Electronic Supplementary Material (ESI) for Environmental Science: Processes & Impacts.

Supporting Information for:

Use of Sequential Extraction and Mercury Stable Isotope Analysis to Assess Remobilization of Sediment-Bound Legacy Mercury

Elizabeth R. Crowther,*[a] Jason D. Demers,[a] Joel D. Blum,[a]
Scott C. Brooks,[b] Marcus W. Johnson[a]

[a]Department of Earth and Environmental Sciences, University of Michigan,
1100 N. University Ave., Ann Arbor, MI 48109, USA.

[b]Environmental Science Division, Oak Ridge National Laboratory,
P.O Box 2008, Oak Ridge, TN 37831-6038, USA.

*Corresponding author: Elizabeth R. Crowther, e-mail: ecrowth@umich.edu

S1. Quality Control for Total Mercury Analyses

Cold vapor atomic fluorescence spectrometry (CVAFS)[1] was used to measure total mercury (THg) concentrations of sequential extraction solutions and combustion 1% $KMnO_4$ trap solutions, as well as to determine percent recoveries after purge and trap and transfer procedures. Across all analytical sessions, the percent relative standard deviation (%RSD) of the mean calibration factor (CF_m) for calibration standards (2.5 to 500 pg Hg, or 0.5 to 100 ng L^{-1} on a volumetric basis) was 1.5% (SE = 0.2%, n=18). Average recovery of the lowest calibration standard relative to its expected value was 99.6% (SE = 0.7%, n=18). All initial and calibration verification blanks were below the reporting limit (i.e., the lowest calibration standard) (n=328). Initial calibration verification standards using a certified secondary Hg standard (100, 200, or 300 pg Hg) had an average recovery of 101.1% (SE = 0.3%, n=18). Instrument precision and recovery checks (25 pg Hg, or 5 ng L^{-1}) had an average recovery of 99.9% (SE = 0.3%, n=18). Continuing calibration verification standards (100, 200, or 300 pg Hg) had an average recovery of 101.2% (SE < 0.1%, n=268).

The method detection limit (MDL) was approximately 0.5 pg Hg, or 0.1 ng L^{-1}. The MDL was calculated using the equation MDL = t * SD, where t is the student's t-value appropriate for the single-tailed 99th percentile and a standard deviation estimate with n-1 degrees of freedom, and SD is the standard deviation associated with the average mass or concentration of Hg detected in replicate MDL check standards, which should be analyzed at a concentration ~5 times higher than the expected MDL value.[2] Our MDL check standards were analyzed at 2.5 pg Hg, or 0.5 ng L^{-1}. In our calculation of the MDL, the t-value was 2.57 (based on 17 degrees of freedom), and the SD value was 0.2 pg, or 0.04 ng L^{-1} (n=18).

To assess recovery for sequential extraction samples, matrix spike samples were analyzed, in which an aliquot of spiking solution was added to an aliquot of sample which had undergone UV treatment, which had an average recovery of 100.5% (SE = 0.1%, n=109). Additionally, aliquots of spiking solution were added to a subset of sequential extraction samples prior to undergoing UV treatment, which had an average recovery of 98.4% (SE = 0.5%, n=21). Relative percent difference among duplicate UV-treated sequential extraction sample analyses averaged 1.2% (SE = 0.1%, n=17).

S2. Sequential Extraction Reagents

Based on the sequential extraction procedure developed by Bloom *et al.*,[3] our reagents were made as follows. F1 (deionized water) was purged overnight with argon gas. F2 (0.1 M acetic acid + 0.01 M HCl) was made by diluting 6 mL glacial acetic acid + 0.8 mL trace metal grade HCl to 1000 mL with deionized water. F3 (1 M KOH) was made by diluting 86 mL of 45% (w/w) reagent grade KOH solution to 1000 mL with deionized water. F4 (12 M HNO_3) was made by diluting 750 mL trace metal grade HNO_3 to 1000 mL with deionized water. F5 (aqua regia) was made by adding 10 mL trace metal grade HCl, followed by 3 mL trace metal grade HNO_3, directly into centrifuge tubes. Reagents were diluted in trace metal clean glass volumetric flasks and stored in trace metal clean glass or Teflon bottles.

S3. Comparison of Sequential Extractions of Reference Materials across Multiple Studies

Our study is one of three that has measured the isotopic composition of sequential extractions of NIST SRM 2711 (Montana Soil), each of which used different reagents to target organically-bound Hg. Wiederhold *et al.*[4] performed a two-step extraction in which 6 M HNO_3 released 19% of the THg from NIST SRM 2711, and Grigg *et al.*[5] performed a four-step extraction in which 0.1 M $Na_4P_2O_7$ released 6.0% of the THg from NIST SRM 2711a. In our study, the 1 M KOH (F3) extraction step was used to target organically-bound Hg, which released 2.6 ± 0.2% (1SD, n=3) of the THg from NIST SRM 2711 and was isotopically similar to the residual Hg. For our analysis of NIST SRM 2711, δ^{202}Hg values of the F3 extraction and the weighted average of the F4 and F5 extractions were -0.25‰ and -0.19‰, respectively (± 0.08‰, 2SD) (Table S5). In the other two studies, the organically-bound fractions tended to be isotopically heavier than the residual Hg, although their delta values were also within analytical uncertainty. For Wiederhold *et al.*,[4] δ^{202}Hg values of the 6 M HNO_3 and aqua regia extractions of NIST SRM 2711 were 0.05‰ and -0.33‰, respectively (± 0.15‰, 2SD), and for Grigg *et al.*,[5] δ^{202}Hg values of the 0.1 M $Na_4P_2O_7$ and aqua regia extractions of NIST SRM 2711a were -0.01‰ and -0.13‰, respectively (± 0.11‰, 2SD). These differences among various study results are likely due to different reagents (6 M HNO_3, 0.1 M $Na_4P_2O_7$, and 1 M KOH) targeting different pools of Hg. Given the large range in the amount of Hg released by these various methods (2.6 to 19%), it seems plausible that different pools of organically-bound Hg with unique isotopic compositions could have been accessed. For example, soil and sediment are

known to contain acid-soluble and base-soluble organic constituents.[6] Additionally, the 6 M HNO_3 step for Wiederhold *et al.*[4] was isotopically more similar to our 12 M HNO_3 step (F4) and released ~7 times more Hg than to our 1 M KOH step (F3), further suggesting that the Hg species found in operationally-defined pools of organically-bound Hg likely differ slightly depending on which reagent is used. Alternatively, differences in the isotopic composition of organically-bound Hg may be due to differences between individual batches of NIST SRM 2711 and NIST SRM 2711a, as reported δ^{202}Hg values of individual batches of NRC MESS-3 have differed by as much as 0.38‰,[7] although the bulk isotopic composition of individual batches of NIST SRM 2711 and NIST SRM 2711a has been shown to be fairly consistent.[4,5,7]

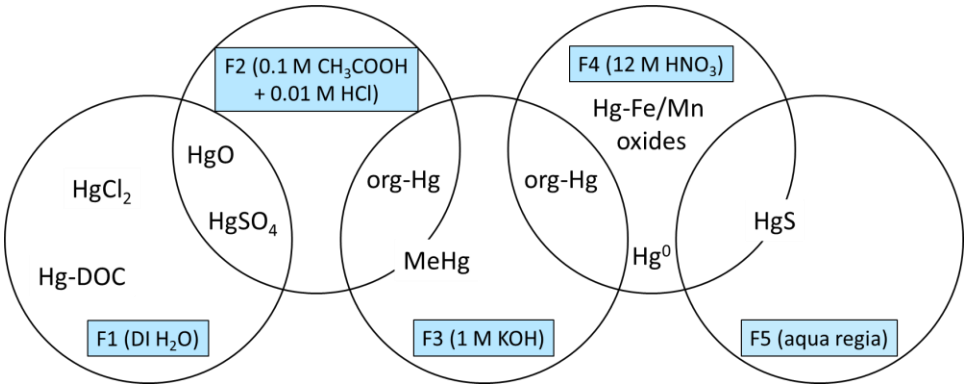

Figure S1: Schematic diagram showing Hg compounds that may be released during sequential extractions. Blue boxes indicate the reagents used for the five-step sequential extraction procedure. Hg-DOC represents Hg bound to highly soluble or easily desorbable organic matter, while org-Hg represents Hg bound to less soluble or more strongly sorbed organic matter. Note that methylmercury (MeHg) makes up a small percentage (typically <0.05%) of THg in EFPC streambed sediment.[8] Also note that some Hg(0) may be released during earlier extraction steps. Placement of Hg compounds is approximate, and is based on information found in the literature.[3,9-11]

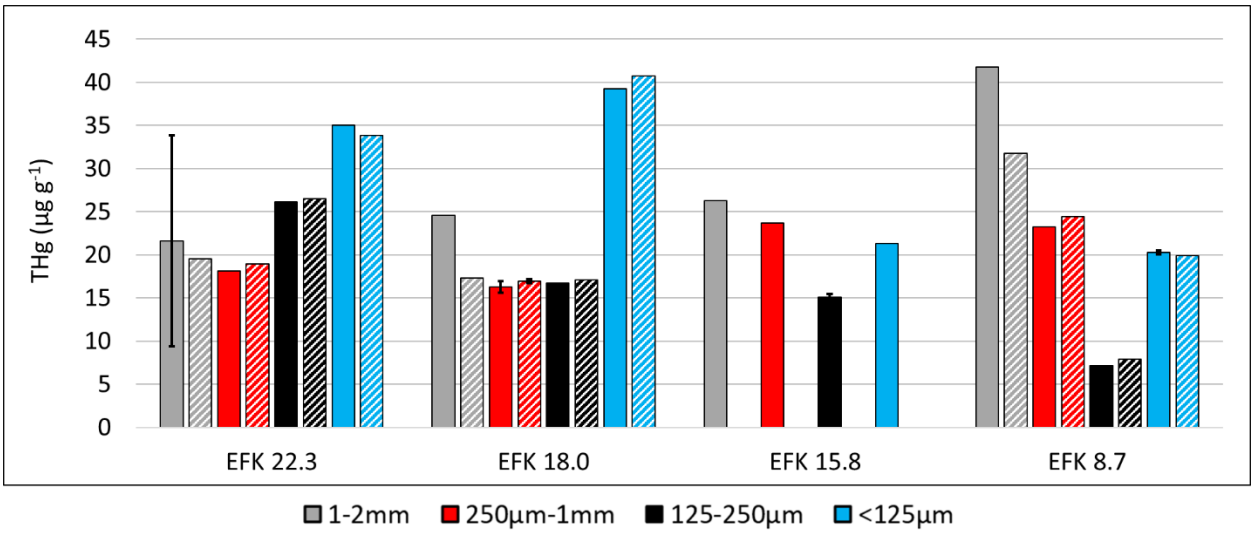

Figure S2: Total Hg concentration (µg g^{-1}) of EFPC streambed sediment, measured via combustion (solid bars) and the sum of sequential extractions (striped bars). Error bars represent 1SD for combustion sample replicates (n=2) or sequential extraction replicates (n=2).

