Electronic Supporting Information

Spectroscopic and Computational Comparisons of Thiolate-Ligated Ferric Nonheme Complexes to Cysteine Dioxygenase: Second-Sphere Effects On Substrate (Analogue) Positioning

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Additional Experimental Details

X-ray Crystallography. X-ray crystallographic measurements were conducted at 100K using an Oxford Diffraction SuperNova kappa-diffractometer (Rigaku Corp.) equipped with both Cu and Mo X-ray sources, X-ray mirror optics, an Atlas CCD detector, and a low-temperature Cryojet device manufactured by Oxford Instruments. The data was analyzed using the CrysAlisPro program package and the numerical absorption correction was derived from Gaussian integration over a multifaceted crystal model. An empirical absorption correction based on spherical harmonics was applied with the SCALE3 ABSPACK scaling algorithm. The structure was solved using the SHELXS program and refined with the SHELXL program[1] contained within the Olex2 crystallographic package.[2] Non-hydrogen atoms were refined with anisotropic displacement parameters. Hydrogen atoms were generally positioned geometrically and refined using appropriate restrictions on bond lengths and angles, and the torsion angles of methyl hydrogens were optimized to fit residual electron density.

Protein Purification. In order to create the C93G CDO variant, SDM was employed to make a point mutation in the cdo gene. This mutation changed the codon that encodes for cysteine, TGC, to one that encodes for glycine, GGC. The primers used to make this point mutation can be seen below (point mutation bolded):

Forward Primer: 5'-GGACTCCCAC**G**GCTTTCTGAAGCTGC-3'

Reverse Primer: 5'-GCAGCTTCAGAAAGC**C**GTGGGAGTCC-3'

The iron content of WT and C93G CDO samples could be determined using a colorimetric assay which employed the chelator agent 2,4,6-tris(2-pyridyl)-s-triazine (TPTZ).[3] In brief, a CDO sample was denatured to release any bound iron to solution. Then, TPTZ was added to produce a complex with Fe^{II} that has a molar extinction coefficient at 593 nm of 22.6 $mM^{-1}cm^{-1}$, which is amenable to quantization via UV-vis absorption spectroscopy. Denatured protein solutions were treated with hydroxylamine to determine the total Fe content or without to selectively assay the Fe^{II} content. The difference between the total Fe and Fe^{II} content was used to determine the Fe^{III} content.

1. Sheldrick, G. M. A short history of SHELX. *Acta Crystallogr. Sect. A* **2008,** *64*, 112-122.
2. Dolomanov, O. V.; Bourhis, L. J.; Gildea, R. J.; Howard, J. A. K.; Puschmann, H. OLEX2: a complete structure solution, refinement and analysis program. *J. Appl. Crystallogr.* **2009,** *42*, 339-341.
3. Carter, P. Spectrophotometric determination of serum iron at the submicrogram level with a new reagent (ferrozine). *Anal. Biochem.* **1971,** *40*, 450-8.

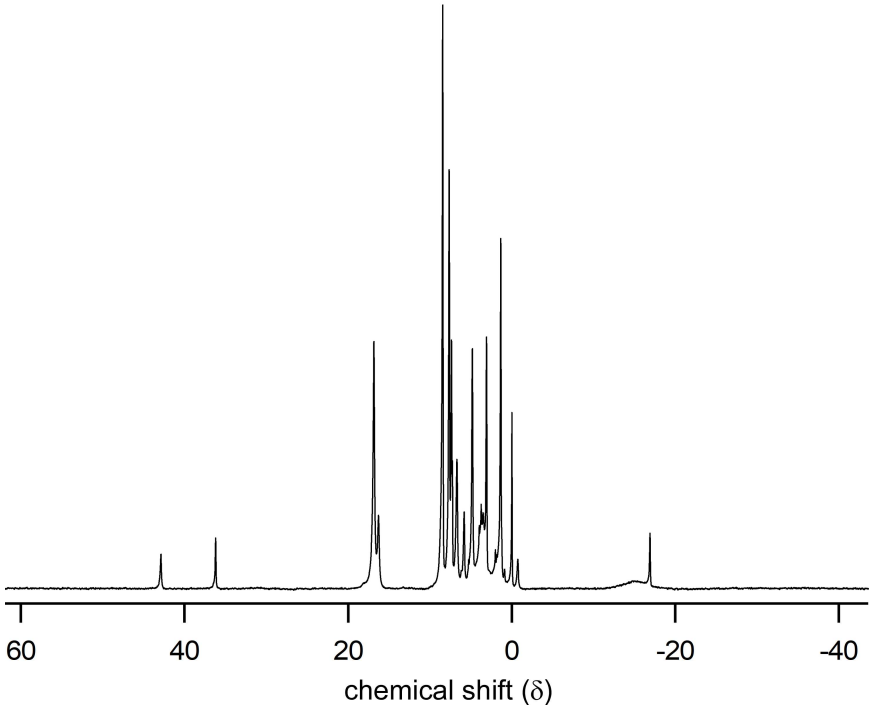

Figure S1. ^{1}H NMR spectrum of **2** measured in $CDCl_3$ at room temperature.

