

# On the propagation of the OH radical produced by Cu-Amyloid Beta peptide model complexes. Insight from molecular modelling. Supplementary Informations

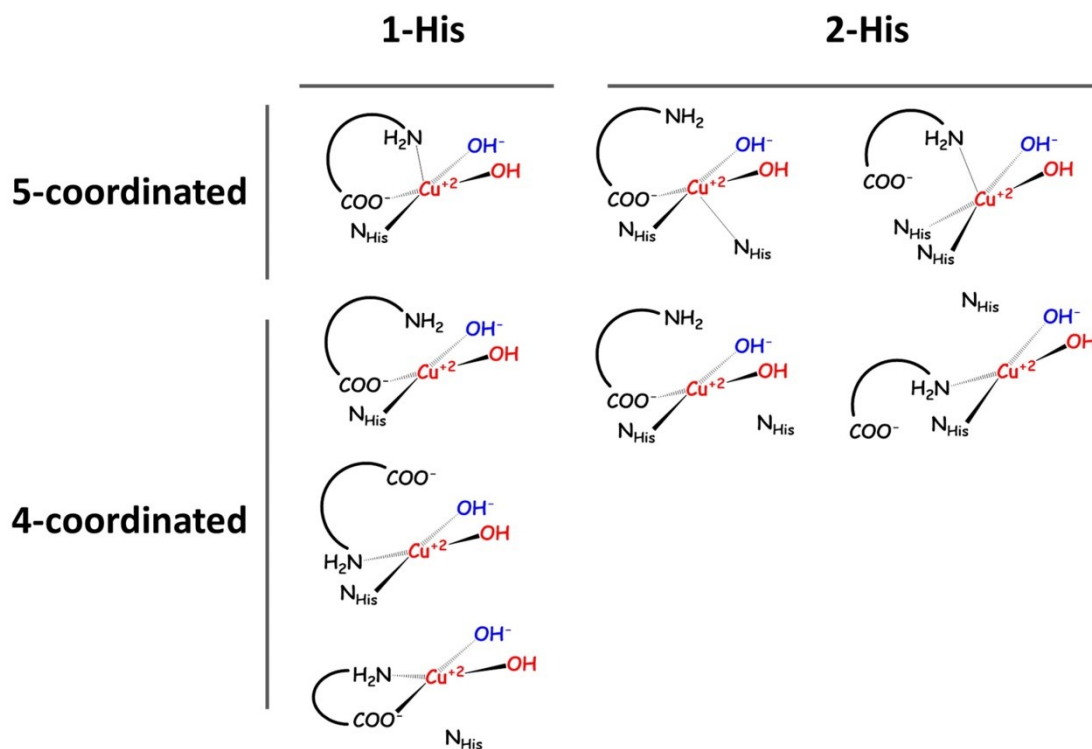
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## Outline

1. Scheme S1. Cu(II)(OH)<sub>2</sub><sup>-</sup>Aβ coordinations scheme;
2. Table S1/S2. DFT Optimized 1-His and 2-His model coordinations;
3. Scheme S1a. Geometry optimization of the **1** oxyl form;
4. Scheme S1b. Protonation of **1** Cu(II)(OH)<sub>2</sub><sup>-</sup>Aβ form.
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16. Scheme S4. MP2/6-31G\* charges for CHARMM36m force field parametrization of the Cu(II) (OH)<sub>2</sub><sup>-</sup>Aβ four-coordinated model.
17. Table S7. Salt-bridge interaction data from molecular dynamic simulation.