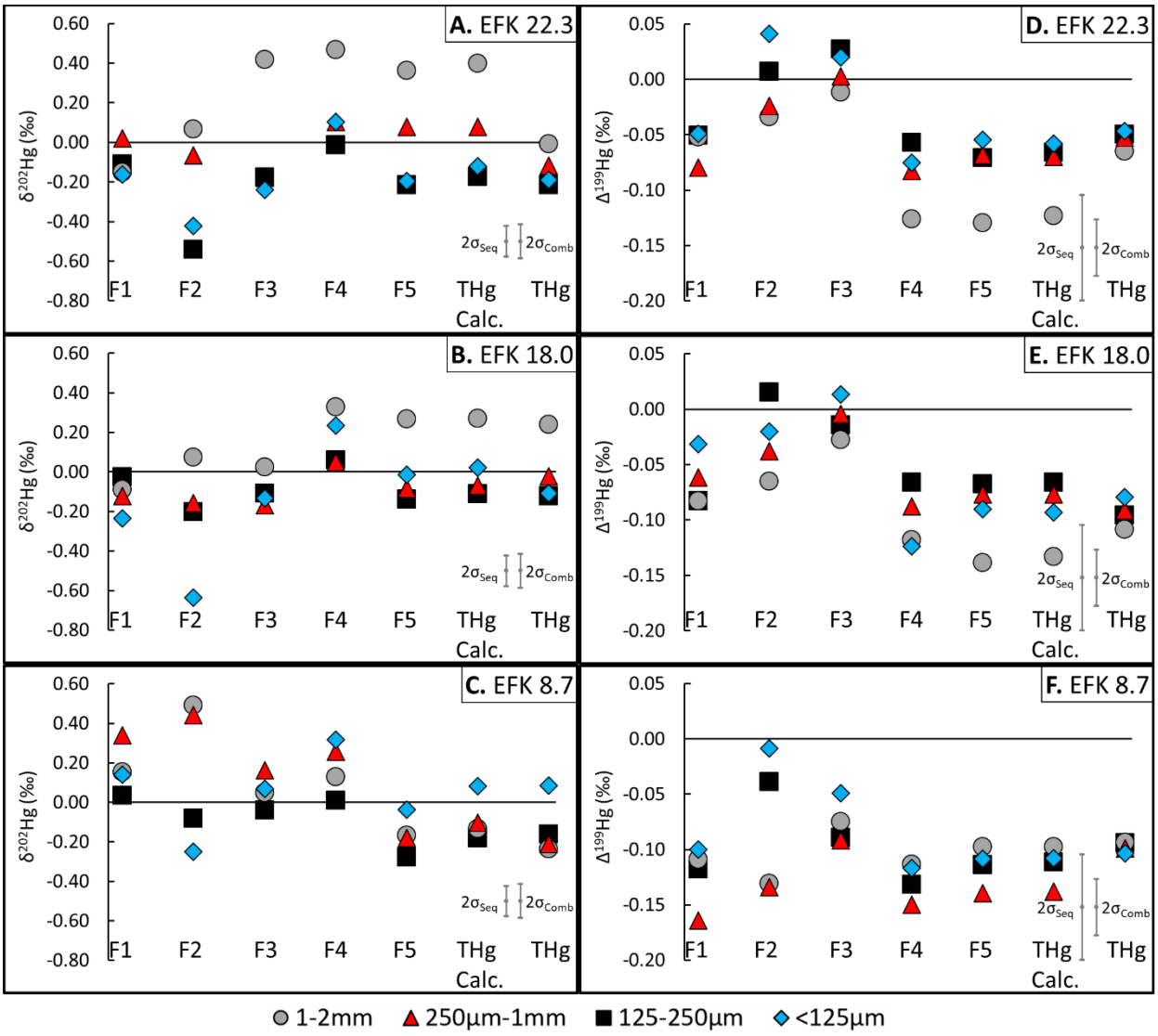

Figure S3 Mercury isotopic composition of sequential extractions of EFPC streambed sediment. Shown are (A-C) δ^{202}Hg and (D-F) Δ^{199}Hg of sequentially extracted Hg pools, the calculated total Hg based on the weighted average of sequential extraction concentrations (THg Calc.), and bulk sediment measured via combustion (THg). Some of the THg points represent an average of combustion replicates (Table S3, Table S4). Analytical uncertainty in sequential extraction delta values is shown as the average uncertainty (2SD) across all UM-Almadén analyses ($2\sigma_{seq}$). Analytical uncertainty in combustion sample delta values is shown as the average uncertainty (2SD) across combustion reference material analyses ($2\sigma_{comb}$) (see Section 2.5).

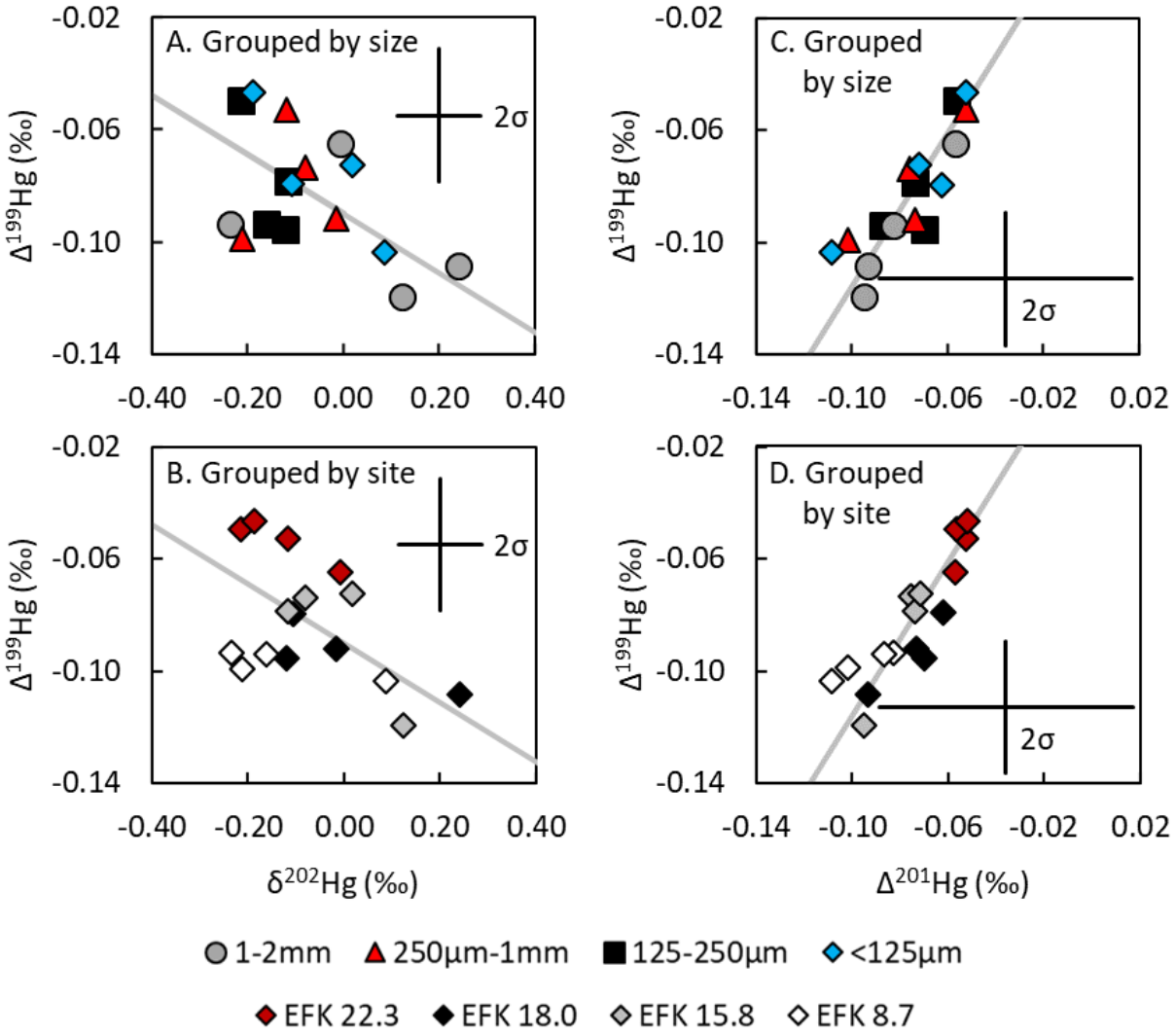

Figure S4 Mercury isotopic composition of EFPC streambed sediment, measured via combustion. Shown are (A and B) Δ^{199}Hg versus δ^{202}Hg and (C and D) Δ^{199}Hg versus Δ^{201}Hg for samples grouped by (A and C) sediment size fraction and (B and D) sampling site. Analytical uncertainty in delta values is shown as the average uncertainty (2SD) across combustion reference material analyses (see Section 2.5). Using the York regression,[12] the Δ^{199}Hg/δ^{202}Hg slope is -0.11 (± 0.01, 1SE, n=16), and the Δ^{199}Hg/Δ^{201}Hg slope is 1.37 (± 0.21, 1SE, n=16). Slopes were generated using IsoplotR.[13]

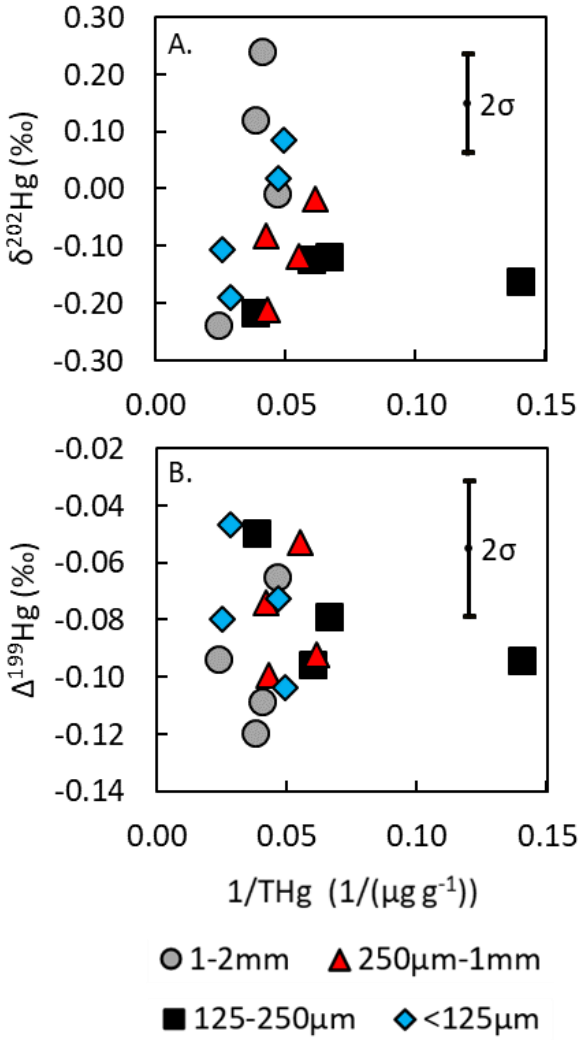

Figure S5: Mercury isotopic composition of EFPC streambed sediment, measured via combustion. Shown are (A) δ^{202}Hg and (B) Δ^{199}Hg versus inverse total Hg concentration. Analytical uncertainty in delta values is shown as the average uncertainty (2SD) across combustion reference material analyses (see Section 2.5).

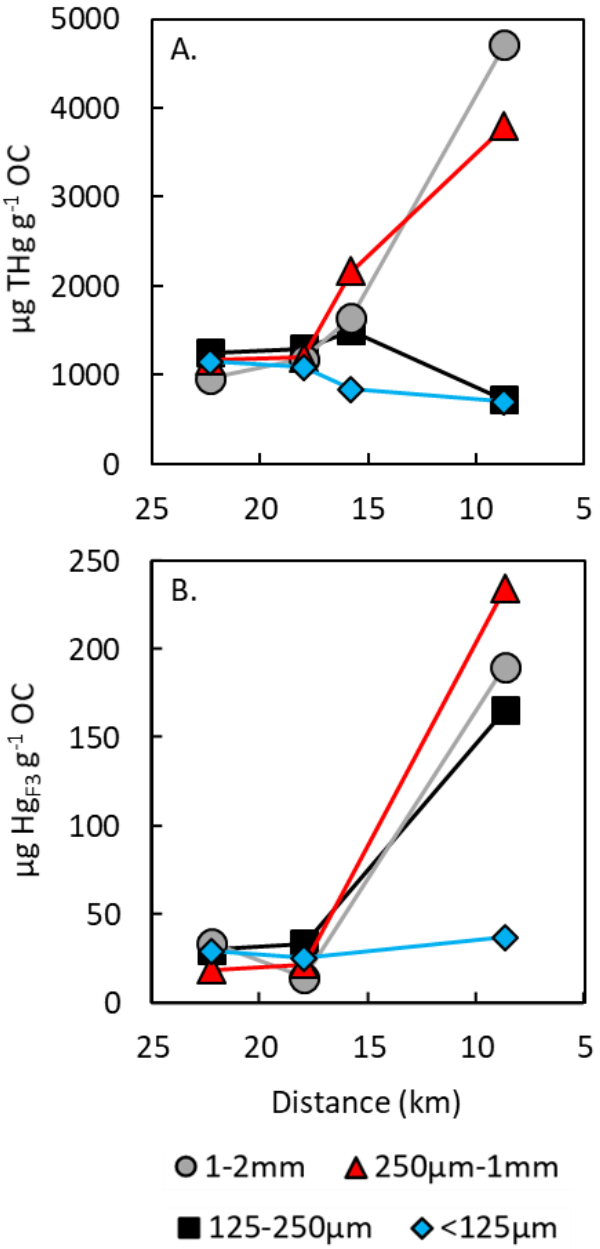

Figure S6: Concentrations of (A) total Hg and (B) F3-extracted Hg per mass of organic carbon for EFPC streambed sediment versus distance upstream of the confluence with Poplar Creek.

