MERCURY CONCENTRATION AND SPECIATION IN SEDIMENTS

THROUGHOUT THE WATERSHED AFFECTED BY BLACK BUTTE MINE

IN OREGON

by

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THE WATERSHED AFFECTED BY BLACK BUTTE MINE IN OREGON

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Matthew Polizzotto

The history of mining in Oregon has left a legacy of contaminated and abandoned sites that threatens environmental and human health. The goal of this research is to better understand mercury (Hg) cycling within an Oregon watershed that is being contaminated by an abandoned mine. The chosen site is the Superfund site at Black Butte Mine, where contaminated sediment has washed down a series of streams that drain into Cottage Grove Reservoir. The specific objectives of this work are to determine how Hg sediment concentrations and forms (reactivity) change from the mine site to the reservoir and how these factors change within the different areas of the reservoir. Sediment sampling was conducted at the Black Butte Mine site, along drainage creeks and streams, and throughout the Cottage Grove Reservoir. Sediment composites were collected and used for total Hg and speciation analysis. The concentrations of mercury decreased with distance from the mine site to the reservoir. Mercury in sediment was found in progressively more reactive forms from the mine site to the reservoir. Within the reservoir, reactivity of Hg was higher in the floodplains that experience seasonal variations than the permanently inundated areas. The creek immediately adjacent to the mine site posed the most dangers to human and environmental health in terms of total Hg. Above the mine tailings and at the reservoir, conditions are supporting the transformation of inorganic Hg to organic Hg. This information, particularly the forms of mercury found, has significant implications for Hg bioaccumulation and the resulting safety of humans and species that live within the watershed. Data gaps in the US Environmental Protection Agency and Oregon Department of Environmental Quality's previous work on mercury analysis have been filled, which will inform future work at this site and allow them to target areas of most concern. A new site of organic mercury transformation has been identified, and future work can address this site. Additionally, mercury cycling in sediment is now better understood at this site, which will inform future research looking at mercury cycling at the watershed scale.

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<u>Introduction</u>

Mercury Contamination from Abandoned Mines in Oregon

Anthropogenic activity has tripled the amount of mercury found in the environment compared to background levels (Eckley & Hintelmann, 2006). Mines for Hg-bearing ore are significant sources of mercury for water systems downstream of these mines and can cause adverse effects for aquatic environments (Rimondi et al., 2012; Zhang et al., 2010). Abandoned mines are of particular concern because their abandoned wastes can have incredibly high levels of mercury and can greatly impact the local environment (Gray et al., 2012). Even years after mining stops, stream sediments can have uncharacteristically high levels of mercury downstream from these sites (Lecce & Pavlowsky, 2014; Rimondi et al., 2012). Aquatic systems downstream from these sites have the potential to transport both inorganic and organic forms of mercury (Zhang et al., 2010), and the different forms have different environmental impacts on the surrounding area.

Black Butte Mine: A Superfund Site Contaminating Cottage Grove with Mercury

Black Butte Mine is an abandoned mercury mine in Oregon, USA. Its operations stopped in 1969 (Curtis et al., 2013), and in 2010 it was added to the EPA's National Priorities List (Superfund) (Eckley et al., 2015). While in operation from the 1890s to the 1960s, this mine produced about 635,000 kg of Hg and over 200,000 m^3 of mine tailings, which were deposited on-site (Curtis et al., 2013; Eckley et al., 2015; Eckley et al., 2017). Cinnabar roasting is the principle source of local pollution at this site (Curtis et al., 2013) . The mine site is surrounded primarily by silviculture-managed coniferous forests and is about 15 km from Cottage Grove Reservoir, which was constructed in 1942 for flood control (Eckley et al., 2015; Eckley et al.,

2017). Studies have shown that this reservoir continues to be contaminated with Hg from Black Butte Mine (Curtis et al., 2013; Eckley et al., 2015; Eckley et al., 2017). Currently, the reservoir has a public health fish advisory since fish within the reservoir contain high levels of mercury (Curtis et al., 2013; Eckley et al., 2015).

Human and Environmental Health Concerns Due to Mercury

The reactivity and toxicity of mercury depend on the form in which it is found. Organic forms, mainly methylmercury, are much more bioavailable than inorganic forms. Thus, methylmercury and other organic forms are much more toxic to organisms and humans, and have the potential to bioaccumulate (Gray et al., 2012; Rimondi et al., 2012; Różański et al., 2016; Zhang et al., 2010). Methylmercury is the most common form of organic mercury found in the environment (Liu et al., 2012). Mercury is a neurotoxin and can cause a host of problems for the organisms and humans that are exposed to it, including deafness, blindness, promotion of cardiovascular disease, damage to the central nervous system, coordination problems, and sometimes death (Curtis et al., 2013; Eckley & Hintelmann, 2006; Gray et al., 2012; Rimondi et al., 2012). Organisms in the early stages of development are at particular risk when exposed to mercury (Curtis et al., 2013; Gray et al., 2012). Because of its ability to easily bioaccumulate (Eckley et al., 2017; Rimondi et al., 2012; Różański et al., 2016), humans are typically exposed to mercury through the consumption of fish (Eckley et al., 2015; Eckley & Hintelmann, 2006; Kim et al., 2004; Rimondi et al., 2012). Because of its accumulative nature, solubility, persistence, and toxicity when in organic forms, understanding the different forms of mercury found within a system is crucial for determining potential organism and human health impacts (Daye et al., 2015; Zhang et al., 2010).

Mercury Transformations in the Environment

The release and transport of Hg from its source is dependent on its species. Speciation determines solubility, and thus mobility, reactivity, and its potential to bioaccumulate (Kim et al., 2004). Because the species of Hg determines these qualities, species type, along with total concentration, determines the overall impact of Hg on environmental and human health. Previous studies of Hg concentrations in watersheds found that Hg concentrations were highest in the lake/reservoir sediments downstream from mercury mine sites, including our site (Curtis et al., 2013; Rimondi et al., 2012). Many studies have found that Hg concentration decreases as distance from the source site increases and that though mining has ceased, abandoned mine sites are still active sources of Hg pollution (Curtis et al., 2013; Gray et al., 2012; Lecce & Pavlowsky, 2014; Rimondi et al., 2012). This has also been found the case for the Black Butte Mine site (Curtis et al., 2013). However, the consistency of concentration trends begins to vary as distance from the site increases (Gray et al., 2012).

