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2 **Photochemical Reactions between Mercury (Hg) and Dissolved Organic**
3 **Matter Decrease Hg Bioavailability and Methylation**

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Abstract

Atmospheric deposition of mercury (Hg) to surface water is one of the dominant sources of Hg in aquatic environments and ultimately drives methylmercury (MeHg) toxin accumulation in fish. It is known that freshly deposited Hg is more readily methylated by microorganisms than aged or preexisting Hg; however the underlying mechanism of this process is unclear. We report that Hg bioavailability is decreased by photochemical reactions between Hg and dissolved organic matter (DOM) in water. Photo-irradiation of Hg-DOM complexes results in loss of Sn(II)-reducible (i.e. reactive) Hg and up to an 80% decrease in MeHg production by the methylating bacterium *Geobacter sulfurreducens* PCA. Loss of reactive Hg proceeded at a faster rate with a decrease in the Hg to DOM ratio and is attributed to the possible formation of mercury sulfide (HgS). These results suggest a new pathway of abiotic photochemical formation of HgS in surface water and provide a mechanism whereby freshly deposited Hg is readily methylated but, over time, progressively becomes less available for microbial uptake and methylation.

Capsule: Photolysis of Hg-DOM complexes in water leads to decreased Hg bioavailability and methylation, likely due to photochemical formation of HgS.

Keywords: Dissolved organic matter, Hg-DOM photolysis, methylmercury, biogeochemical transformation, HgS precipitation

1. Introduction

Microbial conversion of inorganic mercury (Hg) to organometallic methylmercury, CH_3Hg^+ (MeHg), is a global health concern due to the bioaccumulation and biomagnification of MeHg in the food chain (Harris et al., 2007; Krabbenhoft and Sunderland, 2013; Parks et al., 2013; Podar et al., 2015). Microorganisms preferentially utilize freshly deposited inorganic Hg(II) to form MeHg (Hintelmann et al., 2002; Paterson et al., 2006), usually within a short time period, but the longer Hg remains in the aquatic environment, the less likely it is to be involved in methylation and bioaccumulation (Harris et al., 2007; Hrabik and Watras, 2002; Jonsson et al., 2014; Paterson et al., 2006). For example, addition of enriched Hg stable isotopes in a large enclosure study (Paterson et al., 2006) and a whole lake experiment (Harris et al., 2007) show that fish MeHg concentrations increased rapidly following fresh Hg input at the lake surface. Independent studies of simultaneous decline in Hg deposition and fish MeHg concentration in Little Rock Lake in Wisconsin, United States, support the view that freshly deposited Hg is a dominant contributor to Hg bioaccumulation in lake fish (Hrabik and Watras, 2002).

Decreases in Hg reactivity (defined as Sn(II)-reducible Hg) (Lamborg et al., 2003; Miller et al., 2013; Miller et al., 2009) and bioavailability (Barkay et al., 1997; Jonsson et al., 2014) by DOM in the dark have been attributed to Hg binding with reduced sulfur or thiol ($-\text{S}$) functional groups in DOM. However, formation of Hg-thiol complexes does not necessarily prevent Hg from being taken up by microorganisms for methylation (Chiasson-Gould et al., 2014; Graham et al., 2012b; Schaefer et al., 2011), and Hg-cysteine complexes are known to enhance microbial methylation (Graham et al., 2012b; Lin et al., 2015; Schaefer et al., 2011).