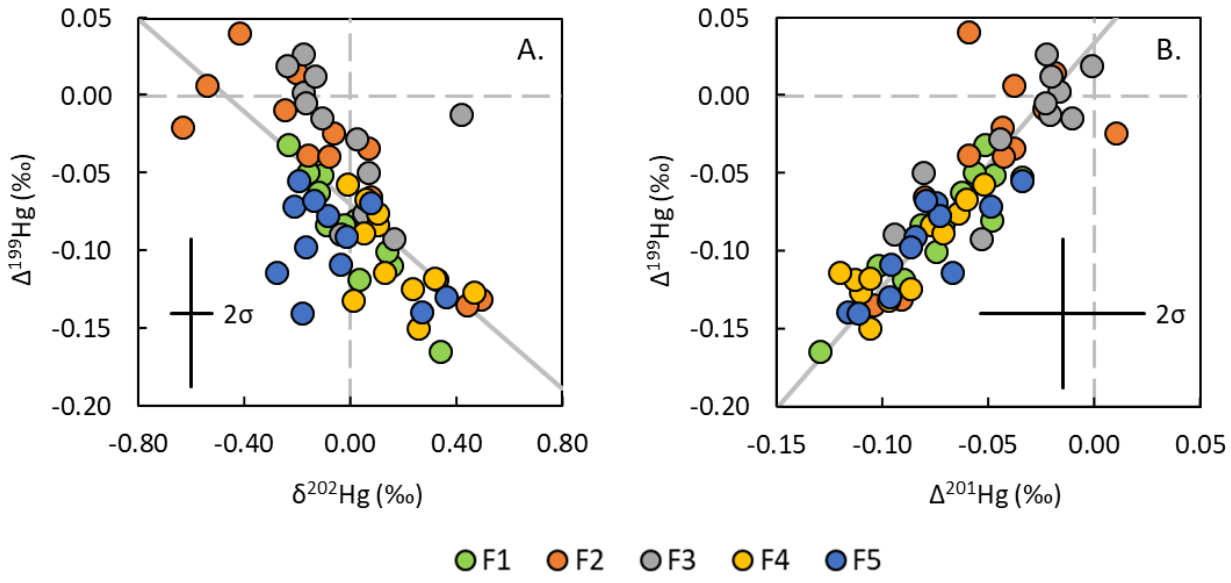

Figure S7: Mercury isotopic composition of sequential extractions of EFPC streambed sediment. Shown are (A) Δ^{199}Hg versus δ^{202}Hg and (B) Δ^{199}Hg versus Δ^{201}Hg across all sampling sites and size fractions. Analytical uncertainty in delta values is shown as the average uncertainty (2SD) across all UM-Almadén analyses (see Section 2.5). Using the York regression,[12] the Δ^{199}Hg/δ^{202}Hg slope is -0.15 (± 0.01, 1SE, n=60), and the Δ^{199}Hg/Δ^{201}Hg slope is 1.57 (± 0.16, 1SE, n=60). Slopes were generated using IsoplotR.[13]

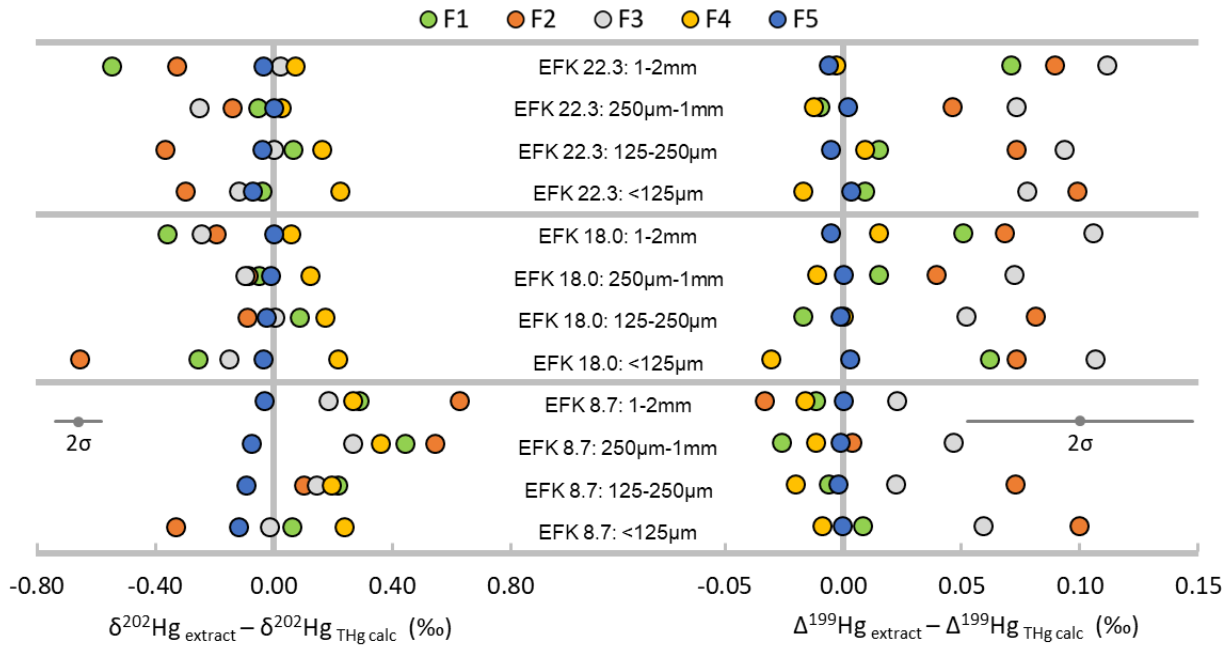

Figure S8: Offsets in isotopic composition ($\delta^{202}Hg$ and $\Delta^{199}Hg$) of sequential extractions relative to the isotopic composition of bulk sediment calculated via the weighted average of the five sequential extractions for each sediment size fraction (see Section 2.3). Analytical uncertainty in delta values is shown as the average uncertainty (2SD) across all UM-Almadén analyses (see Section 2.5).

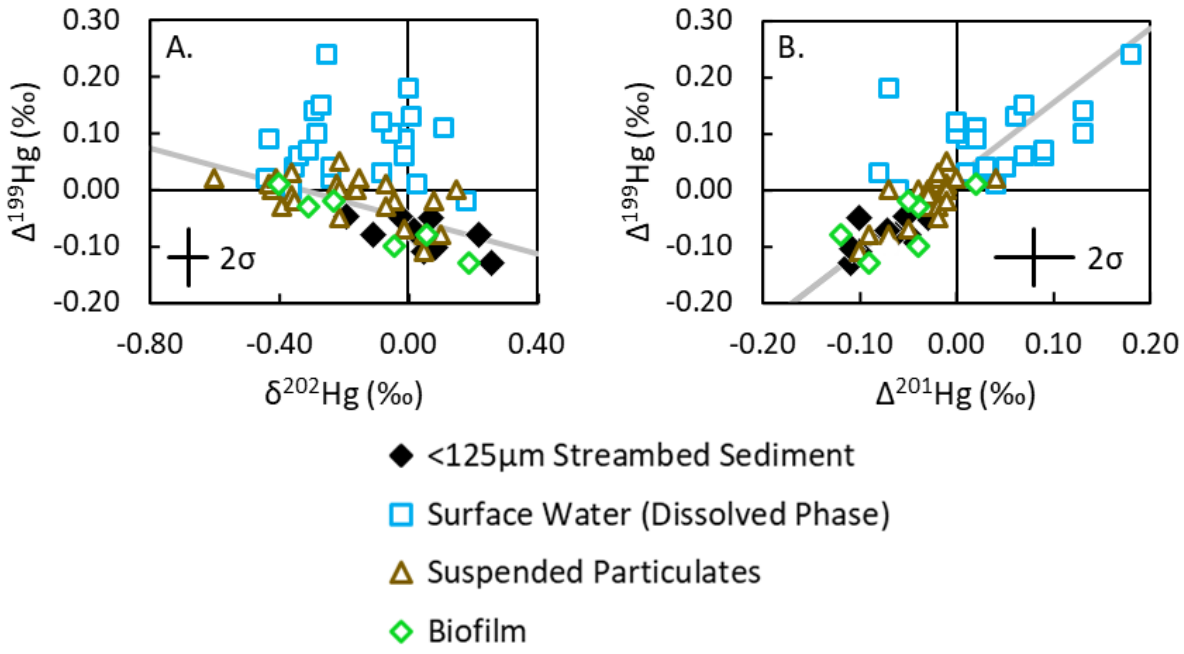

Figure S9: Mercury isotopic composition of <125µm streambed sediment (including sediment analyzed in this study as well as by Donovan *et al.*[14]), along with EFPC surface water dissolved phase, suspended particulates, and biofilm analyzed by Demers *et al.*[15] Shown are (A) Δ^{199}Hg versus δ^{202}Hg and (B) Δ^{199}Hg versus Δ^{201}Hg. Analytical uncertainty in delta values is shown as the average uncertainty (2SD) across all UM-Almadén analyses as defined by Demers *et al.*,[15] which is similar to this study, though the analytical uncertainty for sediment analyzed by Donovan *et al.*[14] is somewhat larger. Using the York regression,[12] the Δ^{199}Hg/δ^{202}Hg slope for suspended particulates and <125µm streambed sediment combined is -0.16 (± 0.02, 1SE, n=34), and the Δ^{199}Hg/Δ^{201}Hg slope is 1.32 (± 0.20, 1SE, n=34). Slopes were generated using IsoplotR.[13]

Table S1: Mass and Hg fractions of each grain size of EFPC streambed sediment within each sampling site.[a]

Site ID	1-2mm		250µm-1mm		125-250µm		<125µm	
	Mass Fraction	Hg Fraction	Mass Fraction	Hg Fraction	Mass Fraction	Hg Fraction	Mass Fraction	Hg Fraction
EFK 22.3	42%	41%	40%	33%	4%	5%	14%	22%
EFK 18.0	42%	49%	50%	39%	3%	2%	5%	9%
EFK 15.8	40%	43%	53%	51%	2%	1%	5%	4%
EFK 8.7	43%	59%	45%	34%	3%	1%	9%	6%

[a]Mass fraction is based on freeze dried masses of each sediment size fraction within each site. Hg fraction is based on THg concentrations (measured via combustion) normalized to the mass fraction of each sediment size fraction within each site. Excludes >2mm sediment.

Table S2: Mercury isotopic composition of UM-Almadén and procedural standards.[a]

Standard Type	n	δ^{204}Hg (‰)	2σ (‰)	δ^{202}Hg (‰)	2σ (‰)	δ^{201}Hg (‰)	2σ (‰)	δ^{200}Hg (‰)	2σ (‰)	δ^{199}Hg (‰)	2σ (‰)	Δ^{204}Hg (‰)	2σ (‰)	Δ^{201}Hg (‰)	2σ (‰)	Δ^{200}Hg (‰)	2σ (‰)	Δ^{199}Hg (‰)	2σ (‰)
UM-Almadén	25	-0.83	0.01	-0.55	0.01	-0.45	0.01	-0.27	0.00	-0.16	0.00	-0.01	0.01	-0.04	0.00	0.01	0.00	-0.03	0.00
Purge & Trap NIST SRM 3133	21	0.00	0.02	0.00	0.02	0.00	0.01	0.00	0.01	0.00	0.01	0.00	0.01	0.00	0.01	0.00	0.01	0.00	0.01
NIST SRM 2711 (Montana Soil)	2	-0.27	0.01	-0.21	0.02	-0.35	0.02	-0.12	0.02	-0.29	0.03	0.04	0.05	-0.19	0.01	-0.01	0.01	-0.23	0.02
NIST SRM 2711 (Montana Soil) Long-term average	11	-0.28	0.08	-0.18	0.05	-0.32	0.04	-0.10	0.03	-0.28	0.02	-0.01	0.03	-0.19	0.02	-0.01	0.01	-0.24	0.01
NIST SRM 1944 (NY/NJ Sediment)	1	-0.65	-	-0.43	-	-0.34	-	-0.21	-	-0.11	-	-0.01	-	-0.01	-	0.01	-	0.00	-
NIST SRM 1944 (NY/NJ Sediment) Long-term average	62	-0.65	0.02	-0.43	0.01	-0.34	0.01	-0.21	0.01	-0.11	0.01	-0.01	0.01	-0.02	0.01	0.01	0.01	0.00	0.01

[a] For UM-Almadén, n is the number of preparations (i.e. the number of session averages, with preparations at different concentrations counted separately). For procedural standards, n is the number of completely independent preparations of the material (via purge and trap or combustions). Isotope ratios represent the average value (± 2SE) across independent preparations for each standard type. Long-term average isotopic compositions are included for comparison.[7]

Table S3: Mercury concentration and isotopic composition of EFPC streambed sediment measured via combustion.[a]