Mercury is typically found in three different states: elemental (Hg0), inorganic, and organic (Liu et al., 2012; Morel & Amyot, 1998). The typical transformation pathway is as follows: elemental mercury is oxidized to form inorganic mercury, which can then be methylated to form organic mercury (Daye et al., 2015; Liu et al., 2012; Rimondi et al., 2012). Inorganic and organic mercury can also be reduced to elemental mercury through photoreduction or microbial reduction (Morel & Amyot, 1998). Inorganic and elemental mercury are the typical Hg forms input into the environment, but organic mercury, particularly methylmercury (MeHg), is of most concern to humans though it accounts for only a small portion of total Hg found in the environment (Li & Cai, 2013). At mercury mines like Black Butte Mine, cinnabar (HgS) is heated to 600–700 °C, leading to the transformation of cinnabar to elemental mercury, which is

then collected. However, this process can be inefficient, leading to mine-wastes containing unconverted cinnabar, Hg0, and various Hg salts. This leftover Hg can be transformed to other forms of inorganic mercury, and then into methylmercury and other organic forms, as it is transported throughout the watershed and encounters different chemical and biological conditions. Methylmercury has also been found to form directly at these mine waste sites, providing another source of methylmercury for the environment (Rimondi et al., 2012).

In natural ecosystems, anaerobic microbial activity is the main driver of Hg methylation (Rimondi et al., 2012). These microbes are typically sulfur or iron reducing (Curtis et al., 2013; Eckley et al., 2015; Eckley & Hintelmann, 2006; Eckley et al., 2017; Gray et al., 2012). The speciation and methylation of Hg is affected by this microbial activity and the bioavailability of inorganic Hg to the microbes. Various factors influence the bioavailability of inorganic Hg and the activity of these microbes, such as the quality and quantity of organic matter in the sediment, the extent of anoxic conditions in the sediment, temperature, pH, redox conditions, and concentrations of sulfate, ferric iron, and other electron acceptors (Daye et al., 2015; Eckley et al., 2015; Eckley et al., 2017; Graham et al., 2013; Gray et al., 2012; Rimondi et al., 2012; Zhang et al., 2010). Other factors that affect Hg speciation include particle size, the ore roasting process used, and weathering. Distribution from the source alone was not found to affect mercury speciation (Kim et al., 2004).

Previous studies conducted by Eckley et al. at our site have also found some unique ways in which mercury is changing form throughout the watershed affected by Black Butte Mine. First, water-level fluctuations present at Cottage Grove Reservoir increase methylation of mercury. This is because when water levels drop, sediments are exposed to air and reduced compounds are re-oxidized, creating a new source of electron-accepting compounds for microbes

to use once the areas are inundated again. This increased microbial activity then leads to increased methylation. Water-level fluctuations may also lead to partitioning of Hg to the aqueous phase, which is more bioavailable for microbes. Furthermore, when the area is re-flooded, there is an influx of organic material, sulfates, and Hg, and anoxic conditions are developed, which allows for more microbial activity and methylation. Water fluctuations also promote the cycling of sulfide to sulfate, further promoting microbial activity. Thus, areas that experience water-level fluctuations will have more methylmercury than areas that are permanently inundated (Eckley et al., 2015; Eckley et al., 2017).

<u>Research Questions, Project Goals, and Objectives</u>

The history of mining in Oregon has led to a legacy of contaminated and abandoned sites that threatens environmental and human health. My research goal was to better understand mercury (Hg) cycling within an Oregon watershed that is being contaminated by an abandoned mine. The chosen site was the Superfund site at Black Butte Mine, where contaminated sediment has washed down a series of streams that drain into Cottage Grove Reservoir. I asked three main research questions: How do Hg concentrations change in sediment from the mine site to the reservoir? How do the forms and reactivity of Hg change from the mine site to the reservoir? Do form and concentration change in different areas of the reservoir? To answer these questions, I established two objectives: first, measure total concentrations of Hg in sediments that have accumulated along the watershed from Black Butte Mine to Cottage Grove Reservoir, and second, quantify the Hg species in sediments along the watershed from the mine source to the reservoir. Although the Department of Environmental Quality (DEQ) and the Environmental Protection Agency (EPA) have conducted preliminary Hg analyses in the area, no studies have

looked at how the forms and concentrations of mercury change in the sediment from the mine site to the reservoir. This information, particularly the forms of mercury found, has significant implications for Hg bioaccumulation and the resulting safety of humans and species that live within the watershed.

To attempt to answer my research questions, I tested the following hypotheses concerning Hg in sediment within the watershed:

- Concentrations of mercury will be highest at the mine site, decrease in concentration along the river channels, and then increase within the reservoir as Hg is deposited and accumulates.

- Hg in sediment will become progressively more labile (i.e. reactive) from the mine site to the reservoir.

- Within the reservoir, lability and concentrations of Hg will be higher in the floodplains that experience seasonal variations than the permanently inundated areas.

To test these hypotheses, I worked in partnership with members of DEQ, US EPA, and UO Earth Sciences Department.

Methods

Sample Collection

Sampling was conducted at the Black Butte Mine site, along Furnace Creek, Garoutte Creek, the Coast Fork of the Willamette River, and throughout Cottage Grove Reservoir (Figure 1). Sediment composites were collected by taking five sub-samples at each site with stainless-steel spoons at depths of 1-10 cm. The sediment was mixed thoroughly and then split in half, with each half stored in a separate plastic vial. The first half of the sediment was used for total

Hg analysis and the second half was used for speciation analysis. There was one instance where the sediment composite was collected at two nearby sites due to lack of sediment. Roughly 50-100g of sediment per mixed sample was collected before splitting the sample in half for each analysis. The mixed and split samples were stored in vials held in a cooler and then a refrigerator at $4^{\circ}C$ until analyzed. The samples meant for speciation analysis were moved to the freezer, once available, before analysis. This sampling method is based off of standard protocol for studies conducted by DEQ and the EPA at this site (Curtis et al., 2013; Eckley et al., 2015; Eckley et al., 2017).
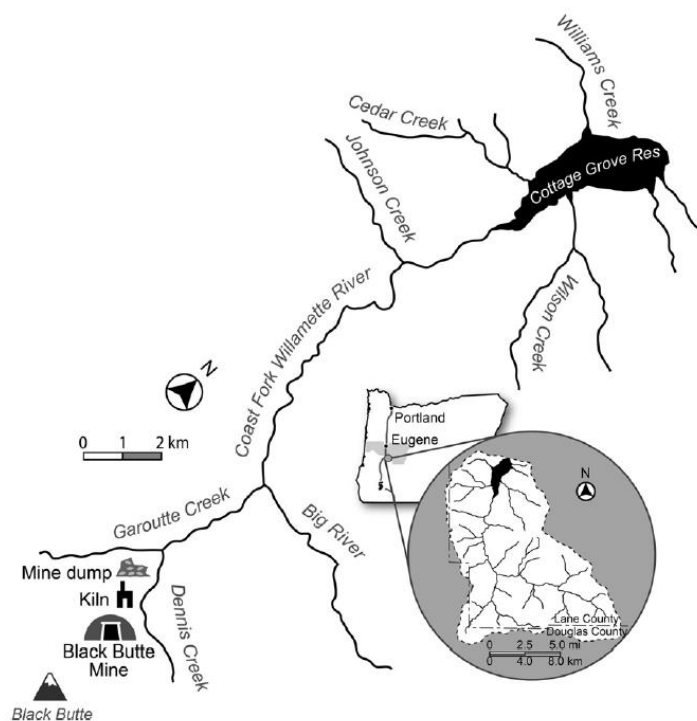

Figure 1: Map of watershed affected by Black Butte Mine, including the mine site and Cottage Grove Reservoir (Curtis et al., 2013).