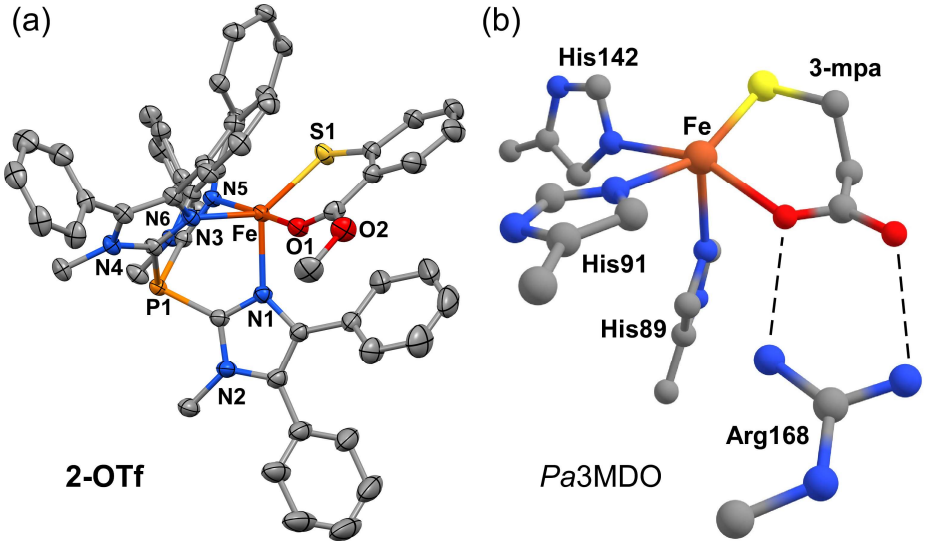

Figure S2. (a) Thermal ellipsoid plot (50% probability) obtained from the X-ray crystal structure of **2-OTf**. The triflate counteranion, non-coordinating solvent molecules, and all hydrogen atoms are omitted for clarity. (b) Model of substrate-bound MDO generated by docking 3-mpa within the active-site of the *Pa*3MDO crystal structure (PDB entry 4TLF).

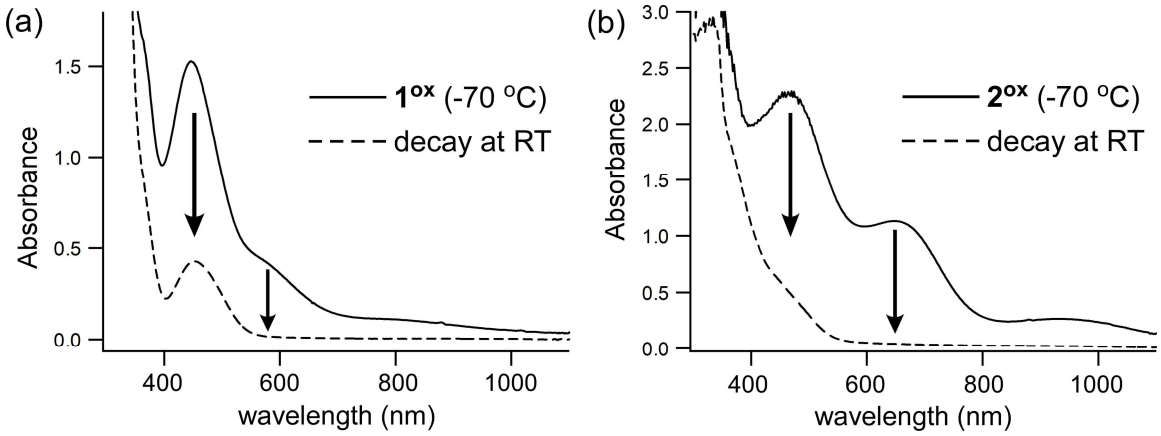

Figure S3. UV-vis absorption spectra showing the decay of (a) **1**^{ox} and (b) **2**^{ox} upon warming the sample from -70 °C to room temperature in CH_2Cl_2.

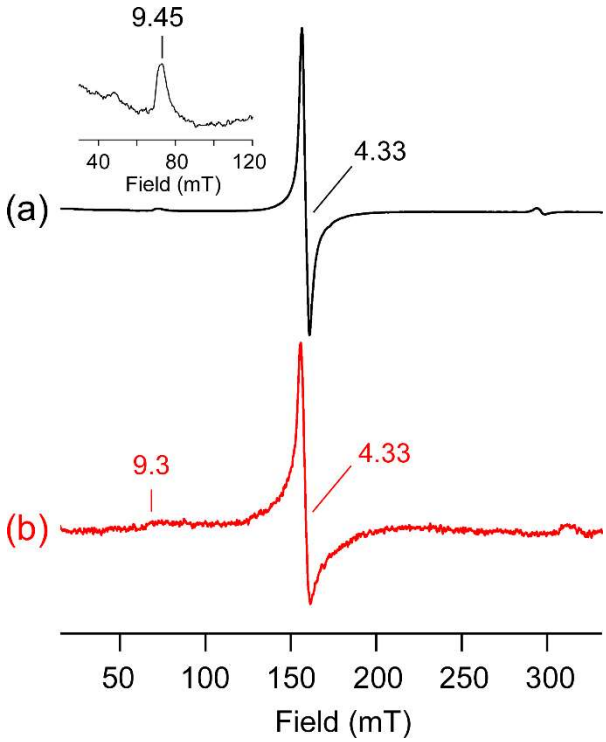

Figure S4. X-band EPR spectra of (a) **1**^{ox} in frozen THF and (b) **2**^{ox} in frozen CH_2Cl_2. Samples were prepared by addition of $[AcFc]BF_4$ (1 equiv.) to solutions of the precursor complexes cooled to -70 °C. EPR data collection: frequency = 9.63 GHz; power = 2.0 mW; T = 10 K.

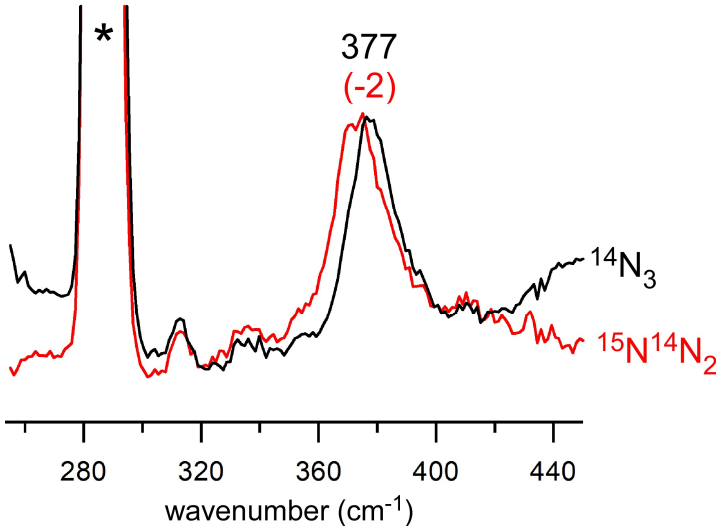

Figure S5. Resonance Raman spectra of **1^{ox}-N_3** prepared with $^{14}N_3^-$ (black line) or $^{15}N^{14}N_2^-$ (red line). Spectra were collected in CD_2Cl_2 using 488.0 nm laser excitation (55 mW). Samples were prepared by treating **1^{ox}** at 20 °C with KN_3 or $K^{15}N^{14}N_2$ (5 equiv.) solubilized with 13-crown-6-ether. Frequencies and isotope shifts are provided in cm^{-1}. Solvent-derived peaks are indicated with an asterisk (*).