Site ID	Size Fraction	THg (μg g^{-1})	n_1	n_2	n_3	% Rec.	δ^{204}Hg (‰)	2σ (‰)	δ^{202}Hg (‰)	2σ (‰)	δ^{201}Hg (‰)	2σ (‰)	δ^{200}Hg (‰)	2σ (‰)	δ^{199}Hg (‰)	2σ (‰)	Δ^{204}Hg (‰)	2σ (‰)	Δ^{201}Hg (‰)	2σ (‰)	Δ^{200}Hg (‰)	2σ (‰)	Δ^{199}Hg (‰)	2σ (‰)
EFK 22.3	1-2mm	21.60	2	3	9	92.8	0.00	0.12	-0.01	0.09	-0.06	0.07	-0.01	0.04	-0.07	0.02	0.01	0.01	-0.06	0.05	-0.01	0.02	-0.06	0.02
	250μm-1mm	18.14	1	1	3	93.5	-0.18	0.12	-0.12	0.09	-0.14	0.07	-0.05	0.04	-0.08	0.02	0.00	0.01	-0.05	0.05	0.01	0.02	-0.05	0.02
	125-250μm	26.15	1	1	2	97.2	-0.33	0.12	-0.22	0.09	-0.22	0.07	-0.09	0.04	-0.10	0.02	-0.01	0.01	-0.06	0.05	0.01	0.02	-0.05	0.02
	<125μm	35.00	1	1	2	95.5	-0.26	0.12	-0.19	0.09	-0.19	0.07	-0.09	0.04	-0.09	0.02	0.02	0.01	-0.05	0.05	0.01	0.02	-0.05	0.02
EFK 18.0	1-2mm	24.58	1	1	2	94.1	0.34	0.12	0.24	0.09	0.09	0.07	0.10	0.04	-0.05	0.02	-0.02	0.01	-0.09	0.05	-0.02	0.02	-0.11	0.02
	250μm-1mm	16.28	2	3	7	96.6	-0.03	0.12	-0.02	0.09	-0.09	0.07	-0.02	0.04	-0.10	0.02	-0.01	0.01	-0.07	0.05	-0.01	0.02	-0.09	0.02
	125-250μm	16.75	1	1	2	97.4	-0.18	0.12	-0.12	0.09	-0.16	0.07	-0.06	0.04	-0.13	0.02	0.00	0.01	-0.07	0.05	0.00	0.02	-0.10	0.02
	<125μm	39.27	1	1	3	97.7	-0.14	0.12	-0.11	0.09	-0.14	0.07	-0.05	0.04	-0.11	0.02	0.02	0.01	-0.06	0.05	0.00	0.02	-0.08	0.02
EFK 15.8	1-2mm	26.30	1	1	3	96.1	0.18	0.12	0.12	0.09	0.00	0.07	0.06	0.04	-0.09	0.02	-0.01	0.01	-0.10	0.05	0.00	0.02	-0.12	0.02
	250μm-1mm	23.68	1	1	2	96.9	-0.13	0.12	-0.08	0.09	-0.14	0.07	-0.03	0.04	-0.09	0.02	-0.01	0.01	-0.08	0.05	0.01	0.02	-0.07	0.02
	125-250μm	15.07	2	3	8	95.2	-0.17	0.12	-0.12	0.09	-0.16	0.07	-0.05	0.04	-0.11	0.02	0.00	0.01	-0.07	0.05	0.00	0.02	-0.08	0.02
	<125μm	21.32	1	1	2	97.3	0.01	0.12	0.02	0.09	-0.06	0.07	0.01	0.04	-0.07	0.02	-0.02	0.01	-0.07	0.05	0.00	0.02	-0.07	0.02
EFK 8.7	1-2mm	41.77	1	1	2	92.1	-0.35	0.12	-0.24	0.09	-0.26	0.07	-0.10	0.04	-0.15	0.02	0.01	0.01	-0.08	0.05	0.02	0.02	-0.09	0.02
	250μm-1mm	23.22	1	1	4	96.2	-0.28	0.12	-0.21	0.09	-0.26	0.07	-0.09	0.04	-0.15	0.02	0.03	0.01	-0.10	0.05	0.01	0.02	-0.10	0.02
	125-250μm	7.14	1	1	2	97.6	-0.23	0.12	-0.16	0.09	-0.21	0.07	-0.08	0.04	-0.13	0.02	0.01	0.01	-0.09	0.05	0.00	0.02	-0.09	0.02
	<125μm	20.26	2	3	6	96.2	0.14	0.12	0.09	0.09	-0.04	0.07	0.03	0.04	-0.08	0.02	0.01	0.01	-0.11	0.05	-0.01	0.02	-0.10	0.02

[a] Sieved streambed sediment was collected from four sites along EFPC. Site ID refers to the sampling location identified by the number of kilometers upstream of the confluence of Poplar Creek and EFPC. Here, n_1 denotes the number of sample replicates that were ground independently prior to combustions, n_2 denotes the number of combustion preparations, and n_3 denotes the number of separate isotopic analyses on an individual preparation(s). The percent recovery (% Rec.) shows recovery of Hg during the transfer procedure for preparation of combustion samples for isotope analysis. The uncertainty in the isotopic composition of Hg in combustion samples is represented by the average uncertainty (2SD) across combustion reference material analyses.

Table S4: Comparison of combustion replicates.[a]

Sample ID	Replicates	THg (μg g^{-1})	n_1	% Rec.	δ^{204}Hg (‰)	2σ (‰)	δ^{202}Hg (‰)	2σ (‰)	δ^{201}Hg (‰)	2σ (‰)	δ^{200}Hg (‰)	2σ (‰)	δ^{199}Hg (‰)	2σ (‰)	Δ^{204}Hg (‰)	2σ (‰)	Δ^{201}Hg (‰)	2σ (‰)	Δ^{200}Hg (‰)	2σ (‰)	Δ^{199}Hg (‰)	2σ (‰)
EFK 22.3 1-2mm	Rep 1, C1	30.32	3	94.1	-0.19	0.12	-0.14	0.09	-0.16	0.07	-0.07	0.04	-0.09	0.02	0.01	0.01	-0.05	0.05	0.00	0.02	-0.05	0.02
	Rep 1, C2	30.20	3	92.7	-0.30	0.12	-0.20	0.09	-0.20	0.07	-0.10	0.04	-0.08	0.02	0.00	0.01	-0.05	0.05	0.00	0.02	-0.03	0.02
	Rep 2	12.94	3	92.2	0.25	0.12	0.15	0.09	0.05	0.07	0.07	0.04	-0.05	0.02	0.02	0.01	-0.06	0.05	-0.01	0.02	-0.09	0.02
EFK 18.0 250μm-1mm	Rep 1, C1	15.98	2	99.3	0.08	0.12	0.04	0.09	-0.04	0.07	0.02	0.04	-0.09	0.02	0.01	0.01	-0.08	0.05	0.00	0.02	-0.10	0.02
	Rep 1, C2	15.65	3	96.2	-0.08	0.12	-0.06	0.09	-0.11	0.07	-0.03	0.04	-0.10	0.02	0.01	0.01	-0.07	0.05	0.00	0.02	-0.08	0.02
	Rep 2	16.75	2	95.4	-0.06	0.12	-0.02	0.09	-0.09	0.07	-0.03	0.04	-0.10	0.02	-0.03	0.01	-0.08	0.05	-0.01	0.02	-0.09	0.02
EFK 15.8 125-250μm	Rep 1, C1	14.90	3	97.6	-0.19	0.12	-0.13	0.09	-0.16	0.07	-0.06	0.04	-0.11	0.02	0.00	0.01	-0.06	0.05	0.00	0.02	-0.08	0.02
	Rep 1, C2	14.69	3	94.2	-0.19	0.12	-0.12	0.09	-0.17	0.07	-0.05	0.04	-0.10	0.02	-0.01	0.01	-0.08	0.05	0.01	0.02	-0.07	0.02
	Rep 2	15.35	2	94.6	-0.15	0.12	-0.11	0.09	-0.16	0.07	-0.05	0.04	-0.11	0.02	0.01	0.01	-0.07	0.05	0.01	0.02	-0.08	0.02
EFK 8.7 <125μm	Rep 1, C1	20.30	2	98.5	0.11	0.12	0.06	0.09	-0.04	0.07	0.04	0.04	-0.09	0.02	0.02	0.01	-0.09	0.05	0.01	0.02	-0.11	0.02
	Rep 1, C2	19.92	2	93.7	0.21	0.12	0.13	0.09	0.02	0.07	0.06	0.04	-0.07	0.02	0.02	0.01	-0.08	0.05	-0.01	0.02	-0.10	0.02
	Rep 2	20.41	2	96.3	0.12	0.12	0.08	0.09	-0.07	0.07	0.02	0.04	-0.09	0.02	0.01	0.01	-0.13	0.05	-0.02	0.02	-0.11	0.02
NIST SRM 2711 (Montana Soil)	Rep 1	6.56	3	95.9	-0.27	0.12	-0.22	0.09	-0.36	0.07	-0.13	0.04	-0.30	0.02	0.06	0.01	-0.19	0.05	-0.02	0.02	-0.24	0.02
	Rep 2	6.57	3	92.7	-0.28	0.12	-0.20	0.09	-0.34	0.07	-0.11	0.04	-0.27	0.02	0.01	0.01	-0.19	0.05	-0.01	0.02	-0.22	0.02

[a] Sieved streambed sediment was collected from four sites along EFPC. Sample ID refers to the sediment size fraction and the sampling location identified by the number of kilometers upstream of the confluence of Poplar Creek and EFPC. Rep 1 and Rep 2 refer to sample replicates that were ground independently prior to combustions. C1 and C2 refer to combustion process replicates, which used aliquots of ground sediment from the same vial. Here, n_1 denotes the number of separate isotopic analyses on an individual preparation. The percent recovery (% Rec.) shows recovery of Hg during the transfer procedure for preparation of combustion samples for isotope analysis. The uncertainty in the isotopic composition of Hg in combustion samples is represented by the average uncertainty (2SD) across combustion reference material analyses.

Table S5: Mercury concentration and isotopic composition of sequential extractions of standard reference materials.[a]

Reference Material	Sequential Extraction	THg (μg g^{-1})	% of THg	n_1	n_2	n_3	% Rec.	δ^{204}Hg (‰)	2σ (‰)	δ^{202}Hg (‰)	2σ (‰)	δ^{201}Hg (‰)	2σ (‰)	δ^{200}Hg (‰)	2σ (‰)	δ^{199}Hg (‰)	2σ (‰)	Δ^{204}Hg (‰)	2σ (‰)	Δ^{201}Hg (‰)	2σ (‰)	Δ^{200}Hg (‰)	2σ (‰)	Δ^{199}Hg (‰)	2σ (‰)
NIST SRM 2711 (Montana Soil)	F1	0.07	1.2	3	4	5	98.4	-0.30	0.12	-0.21	0.08	-0.32	0.07	-0.10	0.05	-0.25	0.05	0.02	0.08	-0.16	0.04	0.00	0.04	-0.20	0.05
	F2	0.00	0.1	2	2	2	96.0	-1.18	0.12	-0.81	0.08	-0.67	0.07	-0.35	0.05	-0.31	0.05	0.03	0.08	-0.06	0.04	0.06	0.04	-0.10	0.05
	F3	0.15	2.6	3	5	9	97.5	-0.36	0.12	-0.25	0.08	-0.33	0.07	-0.13	0.05	-0.25	0.05	0.00	0.08	-0.15	0.04	-0.01	0.04	-0.19	0.05
	F4	4.07	68.3	3	4	7	97.7	-0.13	0.12	-0.09	0.08	-0.27	0.07	-0.05	0.05	-0.25	0.05	0.01	0.08	-0.20	0.04	0.00	0.04	-0.23	0.05
	F5	1.66	27.8	3	4	7	98.9	-0.63	0.12	-0.43	0.08	-0.50	0.07	-0.22	0.05	-0.32	0.05	0.02	0.08	-0.18	0.04	-0.01	0.04	-0.22	0.05
	Calc. THg	5.96					95.4	-0.27	0.12	-0.19	0.08	-0.34	0.07	-0.10	0.05	-0.27	0.05	0.01	0.08	-0.19	0.04	0.00	0.04	-0.22	0.05
NIST SRM 1944 (NY/NJ Waterway Sediment)	F1	0.02	0.4	1	1	1	95.1	-0.48	0.12	-0.41	0.08	-0.30	0.07	-0.07	0.05	-0.19	0.05	0.13	0.08	0.00	0.04	0.13	0.04	-0.09	0.05
	F2	0.00	<0.1	Insufficient mercury for isotopic analysis.																					
	F3	0.31	8.8	1	2	3	95.7	-0.61	0.12	-0.39	0.08	-0.29	0.07	-0.19	0.05	-0.06	0.05	-0.02	0.08	0.01	0.04	0.00	0.04	0.04	0.05
	F4	2.77	79.6	1	3	4	97.5	-0.58	0.12	-0.40	0.08	-0.31	0.07	-0.21	0.05	-0.11	0.05	0.02	0.08	-0.01	0.04	-0.01	0.04	-0.01	0.05
	F5	0.39	11.1	1	2	3	95.6	-2.17	0.12	-1.46	0.08	-1.19	0.07	-0.72	0.05	-0.49	0.05	0.00	0.08	-0.10	0.04	0.01	0.04	-0.12	0.05
	Calc. THg	3.48					102.3	-0.75	0.12	-0.52	0.08	-0.40	0.07	-0.27	0.05	-0.15	0.05	0.02	0.08	-0.02	0.04	-0.01	0.04	-0.02	0.05

[a] Five-step sequential extractions were performed on two standard reference materials. Calc. THg refers to the calculated THg concentration and isotopic composition of the material based on the weighted average of sequential extractions. Here, n_1 denotes the number of sequential extraction sets, n_2 denotes the number of purge and trap preparations, and n_3 denotes the number of separate isotopic analyses on an individual preparation(s). The percent recovery (% Rec.) for F1 – F5 shows recovery of Hg during the purge and trap procedure for preparation of sequential extraction samples for isotope analysis. The percent recovery for Calc. THg is based on the sum of sequential extractions relative to certified values. The uncertainty in the isotopic composition of Hg in sequential extraction samples is represented by the average uncertainty (2SD) across all UM-Almadén analyses.

