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Supporting Information

Probing the Coordination Environment of the Human Copper Chaperone HAH1: Characterization of Hg^{II}-Bridged Homodimeric Species in Solution

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Table S1. Buffer composition for each pH of the Hg^{II}-HAH1 gel filtration studies. Note: all buffers were degassed/filtered prior to use, and the pH was adjusted for temperature dependent changes (studies done at 4°C).

Buffer (pH):	Composition:
Phosphate Buffer (7.5)	100 mM phpsphate buffer (Na_2HPO_4, and NaH_2PO_4), 200 mM NaCl, 1 mM TCEP
Phosphate Buffer (8.5)	100 mM phosphate buffer (Na_2PO_4, and NaH_2PO_4), 200 mM NaCl, 1 mM TCEP
CHES Buffer (9.4)	100 mM CHES, 200 mM NaCl, 1 mM TCEP

Table S2. List of calibration standards used on the Superdex 75 HiLoad 16/600 gel filtration column to determine apparent molecular weights of the different aggregation states of apo and Hg^{II}-HAH1.

Calibration Standard	Molecular Weight (Daltons)
Aprotinin	6,500
Ribonuclease A	13,700
Carbonic Anhydrase	29,000
Ovalbumin	43,000
Blue Dextran	2,000,000

Table S3. Compilation of the elution volumes and apparent masses of Hg^{II}-HAH1 at the various pH values.

pH	Elution Volume (mL)	Apparent Mass (kDa)
7.5	84.6	9.7
8.5	83.1	10.5
9.4	81.7	11.4

Table S4. Parameters fitted to the ^{199m}Hg PAC data. The numbers in the parenthesis are the standard deviations of the fitted parameters. Initially, the data recorded at pH 7.5 were analyzed, giving one nuclear quadrupole interaction (NQI). Next, the parameters for this NQI were fixed, and the data recorded at pH 9.4 were analyzed with two additional NQIs, giving in total three NQIs. Finally, the data recorded at pH 8.5 were analyzed with these three NQIs keeping all parameters except the amplitudes (A) fixed. There are other ways of analyzing the data that give equally good fits.

pH	HAH1/ Hg^{II}	ν_Q [rad/ns]	η	δ ×100	$1/\tau_C$ [μs^{-1}]	A ×100	χ_r^2
7.5	2:1	1.42(2)	0f	5(2)	51(25)	15(2)	0.65
8.5	2:1	1.42f 0.9f 0.4f	0f 1f 0.7f	5f 21f 41f	51f 51f 51f	8(1) 4(1) 2.2(5)	0.84
9.4	2:1	1.42f 0.9(3) 0.4(4)	0f 1f 0.7(7)	5f 21(27) 41(63)	51f 51f 51f	5(3) 3(4) 5(3)	0.83