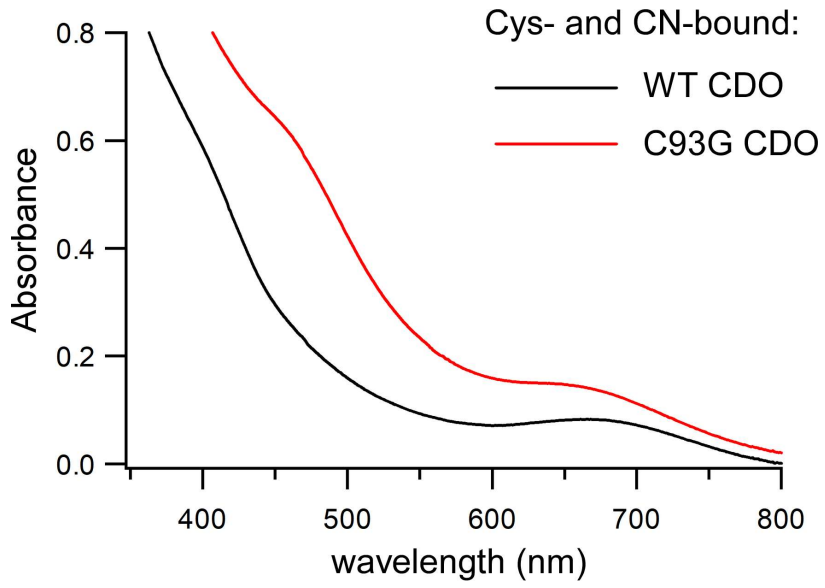

Figure S6. UV-vis absorption spectra of Cys- and CN-treated Fe^{III}-CDO measured at room temperature. *Black line*: WT CDO; *red line*: C93G CDO variant.

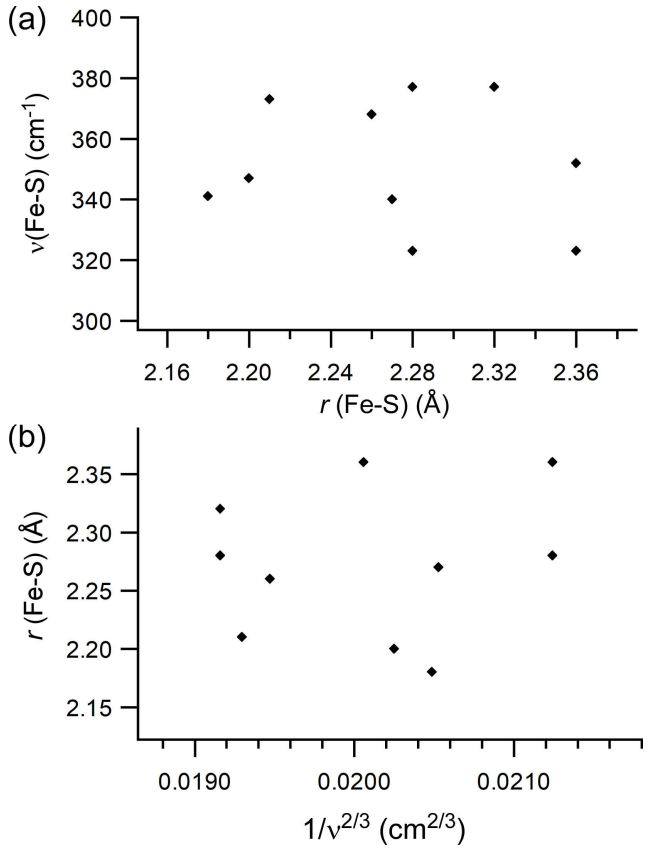

Figure S7. Relationship between Fe–S bond distances (*r*, in Å) and stretching frequencies (ν, in cm^{-1}) for nonheme Fe^{III} species with mixed N/S ligation (see Table 4 for the raw data). *Top*: Plot of Fe–S stretching frequencies versus Fe–S bond distances. *Bottom:* Plot of Fe–S bond distance versus $1/\nu^{2/3}$. The high degree of scatter in both plots indicate that the values do not follow Badger's rule.

Table S1. Selected Bond Distances (Å) and Bond Angles (°) for Complexes **1** and **2-OTf** [a] Measured by X-ray Crystallography. Metric parameters for the Cys-Bound CDO Active Site are Provided for Comparison.

Bond Distances	**1**	**2-OTf** (A)	**2-OTf** (B)	CDO [b]
Fe1-N1	2.170(2)	2.1353(19)	2.1294(19)	1.893
Fe1-N3	2.153(2)	2.0914(18)	2.1294(19)	2.199
Fe1-N5	2.195(2)	2.2487(17)	2.193(2)	2.110
Fe1-S1	2.3107(6)	2.3233(6)	2.3227(7)	2.291
Fe1-N7/O1	2.245(2)	2.1200(16)	2.1336(19)	2.262
Bond Angles				
N1-Fe1-N3	96.73(6)	92.28(7)	93.92(7)	99.9
N1-Fe1-N5	87.90(6)	92.40(7)	86.76(8)	98.6
N1-Fe1-S1	125.08(5)	128.34(6)	135.62(6)	112.9
N1-Fe1-N7/O1	89.01(6)	87.45(7)	85.02(7)	85.7
N3-Fe1-N5	83.02(6)	83.81(7)	86.92(8)	86.0
N3-Fe1-S1	137.22(5)	139.04(5)	129.33(5)	146.8
N3-Fe1-N7/O1	87.80(6)	88.42(7)	86.03(7)	96.9
N5-Fe1-S1	104.99(4)	97.10(5)	102.96(6)	94.1
N5-Fe1-N7/O1	169.90(6)	172.21(7)	168.77(7)	174.3
S1-Fe1-N7/O1	84.63(5)	88.40(5)	88.27(5)	80.8
tau(τ)–value	0.54	0.59	0.55	0.46

[a] The unit cell of **2-OTf** contains two symmetrically-independent Fe^{II} complexes, labeled A and B.

