Supporting Information for

Enhanced removal of dissolved Hg(II), Cd(II) and Au(III) from water by Bacillus subtilis

bacterial biomass containing an elevated concentration of sulfhydryl sites

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Figure S2. Langmuir model fits to the measured extents of Hg(II), Cd(II) and Au(III) sorption onto HSBS (blue dash lines) and LSBS (pink dash lines) as a function of metal concentration.

Table S1. Characteristics of the Dowex[®] Marathon C cation exchange resin provided by the manufacturer. The value of total exchange sites is in terms of dry weight of the resin.

Matrix	Functional Group	Total Exchange Sites	Operating pH	Resin Diameter
Polystyrene divinylbenzene	$-SO_3$ Na ⁺	$\geq 1810 \ \mu mol/g$	0-14	0.6 mm

Table S2. Sulfhydryl site and total binding site concentrations within the cell envelopes of *B*. *subtilis* biomass that was grown in the TSB medium with (HSBS) or without (LSBS) the addition of 50 g/L of glucose.^{1, 2} Concentrations are reported in terms of dry weight of the biomass.

	Sulfhydryl Sites	Total Binding Sites
	µmol/g	µmol/g
LSBS	108±33	1205±80
HSBS	436±38	1290±47

Table S3. Langmuir modeling results for the sorption isotherms for Hg(II), Cd(II) and Au(III) onto HSBS and LSBS. The Langmuir model is expressed as $q_e=bq_mC_e/(1+bC_e)$, where q_e is the equilibrium sorption concentration for each metal (µmol/g), C_e is the equilibrium concentration of each metal, and q_m is the maximum monolayer sorption capacity (µmol/g).

Metal	Sorbent	q _m µmol∕g	R^2
Au	HSBS	605	0.9779
	LSBS	368	0.9402
C.1	HSBS	111	0.9818
Cu	LSBS	61	0.9901
Hg	HSBS	59	0.6079
	LSBS	11	0.9623

Table S4. Summary of the calculated percentage of each major species for each metal in sorbentfree systems at pH 6.0 and in the presence of 0.1 M NaCl. The speciation calculations for the Hg(II), Cd(II) and Au(III) systems were conducted using FITEQL³ considering all pertinent aqueous metal-ligand complexes.^{4, 5} The relative percentages of the dominant aqueous species do not vary significantly over the metal concentration range that was used in sorption experiments (10 to 500 μ M).

Metal	Major Species (percentage)
Hg	HgCl ₂ ⁰ (41%), HgCl ₃ ⁻ (35%), HgCl ₄ ²⁻ (23%)
Cd	$CdCl^{+}(64\%), Cd^{2+}(18\%), CdCl_{2}^{0}(18\%)$
Au	Au(OH) ₂ Cl ₂ ⁻ (51%), Au(OH)Cl ₃ ⁻ (44%)



Figure S1. Reaction of qBBr with sulfhydryl binding sites within bacterial cell envelopes.^{1,6}



Figure S2. Langmuir model fits to the measured extents of Hg(II), Cd(II) and Au(III) sorption onto HSBS (blue dash lines) and LSBS (pink dash lines) as a function of metal concentration.

References

1. Yu, Q.; Szymanowski, J.; Myneni, S. C. B.; Fein, J. B., Characterization of sulfhydryl sites within bacterial cell envelopes using selective site-blocking and potentiometric titrations. *Chem Geol* **2014**, *373*, 50-58.

2. Yu, Q.; Fein, J. B., Controls on Bacterial Cell Envelope Sulfhydryl Site Concentrations: The Effect of Glucose Concentration During Growth. *Environ Sci Technol* **2017**, *51*, (13), 7395-7402.

3. Westall, J. C., *FITEQL*, A computer program for determination of chemical equilibrium constants from experimental data. Version 2.0. Report 82-02. Dept. Chem., Oregon St. Univ., Corvallis, OR, USA., 1982.

4. Smith, R. M.; Martell, A. E. Critical Stability Constants; Plenum Press: New York, 1976

5. Usher, A.; McPhail, D. C.; Brugger, J., A spectrophotometric study of aqueous Au(III)halidehydroxide complexes at 25-80 degrees C. *Geochim Cosmochim Ac* **2009**, 73, (11), 3359-3380.

6. Kosower, N.S., Kosower, E.M., Newton, G.L., Ranney, H.M., Bimane fluorescent labels: labeling of normal human red cells under physiological conditions. *Proc. Natl. Acad. Sci. U. S. A.* **1979**, 76, 7, 3382–3386.