In total, 29 samples were collected. Twelve samples were collected along Furnace and Garoutte Creek, nine were collected along the Coast Fork of the Willamette, and eight were collected within the Cottage Grove Reservoir (Figure 2). Creek and river sediment samples were

collected at spots in stream bends where ample sediment had accumulated. Within the Cottage

Grove Reservoir, samples were collected from areas that are seasonally flooded and always

flooded. Three additional samples were collected for methylmercury analysis, with each site at a

location already sampled for total mercury analysis.

Figure 2: Map of sampling sites. Each point represents a sampling site.

Analytical Approaches

To analyze total Hg concentrations, sediment samples were decanted, then a Lumex total

Hg analyzer was used to determine Hg concentrations via EPA method 7473 (US EPA, 2007).

Ten samples contained mercury outside of the linear range of this method, so these samples were

sent to Brooks Applied Labs for analysis. Samples were at room temperature at time of analysis.

Sediments were directly analyzed, and dry weight was determined in triplicate by drying ~1 g of sediment in an oven at 100°C for 24 hours and then weighing to determine lost moisture content.

A five-step sequential extraction procedure that targets different sediment-bound Hg phases was conducted to determine Hg forms and reactivity in the sediment. Seven of the collected samples were chosen for analysis. The extractants for each step were: deionized water (F1, water-soluble), a synthetic "stomach acid" (F2, weak acid soluble), 1M potassium hydroxide solution (F3, organo-chelated), 12M nitric acid (F4, elemental Hg), and aqua regia (F5, mercuric sulfide). Each step determined how much Hg of each behavioral class was in the sediment sample. (Bloom et al., 2003; Brooks Applied Labs, 2017). To determine the total concentration of each Hg species for Table 1, which were initially reported in wet weight, total percent solids was used to convert to dry weight. For samples FC-01, FC-09, and CG-01, total percent solids were determined using the sample and then directly converted to dry weight. For all other samples, an average of calculated total percent solids was used to convert to dry weight, as the total percent solids for each of these samples were unavailable. Sequential extractions and analyses of the selected samples were performed by Brooks Applied Labs, a third-party lab in Washington state that has previously done work for the EPA at Black Butte Mine, using a modified version of EPA method 3200 (US EPA, 2014).

Methylmercury was analyzed using solvent extraction followed by back extraction into water (Bloom et al., 1997), then quantification by gas-chromatography cold vapor atomic fluorescence spectrometry using EPA method 1630 (US EPA, 2001). Soil samples (0.05 g) were leached for one hour in 5 mL of 18% (w/v) potassium bromide + 5% (v/v) H_2SO_4, and 1 mL of 1 M copper sulfate solution. Then 10 mL of dichloromethane (CH_2Cl_2) was added, and samples were shaken for one hour, then centrifuged (4000 RPM, 30 min), and phases separated

(Whatman 1-PS). The CH_2Cl_2 layer was evaporated by heating vials in a water bath (60-70°C, 1.5 hours), and the final volume was raised to 40 mL using Milli-Q H_2O. Following solvent extraction and back extraction into water, MeHg was quantified using gas chromatography (GC)-CVAFS, including ethylation with sodium tetraethylborate, following U.S. EPA Method 1630 (Model-III Detector, Brooks Rand Instruments, Seattle, WA).

Results

Total Concentration of Mercury

Watershed sediment Hg concentrations with distance from the mine site are shown in Figure 3. Lines separate the boundaries between creeks, streams, and the reservoir. Starting at the mine site, concentrations of mercury were very high, compared to the rest of the sediment samples. After the first boundary was passed, from Furnace Creek to Garoutte Creek, there was a steep drop-off in sediment Hg concentrations. Concentrations remained relatively steady until Cottage Grove Reservoir, where they began to decrease again. Samples collected at Furnace Creek were clustered but were much higher in concentration than all other samples collected. Between Furnace Creek and the reservoir, Hg sediment concentrations varied, with no obvious trend. At the reservoir, sample concentration decreased with a steady downward trend.
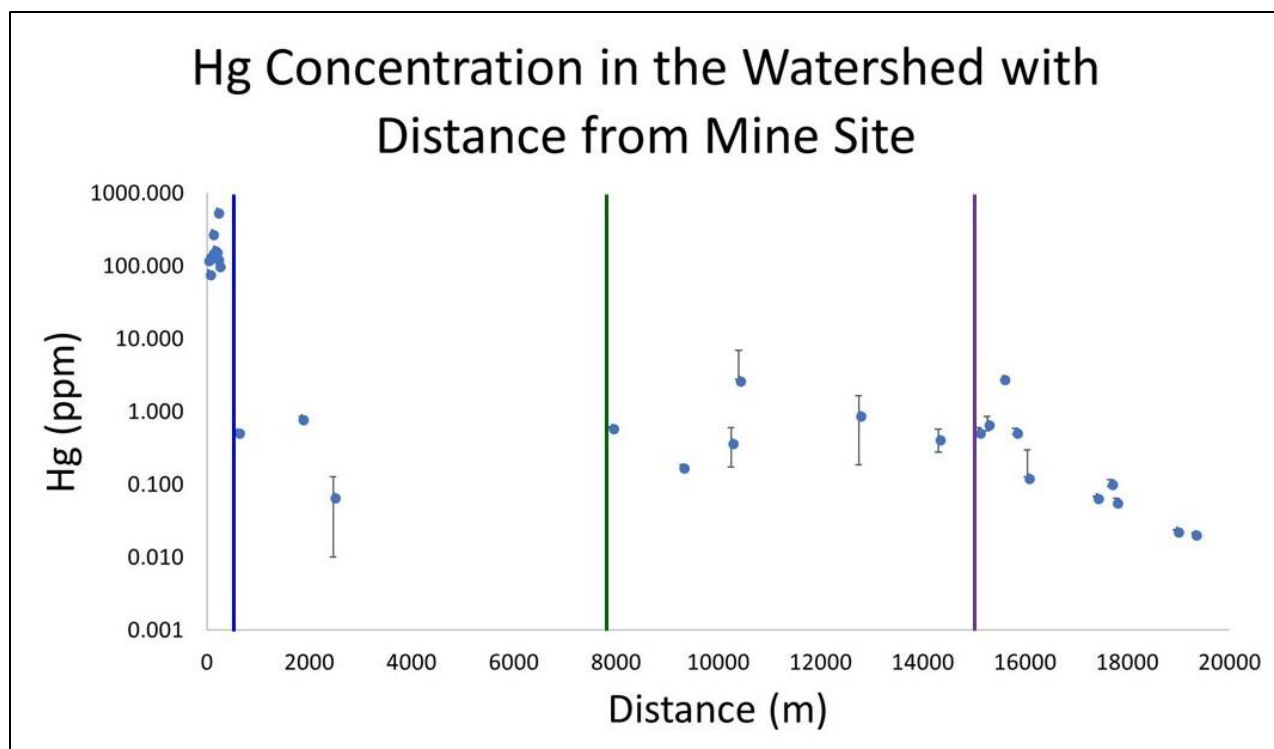
Hg Concentration in the Watershed with Distance from Mine Site

Figure 3: Concentration of mercury in sediment from the Black Butte Mine site (condenser, 0 m), through the watershed, to Cottage Grove Reservoir (> 15,000 m). Error bars represent standard deviation of triplicate measurements. Blue line represents boundary between Furnace and Garoutte Creek. Green line represents boundary between Garoutte Creek and the Coast Fork Willamette. Purple line represents boundary between the Coast Fork Willamette and Cottage Grove Reservoir.