[b] CDO parameters were obtained from an X-ray structure of Cys-bound CDO (PDB 4JTO).

Table S2. CASSCF/NEVTP2 Computed *g*-Values for **1^{ox}-CN**, Along with Experimental Values

Active Space	Basis set [a]	g_1	g_2	g_3	rms dev [b]	Δ	V
CAS(5,5)	SVP	1.932	2.272	2.334	0.104	4234	577
CAS(5,10)	SVP	1.920	2.284	2.365	0.123	3732	601
CAS(11,8)	SVP	1.946	2.246	2.306	0.080	3926	297
CAS(11,13)	SVP	1.960	2.186	2.219	0.023	4209	503
CAS(11,13)	TZVP	1.946	2.183	2.234	0.032	4321	512
exper		1.988	2.161	2.204		4731 [c]	1094 [c]

[a] SVP = split valence basis set with polarization; TZVP = triple zeta basis set with polarization. [b] rms dev = root-mean-square deviation between experimental and computed g-values. [c] The experimental Δ- and V-values were calculated using Taylor's equations for low-spin Fe^{III} complexes in the absence of iron-ligand covalency, assuming a SOC parameter (λ) of 400 cm^{-1}.

Table S3. DFT Computed Fe-S Bond Distances (in Å) for **1^{ox}-CN** and **2^{ox}-CN** Using Different Functionals.

	1^{ox}-CN	**2^{ox}-CN**	
functional [a]	r(Fe-S), Å	r(Fe-S), Å	Δr(Fe-S), Å
BP86	2.163	2.160	-0.003
TPSS	2.169	2.166	-0.003
B3LYP	2.182	2.195	+0.013
TPSSh	2.169	2.170	+0.001
PBE0	2.172	2.174	+0.002

[a] BP86 = Becke 88 exchange and Perdew 86 correlation; TPSS = TPSS meta-GGA functional; B3LYP = conventional B3LYP functional (20% HF exchange); TPSSh = The hybrid version of TPSS (10% HF exchange); PBE0 = One-parameter hybrid version of Perdew-Burke-Erzerhoff GGA functional (25% HF exchange).

Table S4. Cartesian Coordinates (Å) of the DFT-Optimized Model of **1^{ox}-CN**.

Fe	0.194564000000	0.099353000000	-0.209956000000
C	0.395756000000	-0.123947000000	1.673442000000
N	0.455921000000	-0.220583000000	2.834232000000
S	2.351505000000	0.213600000000	-0.523780000000
N	0.389085000000	-1.928476000000	-0.397863000000
C	1.772547000000	-2.447990000000	-0.562628000000
C	1.876859000000	-3.856912000000	0.020348000000
O	0.975996000000	-4.415534000000	0.598765000000
C	2.785354000000	-1.472071000000	0.040177000000
O	3.084743000000	-4.379909000000	-0.189861000000
H	-0.185191000000	-2.272680000000	-1.170802000000
H	-0.013338000000	-2.368654000000	0.439506000000
H	1.985591000000	-2.524780000000	-1.637050000000
H	3.800375000000	-1.720378000000	-0.301678000000
H	2.768170000000	-1.496350000000	1.141511000000
C	-2.753502000000	0.734099000000	-0.608388000000
P	-2.228178000000	2.278810000000	-1.432482000000
N	-4.057222000000	0.365811000000	-0.528042000000
H	-5.055374000000	-1.346427000000	0.336830000000
C	-4.120254000000	-0.838274000000	0.115484000000
N	-1.971944000000	-0.186921000000	-0.043008000000
C	-2.818129000000	-1.191998000000	0.422879000000
H	-4.843375000000	0.907265000000	-0.873430000000
N	-1.133663000000	4.024283000000	0.457407000000
C	-1.030491000000	2.835744000000	-0.188161000000
N	0.060411000000	2.177092000000	0.204539000000
C	-0.056715000000	4.143211000000	1.290544000000
N	-0.941863000000	1.880421000000	-3.904280000000
C	0.702731000000	2.998568000000	1.132429000000
N	-0.240760000000	0.486359000000	-2.337355000000
C	-1.043899000000	1.516731000000	-2.598945000000
C	-0.049045000000	1.037722000000	-4.510096000000
C	0.373995000000	0.152178000000	-3.537843000000
H	0.207389000000	1.105959000000	-5.564532000000
H	-1.459095000000	2.630140000000	-4.352399000000
H	0.122733000000	5.019984000000	1.906882000000
H	-1.870265000000	4.709020000000	0.320454000000
C	-2.464441000000	-2.469722000000	1.089681000000
C	-2.901487000000	-3.672334000000	0.497912000000
H	-3.425078000000	-3.642109000000	-0.461458000000
C	-2.668670000000	-4.899598000000	1.121933000000
H	-3.013913000000	-5.821840000000	0.647461000000
C	-1.993629000000	-4.943104000000	2.344871000000
H	-1.795317000000	-5.901557000000	2.831557000000
C	-1.569928000000	-3.755087000000	2.945424000000