81 Additionally, DOM can enhance MeHg production under sulfidic conditions since DOM can
82 inhibit precipitation and crystallization of HgS in water (Deonarine and Hsu-Kim, 2009;
83 Gerbig et al., 2011; Graham et al., 2012a). DOM may also affect Hg redox reactions
84 depending on the Hg-to-DOM ratio and the reaction kinetics (Gu et al., 2011; Jiang et al.,
85 2015; Miller et al., 2009). As fresh Hg enters aquatic environments, it can initially react with
86 DOM via abundant oxygen moieties and form weak Hg-DOM species that are readily
87 reduced to Hg(0) (Zheng and Hintelmann, 2009). However, over time or at increasing DOM
88 concentration, stronger Hg-DOM complexes via bonding with thiol moieties may
89 progressively dominate and inhibit Hg reduction (Gu et al., 2011; Jiang et al., 2015; Miller et
90 al., 2009; Zheng and Hintelmann, 2009). In illuminated conditions, photochemical reactions
91 can also affect Hg chemical speciation (Amyot et al., 1997b; He et al., 2014; Lalonde et al.,
92 2001; O'Driscoll et al., 2004; Poulain et al., 2004; Si and Ariya, 2015) as well as MeHg
93 photodegradation in water (Fernandez-Gomez et al., 2013; Mostofa et al., 2015; Qian et al.,
94 2014; Zhang and Hsu-Kim, 2010), but it remains unclear how photochemical reactions
95 influence Hg reactivity, bioavailability and/or MeHg biosynthesis.

96 In order to elucidate the mechanisms and chemical species involved in decreasing Hg
97 bioavailability in water, we examined the influence of photolysis on Hg-DOM species
98 transformation and subsequent changes in Hg availability for microbial methylation. Abiotic
99 formation of HgS following photo-irradiation of Hg-DOM complexes is proposed as a
100 mechanism responsible for decreasing Hg reactivity and bioavailability. We determined Hg
101 reactivity changes and the reaction products, as well as Hg bioavailability, assayed by MeHg
102 biosynthesis, following photo-irradiation of Hg-DOM solutions in water.

2. Materials and methods

2.1. Photolysis of Hg-DOM complexes

Two DOM isolates, representing both an aquatic and a soil-derived organic matter, were used in the photolysis experiments. They include a DOM isolated from East Fork Poplar Creek (EFPC-DOM) natural freshwater in Oak Ridge, Tennessee (Dong et al., 2011), and a reference Elliott soil humic acid (HA) from International Humic Substances Society (Gu et al., 2011; Zheng et al., 2012). The reactant solutions were prepared by mixing an aliquot of stock solutions of Hg (5 μM , as HgCl_2 in water) and DOM (2.1 mM DOC), either in simulated freshwater or in a 10 mM phosphate buffered solution (PBS). The simulated freshwater consisted of 1 mM NaHCO_3 , 0.5 mM NaCl , and 0.3 mM Na_2SO_4 at pH 8.0, similar to that observed in EFPC water (Dong et al., 2010; Miller et al., 2009). Final concentrations ranged from 1 to 12.5 nM Hg and 0.008 to 0.21 mM DOC, unless otherwise specified. Sample solutions were then transferred to a series of quartz vials (~6 mL, no headspace), which were placed either in a solar chamber (Suntest XLS+, Atlas) or an ultraviolet (UV) photoreactor (Luzchem Research) for photo-irradiation (He et al., 2012; Qian et al., 2014). The solar chamber is equipped with an air-cooled xenon lamp and a daylight filter (wavelength cutoff at 300 nm). The light irradiance was 40 W m^{-2} (or ~1.18 $\text{E s}^{-1} \text{ m}^{-2}$ photon flux) for UV (300–400 nm) and ~373.6 W m^{-2} for the full spectral range, 300–800 nm, typical of mid-summer conditions in eastern Tennessee (He et al., 2012; Qian et al., 2014). Total irradiance in the UV photoreactor was ~45 W m^{-2} (260–375 nm) (or ~1.34 $\text{E s}^{-1} \text{ m}^{-2}$ photon flux), with peak intensity at ~312 nm. Sample vials were placed upright at the bottom center of the reactor, and the reaction temperature was maintained at $30 \pm 1^\circ\text{C}$. Dark

controls were performed in the same manner in vials wrapped with aluminum foil.