Box and whisker plots, comparing the average Hg concentration in sediment found in three main water bodies, are shown in Figure 4. Sediment from Garoutte Creek has been grouped with sediment from the Coast Fork Willamette. The overall trend was a decrease in average mercury concentration as sediments from the water bodies were farther from the pollution source. Concentrations of mercury were significantly higher in Furnace Creek (mean= 176.53 ppm) than all other streams (mean= 0.72 ppm) and Cottage Grove Reservoir (mean=0.48 ppm) ($p = 0.000173$); significance was calculated with one-way ANOVA. Although Hg concentrations of sediment from the Coast Fork Willamette were generally higher than those from Cottage Grove Reservoir, differences between the sample groups were not significant.
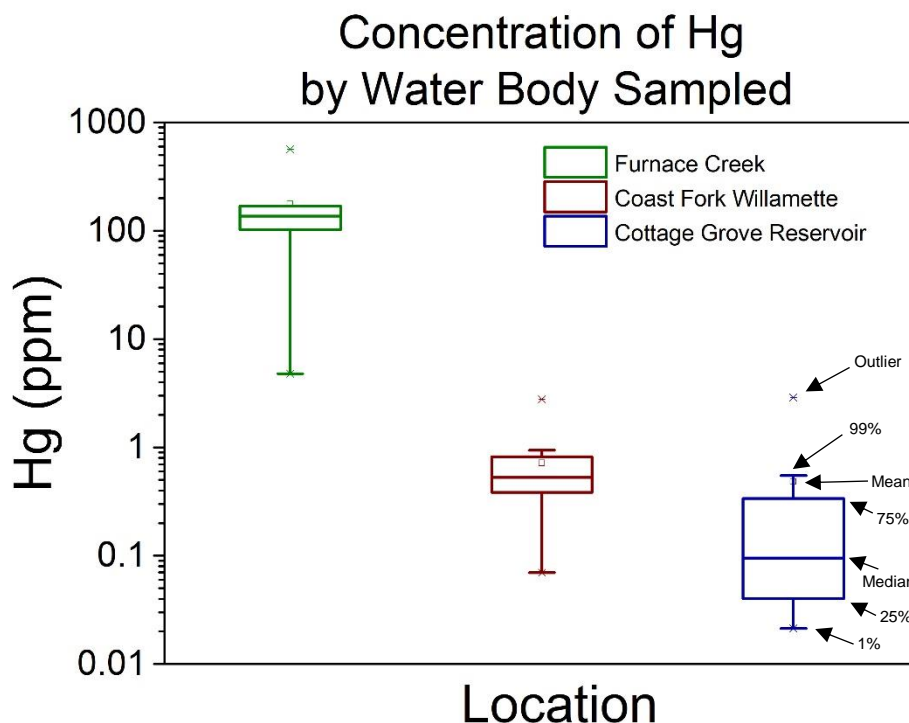
Figure 4: Average Hg concentrations of sediments among the three major water bodies sampled, each a major section of the watershed. Furnace Creek (n=10) is the small stream that runs through the mine site, Coast Fork Willamette (n=11) is the river that connects Furnace Creek to the reservoir, and Cottage Grove Reservoir (n=8) is the reservoir impacted by the mercury pollution coming from the mine site.

Mercury Speciation

The Hg species concentrations for each sediment sample are shown in Table 1. Because some mercury may be lost during the sequential extraction process, the five-step sequential extraction method is better used to show the percent of total mercury for each behavioral class; however, the species concentration table is included here to emphasize that each sample had varying degrees of total mercury. The raw data of mercury fraction percentages for each behavioral class can be seen in Table 2. For more details on the specific Hg species for each class, see Figure 5. For a graphical representation of Table 2, see Figure 6.

Table 1: Data of total mercury for each behavioral class (F1-F5) by sample. F1 is the most labile, each class after is progressively less labile, and F5 is the least labile. Samples are ordered from closest to the mine site to farthest from the mine site. See details of each behavioral class below (Figure 5).

Sample ID	F1	F2	F3	F4	F5	Sum	Position in Watershed	Distance from Mine Site (m)
FC-00	0.038	0.0031	3.2	0.53	0.079	3.8	Furnace Creek	0
FC-01	1.1	12	3.8	40	39	95	Furnace Creek	0
FC-09	0.43	3.9	4.2	76	27	111	Furnace Creek	219.3
GC-02	0.0077	0.0012	0.44	0.18	0.061	0.69	Garoutte Creek	1854.3
W-02	0.0012	0.00069	0.15	0.047	0.21	0.40	Coast Fork Willamette	9302.3
CG-01	0.014	0.0011	1.9	0.23	0.65	2.8	Cottage Grove Reservoir	15570.3
CG-06	0.00037	0.00029	0.063	0.0056	0.00079	0.070	Cottage Grove Reservoir	17395.3

Table 2: Mercury fraction percentages for each behavioral class (F1-F5). F1 is the most labile, each class after is progressively less labile, and F5 is the least labile. Samples are ordered from closest to the mine site to farthest from the mine site. See details of each behavioral class below (Figure 5).