H	-1.047489000000	-3.780934000000	3.904818000000
C	-1.811247000000	-2.521944000000	2.333873000000
H	-1.487257000000	-1.605374000000	2.825396000000
C	2.000279000000	2.809556000000	1.829112000000
H	3.204748000000	3.047402000000	0.046597000000
C	3.214166000000	2.918269000000	1.130508000000
H	5.366690000000	2.966748000000	1.260875000000
C	4.429569000000	2.880130000000	1.816276000000
H	5.397196000000	2.725618000000	3.747063000000
C	4.446253000000	2.748513000000	3.208698000000
H	3.247709000000	2.551143000000	4.999835000000
C	3.242221000000	2.654354000000	3.911925000000
H	1.086865000000	2.588542000000	3.777506000000
C	2.024993000000	2.684415000000	3.227539000000
C	1.221765000000	-1.040944000000	-3.792809000000
H	-0.477503000000	-2.385397000000	-3.869226000000
C	0.609832000000	-2.298845000000	-3.952139000000
C	1.374994000000	-3.431399000000	-4.248511000000
H	0.885428000000	-4.400459000000	-4.377056000000
H	3.361880000000	-4.197451000000	-4.640608000000
C	2.760692000000	-3.315971000000	-4.404490000000
C	3.372708000000	-2.064218000000	-4.277452000000
H	4.452065000000	-1.966713000000	-4.419087000000
H	3.093748000000	0.040333000000	-3.866853000000
C	2.610105000000	-0.932345000000	-3.974672000000
C	3.310315000000	-5.707268000000	0.314576000000
H	3.158466000000	-5.738325000000	1.403662000000
H	2.619457000000	-6.419614000000	-0.160253000000
H	4.348205000000	-5.955855000000	0.063179000000

Table S5. Cartesian Coordinates (Å) of the DFT-Optimized Model of **2^{ox}-CN**.

Fe	0.000000000000	0.000000000000	0.000000000000
C	0.011247000000	-0.125381000000	1.896624000000
N	-0.062108000000	-0.171513000000	3.059013000000
S	2.194632000000	-0.024144000000	-0.035727000000
P	-2.095128000000	2.282770000000	-1.528747000000
O	-0.047385000000	-1.949040000000	-0.194291000000
O	0.440441000000	-4.104233000000	-0.083427000000
N	-1.163415000000	1.222870000000	-3.935379000000
N	-0.305683000000	0.177957000000	-2.182028000000
N	-4.165567000000	0.869084000000	-0.305504000000
N	-2.190936000000	-0.051294000000	0.082812000000
N	-0.577639000000	4.155860000000	-0.132926000000
N	0.123229000000	2.082054000000	0.191767000000
C	-1.073542000000	1.192286000000	-2.580716000000

C	-0.437816000000	0.177634000000	-4.429084000000
C	0.085311000000	-0.496064000000	-3.336084000000
C	-2.823876000000	0.976682000000	-0.482548000000
C	-3.176345000000	-0.872541000000	0.630548000000
C	-4.406369000000	-0.276655000000	0.395397000000
C	-0.789282000000	2.846591000000	-0.408962000000
C	0.518712000000	4.242540000000	0.677716000000
C	0.968800000000	2.953264000000	0.889339000000
C	0.807450000000	-1.782781000000	-3.483172000000
C	0.250770000000	-2.731807000000	-4.366777000000
H	-0.711458000000	-2.524628000000	-4.841733000000
C	0.902467000000	-3.938353000000	-4.630211000000
H	0.452787000000	-4.659279000000	-5.317674000000
C	2.120981000000	-4.222752000000	-4.006883000000
H	2.635078000000	-5.165096000000	-4.212550000000
C	2.679424000000	-3.292163000000	-3.124253000000
H	3.633384000000	-3.508611000000	-2.636697000000
C	2.036653000000	-2.076779000000	-2.870274000000
H	2.490050000000	-1.345743000000	-2.200332000000
C	-3.086282000000	-2.211116000000	1.268372000000
C	-3.999325000000	-3.188152000000	0.814221000000
H	-4.648773000000	-2.964415000000	-0.035809000000
C	-4.082952000000	-4.439550000000	1.428103000000
H	-4.803308000000	-5.176389000000	1.063876000000
C	-3.244674000000	-4.745137000000	2.504433000000
H	-3.315303000000	-5.718757000000	2.996770000000
C	-2.329759000000	-3.789397000000	2.956261000000
H	-1.678648000000	-4.014043000000	3.805137000000
C	-2.253928000000	-2.528226000000	2.356063000000
H	-1.567745000000	-1.785273000000	2.758321000000
C	2.201240000000	2.687511000000	1.679989000000
C	3.429338000000	3.121364000000	1.145863000000
H	3.460623000000	3.536957000000	0.135585000000
C	4.606580000000	3.012145000000	1.887550000000
H	5.551088000000	3.352673000000	1.454540000000
C	4.568786000000	2.476870000000	3.179727000000
H	5.487563000000	2.390807000000	3.765726000000
C	3.349304000000	2.061040000000	3.723374000000
H	3.309456000000	1.648757000000	4.734857000000
C	2.167657000000	2.171929000000	2.985374000000
H	1.225192000000	1.845958000000	3.424343000000
C	0.716451000000	-2.848054000000	0.199363000000
C	1.967885000000	-2.665216000000	0.951917000000
C	2.681805000000	-1.439540000000	0.907514000000

C	3.899332000000	-1.345879000000	1.608527000000
H	4.446744000000	-0.401564000000	1.592746000000
C	4.391854000000	-2.425653000000	2.338478000000
H	5.333366000000	-2.321361000000	2.883905000000
C	3.683225000000	-3.633288000000	2.386697000000
H	4.067487000000	-4.474870000000	2.967410000000
C	2.485724000000	-3.751003000000	1.689896000000
H	1.923834000000	-4.685240000000	1.715638000000
C	-0.731506000000	-4.392004000000	-0.863824000000
H	-0.716965000000	-5.475564000000	-1.028243000000
H	-0.676783000000	-3.859706000000	-1.823094000000
H	-1.635394000000	-4.094592000000	-0.315899000000
H	-1.145264000000	4.925847000000	-0.472609000000
H	0.905133000000	5.184072000000	1.058951000000
H	-1.685115000000	1.904782000000	-4.476749000000
H	-0.319527000000	-0.012387000000	-5.492431000000
H	-4.859818000000	1.534658000000	-0.629907000000
H	-5.402303000000	-0.588354000000	0.698773000000

Table S6. Cartesian Coordinates (Å) of the DFT-Optimized Model of **1^{ox}-N_3**.