At each selected time point, a set of sample vials ($n = 2-3$) was removed from the chamber and sacrificed for Hg species analysis. An aliquot (1 mL) was transferred into a 4-mL amber glass vial, immediately capped (with septum) and analyzed for dissolved gaseous Hg(0) by direct purging and analysis with a Hg(0) analyzer (Lumex RA-915+) (Gu et al., 2011; Hu et al., 2013; Lin et al., 2014). After purging, an aliquot (0.5 mL, from the purged sample vial) was transferred to a separate vial, where an excess amount of SnCl_2 (10% w/v in 5% HCl, 0.1 mL) was added, and the Sn(II)-reducible Hg was immediately determined. As stated earlier, this fraction of reducible Hg was operationally defined as the reactive Hg (HgR) (Lamborg et al., 2003; Miller et al., 2013; Miller et al., 2009). For mass balance analysis, the remaining aliquot (0.5 mL) was mixed with BrCl (5% v/v), allowed to react overnight at 4°C, and then determined for total non-purgeable Hg (Hg_{NP}) (Hu et al., 2013; Lin et al., 2015; Lin et al., 2014). Variations in duplicate samples were usually within $\pm 5\%$, and for triplicate samples error bars represent one standard deviation.

2.2. Hg methylation assays with *Geobacter sulfurreducens* PCA

Separate experiments were performed to evaluate changes in Hg bioavailability after Hg-DOM solutions were photo-irradiated. Assays were conducted with or without the photo-irradiated Hg-DOM by measuring MeHg production with washed cells of *Geobacter sulfurreducens* PCA in an anoxic chamber in the dark. Cell growth conditions and experimental details have been described previously (Hu et al., 2013; Lin et al., 2015; Lin et al., 2014), except that in this study washed cells were prepared in PBS containing 10 mM NaCl at pH 7.4. Hg-DOM solutions in PBS were first irradiated with UV (2 h) or solar light

(0.5 h or 5 h). An aliquot of the irradiated solution was then transferred to a series of 4-mL amber glass vials, to which washed *G. sulfurreducens* PCA cells were added. Acetate and fumarate (1 mM each) were added once to the cells as the respective electron donor and acceptor. The final Hg concentration was 12.5 nM, DOC was 0.21 mM, and the cell concentration was 10^8 cells mL⁻¹. Control assays (cells plus Hg in PBS, and cells plus Hg-DOM in PBS without prior photo-irradiation) were prepared in the same manner. Samples were then placed on an orbital shaker and kept in the dark in an anoxic chamber. At selected time points, triplicate sample vials were removed from the chamber and an aliquot was analyzed as above for purgeable Hg(0) and non-purgeable Hg_{NP}. Although ~25–50% of the Hg was reduced to Hg(0) during photo-irradiation and bio-incubation, no significant loss of Hg(0) occurred during analysis, as described in previous studies (Hu et al., 2013; Lin et al., 2014).

The remaining sample was preserved in 0.2% (v/v) H₂SO₄ and kept at –20°C until MeHg analysis. Total MeHg was determined using a modified EPA Method 1630, with Me²⁰⁰Hg added as an internal standard, as described in detail elsewhere (Hu et al., 2013; Lin et al., 2014; Parks et al., 2013). MeHg was first extracted from the sample matrix via distillation, ethylation, and trapping on a Tenax column via N₂-puring. Thermal desorption and separation by gas chromatography were applied prior to the detection of Hg by ICP-MS. The recovery of spiked MeHg standards was 100±10%, and the detection limit was $\sim 3 \times 10^{-5}$ nM MeHg.

2.3. Molecular characterization of Hg-DOM photolysis products

To produce sufficient quantities of the photo-reaction products (e.g., HgS) for compositional and mineralogical analyses, this experiment was performed at a relatively high