Sample	F1	F2	F3	F4	F5
	% of sum	% of sum	% of sum	% of sum	% of sum
FC-00	1.0%	0.1%	83.0%	13.9%	2.1%
FC-01	1.1%	12.1%	4.0%	42.3%	40.5%
FC-09	0.4%	3.5%	3.8%	68.2%	24.1%
GC-02	1.1%	0.2%	64.1%	25.8%	8.8%
W-02	0.3%	0.2%	36.5%	11.7%	51.3%
CG-01	0.5%	0.0%	67.9%	8.1%	23.4%
CG-06	0.5%	0.4%	89.9%	8.0%	1.1%

	Mercury Classification	Primary Compounds Extracted
F1	Water-soluble, i.e. salts	$HgCl_2$
F2	Weak acid-soluble/ "stomach acid" soluble	$HgSO_4$ HgO
F3	Organo-complexed	Hg-humics Hg_2Cl_2 CH_3Hg (MeHg)
F4	Strongly-complexed	mineral lattice bound Hg_2Cl_2 Hg^0 (liquid elemental)
F5	Mineral-bound	HgS (cinnabar) m-HgS (meta-cinnabar) HgSe (amalgam) HgAu (amalgam)

Figure 5: Detailed description of the forms of mercury found in each behavioral class (F1-F5) (Brooks Applied Labs).
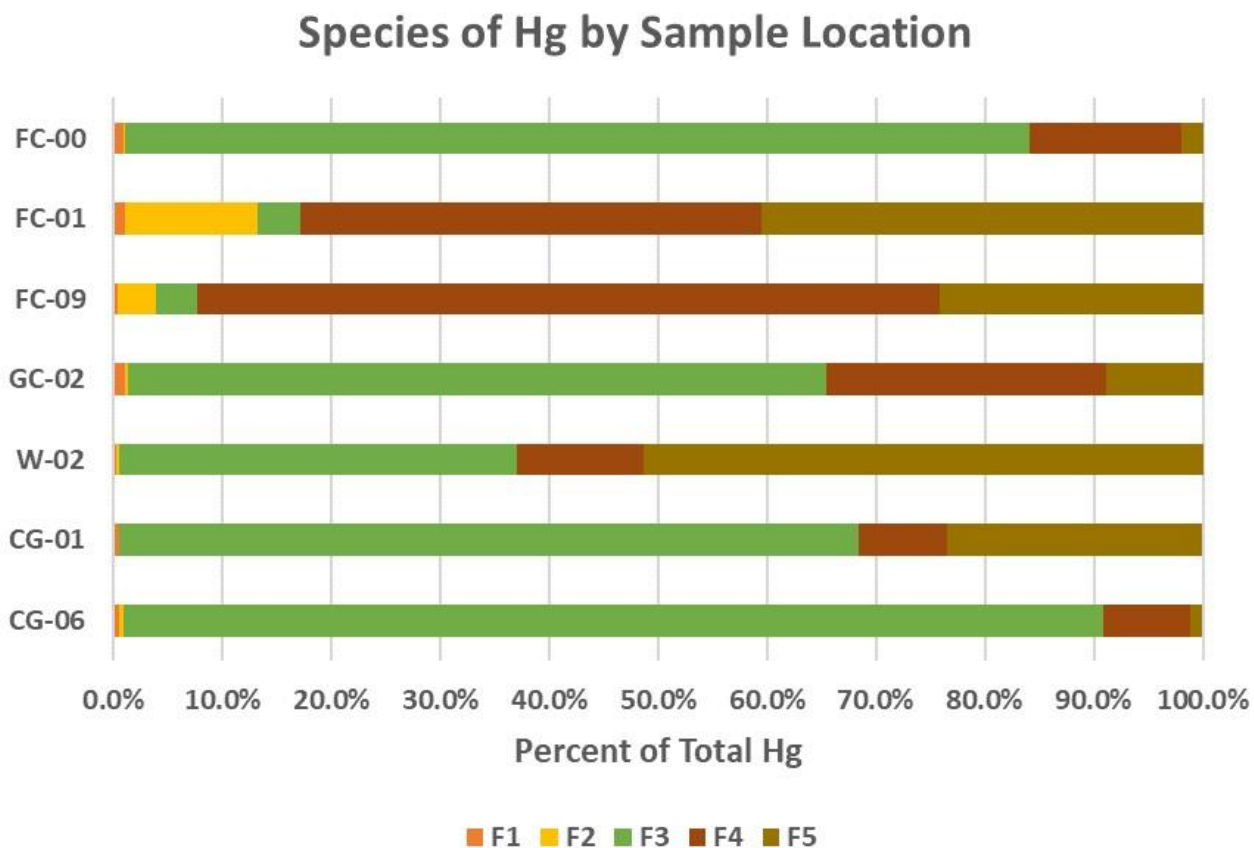
Species of Hg by Sample Location

Figure 6: Mercury fraction percentages for each behavioral class (F1-F5) by sample location. Samples are ordered from closest to the mine site (top) to farthest from the mine site (bottom).

More labile organic Hg species, F3, took up the greatest percentage of Hg in sediments above the condenser (FC-00). The least labile inorganic Hg species, F4 and F5, which are indicative of raw mine tailings, took up a greater percentage of the total mercury at Furnace Creek. More labile species, F3, took up the greatest percentage in Garoutte Creek. The least labile species, F4 and F5, took up a greater percentage of the total mercury at the Coast Fork Willamette. More labile species, F3, took up the greatest percentage in Cottage Grove Reservoir. For all samples, there was very little species from class F1 and F2, the most labile inorganic species. These took up the greatest percentage in sample FC-01, from Furnace Creek, but overall still took up a comparatively small percentage of the total mercury.
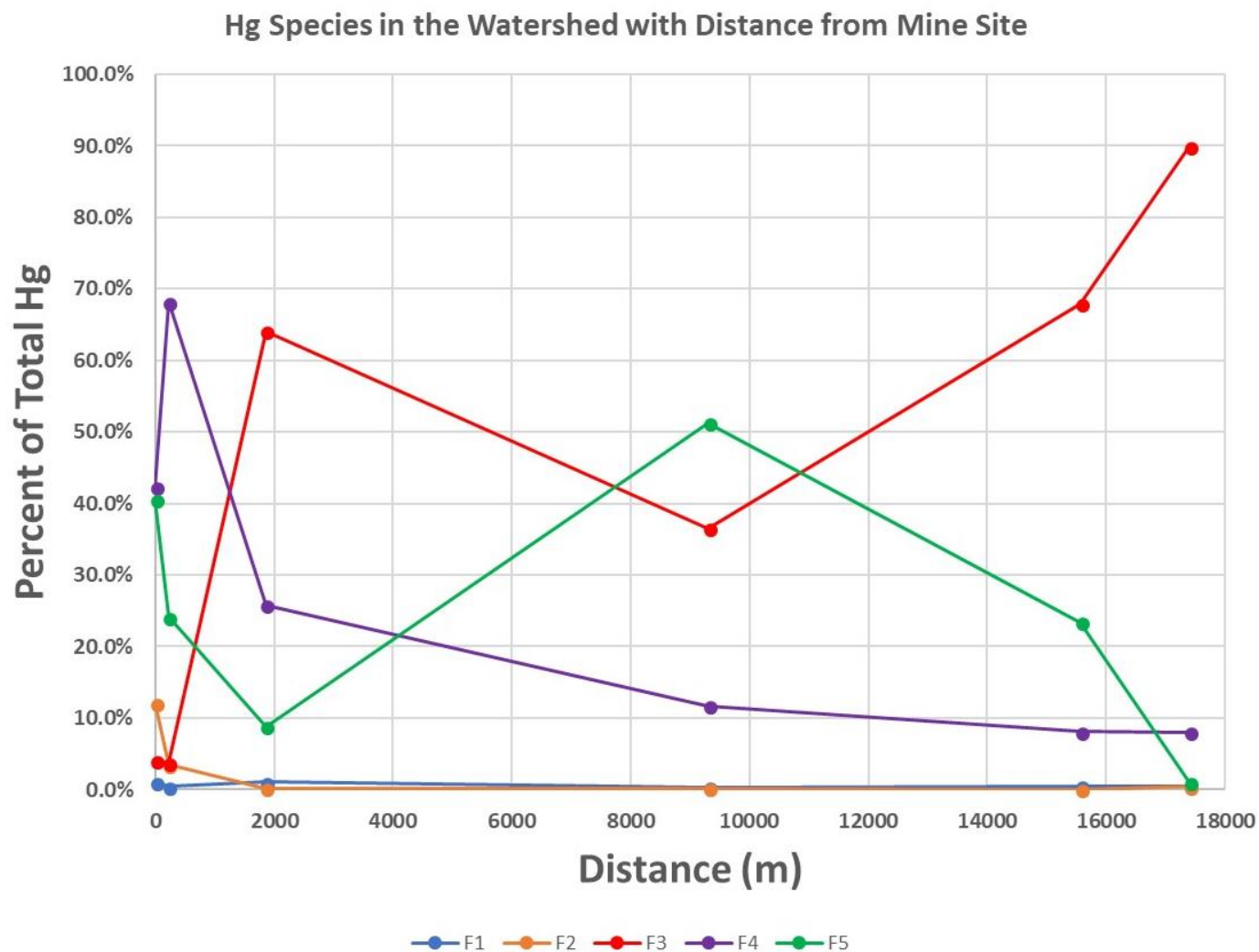
Hg Species in the Watershed with Distance from Mine Site