Table S6.1: Mercury concentration and isotopic composition of sequential extractions of EFPC streambed sediment at EFK 22.3.[a]

Sample ID	Sequential Extraction	THg (μg g^{-1})	% of THg	n_1	n_2	n_3	% Rec.	δ^{204}Hg (‰)	2σ (‰)	δ^{202}Hg (‰)	2σ (‰)	δ^{201}Hg (‰)	2σ (‰)	δ^{200}Hg (‰)	2σ (‰)	δ^{199}Hg (‰)	2σ (‰)	Δ^{204}Hg (‰)	2σ (‰)	Δ^{201}Hg (‰)	2σ (‰)	Δ^{200}Hg (‰)	2σ (‰)	Δ^{199}Hg (‰)	2σ (‰)
EFK 22.3 1-2mm	F1	0.08	0.4	1	2	4	91.5	-0.23	0.12	-0.15	0.08	-0.15	0.07	-0.09	0.05	-0.09	0.05	0.00	0.08	-0.03	0.04	-0.01	0.04	-0.05	0.05
	F2	0.09	0.5	1	2	5	96.0	0.07	0.12	0.07	0.08	0.01	0.07	0.05	0.05	-0.02	0.05	-0.03	0.08	-0.04	0.04	0.01	0.04	-0.03	0.05
	F3	0.75	3.8	1	1	2	101.5	0.57	0.12	0.42	0.08	0.29	0.07	0.22	0.05	0.09	0.05	-0.06	0.08	-0.02	0.04	0.01	0.04	-0.01	0.05
	F4	6.73	34.4	1	1	2	96.7	0.71	0.12	0.47	0.08	0.24	0.07	0.22	0.05	-0.01	0.05	0.01	0.08	-0.11	0.04	-0.02	0.04	-0.13	0.05
	F5	11.92	60.9	1	1	2	93.7	0.55	0.12	0.36	0.08	0.18	0.07	0.17	0.05	-0.04	0.05	0.01	0.08	-0.10	0.04	-0.01	0.04	-0.13	0.05
	Calc. THg	19.57					90.6	0.60	0.12	0.40	0.08	0.20	0.07	0.19	0.05	-0.02	0.05	0.00	0.08	-0.10	0.04	-0.01	0.04	-0.12	0.05
EFK 22.3 250μm-1mm	F1	0.31	1.6	1	1	2	96.8	0.01	0.12	0.02	0.08	-0.03	0.07	0.00	0.05	-0.07	0.05	-0.02	0.08	-0.05	0.04	-0.01	0.04	-0.08	0.05
	F2	0.01	<0.1	1	1	1	97.2	-0.08	0.12	-0.07	0.08	-0.04	0.07	-0.05	0.05	-0.04	0.05	0.02	0.08	0.01	0.04	-0.01	0.04	-0.02	0.05
	F3	0.29	1.5	1	2	5	96.6	-0.28	0.12	-0.18	0.08	-0.15	0.07	-0.07	0.05	-0.04	0.05	-0.01	0.08	-0.02	0.04	0.02	0.04	0.00	0.05
	F4	3.49	18.4	1	1	2	95.8	0.14	0.12	0.10	0.08	0.00	0.07	0.03	0.05	-0.06	0.05	-0.02	0.08	-0.08	0.04	-0.02	0.04	-0.08	0.05
	F5	14.83	78.4	1	1	2	99.5	0.11	0.12	0.08	0.08	-0.02	0.07	0.04	0.05	-0.05	0.05	0.00	0.08	-0.07	0.04	0.00	0.04	-0.07	0.05
	Calc. THg	18.92					104.3	0.11	0.12	0.08	0.08	-0.02	0.07	0.03	0.05	-0.05	0.05	-0.01	0.08	-0.07	0.04	-0.01	0.04	-0.07	0.05
EFK 22.3 125-250μm	F1	0.15	0.6	1	1	2	98.5	-0.18	0.12	-0.11	0.08	-0.13	0.07	-0.04	0.05	-0.08	0.05	-0.02	0.08	-0.05	0.04	0.01	0.04	-0.05	0.05
	F2	0.00	<0.1	1	1	1	98.9	-0.82	0.12	-0.54	0.08	-0.45	0.07	-0.26	0.05	-0.13	0.05	-0.01	0.08	-0.04	0.04	0.01	0.04	0.01	0.05
	F3	0.62	2.4	1	1	2	96.3	-0.28	0.12	-0.18	0.08	-0.16	0.07	-0.06	0.05	-0.02	0.05	-0.02	0.08	-0.02	0.04	0.03	0.04	0.03	0.05
	F4	5.22	19.7	1	1	2	97.6	-0.04	0.12	-0.01	0.08	-0.06	0.07	0.01	0.05	-0.06	0.05	-0.02	0.08	-0.05	0.04	0.01	0.04	-0.06	0.05
	F5	20.48	77.3	1	2	5	95.3	-0.29	0.12	-0.21	0.08	-0.21	0.07	-0.10	0.05	-0.13	0.05	0.03	0.08	-0.05	0.04	0.01	0.04	-0.07	0.05
	Calc. THg	26.48					101.3	-0.24	0.12	-0.17	0.08	-0.18	0.07	-0.08	0.05	-0.11	0.05	0.02	0.08	-0.05	0.04	0.01	0.04	-0.07	0.05
EFK 22.3 <125μm	F1	0.07	0.2	1	2	4	97.5	-0.25	0.12	-0.16	0.08	-0.18	0.07	-0.08	0.05	-0.09	0.05	-0.01	0.08	-0.06	0.04	0.00	0.04	-0.05	0.05
	F2	0.01	<0.1	1	1	1	96.5	-0.67	0.12	-0.42	0.08	-0.38	0.07	-0.20	0.05	-0.07	0.05	-0.04	0.08	-0.06	0.04	0.02	0.04	0.04	0.05
	F3	0.89	2.6	1	1	2	102.5	-0.33	0.12	-0.24	0.08	-0.18	0.07	-0.11	0.05	-0.04	0.05	0.03	0.08	0.00	0.04	0.01	0.04	0.02	0.05
	F4	8.57	25.3	1	2	4	96.8	0.14	0.12	0.10	0.08	0.01	0.07	0.05	0.05	-0.05	0.05	-0.02	0.08	-0.06	0.04	0.00	0.04	-0.08	0.05
	F5	24.31	71.8	1	1	2	95.9	-0.30	0.12	-0.19	0.08	-0.18	0.07	-0.10	0.05	-0.10	0.05	-0.01	0.08	-0.03	0.04	0.00	0.04	-0.05	0.05
	Calc. THg	33.85					96.7	-0.19	0.12	-0.12	0.08	-0.13	0.07	-0.06	0.05	-0.09	0.05	-0.01	0.08	-0.04	0.04	0.00	0.04	-0.06	0.05

[a] See Table S6.3 for notes.

Table S6.2: Mercury concentration and isotopic composition of sequential extractions of EFPC streambed sediment at EFK 18.0.[a]

Sample ID	Sequential Extraction	THg (μg g^{-1})	% of THg	n_1	n_2	n_3	% Rec.	δ^{204}Hg (‰)	2σ (‰)	δ^{202}Hg (‰)	2σ (‰)	δ^{201}Hg (‰)	2σ (‰)	δ^{200}Hg (‰)	2σ (‰)	δ^{199}Hg (‰)	2σ (‰)	Δ^{204}Hg (‰)	2σ (‰)	Δ^{201}Hg (‰)	2σ (‰)	Δ^{200}Hg (‰)	2σ (‰)	Δ^{199}Hg (‰)	2σ (‰)
EFK 18.0 1-2mm	F1	0.10	0.6	1	1	2	99.6	-0.13	0.12	-0.09	0.08	-0.15	0.07	-0.07	0.05	-0.11	0.05	0.01	0.08	-0.08	0.04	-0.03	0.04	-0.08	0.05
	F2	0.02	0.1	1	2	2	96.6	0.10	0.12	0.08	0.08	-0.02	0.07	0.04	0.05	-0.05	0.05	-0.01	0.08	-0.08	0.04	0.00	0.04	-0.06	0.05
	F3	0.30	1.7	1	1	2	101.4	0.03	0.12	0.02	0.08	-0.03	0.07	0.01	0.05	-0.02	0.05	-0.01	0.08	-0.04	0.04	0.00	0.04	-0.03	0.05
	F4	2.70	15.6	1	1	2	101.0	0.47	0.12	0.33	0.08	0.13	0.07	0.14	0.05	-0.04	0.05	-0.02	0.08	-0.11	0.04	-0.02	0.04	-0.12	0.05
	F5	14.19	82.0	1	1	2	104.0	0.37	0.12	0.27	0.08	0.09	0.07	0.12	0.05	-0.07	0.05	-0.03	0.08	-0.12	0.04	-0.02	0.04	-0.14	0.05
	Calc. THg	17.31					70.4	0.38	0.12	0.27	0.08	0.09	0.07	0.12	0.05	-0.06	0.05	-0.03	0.08	-0.11	0.04	-0.02	0.04	-0.13	0.05
EFK 18.0 250μm-1mm	F1	0.23	1.3	2	2	4	100.4	-0.16	0.12	-0.12	0.08	-0.15	0.07	-0.06	0.05	-0.09	0.05	0.02	0.08	-0.06	0.04	0.01	0.04	-0.06	0.05
	F2	0.01	0.1	2	2	2	97.2	-0.25	0.12	-0.16	0.08	-0.18	0.07	-0.08	0.05	-0.08	0.05	-0.01	0.08	-0.06	0.04	0.00	0.04	-0.04	0.05
	F3	0.29	1.7	2	2	4	99.9	-0.26	0.12	-0.17	0.08	-0.15	0.07	-0.06	0.05	-0.05	0.05	-0.01	0.08	-0.02	0.04	0.02	0.04	0.00	0.05
	F4	1.97	11.6	2	3	7	98.7	0.08	0.12	0.05	0.08	-0.03	0.07	0.02	0.05	-0.08	0.05	0.01	0.08	-0.07	0.04	0.00	0.04	-0.09	0.05
	F5	14.43	85.2	2	2	4	100.2	-0.14	0.12	-0.08	0.08	-0.14	0.07	-0.04	0.05	-0.10	0.05	-0.02	0.08	-0.07	0.04	0.00	0.04	-0.08	0.05
	Calc. THg	16.93					104.0	-0.12	0.12	-0.07	0.08	-0.12	0.07	-0.04	0.05	-0.09	0.05	-0.01	0.08	-0.07	0.04	0.00	0.04	-0.08	0.05
EFK 18.0 125-250μm	F1	0.11	0.7	1	2	5	99.5	-0.05	0.12	-0.02	0.08	-0.09	0.07	-0.01	0.05	-0.09	0.05	-0.01	0.08	-0.07	0.04	0.00	0.04	-0.08	0.05
	F2	0.00	<0.1	1	1	1	96.7	-0.34	0.12	-0.20	0.08	-0.17	0.07	-0.11	0.05	-0.04	0.05	-0.04	0.08	-0.02	0.04	0.00	0.04	0.02	0.05
	F3	0.43	2.5	1	1	2	99.0	-0.14	0.12	-0.11	0.08	-0.09	0.07	-0.04	0.05	-0.04	0.05	0.02	0.08	-0.01	0.04	0.02	0.04	-0.01	0.05
	F4	2.16	12.6	1	1	2	97.3	0.07	0.12	0.06	0.08	-0.02	0.07	0.03	0.05	-0.05	0.05	-0.01	0.08	-0.06	0.04	0.00	0.04	-0.07	0.05
	F5	14.40	84.2	1	1	2	100.0	-0.19	0.12	-0.14	0.08	-0.18	0.07	-0.08	0.05	-0.10	0.05	0.01	0.08	-0.08	0.04	-0.01	0.04	-0.07	0.05
	Calc. THg	17.11					102.1	-0.16	0.12	-0.11	0.08	-0.16	0.07	-0.06	0.05	-0.09	0.05	0.01	0.08	-0.08	0.04	-0.01	0.04	-0.07	0.05
EFK 18.0 <125μm	F1	0.04	0.1	1	1	2	99.2	-0.37	0.12	-0.24	0.08	-0.23	0.07	-0.11	0.05	-0.09	0.05	-0.02	0.08	-0.05	0.04	0.01	0.04	-0.03	0.05
	F2	0.01	<0.1	1	1	1	96.9	-0.95	0.12	-0.64	0.08	-0.52	0.07	-0.33	0.05	-0.18	0.05	-0.01	0.08	-0.04	0.04	-0.01	0.04	-0.02	0.05
	F3	0.91	2.2	1	1	2	98.7	-0.23	0.12	-0.13	0.08	-0.12	0.07	-0.04	0.05	-0.02	0.05	-0.03	0.08	-0.02	0.04	0.02	0.04	0.01	0.05
	F4	6.29	15.4	1	1	2	99.8	0.34	0.12	0.24	0.08	0.09	0.07	0.11	0.05	-0.06	0.05	-0.01	0.08	-0.09	0.04	-0.01	0.04	-0.12	0.05
	F5	33.52	82.2	1	2	5	100.6	-0.03	0.12	-0.01	0.08	-0.10	0.07	-0.02	0.05	-0.09	0.05	-0.01	0.08	-0.08	0.04	-0.01	0.04	-0.09	0.05
	Calc. THg	40.76					103.8	0.02	0.12	0.02	0.08	-0.07	0.07	0.00	0.05	-0.09	0.05	-0.01	0.08	-0.08	0.04	-0.01	0.04	-0.09	0.05