Fe	0.374009000000	0.067130000000	-0.113576000000
N	0.408205000000	-0.099689000000	1.836872000000
N	-0.161984000000	0.411744000000	2.761921000000
N	-0.703786000000	0.884418000000	3.662111000000
S	2.633416000000	-0.004940000000	-0.655979000000
N	0.521717000000	-2.155606000000	-0.190996000000
C	1.870643000000	-2.669889000000	-0.492309000000
C	2.055390000000	-4.049790000000	0.144326000000
O	1.491102000000	-4.397208000000	1.154490000000
C	2.940321000000	-1.686048000000	0.011259000000
O	2.919704000000	-4.797041000000	-0.544285000000
H	-0.171069000000	-2.609285000000	-0.789415000000
H	0.276775000000	-2.427609000000	0.769375000000
H	1.976882000000	-2.769137000000	-1.579139000000
H	3.934866000000	-2.018608000000	-0.320772000000
H	2.936326000000	-1.640227000000	1.113074000000
C	-2.789437000000	0.729311000000	-0.669885000000
P	-2.212444000000	2.287736000000	-1.441786000000
N	-4.099863000000	0.359279000000	-0.702739000000
H	-5.165125000000	-1.365284000000	0.048182000000
C	-4.214038000000	-0.856484000000	-0.087891000000
N	-2.060193000000	-0.201349000000	-0.059586000000
C	-2.936780000000	-1.207540000000	0.316658000000
H	-4.857570000000	0.905147000000	-1.100899000000
N	-1.332125000000	4.016957000000	0.569601000000
C	-1.083094000000	2.886710000000	-0.143193000000
N	0.074452000000	2.344667000000	0.234960000000
C	-0.293234000000	4.210236000000	1.436765000000
N	-0.787053000000	2.012724000000	-3.850350000000
C	0.596473000000	3.170999000000	1.222951000000
N	-0.217587000000	0.489916000000	-2.355617000000
C	-0.973917000000	1.563115000000	-2.580386000000
C	0.111921000000	1.184221000000	-4.468485000000
C	0.450855000000	0.217243000000	-3.540488000000
H	0.417538000000	1.314998000000	-5.503829000000
H	-1.258449000000	2.806497000000	-4.273033000000
H	-0.231564000000	5.068056000000	2.101168000000
H	-2.142622000000	4.618665000000	0.461758000000
C	-2.575097000000	-2.468043000000	1.005114000000
C	-2.932561000000	-3.698561000000	0.421916000000
H	-3.448928000000	-3.708290000000	-0.541821000000
C	-2.629420000000	-4.904983000000	1.060295000000
H	-2.920345000000	-5.850354000000	0.594564000000

C	-1.954100000000	-4.897722000000	2.283684000000
H	-1.697832000000	-5.837167000000	2.779595000000
C	-1.610942000000	-3.679797000000	2.879182000000
H	-1.097380000000	-3.668636000000	3.843417000000
C	-1.930076000000	-2.471837000000	2.254771000000
H	-1.709428000000	-1.530550000000	2.757418000000
C	1.901470000000	3.031945000000	1.901967000000
H	3.055693000000	2.882775000000	0.077549000000
C	3.094506000000	2.930933000000	1.166964000000
H	5.248014000000	2.849232000000	1.239474000000
C	4.326895000000	2.920035000000	1.822793000000
H	5.348862000000	3.004983000000	3.730000000000
C	4.383494000000	3.008748000000	3.217550000000
H	3.238236000000	3.174091000000	5.046613000000
C	3.200700000000	3.107713000000	3.956374000000
H	1.042306000000	3.187024000000	3.884516000000
C	1.966012000000	3.124712000000	3.304200000000
C	1.251219000000	-1.001966000000	-3.810188000000
H	-0.465380000000	-2.311303000000	-3.646309000000
C	0.609595000000	-2.253913000000	-3.838707000000
C	1.327577000000	-3.416088000000	-4.135631000000
H	0.815564000000	-4.381695000000	-4.161278000000
H	3.262868000000	-4.244886000000	-4.643026000000
C	2.696841000000	-3.338913000000	-4.412965000000
C	3.336746000000	-2.095342000000	-4.410921000000
H	4.402821000000	-2.028747000000	-4.642351000000
H	3.127106000000	0.034409000000	-4.109922000000
C	2.619397000000	-0.932168000000	-4.118480000000
C	3.203013000000	-6.111481000000	-0.038637000000
H	3.517877000000	-6.066436000000	1.014371000000
H	2.310395000000	-6.750177000000	-0.119809000000
H	4.010758000000	-6.509965000000	-0.663277000000

Table S7. Cartesian Coordinates (Å) of **α-CDO** Derived from QM/MM Calculations.[a]