Hg concentration (0.1 mM). However, changes in HgR were measured independently at both high and low Hg (1–12.5 nM) concentrations for comparison; experiments were also performed with Hg-cysteine complexes using cysteine as an analog for thiol functional groups in DOM. Control experiments in the absence of DOM or in the presence of sulfate were performed in the same manner, and no HgS precipitates were observed. High-resolution Fourier transform ion cyclotron resonance mass spectrometry (FTICR-MS) coupled with electrospray ionization was used to determine the molecular formulae of DOM before and after light exposure. Prior to MS analysis, DOM samples were desalted using styrene divinyl benzene polymer (PPL) cartridges (Dittmar et al., 2008), and molecular formulae were assigned using previously established methods (Chen et al., 2014). Precipitates of HgS particles were further analyzed by scanning electron microscopy (SEM) (Zeiss Merlin) and energy-dispersive X-ray (EDX) spectroscopy operated at 20 kV with a Quantax microanalysis system (Bruker-AXS Microanalysis GmbH) (Gu et al., 2014).

3. Results and Discussion

3.1. Effects of photolysis on Hg reduction, oxidation, and reactivity

To determine the influence of Hg-DOM complexation and photo-redox transformations on Hg bioavailability, we first examined Hg photo-reduction, oxidation, and reactivity changes either in simulated freshwater, in PBS containing a reference HA (0.21 mM DOC), or directly in EFPC water containing ~ 0.13 mM DOC (Dong et al., 2010; Gu et al., 2011; Zheng et al., 2012). Hg (12.5 nM) was added to give a Hg/DOC molar ratio of 1×10^{-4} to 6×10^{-5} , at which Hg is expected to be predominantly complexed with reduced sulfur (–S)

functional groups (Gu et al., 2011; Jiang et al., 2015; Zheng and Hintelmann, 2009). Consistent with previous studies (Amyot et al., 1997a; Jeremiason et al., 2015; O'Driscoll et al., 2004; Zheng and Hintelmann, 2009), rapid photoreduction of Hg was observed with concurrent production of Hg(0) in 1 h (Fig. 1) due to direct photolysis of Hg-DOM in water upon irradiation with either UV (260–375 nm) or solar light (300–800 nm) (Jeremiason et al., 2015). In contrast, only a small amount of Hg (< 10%) was reduced in the dark (Fig. S1) because, in this case, the oxidized HA or creek DOM were used at a low, fixed concentration (0.13–0.21 mM DOC). The result thus differed from those observed previously using the reduced DOM at increasing DOM concentrations (Gu et al., 2011; Zheng et al., 2012). Importantly, we found that a large percentage of the Hg(0) product disappeared after a few hours of continued light exposure, indicating rapid photo-reoxidation of Hg(0), particularly under UV irradiation. The production of Hg(0) reached a maximum of ~6.9 nM (or 55%), 3 nM (24%), and 4.7 nM (38%) in 1 h in simulated freshwater, PBS, and creek water, respectively, and then decreased to near zero after 6 h (Fig. 1a,b,c). Although, to a lesser extent, solar light also caused photoreduction and photooxidation of Hg in simulated freshwater and EFPC creek water (Fig. 1d,f), no photooxidation of Hg(0) was observed in PBS (Fig. 1e) due to the relatively high concentration of phosphate (10 mM) used and its possible quenching effect (He et al., 2014). Hg(0) production reached a maximum of ~2.5–3 nM in simulated and creek waters (Fig. 1d,f), and reoxidation occurred after 1 h but was slower than that under UV irradiation. These observations agree with several (He et al., 2014; Lalonde et al., 2001), but not all (Jeremiason et al., 2015; Rose et al., 2015; Zheng and Hintelmann, 2009), previous studies, in which Hg(0) photo-reoxidation was not observed.

The disagreement is due mainly to previous experimental methods where evasion or continuous purging was used during photoreaction, i.e., not allowing Hg(0) to remain in the water. Our results suggest that, without purging, Hg(0) photooxidation could be an important natural process (He et al., 2014; Lalonde et al., 2001), given the high solubility of Hg(0) in water (~300 nM) and its rapid photooxidation kinetics.