Figure 7: Mercury fraction percentages for each behavioral class (F1-F5) from the Black Butte Mine site (condenser, 0 m), through the watershed, to Cottage Grove Reservoir (> 15,000 m).

Mercury fraction percentages for each behavioral class as they change throughout the watershed can be seen in Figure 7. In general, behavioral classes F4 and F5 went down with distance from the mine site and F3 went up with distance from the mine site. Class F5 and F4 started at 40.5% and 42.3% at Furnace Creek and decreased to 1.1% and 8.0% at Cottage Grove Reservoir, respectively, whereas class F3 started at 4.0% at Furnace Creek and increased to 89.9% at Cottage Grove Reservoir. Behavioral class F1 remained low throughout the watershed (mean= 0.7%). F2 was slightly elevated at the mine site, but quickly reduced to almost nothing

throughout the rest of the watershed. Overall, F2 remained comparatively low throughout the watershed (mean= 2.4%). F3 started off low, increased greatly, decreased, and then greatly increased again. The point of major increase was in Garoutte Creek. However, the overall trend as it moved throughout the watershed was increasing as it progressed from the mine site to the reservoir. F4 had a spike at the lower Furnace Creek sample, but otherwise decreased as it moved from the mine site to the reservoir. F5 decreased at first, saw a spike at the Coast Fork Willamette, and then decreased again, reaching its lowest point at the second site in Cottage Grove Reservoir.

Methylmercury

Methylmercury sediment concentration and MeHg percent of total Hg sediment concentration by location are shown in Figure 8. Percent MeHg, shown in green, was highest above the condenser, slightly lower in Cottage Grove Reservoir, and lowest below the condenser. Percent MeHg below the condenser (0.011%) was considerably lower than the two other sites (0.11%, 0.083%). MeHg concentration, shown in red, was lowest in the reservoir (1.6×10^{-5} ppm). Sites above and below the condenser had similar MeHg concentrations (1.7×10^{-3} ppm, 2.3×10^{-3} ppm, respectively).
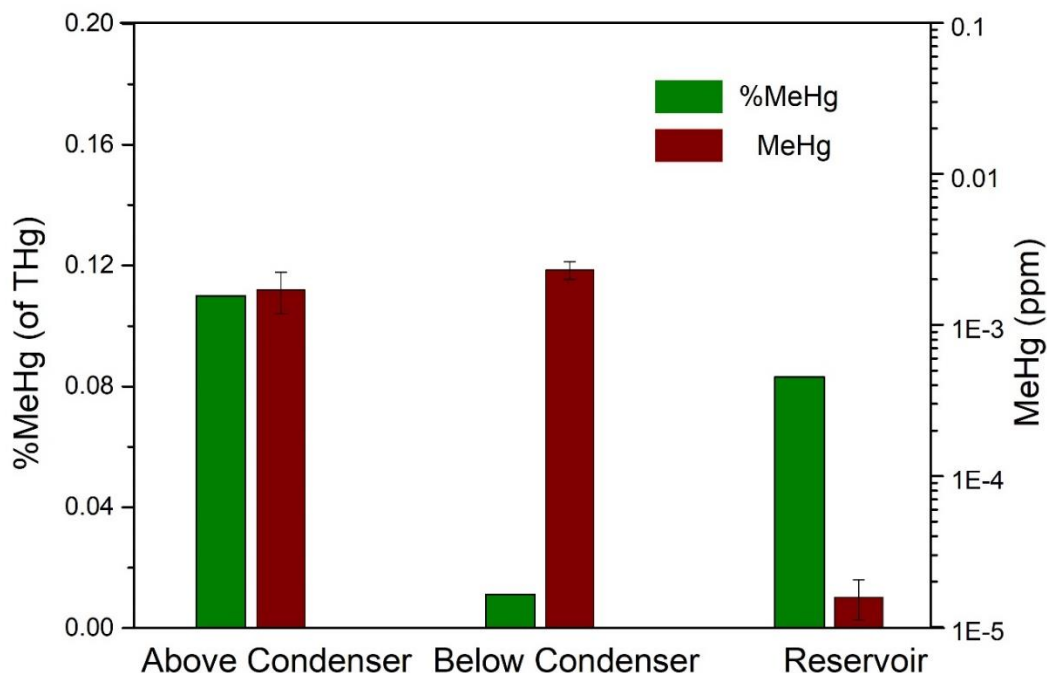
Methylation of Hg

Figure 8: Methylation of mercury at three sites by percent of total mercury in the sediments (green bars) and absolute methylmercury concentration of sediments (red bars). MeHg is methylmercury and THg is total mercury. Error bars represent standard deviation of results from triplicate measurements.

Discussion

Total Concentration of Mercury

As predicted, sediments from Furnace Creek have the highest total concentrations of mercury (mean= 176.53 ppm). Previous studies conducted by the DEQ found lower ranges of total concentration at Furnace Creek (70.2-173 ppm). Mercury concentrations of sediments from Garoutte Creek (mean= 0.47 ppm) were in the lower end of the range found in previous studies, but past studies had a very large range of Hg concentrations (0.34-159 ppm) (Thoms, 2008). Mercury concentrations of sediments from the Coast Fork Willamette were slightly higher than a sample taken during a previous study (mean= 0.82 ppm, compared to 0.30 ppm), and now that concentrations throughout the Coast Fork Willamette have been quantified, future studies can use this mean as a comparison rather than the single sample value. Mercury concentrations of

sediments from Cottage Grove Reservoir (mean= 0.48 ppm) were slightly lower than expected compared to previous studies conducted at this site (0.68-3.6 ppm) (Thoms, 2008; US EPA, 2012).

