Electronic Supplementary Material (ESI) for Faraday Discussions.

Supplementary Information

Fine-tuning non-covalent interactions between hybrid metal-oxo clusters and proteins

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Table of Contents

NMR

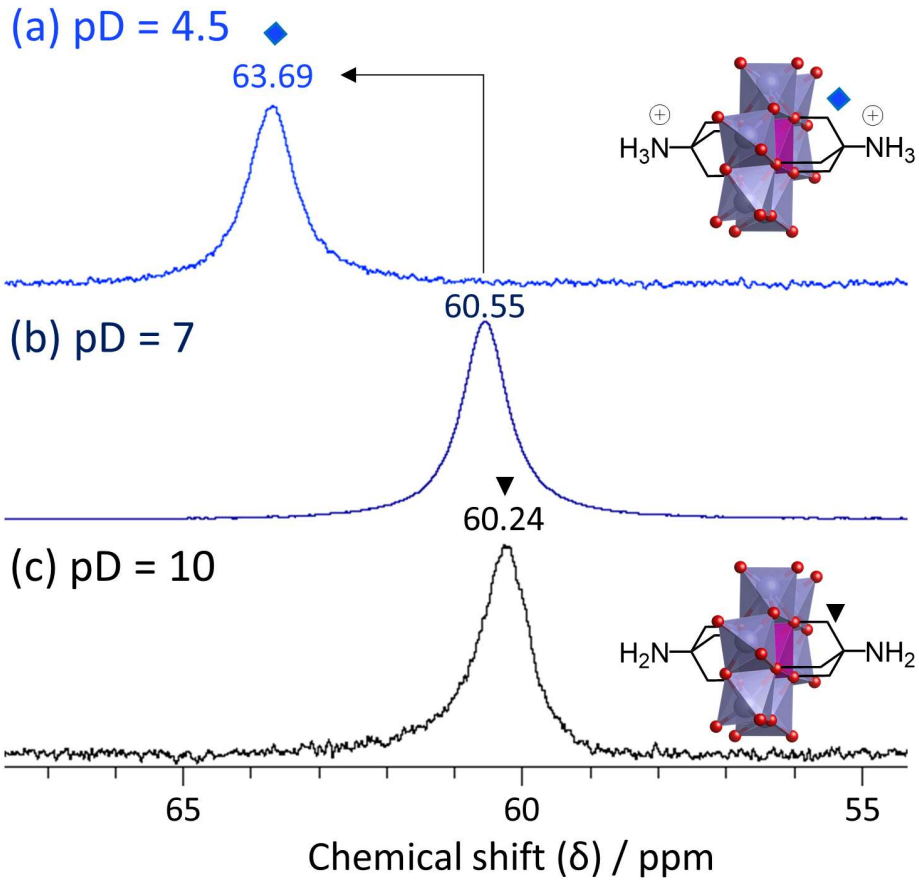

Figure S1 (a) ^{1}H NMR spectra of AE-NH_2 in (a) 100 mM acetate buffer at pD 4.5, (b) D_2O at pD 7 and (c) 100 mM carbonate buffer at pD 10 showing the peak corresponding to the paramagnetically deshielded -OCH_2- groups bound to the POM.