Coinciding with Hg photoreduction and oxidation, HgR decreased sharply in all cases following photo-irradiation in the presence of DOM (Fig. 2), and UV was more effective than solar light in decreasing Hg reactivity. When Hg(0) was excluded by normalizing HgR to the non-purgeable Hg_{NP} (Fig. 1), HgR decreased by ~88% and 54%, following 1 h exposure to the UV and solar light, respectively, in simulated freshwater (Fig. 2a). After 6 h, only ~7% (under UV) and 36% (under solar) of the Hg was reducible by Sn(II). In the dark control experiment, HgR decreased by ~20%, as observed previously with HA at the same DOC concentration due to strong binding between Hg and certain fractions of DOM (Miller et al., 2013; Miller et al., 2009). Similar results were also obtained for Hg reactivity changes in PBS (Fig. 2b) and in natural EFPC water (Fig. 2c). After 6-h exposure to UV and solar light, HgR decreased by ~97% and 77%, respectively, in PBS; whereas in the creek water, HgR decreased by more than 99% and 75% (Fig. 2c), respectively.

Separate solar irradiation experiments were performed with an EFPC-DOM isolate (Dong et al., 2011) at a low Hg concentration (1 nM) (or Hg/DOC ratio = 7.7×10^{-6}) (Fig. 3a,b), similar to the conditions in the natural aquatic environment (Dong et al., 2010; Miller et al., 2009). Additionally, we evaluated the effects of Hg/DOC ratios (1.5×10^{-3} – 6×10^{-5}) on HgR changes in simulated freshwater (Fig. 3c). Results show that HgR decreased faster at

lower Hg/DOC ratios. Following 1 h sunlight exposure, >90% of the Hg became non-reducible at a Hg/DOC ratio $< 6 \times 10^{-5}$ (Fig. 3), compared to ~50% reducible at relatively high Hg/DOC ratios (1.5×10^{-3}). The initial slope of HgR increased substantially with decreasing Hg/DOC ratios (Fig. 3c), suggesting that photo-irradiation can induce rapid speciation changes of freshly deposited Hg in surface water (typically with a low Hg/DOC $< 10^{-5}$). Furthermore, the general patterns of HgR decrease upon photo-exposure are similar in natural creek water, in PBS, or in simulated freshwater containing HA or EFPC-DOM. This observation may be partially attributed to similar binding strength between Hg and thiolate functional groups in all natural organic matter, and also the relatively low Hg/DOC ratios (10^{-4} – 10^{-6}) used in this study (Dong et al., 2011; Dong et al., 2010; Haitzer et al., 2003). Together, these results demonstrate that photo-irradiation of Hg in the presence of DOM has altered the Hg or Hg-DOM chemical species to an extent that a portion of the Hg species cannot be reduced by Sn(II). In contrast, Hg complexes with simple thiols, e.g., Hg-cysteine and Hg-glutathione, are all reducible in the dark (Lamborg et al., 2003; Miller et al., 2009).

3.3. Photolysis of Hg-DOM on Hg bioavailability

We subsequently investigated whether decreasing Hg reactivity results in changes to Hg bioavailability, assayed by MeHg production using washed cells of *Geobacter sulfurreducens* PCA as a model organism (Hu et al., 2013; Lin et al., 2014; Parks et al., 2013; Schaefer et al., 2011). After irradiating Hg-DOM in PBS with either UV or solar light, we added cells (at 10^8 mL⁻¹) to initiate the biosynthesis of MeHg in the dark (Fig. 4). In control experiments without DOM, cells produced 0.52 nM MeHg in 4 days, equivalent to about 6.6% normalized to the total non-purgeable Hg_{NP}, as shown previously (Lin et al., 2014). Here we assume that Hg(0),

