, pure HA was produced by reduced pressure distillation.

2.4. Experimental design

In the field, the deposited HA and their influences on mercury methylation were studied in top-layer sediments at different