Fluorescence Emission Spectra

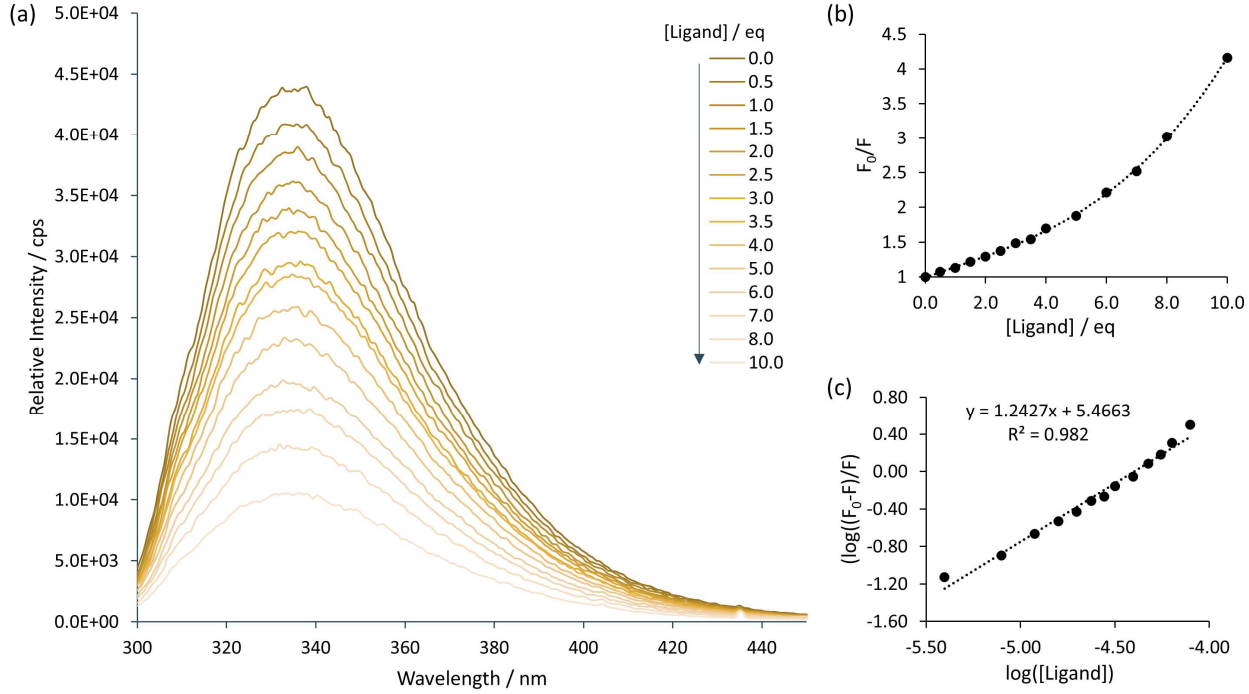

Figure S2 (a) Fluorescence emission spectra of 8 µM HEWL in PBS(1X) buffer at pH 7.4 with increasing amounts of AE-Biot (0 – 10 eq). (b) Stern-Volmer and (c) derived Stern-Volmer plots.