Fe	0.000000000000	0.000000000000	0.000000000000
C	0.000000000000	-0.000031000000	1.947906000000
N	0.125687000000	0.176956000000	3.087082000000
S	2.257965000000	0.032974000000	0.315140000000
C	-0.519180000000	0.244965000000	-5.684418000000
C	0.095795000000	0.191818000000	-4.325958000000
C	-0.375488000000	-0.339264000000	-3.146713000000
N	1.222061000000	0.922119000000	-3.948502000000
C	1.363159000000	0.838226000000	-2.595062000000
N	0.412491000000	0.069122000000	-2.079742000000
H	-1.102295000000	1.228790000000	-5.875473000000
H	-1.232529000000	-0.579315000000	-5.788849000000
H	0.223007000000	0.114243000000	-6.476624000000
H	-1.193466000000	-1.012680000000	-2.994019000000
H	1.767761000000	1.541443000000	-4.532150000000
H	2.148865000000	1.316605000000	-2.039871000000
C	-5.604584000000	-1.376831000000	-0.361481000000
C	-4.286880000000	-0.698151000000	-0.111679000000
C	-2.987488000000	-1.142242000000	0.024139000000
N	-4.164642000000	0.688873000000	-0.193939000000
C	-2.846695000000	1.029083000000	-0.148605000000
N	-2.095963000000	-0.062332000000	-0.015137000000
H	-5.758469000000	-1.631500000000	-1.485367000000
H	-6.427719000000	-0.738525000000	-0.022751000000
H	-5.673660000000	-2.303635000000	0.211777000000
H	-2.648712000000	-2.151871000000	0.160721000000
H	-4.911743000000	1.334778000000	-0.453766000000
H	-2.521027000000	2.050354000000	-0.237991000000
C	-0.356323000000	5.595200000000	-0.759552000000
C	-0.144974000000	4.205048000000	-0.282608000000
C	-0.386856000000	2.957474000000	-0.814713000000
N	0.450684000000	3.948914000000	0.940979000000
C	0.525406000000	2.610733000000	1.134200000000
N	0.034744000000	1.978546000000	0.077759000000
N	0.171890000000	-2.078568000000	-0.058044000000
C	1.570251000000	-2.530960000000	-0.365128000000
C	1.814667000000	-4.047119000000	-0.242142000000
O	1.163681000000	-4.706635000000	0.675461000000
C	2.508331000000	-1.765106000000	0.577591000000
O	2.706696000000	-4.576889000000	-0.983536000000
H	-0.279785000000	5.713623000000	-1.897797000000
H	0.416229000000	6.228271000000	-0.312439000000
H	-1.319992000000	6.004807000000	-0.429764000000
H	-0.850540000000	2.707260000000	-1.751114000000
H	0.750000000000	4.570587000000	1.681778000000

H	0.916580000000	2.155548000000	2.024979000000
H	-0.486481000000	-2.503708000000	-0.719574000000
H	-0.093933000000	-2.406387000000	0.875793000000
H	1.779282000000	-2.243378000000	-1.397964000000
H	3.550705000000	-2.015671000000	0.376770000000
H	2.276688000000	-1.996078000000	1.622757000000

a Coordinates were obtained from Li, W.; Blaesi, E. J.; Pecore, M. D.; Crowell, J. K.; Pierce, B. S. *Biochemistry* **2013,** *52*, 9104-9119.

Table S8. Cartesian Coordinates (Å) of **β-CDO** Derived from QM/MM Calculations.[a]

Fe	0.000000000000	0.000000000000	0.000000000000
C	0.000031000000	-0.000031000000	1.945480000000
N	0.040070000000	-0.062851000000	3.103241000000
S	2.252258000000	0.015533000000	0.187759000000
C	-1.245667000000	-0.377106000000	-5.513306000000
C	-0.426834000000	-0.299042000000	-4.266159000000
C	-0.758209000000	-0.623962000000	-2.971558000000
N	0.802582000000	0.350449000000	-4.151428000000
C	1.139969000000	0.419479000000	-2.829193000000
N	0.209946000000	-0.170303000000	-2.089966000000
H	-1.803482000000	0.610229000000	-5.757690000000
H	-2.008179000000	-1.152893000000	-5.387833000000
H	-0.647598000000	-0.670731000000	-6.379593000000
H	-1.600281000000	-1.178253000000	-2.615692000000
H	1.312744000000	0.819244000000	-4.887619000000
H	2.036300000000	0.878265000000	-2.451157000000
C	-5.624054000000	-0.821457000000	0.665878000000
C	-4.237823000000	-0.265152000000	0.599686000000
C	-2.988098000000	-0.840805000000	0.535355000000
N	-4.023026000000	1.062241000000	0.258545000000
C	-2.705933000000	1.255524000000	-0.002441000000
N	-2.042007000000	0.112198000000	0.152603000000
H	-5.944412000000	-1.222641000000	-0.368942000000
H	-6.337189000000	-0.052551000000	0.978607000000
H	-5.696000000000	-1.630768000000	1.394348000000
H	-2.717560000000	-1.861771000000	0.728836000000
H	-4.747452000000	1.759262000000	0.088486000000
H	-2.319031000000	2.213043000000	-0.297745000000
C	0.052307000000	5.475388000000	-1.494308000000
C	0.140289000000	4.132584000000	-0.843246000000
C	-0.182098000000	2.844009000000	-1.200089000000
N	0.623367000000	4.009323000000	0.458420000000
C	0.577179000000	2.716034000000	0.842026000000
N	0.109070000000	1.971924000000	-0.141098000000

N	0.151794000000	-2.105621000000	0.090515000000
C	1.515381000000	-2.561325000000	-0.336960000000
C	1.833984000000	-4.071457000000	-0.266876000000
O	1.298889000000	-4.820679000000	0.671585000000
C	2.518036000000	-1.770294000000	0.518265000000
O	2.697998000000	-4.499130000000	-1.089249000000
H	-0.096909000000	5.490800000000	-2.638550000000
H	0.982559000000	6.015839000000	-1.281464000000
H	-0.757202000000	6.066635000000	-1.044922000000
H	-0.637512000000	2.486206000000	-2.103470000000
H	1.034027000000	4.690720000000	1.086670000000
H	0.898453000000	2.368607000000	1.811478000000
H	-0.582367000000	-2.571442000000	-0.452774000000
H	0.021652000000	-2.376480000000	1.072205000000
H	1.634766000000	-2.280563000000	-1.384750000000
H	3.545258000000	-2.033936000000	0.266449000000
H	2.341202000000	-1.957016000000	1.582565000000

a Coordinates were obtained from Li, W.; Blaesi, E. J.; Pecore, M. D.; Crowell, J. K.; Pierce, B. S. *Biochemistry* **2013,** *52*, 9104-9119.