[a] See Table S6.3 for notes.

Table S6.3: Mercury concentration and isotopic composition of sequential extractions of EFPC streambed sediment at EFK 8.7.[a]

Sample ID	Sequential Extraction	THg (μg g^{-1})	% of THg	n_1	n_2	n_3	% Rec.	δ^{204}Hg (‰)	2σ (‰)	δ^{202}Hg (‰)	2σ (‰)	δ^{201}Hg (‰)	2σ (‰)	δ^{200}Hg (‰)	2σ (‰)	δ^{199}Hg (‰)	2σ (‰)	Δ^{204}Hg (‰)	2σ (‰)	Δ^{201}Hg (‰)	2σ (‰)	Δ^{200}Hg (‰)	2σ (‰)	Δ^{199}Hg (‰)	2σ (‰)
EFK 8.7 1-2mm	F1	0.20	0.6	1	2	4	104.8	0.24	0.12	0.15	0.08	0.01	0.07	0.08	0.05	-0.07	0.05	0.00	0.08	-0.10	0.04	0.00	0.04	-0.11	0.05
	F2	0.04	0.1	1	2	2	101.0	0.78	0.12	0.49	0.08	0.28	0.07	0.26	0.05	-0.01	0.05	0.05	0.08	-0.09	0.04	0.02	0.04	-0.13	0.05
	F3	1.68	5.3	1	1	2	105.1	0.03	0.12	0.05	0.08	-0.04	0.07	0.03	0.05	-0.06	0.05	-0.04	0.08	-0.07	0.04	0.01	0.04	-0.07	0.05
	F4	2.05	6.5	1	1	2	102.5	0.22	0.12	0.13	0.08	-0.02	0.07	0.07	0.05	-0.08	0.05	0.02	0.08	-0.12	0.04	0.00	0.04	-0.11	0.05
	F5	27.80	87.5	1	1	2	107.7	-0.26	0.12	-0.17	0.08	-0.21	0.07	-0.09	0.05	-0.14	0.05	-0.01	0.08	-0.09	0.04	-0.01	0.04	-0.10	0.05
	Calc. THg	31.77					76.1	-0.21	0.12	-0.13	0.08	-0.19	0.07	-0.07	0.05	-0.13	0.05	-0.01	0.08	-0.09	0.04	0.00	0.04	-0.10	0.05
EFK 8.7 250μm-1mm	F1	0.24	1.0	1	2	5	104.7	0.54	0.12	0.34	0.08	0.13	0.07	0.14	0.05	-0.08	0.05	0.03	0.08	-0.13	0.04	-0.03	0.04	-0.16	0.05
	F2	0.02	0.1	1	1	1	101.0	0.74	0.12	0.44	0.08	0.23	0.07	0.23	0.05	-0.02	0.05	0.08	0.08	-0.10	0.04	0.01	0.04	-0.13	0.05
	F3	1.43	5.9	1	1	2	105.6	0.25	0.12	0.16	0.08	0.07	0.07	0.08	0.05	-0.05	0.05	0.01	0.08	-0.05	0.04	-0.01	0.04	-0.09	0.05
	F4	2.89	11.8	1	1	2	103.9	0.36	0.12	0.26	0.08	0.09	0.07	0.11	0.05	-0.08	0.05	-0.02	0.08	-0.11	0.04	-0.02	0.04	-0.15	0.05
	F5	19.84	81.3	1	1	2	105.3	-0.27	0.12	-0.18	0.08	-0.25	0.07	-0.12	0.05	-0.18	0.05	0.00	0.08	-0.11	0.04	-0.03	0.04	-0.14	0.05
	Calc. THg	24.41					105.2	-0.15	0.12	-0.10	0.08	-0.18	0.07	-0.08	0.05	-0.16	0.05	0.00	0.08	-0.11	0.04	-0.02	0.04	-0.14	0.05
EFK 8.7 125-250μm	F1	0.05	0.6	1	1	2	103.8	0.08	0.12	0.03	0.08	-0.06	0.07	-0.01	0.05	-0.11	0.05	0.03	0.08	-0.09	0.04	-0.02	0.04	-0.12	0.05
	F2	0.00	<0.1	1	1	1	93.9	-0.13	0.12	-0.08	0.08	-0.10	0.07	-0.11	0.05	-0.06	0.05	0.00	0.08	-0.04	0.04	-0.06	0.04	-0.04	0.05
	F3	1.62	20.5	1	1	2	103.9	-0.03	0.12	-0.04	0.08	-0.12	0.07	-0.02	0.05	-0.10	0.05	0.03	0.08	-0.09	0.04	0.00	0.04	-0.09	0.05
	F4	1.22	15.4	1	2	5	105.2	0.04	0.12	0.01	0.08	-0.09	0.07	0.00	0.05	-0.13	0.05	0.02	0.08	-0.10	0.04	0.00	0.04	-0.13	0.05
	F5	4.99	63.4	1	1	2	106.0	-0.37	0.12	-0.28	0.08	-0.28	0.07	-0.14	0.05	-0.18	0.05	0.05	0.08	-0.07	0.04	0.00	0.04	-0.11	0.05
	Calc. THg	7.88					110.4	-0.23	0.12	-0.18	0.08	-0.21	0.07	-0.10	0.05	-0.16	0.05	0.04	0.08	-0.08	0.04	0.00	0.04	-0.11	0.05
EFK 8.7 <125μm	F1	0.08	0.4	1	1	2	105.4	0.20	0.12	0.14	0.08	0.03	0.07	0.07	0.05	-0.06	0.05	-0.01	0.08	-0.07	0.04	0.00	0.04	-0.10	0.05
	F2	0.01	<0.1	1	1	1	99.8	-0.36	0.12	-0.25	0.08	-0.21	0.07	-0.12	0.05	-0.07	0.05	0.02	0.08	-0.02	0.04	0.01	0.04	-0.01	0.05
	F3	1.07	5.4	1	1	2	105.3	0.12	0.12	0.07	0.08	-0.03	0.07	0.02	0.05	-0.03	0.05	0.02	0.08	-0.08	0.04	-0.01	0.04	-0.05	0.05
	F4	6.31	31.7	1	1	2	104.5	0.49	0.12	0.32	0.08	0.13	0.07	0.16	0.05	-0.04	0.05	0.01	0.08	-0.11	0.04	0.00	0.04	-0.12	0.05
	F5	12.43	62.5	1	2	5	102.9	-0.03	0.12	-0.04	0.08	-0.12	0.07	-0.03	0.05	-0.12	0.05	0.02	0.08	-0.10	0.04	-0.01	0.04	-0.11	0.05
	Calc. THg	19.90					98.2	0.14	0.12	0.08	0.08	-0.04	0.07	0.03	0.05	-0.09	0.05	0.02	0.08	-0.10	0.04	-0.01	0.04	-0.11	0.05

[a] Five-step sequential extractions were performed on sieved streambed sediment collected from three sites along EFPC. Sample ID refers to the sediment size fraction and the sampling location identified by the number of kilometers upstream of the confluence of Poplar Creek and EFPC. Calc. THg refers to the calculated THg concentration and isotopic composition of the sediment based on the weighted average of sequential extractions. Here, n_1 denotes the number of sequential extraction sets, n_2 denotes the number of purge and trap preparations, and n_3 denotes the number of separate isotopic analyses on an individual preparation(s). The percent recovery (% Rec.) for F1 – F5 shows recovery of Hg during the purge and trap procedure for preparation of sequential extraction samples for isotope analysis. The percent recovery for Calc. THg is based on the sum of sequential extractions relative to THg concentrations obtained by combustions. The uncertainty in the isotopic composition of Hg in sequential extraction samples is represented by the average uncertainty (2SD) across all UM-Almadén analyses.

Table S7.1: Comparison of sequential extraction replicates.[a]