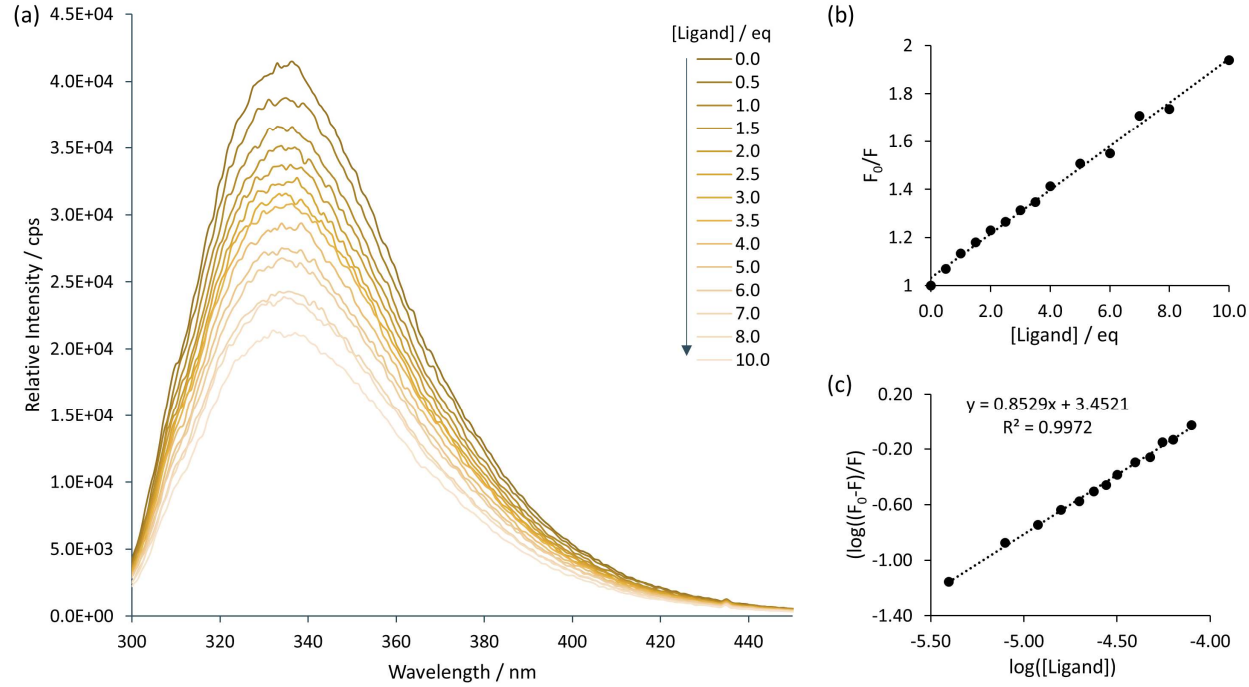

Figure S3 (a) Fluorescence emission spectra of 8 µM HEWL in PBS(1X) buffer at pH 7.4 with increasing amounts of AE-NH_2 (0 – 10 eq). (b) Stern-Volmer and (c) derived Stern-Volmer plots.

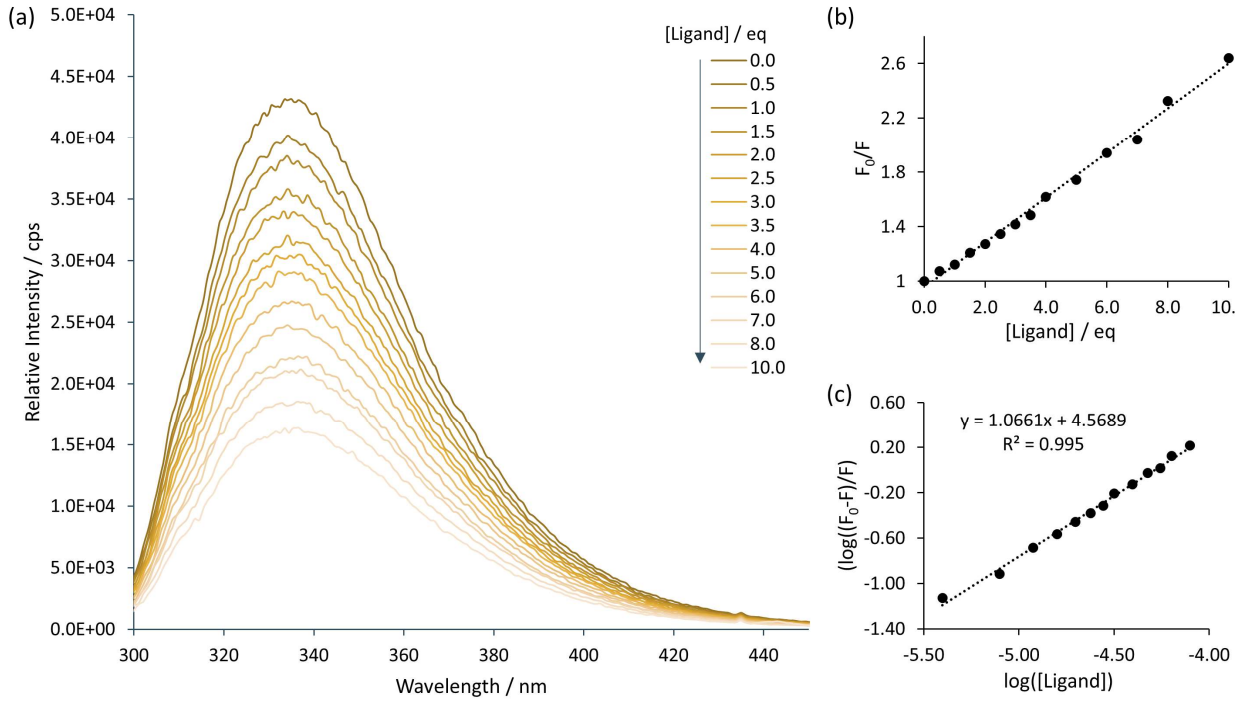

Figure S4 (a) Fluorescence emission spectra of 8 µM HEWL in PBS(1X) buffer at pH 7.4 with increasing amounts of AE-CH_3 (0 – 10 eq). (b) Stern-Volmer and (c) derived Stern-Volmer plots.