Typical background levels of total mercury in this region are 8 ppm (US EPA, 2012), thus the only samples exceeding expected background levels were collected along Furnace Creek. This background level was established by analyzing native soil at the tops of ridges that surround and face Black Butte Mine. In a review of the literature, background levels of the most common form of mercury (HgII) were found to be 0.05-0.5 ppm (Liu et al., 2012), so the mean concentration of samples taken from Furnace Creek and the Coast Fork Willamette were above this background level (FC= 176.53 ppm, W=0.82 ppm). Because we looked at total, and this background range focused on HgII, this only gives us an estimate that total mercury is higher in these parts of the watershed. Overall, Furnace Creek contained sediments with total mercury above typical regional background levels, but the rest of the watershed did not.

Once the sediment exits Furnace Creek, mercury concentration remains about the same until it reaches Cottage Grove Reservoir, where it begins decreasing, similar to the trend we had hypothesized. However, Hg in sediment is not accumulating at the reservoir like we expected. After leaving Furnace Creek, contaminated sediment might be diluted with other sediment, leading to low mercury concentrations further down the watershed until it reaches the reservoir. This may explain the steep drop-off in total concentration once the sediment exits Furnace Creek. Furthermore, sediments are diluted once again as they reach the reservoir and move even father from the source. This would explain the downward trend in total concentration throughout the reservoir. When comparing the Cottage Grove Reservoir samples of permanently inundated areas (n= 5) to those from seasonally inundated areas (n= 4), we found no significant differences

between the two groups. Previous studies found that methylmercury concentrations varied between these two groups (Eckley et al., 2015; Eckley et al., 2017), but this does not appear to be the case for total mercury concentrations.

Mercury Speciation

Because speciation of mercury throughout the watershed has not been looked at in previous studies, there are no studies to compare our results to. However, in a previous study at another site, it was found that distribution from the source alone was not found to affect mercury speciation (Kim et al., 2004). This does not seem to be the case for our site. While there is some variability, discussed in the results, overall, we did see trends in the behavioral species classes F3, F4, and F5. Classes F1 and F2 were low in sediments throughout the watershed, so trends could not be determined. While more investigation is needed, our results suggest that there are qualitative trends in the different Hg behavioral classes.

The overall percentage trends of classes F3, F4, and F5 did match what we had hypothesized. As we expected, the more labile species class F3 started low below the condenser (FC-01) and increased throughout the watershed, its greatest point at Cottage Grove Reservoir. F3 had a higher percentage than expected in Garoutte Creek, indicating that methylation may be occurring at this site. Otherwise, the trend followed what we expected. Another unexpected result with F3 was above the condenser (FC-00). There was a very large percentage of F3, which includes methylmercury, above the condenser, indicating methylation is occurring here too. This is further supported by our methylmercury data. Conditions above the condenser must be promoting methylation, changing less labile inorganic species into organic ones. As expected, F4 started highest in Furnace Creek and decreased throughout the watershed, reaching its lowest point at the reservoir. Class F5 overall followed the trend we expected, decreasing throughout the

watershed, but it did increase unexpectedly in the Coast Fork Willamette before continuing to decrease. Because mercury cycling is still not well understood, it is unclear as to why this is the case. One potential explanation is that conditions in the Coast Fork Willamette caused species from class F4 to transform back to F5, creating the F5 class spike. But considering both F4 and F5 together as one group, as they are both the classes expected close to the mine site, the trend was what we expected.

The Cottage Grove Reservoir sample from the permanently inundated area (CG-01) had a greater percentage of F5 species than the sample from the seasonally inundated area (CG-06). Both samples had about an equal percentage of F4, F1, and F2. The seasonally inundated sample had a higher percentage of F3 than the permanently inundated sample. Thus, these two samples matched our predictions that the seasonally inundated sample would have a greater amount of labile species. While the sample size is small, this does suggest that Hg speciation beyond only methylmercury is affected by water-level fluctuations, and so this should be investigated further in future studies.

Methylmercury

Total methylmercury at Cottage Grove Reservoir (1.58×10^{-5} ppm) was much lower than what was previously found during another study at this site (2.20×10^{-3} - 2.60×10^{-3} ppm) (Eckley et al., 2015). It was also much lower than a national scale lake survey, which found a mean of 1.87×10^{-3} ppm MeHg (Krabbenhoft et al., 1999). Total methylmercury below the condenser (2.3×10^{-3} ppm), which is along Furnace Creek, was much lower than a sample collected in a previous study of this site (0.0127 ppm). Both this study and the previous study found elevated MeHg concentrations above and below the condenser, exceeding DEQ soil screening levels for plants, though our concentrations were lower (US EPA, 2012). Percent

methylmercury (%MeHg) at Cottage Grove Reservoir was slightly lower than results from a previous study at this site (.08%, our study, compared to 0.1%) (US EPA, 2012). There are no previous studies to compare %MeHg above or below the condenser, but above the condenser (0.11%) was within the generally expected range (0.1-10%) (Liu et al., 2012), while below the condenser (0.01%) was not.

Conditions in the reservoir and above the condenser appear to be promoting methylation; more methylation of mercury is occurring above the condenser and in the reservoir as indicated by the higher %MeHg at these sites. Since there is more total methylmercury above and below the condenser, methylmercury appears to be accumulating above and below the condenser after being methylated above it. Also, rates of methylation may have decreased since the previous study was conducted. These results were not what we predicted. We did not expect to find much, if any, methylmercury above the condenser as it is very labile. There must be conditions at this site that are promoting microbial activity and therefore increasing methylation rates.

There are some limitations to this part of our study. The goal of analyzing methylation was exploratory, so our sample size was small. Furthermore, methylmercury concentrations can be highly spatially and temporally variable (Eckley et al., 2015). Thus, more research is needed before we can confirm these preliminary results and establish if methylation rates have in fact decreased.