Sample ID	Sequential Extraction	Rep #	THg ($\mu g\ g^{-1}$)	% of THg	n_1	n_2	% Rec.	δ^{204}Hg (‰)	2σ (‰)	δ^{202}Hg (‰)	2σ (‰)	δ^{201}Hg (‰)	2σ (‰)	δ^{200}Hg (‰)	2σ (‰)	δ^{199}Hg (‰)	2σ (‰)	Δ^{204}Hg (‰)	2σ (‰)	Δ^{201}Hg (‰)	2σ (‰)	Δ^{200}Hg (‰)	2σ (‰)	Δ^{199}Hg (‰)	2σ (‰)
EFK 18.0 250µm-1mm	F1	Rep 1	0.32	1.89	1	2	101.5	-0.23	0.12	-0.18	0.08	-0.18	0.07	-0.08	0.05	-0.08	0.05	0.03	0.08	-0.04	0.04	0.01	0.04	-0.04	0.05
	F2	Rep 1	0.01	0.04	1	1	98.0	-0.22	0.12	-0.13	0.08	-0.15	0.07	-0.06	0.05	-0.07	0.05	-0.02	0.08	-0.05	0.04	0.00	0.04	-0.03	0.05
	F3	Rep 1	0.29	1.71	1	2	99.0	-0.24	0.12	-0.15	0.08	-0.13	0.07	-0.06	0.05	-0.04	0.05	-0.02	0.08	-0.02	0.04	0.01	0.04	-0.01	0.05
	F4	Rep 1	2.13	12.4	2	5	98.9	0.00	0.12	-0.01	0.08	-0.09	0.07	0.00	0.05	-0.09	0.05	0.01	0.08	-0.08	0.04	0.00	0.04	-0.09	0.05
	F5	Rep 1	14.36	83.9	1	2	101.9	-0.23	0.12	-0.14	0.08	-0.16	0.07	-0.07	0.05	-0.10	0.05	-0.02	0.08	-0.06	0.04	0.00	0.04	-0.07	0.05
	Calc. THg	Rep 1	17.11				105.1	-0.20	0.12	-0.12	0.08	-0.15	0.07	-0.06	0.05	-0.10	0.05	-0.02	0.08	-0.06	0.04	0.00	0.04	-0.07	0.05
	F1	Rep 2	0.13	0.78	1	2	99.4	-0.09	0.12	-0.07	0.08	-0.13	0.07	-0.03	0.05	-0.10	0.05	0.01	0.08	-0.08	0.04	0.01	0.04	-0.08	0.05
	F2	Rep 2	0.02	0.11	1	1	96.4	-0.28	0.12	-0.19	0.08	-0.21	0.07	-0.10	0.05	-0.09	0.05	-0.01	0.08	-0.07	0.04	-0.01	0.04	-0.04	0.05
	F3	Rep 2	0.30	1.77	1	2	100.9	-0.29	0.12	-0.19	0.08	-0.17	0.07	-0.07	0.05	-0.05	0.05	-0.01	0.08	-0.02	0.04	0.03	0.04	0.00	0.05
	F4	Rep 2	1.82	10.8	1	2	98.4	0.16	0.12	0.11	0.08	0.02	0.07	0.05	0.05	-0.06	0.05	0.00	0.08	-0.06	0.04	-0.01	0.04	-0.09	0.05
	F5	Rep 2	14.50	86.5	1	2	98.5	-0.06	0.12	-0.03	0.08	-0.11	0.07	-0.02	0.05	-0.09	0.05	-0.01	0.08	-0.09	0.04	0.00	0.04	-0.08	0.05
	Calc. THg	Rep 2	16.76				102.9	-0.04	0.12	-0.02	0.08	-0.09	0.07	-0.01	0.05	-0.09	0.05	-0.01	0.08	-0.08	0.04	0.00	0.04	-0.08	0.05
EFK 8.7 1-2mm	F1	Rep 1	0.20	0.6	2	4	104.8	0.24	0.12	0.15	0.08	0.01	0.07	0.08	0.05	-0.07	0.05	0.00	0.08	-0.10	0.04	0.00	0.04	-0.11	0.05
	F2	Rep 1	0.04	0.1	2	2	101.0	0.78	0.12	0.49	0.08	0.28	0.07	0.26	0.05	-0.01	0.05	0.05	0.08	-0.09	0.04	0.02	0.04	-0.13	0.05
	F1	Rep 2	0.34	-	1	2	105.6	0.23	0.12	0.15	0.08	-0.01	0.07	0.07	0.05	-0.10	0.05	0.01	0.08	-0.12	0.04	0.00	0.04	-0.14	0.05
	F2	Rep 2	0.14	-	1	2	105.3	0.54	0.12	0.35	0.08	0.15	0.07	0.18	0.05	-0.04	0.05	0.02	0.08	-0.11	0.04	0.01	0.04	-0.13	0.05
EFK 8.7 250µm-1mm	F1	Rep 1	0.24	1.0	2	5	104.7	0.54	0.12	0.34	0.08	0.13	0.07	0.14	0.05	-0.08	0.05	0.03	0.08	-0.13	0.04	-0.03	0.04	-0.16	0.05
	F2	Rep 1	0.02	0.1	1	1	101.0	0.74	0.12	0.44	0.08	0.23	0.07	0.23	0.05	-0.02	0.05	0.08	0.08	-0.10	0.04	0.01	0.04	-0.13	0.05
	F1	Rep 2	0.34	-	1	2	102.8	-0.16	0.12	-0.11	0.08	-0.17	0.07	-0.07	0.05	-0.14	0.05	0.01	0.08	-0.08	0.04	-0.01	0.04	-0.11	0.05
	F2	Rep 2	0.04	-	1	1	104.8	0.66	0.12	0.40	0.08	0.20	0.07	0.20	0.05	-0.04	0.05	0.07	0.08	-0.09	0.04	0.00	0.04	-0.14	0.05
EFK 8.7 125-250µm	F1	Rep 1	0.05	0.6	1	2	103.8	0.08	0.12	0.03	0.08	-0.06	0.07	-0.01	0.05	-0.11	0.05	0.03	0.08	-0.09	0.04	-0.02	0.04	-0.12	0.05
	F1	Rep 2	0.04	-	1	1	103.0	0.17	0.12	0.09	0.08	-0.01	0.07	0.06	0.05	-0.09	0.05	0.04	0.08	-0.07	0.04	0.01	0.04	-0.11	0.05
EFK 8.7 <125µm	F1	Rep 1	0.08	0.4	1	2	105.4	0.20	0.12	0.14	0.08	0.03	0.07	0.07	0.05	-0.06	0.05	-0.01	0.08	-0.07	0.04	0.00	0.04	-0.10	0.05
	F1	Rep 2	0.04	-	1	1	104.4	0.09	0.12	0.06	0.08	-0.06	0.07	0.00	0.05	-0.10	0.05	0.00	0.08	-0.11	0.04	-0.03	0.04	-0.12	0.05

[a] See Table S7.2 for notes.

Table S7.2: Comparison of sequential extraction replicates.[a]

Sample ID	Sequential Extraction	Rep #	THg ($\mu g\ g^{-1}$)	% of THg	n_1	n_2	% Rec.	δ^{204}Hg (‰)	2σ (‰)	δ^{202}Hg (‰)	2σ (‰)	δ^{201}Hg (‰)	2σ (‰)	δ^{200}Hg (‰)	2σ (‰)	δ^{199}Hg (‰)	2σ (‰)	Δ^{204}Hg (‰)	2σ (‰)	Δ^{201}Hg (‰)	2σ (‰)	Δ^{200}Hg (‰)	2σ (‰)	Δ^{199}Hg (‰)	2σ (‰)
NIST SRM 2711 (Montana Soil)	F1	Rep 1	0.05	0.9	1	1	98.0	-0.34	0.12	-0.28	0.08	-0.35	0.07	-0.14	0.05	-0.26	0.05	0.07	0.08	-0.14	0.04	0.00	0.04	-0.19	0.05
	F2	Rep 1	0.00	0.1				Insufficient mercury for isotopic analysis.																	
	F3	Rep 1	0.15	2.5	2	2	94.6	-0.29	0.12	-0.22	0.08	-0.32	0.07	-0.12	0.05	-0.26	0.05	0.04	0.08	-0.15	0.04	-0.01	0.04	-0.20	0.05
	F4	Rep 1	4.03	69.0	2	3	97.0	-0.06	0.12	-0.05	0.08	-0.23	0.07	-0.03	0.05	-0.24	0.05	0.01	0.08	-0.19	0.04	-0.01	0.04	-0.22	0.05
	F5	Rep 1	1.61	27.5	2	3	97.3	-0.61	0.12	-0.42	0.08	-0.50	0.07	-0.22	0.05	-0.31	0.05	0.01	0.08	-0.19	0.04	-0.01	0.04	-0.21	0.05
	Calc. THg	Rep 1	5.84				93.4	-0.22	0.12	-0.16	0.08	-0.31	0.07	-0.09	0.05	-0.26	0.05	0.02	0.08	-0.19	0.04	-0.01	0.04	-0.22	0.05
	F1	Rep 2	0.05	0.8	2	2	99.0	-0.32	0.12	-0.21	0.08	-0.33	0.07	-0.11	0.05	-0.25	0.05	-0.01	0.08	-0.17	0.04	0.00	0.04	-0.20	0.05
	F2	Rep 2	0.00	0.1	1	1	93.8	-1.41	0.12	-0.94	0.08	-0.75	0.07	-0.42	0.05	-0.37	0.05	-0.01	0.08	-0.04	0.04	0.05	0.04	-0.13	0.05
	F3	Rep 2	0.15	2.5	1	2	103.3	-0.43	0.12	-0.28	0.08	-0.35	0.07	-0.16	0.05	-0.25	0.05	-0.01	0.08	-0.14	0.04	-0.02	0.04	-0.18	0.05
	F4	Rep 2	4.25	70.5	1	2	95.3	-0.18	0.12	-0.15	0.08	-0.32	0.07	-0.08	0.05	-0.27	0.05	0.03	0.08	-0.21	0.04	0.00	0.04	-0.23	0.05
	F5	Rep 2	1.58	26.2	1	2	96.9	-0.65	0.12	-0.46	0.08	-0.51	0.07	-0.23	0.05	-0.33	0.05	0.04	0.08	-0.16	0.04	0.00	0.04	-0.22	0.05
	Calc. THg	Rep 2	6.03				96.5	-0.31	0.12	-0.23	0.08	-0.37	0.07	-0.12	0.05	-0.29	0.05	0.03	0.08	-0.19	0.04	0.00	0.04	-0.23	0.05
	F1	Rep 3	0.12	2.1	1	2	98.1	-0.24	0.12	-0.15	0.08	-0.30	0.07	-0.06	0.05	-0.25	0.05	-0.01	0.08	-0.19	0.04	0.02	0.04	-0.21	0.05
	F2	Rep 3	0.00	0.1	1	1	98.1	-0.94	0.12	-0.68	0.08	-0.58	0.07	-0.28	0.05	-0.25	0.05	0.07	0.08	-0.07	0.04	0.06	0.04	-0.08	0.05
	F3	Rep 3	0.17	2.8	2	5	94.6	-0.37	0.12	-0.23	0.08	-0.33	0.07	-0.12	0.05	-0.24	0.05	-0.02	0.08	-0.15	0.04	0.00	0.04	-0.19	0.05
	F4	Rep 3	3.92	65.3	1	2	100.9	-0.13	0.12	-0.07	0.08	-0.26	0.07	-0.03	0.05	-0.25	0.05	-0.02	0.08	-0.20	0.04	0.00	0.04	-0.23	0.05
	F5	Rep 3	1.79	29.8	1	2	102.5	-0.62	0.12	-0.41	0.08	-0.50	0.07	-0.22	0.05	-0.32	0.05	0.00	0.08	-0.19	0.04	-0.02	0.04	-0.22	0.05
	Calc. THg	Rep 3	6.01				96.1	-0.28	0.12	-0.18	0.08	-0.33	0.07	-0.09	0.05	-0.27	0.05	-0.02	0.08	-0.20	0.04	0.00	0.04	-0.23	0.05

[a] Five-step sequential extractions were performed on standard reference materials and on sieved streambed sediment collected from three sites along EFPC. Sample ID refers to the sediment size fraction and the sampling location identified by the number of kilometers upstream of the confluence of Poplar Creek and EFPC. Calc. THg refers to the calculated THg concentration and isotopic composition of the sediment based on the weighted average of sequential extractions. Rep # refers to sequential extraction replicates on separate aliquots of sediment. Here, n_1 denotes the number of purge and trap preparations, and n_2 denotes the number of separate isotopic analyses on an individual preparation(s). The percent recovery (% Rec.) for F1 – F5 shows recovery of Hg during the purge and trap procedure for preparation of sequential extraction samples for isotope analysis. The percent recovery for Calc. THg of EFPC sediment is based on the sum of sequential extractions relative to THg concentrations obtained by combustions. The percent recovery for Calc. THg of reference materials is based on the sum of sequential extractions relative to certified values. The uncertainty in the isotopic composition of Hg in sequential extraction samples is represented by the average uncertainty (2SD) across all UM-Almadén analyses.

Table S8: Results of loss-on-ignition.[a]

Site ID	Size Fraction	Percent mass loss after 500ºC (i.e., organic matter) (± 1SD, n=2)	Percent mass loss after 800ºC (i.e., carbonates) (± 1SD, n=2)	Calculated percent organic carbon (± 1SD, n=2)	Total Hg per mass of organic carbon, μg THg g^{-1} OC (± 1SD, n=2)	F3-extracted Hg per mass of organic matter carbon, μg Hg_{F3} g^{-1} OC (± 1SD, n=2)
EFK 22.3	1-2mm	4.39 ± 0.24	2.56 ± 0.65	2.19 ± 0.12	970 ± 504	34.2 ± 1.9
	250μm-1mm	3.09	2.78	1.54	1175	19.0
	125-250μm	4.15	2.28	2.08	1259	30.1
	<125μm	6.04	2.37	3.02	1159	29.4
EFK 18.0	1-2mm	4.14	3.70	2.07	1188	14.6
	250μm-1mm	2.71 ± 0.19	2.55 ± 0.07	1.36 ± 0.09	1205 ± 133	21.8 ± 1.5
	125-250μm	2.57	2.43	1.28	1306	33.6
	<125μm	7.15	2.81	3.57	1099	25.4
EFK 15.8	1-2mm	3.18	1.17	1.59	1656	-
	250μm-1mm	2.18	0.91	1.09	2174	-
	125-250μm	2.02 ± 0.27	0.87 ± 0.08	1.01 ± 0.13	1510 ± 238	-
	<125μm	5.04	1.61	2.52	846	-
EFK 8.7	1-2mm	1.77	0.50	0.89	4715	190
	250μm-1mm	1.22	0.24	0.61	3802	234
	125-250μm	1.95	0.13	0.98	731	166
	<125μm	5.74 ± 0.01	0.87 ± 0.04	2.87 ± 0.01	706 ± 5	37.2 ± 0.1
NIST SRM 1944 (NY/NJ Waterway Sediment)		8.77 ± 0.06	1.73 ± 0.14	4.38 ± 0.03	82 ± 1	7.0 ± 0.0

[a] A loss-on-ignition procedure was performed on sieved streambed sediment collected from four sites along EFPC. Site ID refers to the sampling location identified by the number of kilometers upstream of the confluence of Poplar Creek and EFPC. Percentages of mass loss after 500ºC and 800ºC are thought to primarily represent percentages of organic matter and carbonates, respectively. Percentages of organic carbon were calculated by dividing the percentages of mass lost after 500ºC by 2. Concentrations of total Hg per mass of organic carbon (μg THg g^{-1} OC) were calculated using THg concentrations measured via combustion (Table S3, Table S4), and for values represented by an average ± 1SD, these averages were based on independently ground sediment samples which were each analyzed for THg and OC concentrations. Concentrations of F3-extracted Hg per mass of organic carbon (μg Hg_{F3} g^{-1} OC) were calculated using Hg concentrations of the F3 sequential extractions (Table S6), and for values represented by an average ± 1SD, these averages were based on replicate OC concentrations.