produced through photoreduction or bioreduction (Fig. S2), was unavailable for methylation by *G. sulfurreducens* (Hu et al., 2013; Lin et al., 2014). In comparison, cells produced only 0.1 nM MeHg in 4 days (or ~1.3% of the Hg_{NP}) in the presence of the photo-irradiated Hg-DOM (2 h UV exposure). This represents a decrease in MeHg production of ~80% compared to the no DOM dark control. The observed decrease in MeHg production is attributed to the rapid decline in HgR levels following exposure of the Hg-DOM solution to UV light (Fig. 2a,b). Similarly, exposure of Hg-DOM to solar light for 0.5 h and 5 h resulted in decreased MeHg production by ~40% and 56%, respectively (Fig. 4). Longer light exposure to Hg-DOM solutions led to lower MeHg production, whereas UV is more effective than sunlight in decreasing Hg bioavailability and caused the most dramatic decrease in MeHg production. We also note a decrease in MeHg production (~25% at day 4) in the presence of non-irradiated DOM. This is expected, as reactions of Hg with DOM also lead to decrease in HgR without light exposure (Fig. S1), although HgR decreases at a slower rate (Lamborg et al., 2003; Miller et al., 2009). Results of the methylation assays thus provide direct evidence that photo-irradiation of Hg-DOM complexes in water greatly decreases HgR and bioavailability for MeHg production by *G. sulfurreducens* PCA cells.

3.4. Characterization of Hg-DOM photolysis products

Techniques including FTICR-MS and SEM-EDX spectroscopy were subsequently employed to further understand how photo-irradiation of Hg-DOM in water may result in decreased HgR and bioavailability. For these analyses, it was necessary to use a relatively high Hg concentration to produce sufficient quantities of the photo-reaction products (e.g., HgS), although similar loss of HgR (>85%) was noted in both high (0.1 mM) and low Hg

(12.5 nM) solutions containing DOM (Fig. S3). In the absence of light, HgR loss was much less (~27%) (Fig. S3a,e). Notable amounts of black precipitate were observed only after light exposure at high Hg concentration (Fig. 5a inset) but not in the absence of DOM under the same experimental conditions. These results were further validated by photolysis of Hg-cysteine complexes under identical experimental conditions to the DOM experiments, and similar results were observed (Fig. 5b and Fig. S3b,d). We hypothesize that direct photolysis of Hg-DOM or Hg-cysteine complexes leads to the breakage of R-S bonds and formation of HgS solids or nanoparticles. Similar photochemical formation of HgS particles has been reported for reactions between Hg and thioglycolic acid at low pH (4) (Si and Ariya, 2015). Even in the dark, the formation of HgS from reactions between Hg and thiols was reported to be thermodynamically favorable (Manceau et al., 2015), involving a two-step reaction sequence analogous to the mechanisms for early formation of Hg_xS_y clusters in sulfidic solutions (Charnock et al., 2003). Direct photolysis of Hg bound to thiol moieties in DOM is also proposed as a mechanism for Hg photoreduction (Jeremiason et al., 2015). However the rate and extent of Hg photoreduction may be over-estimated in the study, in light of our findings of HgS formation and the indirect method used by these authors for determining Hg(0) during Hg-DOM photolysis.

FTICR-MS molecular characterization of DOM before and after reaction with Hg under solar irradiation shows that a large portion (~50%) of S-containing molecular formulae disappeared following light exposure (Table S1). Although FTICR-MS analysis cannot provide quantitative and structural information for these DOM molecules, the disappearance of S-containing formulae suggests elimination of S from Hg-DOM complexes via breakage

of the R–S bonds in (DOM-R)–SHg complexes. Further, similar EDX spectra of the black precipitates were observed in both the DOM and cysteine samples and show the dominant Hg and S peaks (Fig. 5a,b) (Note that the Cu and Al signatures in the EDX spectra are artifacts stemming from the use of copper grids and an aluminum sample holder during EDX analyses). These results thus support our hypothesis that HgS precipitates were formed as DOM or cysteine is the only source of S in these systems. In other words, HgS is the only possible precipitate or product that could form in systems containing Hg and cysteine. This also explains the loss of S-containing molecular formulae in DOM after light exposure (Table S1). Taken together, we provide strong evidence of photochemical-driven formation of HgS from reactions between Hg and DOM in water. These reactions thereby decrease Sn(II)-reducible Hg, accounting for the observed decreases in HgR and MeHg production.