Table S9. Cartesian Coordinates (Å) of **Hybrid-1** Model of CDO Active Site

Fe	-0.050513000000	-0.027514000000	0.001855000000
C	-0.050513000000	-0.027514000000	1.947906000000
N	0.075687000000	0.156956000000	3.087082000000
S	2.153239000000	0.092703000000	0.488872000000
C	-0.561092000000	0.175916000000	-5.636262000000
C	0.086317000000	0.152972000000	-4.290048000000
C	-0.381677000000	-0.339886000000	-3.094453000000
N	1.242105000000	0.852707000000	-3.942211000000
C	1.404314000000	0.784869000000	-2.587226000000
N	0.432847000000	0.060908000000	-2.047434000000
H	-1.152465000000	1.152120000000	-5.843664000000
H	-1.276194000000	-0.650949000000	-5.698998000000
H	0.160792000000	0.020219000000	-6.441867000000
H	-1.220309000000	-0.978681000000	-2.916687000000
H	1.806359000000	1.432705000000	-4.548207000000
H	2.210897000000	1.245838000000	-2.045250000000
C	-5.640828000000	-1.236970000000	-0.181900000000
C	-4.300435000000	-0.599546000000	-0.001534000000
C	-3.014808000000	-1.092345000000	0.038962000000
N	-4.139331000000	0.768702000000	-0.164105000000
C	-2.816960000000	1.065179000000	-0.224446000000
N	-2.098231000000	-0.049217000000	-0.107916000000
H	-5.798794000000	-1.531251000000	-1.287737000000
H	-6.440586000000	-0.551512000000	0.114934000000

H	-5.746282000000	-2.128992000000	0.437782000000
H	-2.698875000000	-2.112097000000	0.154240000000
H	-4.884276000000	1.438951000000	-0.352187000000
H	-2.465027000000	2.071016000000	-0.359267000000
C	-0.199917000000	5.579669000000	-0.865752000000
C	-0.099411000000	4.176799000000	-0.359196000000
C	-0.281729000000	2.922950000000	-0.894283000000
N	0.223230000000	3.928929000000	0.973979000000
C	0.221178000000	2.599005000000	1.204587000000
N	-0.065102000000	1.950326000000	0.092554000000
N	0.238082000000	-2.117277000000	-0.118468000000
C	1.673623000000	-2.442087000000	-0.407413000000
C	2.087477000000	-3.929638000000	-0.460645000000
O	1.493002000000	-4.812833000000	0.310675000000
C	2.501605000000	-1.701755000000	0.655043000000
O	3.076505000000	-4.208419000000	-1.202149000000
H	-0.204388000000	5.720926000000	-2.011098000000
H	0.655247000000	6.142919000000	-0.473064000000
H	-1.099624000000	6.067504000000	-0.466541000000
H	-0.592987000000	2.648636000000	-1.883926000000
H	0.501871000000	4.553871000000	1.721942000000
H	0.441555000000	2.158781000000	2.164481000000
H	-0.386703000000	-2.556680000000	-0.802364000000
H	0.004807000000	-2.508451000000	0.801632000000
H	1.903883000000	-2.033938000000	-1.392978000000
H	3.568513000000	-1.875716000000	0.515224000000
H	2.205729000000	-2.021840000000	1.659469000000

Table S10. Cartesian Coordinates (Å) of **Hybrid-1** Model of CDO Active Site

Fe	0.000000000000	0.000000000000	0.000000000000
C	-0.245772000000	-0.236023000000	1.918238000000
N	-0.347660000000	-0.432022000000	3.057169000000
S	2.257965000000	0.032974000000	0.315140000000
C	-0.561092000000	0.175916000000	-5.636262000000
C	0.086317000000	0.152972000000	-4.290048000000
C	-0.381677000000	-0.339886000000	-3.094453000000
N	1.242105000000	0.852707000000	-3.942211000000
C	1.404314000000	0.784869000000	-2.587226000000
N	0.432847000000	0.060908000000	-2.047434000000
H	-1.152465000000	1.152120000000	-5.843664000000
H	-1.276194000000	-0.650949000000	-5.698998000000
H	0.160792000000	0.020219000000	-6.441867000000
H	-1.220309000000	-0.978681000000	-2.916687000000
H	1.806359000000	1.432705000000	-4.548207000000
H	2.210897000000	1.245838000000	-2.045250000000

C	-5.640828000000	-1.236970000000	-0.181900000000
C	-4.300435000000	-0.599546000000	-0.001534000000
C	-3.014808000000	-1.092345000000	0.038962000000
N	-4.139331000000	0.768702000000	-0.164105000000
C	-2.816960000000	1.065179000000	-0.224446000000
N	-2.098231000000	-0.049217000000	-0.107916000000
H	-5.798794000000	-1.531251000000	-1.287737000000
H	-6.440586000000	-0.551512000000	0.114934000000
H	-5.746282000000	-2.128992000000	0.437782000000
H	-2.698875000000	-2.112097000000	0.154240000000
H	-4.884276000000	1.438951000000	-0.352187000000
H	-2.465027000000	2.071016000000	-0.359267000000
C	-0.199917000000	5.579669000000	-0.865752000000
C	-0.099411000000	4.176799000000	-0.359196000000
C	-0.281729000000	2.922950000000	-0.894283000000
N	0.223230000000	3.928929000000	0.973979000000
C	0.221178000000	2.599005000000	1.204587000000
N	-0.065102000000	1.950326000000	0.092554000000
N	0.171890000000	-2.078568000000	-0.058044000000
C	1.570251000000	-2.530960000000	-0.365128000000
C	1.814667000000	-4.047119000000	-0.242142000000
O	1.163681000000	-4.706635000000	0.675461000000
C	2.508331000000	-1.765106000000	0.577591000000
O	2.706696000000	-4.576889000000	-0.983536000000
H	-0.204388000000	5.720926000000	-2.011098000000
H	0.655247000000	6.142919000000	-0.473064000000
H	-1.099624000000	6.067504000000	-0.466541000000
H	-0.592987000000	2.648636000000	-1.883926000000
H	0.501871000000	4.553871000000	1.721942000000
H	0.441555000000	2.158781000000	2.164481000000
H	-0.486481000000	-2.503708000000	-0.719574000000
H	-0.093933000000	-2.406387000000	0.875793000000
H	1.779282000000	-2.243378000000	-1.397964000000
H	3.550705000000	-2.015671000000	0.376770000000
H	2.276688000000	-1.996078000000	1.622757000000