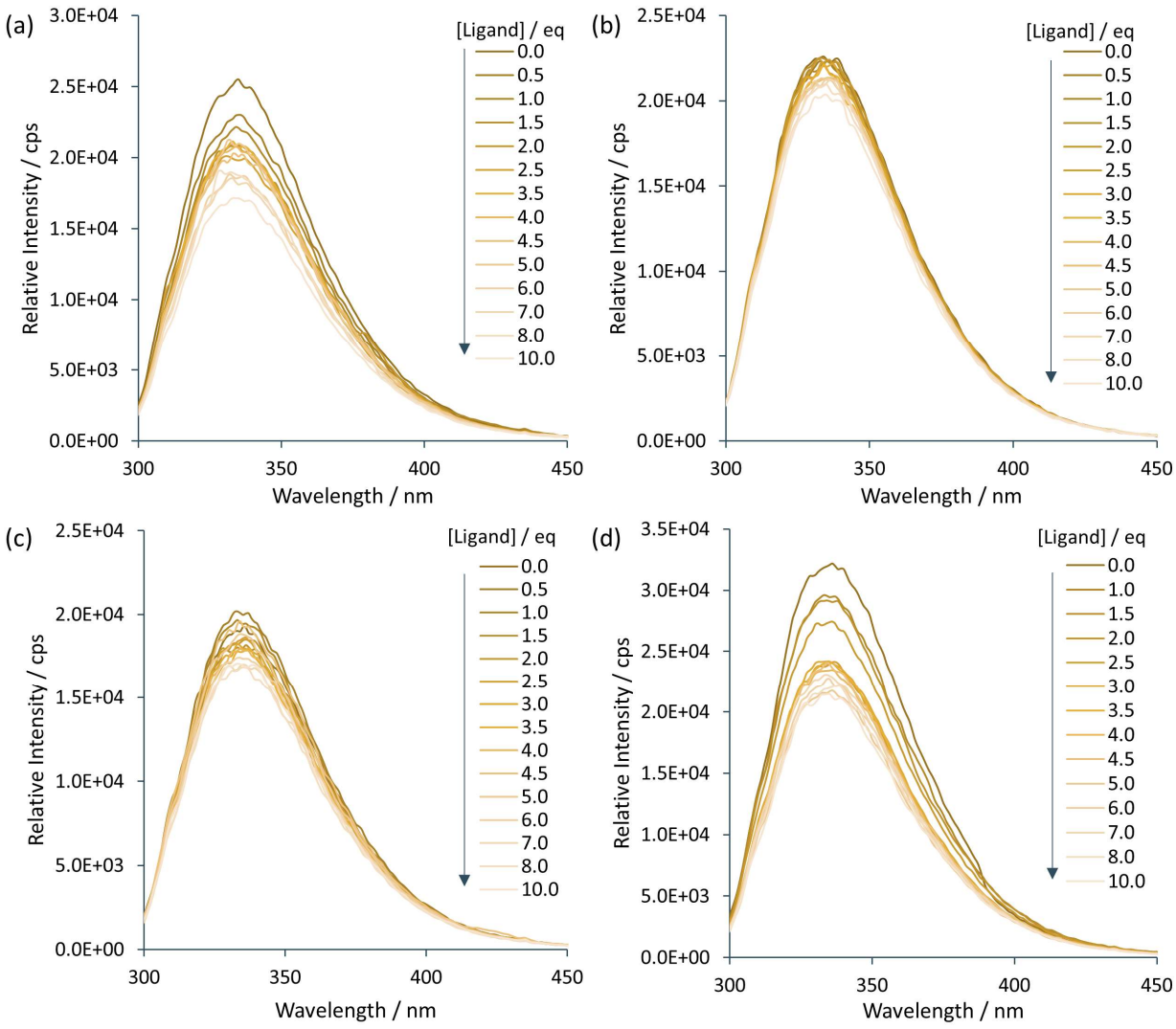

Figure S5 (a) Fluorescence emission spectra of 8 µM HEWL in PBS(1X) buffer at pH 7.4 with increasing amounts of (a) AE(Al), (b) biotin, (c) Tris-NH_2, (d) Tris-CH_3 (0 – 10 eq).