Table S9: Results of two-tailed paired samples t-tests for EFPC sediment sequential extraction results.

Sampling site	Group 1[a]	Group 2[a]	Mean Offset in δ^{202}Hg (1SD)	Mean Offset in Δ^{199}Hg (1SD)	p-value[b] (δ^{202}Hg)	p-value[c] (Δ^{199}Hg)
EFK 22.3	F2	F3	-0.20 ± 0.22‰	-0.01 ± 0.02‰	0.175	0.361
EFK 18.0	F2	F3	-0.13 ± 0.25‰	-0.02 ± 0.03‰	0.366	0.331
EFK 8.7	F2	F3	0.09 ± 0.34‰	0.00 ± 0.06‰	0.631	0.952
EFK 22.3	F4	F5	0.16 ± 0.12‰	0.00 ± 0.02‰	0.075	0.629
EFK 18.0	F4	F5	0.16 ± 0.08‰	-0.01 ± 0.02‰	0.030*	0.651
EFK 8.7	F4	F5	0.34 ± 0.07‰	-0.01 ± 0.00‰	0.002*	0.010*

[a] Group 1 and Group 2 represent different pools of extracted Hg (n=4 for each group).
[b] Null hypothesis: The mean offset in δ^{202}Hg values of different pools of extracted Hg within individual sediment size fractions (i.e., paired samples) is zero.
[c] Null hypothesis: The mean offset in Δ^{199}Hg values of different pools of extracted Hg within individual sediment size fractions (i.e., paired samples) is zero.
* indicates that the mean offset in delta values of different pools of extracted Hg within individual sediment size fractions is significantly different from zero ($p < 0.05$).

Table S10: Results of two-tailed paired samples t-tests for EFPC sediment sequential extraction results using the Bonferroni correction for multiple comparisons.

Sampling site	Group 1[a]	Group 2[a]	Mean Offset in δ^{202}Hg (1SD)	Mean Offset in Δ^{199}Hg (1SD)	p-value$_{adjusted}$[b] (δ^{202}Hg)	p-value$_{adjusted}$[c] (Δ^{199}Hg)
EFK 22.3	F1	F2F3	0.04 ± 0.29‰	-0.06 ± 0.02‰	1.000	0.034*
EFK 22.3	F1	F4F5	-0.19 ± 0.26‰	0.03 ± 0.03‰	0.730	0.960
EFK 22.3	F2F3	F4F5	-0.23 ± 0.05‰	0.09 ± 0.02‰	0.007*	0.011*
EFK 18.0	F1	F2F3	0.04 ± 0.13‰	-0.05 ± 0.02‰	1.000	0.096
EFK 18.0	F1	F4F5	-0.21 ± 0.19‰	0.03 ± 0.04‰	0.354	0.611
EFK 18.0	F2F3	F4F5	-0.25 ± 0.17‰	0.08 ± 0.02‰	0.184	0.011*
EFK 8.7	F1	F2F3	0.06 ± 0.14‰	-0.05 ± 0.03‰	1.000	0.138
EFK 8.7	F1	F4F5	0.16 ± 0.12‰	0.00 ± 0.01‰	0.242	1.000
EFK 8.7	F2F3	F4F5	0.10 ± 0.24‰	0.04 ± 0.04‰	1.000	0.258

[a] Group 1 and Group 2 represent different pools of extracted Hg (n=4 for each group). The isotopic composition of groups F2F3 and F4F5 was calculated as the non-weighted average isotopic composition of the two pools of sequentially extracted Hg within the group.

[b] Null hypothesis: The mean offset in δ^{202}Hg values of different pools of extracted Hg within individual sediment size fractions (i.e., paired samples) is zero.

[c] Null hypothesis: The mean offset in Δ^{199}Hg values of different pools of extracted Hg within individual sediment size fractions (i.e., paired samples) is zero.

* indicates that the mean offset in delta values of different pools of extracted Hg within individual sediment size fractions is significantly different from zero ($p < 0.05$).

Table S11: Results of two-tailed paired samples t-tests for EFPC sediment sequential extraction results using the Bonferroni correction for multiple comparisons, excluding the F2 mercury pool.

Sampling site	Group 1[a]	Group 2[a]	Mean Offset in δ^{202}Hg (1SD)	Mean Offset in Δ^{199}Hg (1SD)	p-value$_{adjusted}$[b] (δ^{202}Hg)	p-value$_{adjusted}$[c] (Δ^{199}Hg)
EFK 22.3	F1	F3	-0.06 ± 0.35‰	-0.07 ± 0.02‰	1.000	0.017*
EFK 22.3	F1	F4F5	-0.19 ± 0.26‰	0.03 ± 0.03‰	0.730	0.960
EFK 22.3	F3	F4F5	-0.13 ± 0.12‰	0.09 ± 0.02‰	0.371	0.011*
EFK 18.0	F1	F3	-0.02 ± 0.10‰	-0.06 ± 0.01‰	1.000	0.004*
EFK 18.0	F1	F4F5	-0.21 ± 0.19‰	0.03 ± 0.04‰	0.364	0.611
EFK 18.0	F3	F4F5	-0.18 ± 0.09‰	0.09 ± 0.03‰	0.087	0.028*
EFK 8.7	F1	F3	0.11 ± 0.05‰	-0.05 ± 0.02‰	0.068	0.055
EFK 8.7	F1	F4F5	0.16 ± 0.12‰	0.00 ± 0.01‰	0.242	1.000
EFK 8.7	F3	F4F5	0.05 ± 0.09‰	0.05 ± 0.02‰	0.921	0.032*

[a] Group 1 and Group 2 represent different pools of extracted Hg (n=4 for each group). The isotopic composition of group F4F5 was calculated as the non-weighted average isotopic composition of the two pools of sequentially extracted Hg within the group.

[b] Null hypothesis: The mean offset in δ^{202}Hg values of different pools of extracted Hg within individual sediment size fractions (i.e., paired samples) is zero.

[c] Null hypothesis: The mean offset in Δ^{199}Hg values of different pools of extracted Hg within individual sediment size fractions (i.e., paired samples) is zero.

* indicates that the mean offset in delta values of different pools of extracted Hg within individual sediment size fractions is significantly different from zero ($p < 0.05$).

Table S12: Results of two-tailed independent samples t-tests for EFPC sediment sequential extraction results.

Sampling site	G1[a]	G2[a]	Mean δ^{202}Hg Group 1 (1SD)	Mean δ^{202}Hg Group 2 (1SD)	Mean Δ^{199}Hg Group 1 (1SD)	Mean Δ^{199}Hg Group 2 (1SD)	p-value[b] (δ^{202}Hg)	p-value[c] (Δ^{199}Hg)
EFK 22.3	F2	F3	-0.24 ± 0.29‰	-0.04 ± 0.31‰	0.00 ± 0.03‰	0.01 ± 0.02‰	0.389	0.553
EFK 18.0	F2	F3	-0.23 ± 0.30‰	-0.10 ± 0.08‰	-0.03 ± 0.03‰	-0.01± 0.02‰	0.416	0.362
EFK 8.7	F2	F3	0.15 ± 0.37‰	0.06 ± 0.08‰	-0.08 ± 0.06‰	-0.08 ± 0.02‰	0.653	0.959
EFK 22.3	F4	F5	0.17 ± 0.21‰	0.01 ± 0.27‰	-0.09 ± 0.03‰	-0.08 ± 0.03‰	0.392	0.853
EFK 18.0	F4	F5	0.17 ± 0.14‰	0.01 ± 0.18‰	-0.10 ± 0.03‰	-0.09 ± 0.03‰	0.207	0.362
EFK 8.7	F4	F5	0.18 ± 0.14‰	-0.17 ± 0.10‰	-0.13 ± 0.02‰	-0.11 ± 0.02‰	0.006*	0.317

[a] Group 1 (G1) and Group 2 (G2), representing different pools of extracted Hg (n=4 for each group).
[b] Null hypothesis: Across all sediment size fractions within a site (i.e., unpaired samples), the difference between mean δ^{202}Hg values of different pools of extracted Hg is zero.
[b] Null hypothesis: Across all sediment size fractions within a site (i.e., unpaired samples), the difference between mean Δ^{199}Hg values of different pools of extracted Hg is zero.
* indicates that the mean delta values of different pools of extracted Hg are significantly different from each other ($p < 0.05$).

Table S13: Results of independent samples Tukey honestly significant difference (HSD) tests for multiple comparisons for EFPC sediment sequential extraction results.

Sampling site	G1[a]	G2[a]	Mean δ^{202}Hg Group 1 (1SD)	Mean δ^{202}Hg Group 2 (1SD)	Mean Δ^{199}Hg Group 1 (1SD)	Mean Δ^{199}Hg Group 2 (1SD)	p-value$_{adj.}$[b] (δ^{202}Hg)	p-value$_{adj.}$[c] (Δ^{199}Hg)
EFK 22.3	F1	F2F3	-0.10 ± 0.08‰	-0.14 ± 0.30‰	-0.06 ± 0.01‰	0.00 ± 0.03‰	0.900	0.018*
EFK 22.3	F1	F4F5	-0.10 ± 0.08‰	0.09 ± 0.24‰	-0.06 ± 0.01‰	-0.08 ± 0.03‰	0.469	0.441
EFK 22.3	F2F3	F4F5	-0.14 ± 0.30‰	0.09 ± 0.24‰	0.00 ± 0.03‰	-0.08 ± 0.03‰	0.336	0.003*
EFK 18.0	F1	F2F3	-0.12 ± 0.09‰	-0.16 ± 0.21‰	-0.06 ± 0.02‰	-0.02 ± 0.03‰	0.900	0.055
EFK 18.0	F1	F4F5	-0.12 ± 0.09‰	0.09 ± 0.17‰	-0.06 ± 0.02‰	-0.10 ± 0.03‰	0.165	0.215
EFK 18.0	F2F3	F4F5	-0.16 ± 0.21‰	0.09 ± 0.17‰	-0.02 ± 0.03‰	-0.10 ± 0.03‰	0.085	0.004*
EFK 8.7	F1	F2F3	0.17 ± 0.13‰	0.11 ± 0.25‰	-0.12 ± 0.03‰	-0.08 ± 0.04‰	0.833	0.126
EFK 8.7	F1	F4F5	0.17 ± 0.13‰	0.01 ± 0.21‰	-0.12 ± 0.03‰	-0.12 ± 0.02‰	0.357	0.900
EFK 8.7	F2F3	F4F5	0.11 ± 0.25‰	0.01 ± 0.21‰	-0.08 ± 0.04‰	-0.12 ± 0.02‰	0.652	0.139

[a] Group 1 (G1) and Group 2 (G2), representing different pools of extracted Hg (n=4 for each group). The isotopic composition of groups F2F3 and F4F5 was calculated as the non-weighted average isotopic composition of the two pools of sequentially extracted Hg within the group.

[b] Null hypothesis: Across all sediment size fractions within a site (i.e., unpaired samples), the difference between mean δ^{202}Hg values of different pools of extracted Hg is zero.

[b] Null hypothesis: Across all sediment size fractions within a site (i.e., unpaired samples), the difference between mean Δ^{199}Hg values of different pools of extracted Hg is zero.

* indicates that the mean delta values of different pools of extracted Hg are significantly different from each other ($p < 0.05$).

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