4. Conclusions

Decreases in HgR and bioavailability are observed upon photo-irradiation of Hg-DOM complexes, leading to the formation of HgS in water. This is an important finding indicating a potentially unrecognized abiotic pathway for HgS formation under oxic conditions, in addition to the commonly recognized microbial formation of HgS under anoxic conditions (Han et al., 2008; Morse et al., 2007). Although a relatively high Hg concentration was used to assist reaction product identification (Fig. 5), similar reactions occur at lower Hg concentrations or lower Hg/DOC ratios (Figs. 2, 3, and S3). In fact, the reaction likely proceeds more completely and more quickly at the lower Hg/DOM ratios (Fig. 3) found in natural aquatic environments.

Our results also provide a mechanism whereby freshly deposited Hg is readily methylated but, over time, progressively becomes less available for microbial uptake and methylation. The photochemical conversion of Hg-DOM to HgS in our study resulted in decreased bioavailable Hg for *G. sulfurreducens* PCA cells to take up and produce MeHg. Similar processes likely occur in surface waters generally, thereby affecting the biogeochemical transformation and cycling of Hg and production of MeHg. These findings highlight the importance of photochemical transformation and removal of Hg via HgS formation, and draw attention to a potential sink for Hg in the aquatic environment.

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Figure Legends

Fig. 1. Effects of light exposure on Hg species distribution: Purgeable dissolved elemental Hg(0) produced, non-purgeable Hg_{NP}, and total Hg_T. Left panel corresponds to experiments performed under UV irradiation (260–375 nm) in (a) simulated freshwater (S. water) and (b) 10-mM phosphate buffered solution (PBS) containing 0.21 mM DOC (as HA), and (c) natural EFPC creek water containing 0.13 mM DOC. Right panel corresponds to similar experiments performed under solar light (300–800 nm) in (d) simulated freshwater, (e) PBS, and (f) the creek water. The initial added Hg concentration was 12.5 nM. Error bars represent one standard deviation where they are larger than the data points.

Fig. 2. Effects of light exposure on reactive Hg (Hg_R), normalized to the total non-purgeable Hg_{NP} (i.e., Hg(0) excluded), as a function of reaction time between Hg and DOM. The experiments were performed separately under ultraviolet, solar light, and dark conditions, either in (a) simulated freshwater, (b) PBS, and (c) natural EFPC creek water (See Fig. 1 for additional details).

Fig. 3. Effects of solar light exposure on reactive Hg (Hg_R), normalized to the total non-purgeable Hg_{NP}, as a function of reaction time between Hg and DOM in (a) simulated freshwater and (b) PBS containing 1 nM Hg and an EFPC-DOM isolate (0.21 mM DOC). (c) Similar experiments were performed at a fixed Hg concentration (12.5 nM) but varying DOM concentrations (0.008 to 0.21 mM DOC) or Hg/DOM ratios (1.5×10^{-3} to 6×10^{-5}). Error bars represent one standard deviation where they are larger than the data points.

Fig. 4. Methylmercury (MeHg) production, normalized to the total non-purgeable Hg, during reactions with washed cells of *Geobacter sulfurreducens* PCA in the presence or absence of photo-irradiated DOM (0.21 mM DOC, as HA). The Hg-DOM solutions were pre-irradiated under either UV or solar light for different time periods before addition to the assay media (consisting of 10 mM phosphate buffer at pH 7.4 and 10 mM NaCl). Cells were added last at 10^8 cells mL⁻¹ and then incubated in the dark. The total Hg concentration was 12.5 nM. Error bars represent one standard deviation of triplicate samples.

Fig. 5. Energy dispersive X-ray spectra of HgS precipitates formed from photochemical reactions between Hg (0.1 mM) and (a) DOM (5 mM DOC, as HA) and (b) cysteine (0.5 mM) under solar light. The insets show the precipitated HgS particles in the vial and the corresponding scanning electron microscopy (SEM) images of the particles.

Fig. 1.

Fig. 2.

519 Fig.3.
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Fig. 4.

Fig. 5.