Significance/Implications for Environmental and Human Health

Table 3: Various recommendations for mercury levels from DEQ, US EPA, and MacDonald, Ingersoll, & Berger (MacDonald et al., 2000; Oregon Department of Environmental Quality, 1998; US EPA, 2012). 1: MacDonald, Ingersoll, & Berger.		
Recommendation	Classification	Total Mercury (ppm)
Environmental Protection Agency Region 9 PRGs	Residential Soil	23
Environmental Protection Agency Region 9 PRGs	Industrial Soil	310
Oregon DEQ Maximum Allowable Soil Concentrations	Residential Soil	80
Oregon DEQ Maximum Allowable Soil Concentrations	Industrial Soil	600
Oregon DEQ Level II Screening Level Values- Plants	Soil	0.3
Oregon DEQ Level II Screening Level Values- Invertebrates	Soil	0.1
Oregon DEQ Level II Screening Level Values- Birds	Soil	1.5
Oregon DEQ Level II Screening Level Values- Mammals	Soil	73
Probable Effect Concentration (PEC): Sediment-dwelling Organisms[1]	Sediment	1.060
Recommendation	Classification	Methylmercury (ppm)
Environmental Protection Agency Region 9 PRGs	Residential Soil	6.1
Environmental Protection Agency Region 9 PRGs	Industrial Soil	6.2
Oregon DEQ Level II Screening Level Values- Plants	Soil	0.0002
Oregon DEQ Level II Screening Level Values- Invertebrates	Soil	----
Oregon DEQ Level II Screening Level Values- Birds	Soil	0.025
Oregon DEQ Level II Screening Level Values- Mammals	Soil	4

Total Concentration of Mercury

Average Hg concentrations at Furnace Creek (176.53 ppm) was not above the concentrations recommended for industrial soils by the US EPA or DEQ, but was above the concentrations recommended for residential soils by the US EPA and DEQ (Table 3). It was also above the probable effect concentration established by MacDonald et al., which established Hg concentration levels that are likely to cause observable harmful effects to sediment-dwelling organisms (Table 3). Furthermore, average sediment Hg concentration at Furnace Creek was above DEQ recommendation levels for plants, invertebrates, birds, and mammals (Table 3), suggesting that sediment throughout Furnace Creek has the potential to harm wildlife. Though

lower than the industrial soil recommendations, environmental sediments do not fall clearly in either the industrial or residential category. Taking all classifications into consideration, Furnace Creek sediment is a potential risk for environmental and human health.

Average Hg concentrations at Garoutte Creek (0.47 ppm) and the Coast Fork Willamette (0.82 ppm) were below all Hg concentration recommendations except for DEQ's recommendations for plants and invertebrates (Table 3). Thus, sediments in Garoutte Creek and the Coast Fork Willamette, compared to Furnace Creek, pose little risk to environmental health and likely no risk to human health. Average Hg concentrations at Cottage Grove Reservoir (0.48 ppm) were also below all Hg concentration recommendations except for DEQ's recommendations for plants and invertebrates (Table 3). Thus, in terms of total concentration of Hg, sediments in Cottage Grove Reservoir pose little risk to environmental health and likely no risk to human health.

Mercury Speciation

Behavioral class F3 contains the most toxic forms of mercury (organic), including methylmercury. Above the condenser (FC-00), Garoutte Creek, and both samples from Cottage Grove Reservoir had class F3 as the highest percentage of total mercury. These areas appear to be potentially problematic to human health and the environment, as they may be sources of methylmercury or other toxic organic forms of mercury. Future studies should investigate the amount of total methylmercury at these sites to determine how much methylmercury is being produced. The other sites, Furnace Creek and the Coast Fork Willamette, appear to be less problematic in terms of the types of mercury present. Overall, the speciation results suggest that there could be multiple areas within the watershed where organic mercury is produced.

Methylmercury

Average total MeHg above the condenser (0.0017 ppm) was below all concentration recommendations except for DEQ's recommendations for plants. There is no established recommendation for invertebrates (Table 3). Thus, sediments above the condenser appear to pose little risk to environmental health and likely no risk to human health in terms of total MeHg present. However, based on percent of total Hg (Figure 8) and the speciation results (Figure 6), it appears that methylation is occurring at this site and could be depositing methylmercury further down the reservoir, thus posing a potential environmental and human health risk due to the bioavailability of methylmercury.

Average total MeHg below the condenser (0.0023 ppm) is also below all concentration recommendations except for DEQ's recommendations for plants (Table 3). Thus, sediments above the condenser also appear to pose little risk to environmental health and likely no risk to human health in terms of total MeHg present. %MeHg is also low here, further suggesting sediments at this site pose little risk to environmental or human health in terms of methylmercury.

Average total MeHg at Cottage Grove Reservoir (1.58×10^{-5} ppm) was below all concentration recommendations (Table 3). Thus, sediments in Cottage Grove Reservoir appear to pose no risk to environmental and human health in terms of total MeHg present. However, based on percent of total Hg (Figure 8), methylation appears to be occurring in the reservoir. Furthermore, there is already a fish advisory for this reservoir (Curtis et al., 2013; Eckley et al., 2015). Thus, even though we found low levels of methylmercury and lower levels of percent total mercury, methylation rates are still high enough to pose a threat to human and

environmental health due to the bioaccumulation of methylmercury. But, as mentioned previously, it is important to note the limitations of this exploratory part of our study.

<u>Future Directions</u>

Since our methylation analysis was mostly exploratory, more research will need to be conducted to fully understand how Hg methylation is impacting this watershed. This includes increasing the sample size and sampling in more areas. While we only analyzed one sample at the reservoir and two at Furnace Creek, studies in the future could collect samples all along the watershed, just as we did to analyze total concentration. More research is also needed to determine why methylation is occurring at the waste site, specifically what conditions at this site are promoting methylation.

Total mercury concentration decreased along the watershed more quickly than we expected. One possible explanation is that the contaminated sediment from Furnace Creek is being diluted with sediment from other tributaries and the Coast Fork Willamette as it moves through the watershed. Analyzing the sediment load of the Coast Fork Willamette and tributaries could confirm if this is the case and help explain our results.

The US EPA plans to continue remediation efforts at this site in a few months (Bryn Thoms, personal communications). Once this remediation occurs, repeating our study could inform us if the remediation efforts are having an impact and successfully controlling the mercury pollution experienced throughout the watershed and at Cottage Grove Reservoir. This would not only be useful within the context of the Black Butte Mine Superfund site, but it would also help establish if the remediation methods used are broadly effective.

As of now, there are many data gaps in the scientific literature involving mercury cycling, particularly at this scale. Furthermore, while there are many studies focusing on methylation of mercury due to its toxicity, much less is understood about the cycling of elemental and inorganic mercury within sediments. While this research will begin to address these data gaps, much more work is needed before the scientific community will fully understand how mercury transforms in the environment, and how these transformations impact the environment and human health.

Conclusions

This study established the trends of Hg in sediments throughout the watershed impacted by Black Butte Mine in Oregon. Total concentrations of Hg in sediments decreased from the mine site to Cottage Grove Reservoir. Mercury in sediments was progressively more reactive from the mine site to Cottage Grove Reservoir. This study also identified an area of the mine site with conditions that appear to promote the transformation of inorganic to organic mercury and methylation. The primary areas of the watershed that are human and environmental health concerns are Furnace Creek due to its elevated mercury levels in sediment, and above the condenser and at Cottage Grove Reservoir due to the increased rates of inorganic to organic Hg transformation. This study has addressed some of the data gaps at this site, which the EPA and DEQ can use to further inform their remediation work at Black Butte Mine, as well as identified an area that requires further investigation. This study has also begun to address the data gaps within the scientific literature on mercury cycling in sediment.

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