Single crystal X-ray structures

Table S1 Data collection and refinement statistics for the co-crystals of HEWL/AE−Biot, HEWL/AE-NH_2 and HEWL/AE-CH_3 (A and B).

Parameter	Value			
Co-crystal	HEWL/AE-Biot	HEWL/AE-NH_2	HEWL/AE-CH_3 (A)	HEWL/AE-CH_3 (B)
PDB deposition code				
Space group	*P* 4_3 2_1 2	*P* 4_3 2_1 2	*P* 4_3 2_1 2	*P* 4_3 2_1 2
Unit cell dimensions [Å]	*a* = *79.39*	*a* = 76.61	*a* = 76.39	*a* = 75.94
	b = *79.39*	*b* = 76.61	*b* = 76.39	*b* = 75.94
	c = *35.14*	*c* = 37.10	*c* = 36.98	*c* = 36.76
Resolution range [Å]	79.39-2.34	38.30-1.57	38.19-1.24	37.97-1.47
	(2.42-2.34)[a]	(1.60-1.57)[a]	(1.26-1.24)[a]	(1.50-1.47)[a]
R_{merge} [%]	8.6 (66.2)[a]	7.2 (79.9)[a]	5.7 (76.4)[a]	7.3 (77.0)[a]
$\langle I/\sigma \rangle$	21.4 (2.4)[a]	24.0 (4.4)[a]	28.4 (4.7)[a]	24.7 (4.7)[a]
No. of unique reflections	5101 (467)[a]	15989 (768)[a]	31633 (1519) [a]	18885 (908)[a]
Multiplicity	19.4 (8.9)[a]	24.0 (23.3)[a]	24.3 (24.0)[a]	24.3 (24.1)[a]
Completeness [%]	100.0 (99.8)[a]	100.0 (100.0)[a]	100.0 (100.0)[a]	100.0 (100.0)[a]
R_{work}/R_{free}[b] [%]	25.1/27.8	19.1/23.2	14.41/16.71	17.1/20.2
RMSD from ideal				
Average isotropic B factors [Å^2]	51.83	25.45	20.28	22.89
No. of water molecules	15	75	154	82
Ramachandran plot [%]				
Residues in favoured regions	90.55	96.85	99.21	98.43
Outliers	0.00	0.00	0.00	0.00

[a] values in parentheses are for the highest resolution shell. [b] Rfree is calculated using a random 5 % of data excluded from the refinement.

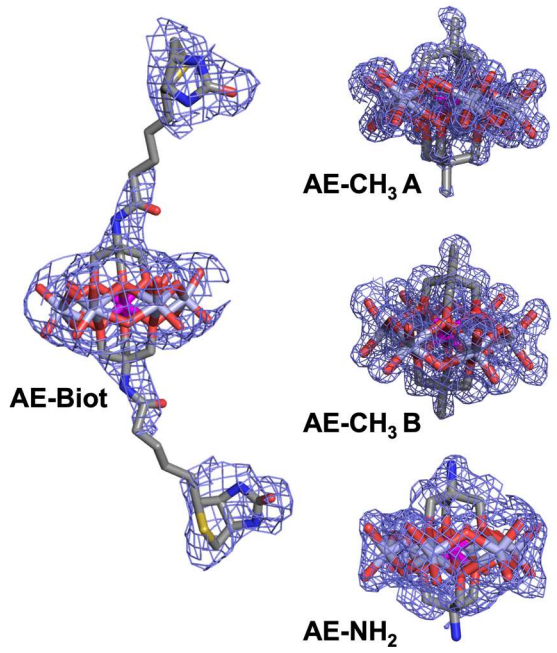

Figure S6 Fit of the HPOMs in the $2F_o$-F_C electron density map (shown as blue mesh) and contoured at 1.0 r.m.s.d.

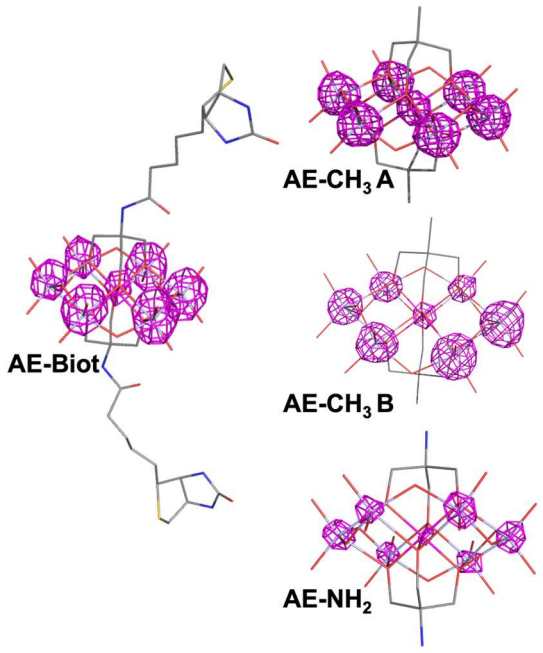

Figure S7 Anomalous difference map of the HPOMs shown as a magenta mesh and contoured at 5.0 r.m.s.d for AE-Biot, AE-CH_3 A and AE-NH_2, and 3.0 r.m.s.d for AE-CH_3 B.

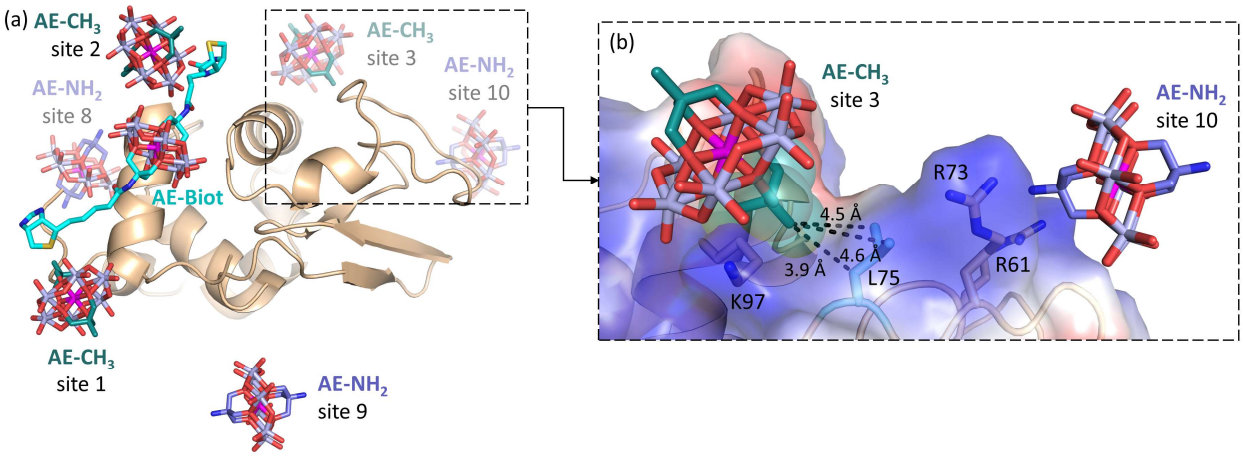

Figure S8 (a) Superposition of the co-crystal structures showing the HPOM binding sites for HEWL/AE-Biot, HEWL/AE-NH_2 and HEWL/AE-CH_3 A. (b) Close-up of AE-CH_3 binding site 3 and AE-NH_2 binding site 10, showing the shortest distances between the -CH_3 carbon of AE-CH_3 and residue Leu75 in black (3.9-4.6 Å), the Van der Waals radius of the C atoms in the Tris-CH_3 ligand as teal spheres, and the protein electrostatic surface charge: blue is positive, white is neutral, and red is negative.