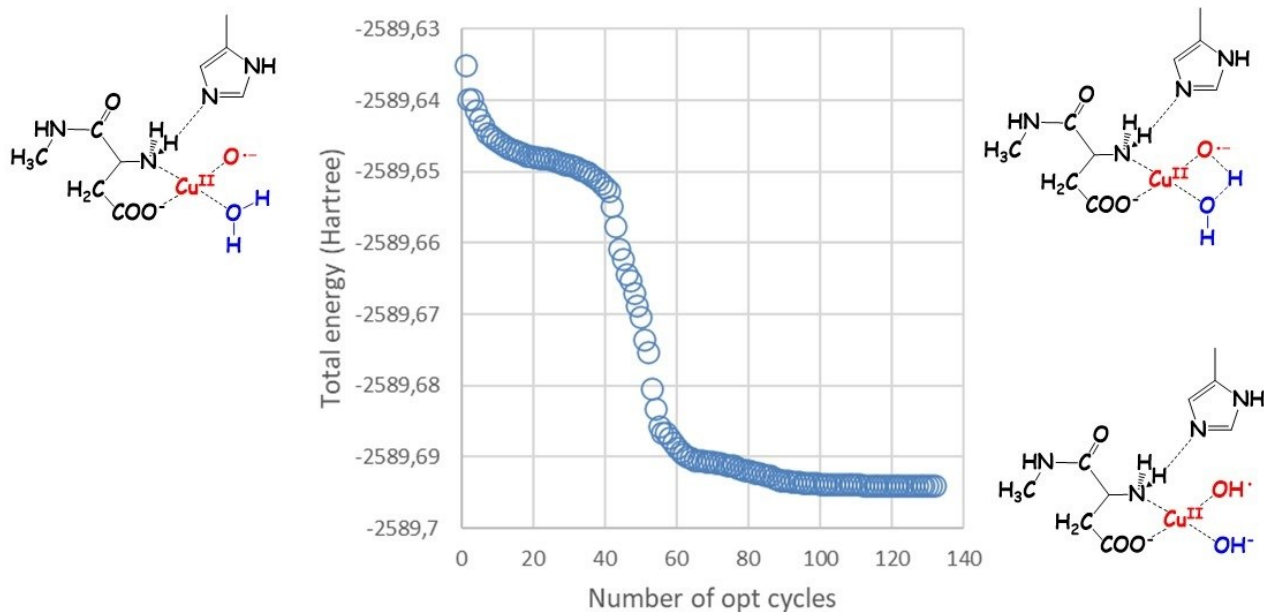
**Scheme S1.** Cu(II)(OH)<sub>2</sub><sup>-</sup>Aβ coordinations. COO<sup>-</sup> and NH<sub>2</sub> belong to Asp1 N-terminal residue. N<sub>His</sub> can belong to His6 and/or His3 and/or His14. Cu(II) can be 4- or 5- coordinated with one or two histidine residue side chains.

**Table S1.** DFT Optimized **1-His** model coordinations.

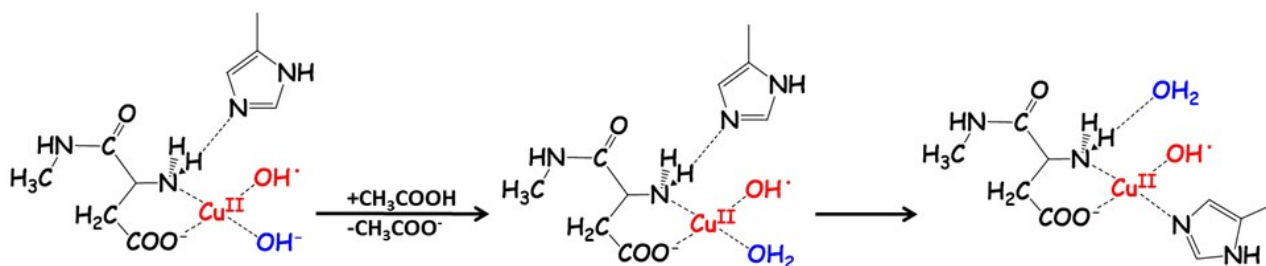
	Cu Coordination Number	Dissociated ligand	Apical Ligand	Apical Cu-L distance (Å)	HO-Cu-OH angle	Total Energy (Hartree)		$\Delta E N\delta$ (kcal/mol)	$\Delta E N\epsilon$ (kcal/mol)
						N $\delta$	N $\epsilon$		
1	4	His			90	-2589,6954	-2589,6945	0,0	0,0
2	4	His			90	-2589,6911	-2589,6917	2,8	1,8
3	4	His			90	-2589,6909	-2589,6892	2,9	3,4
4	4	NH2			90	-2589,6890	-2589,6921	4,1	1,5
5	4	COO-			180	-2589,6882	-2589,6892	4,5	3,3
6	4	NH2			180	-2589,6869	-2589,6887	5,4	3,6
7	4	COO-			90	-2589,6858	-2589,6896	6,1	3,1
8	4	COO-			180	-2589,6858	-2589,6864	6,1	5,1
9	4	NH2			90	-2589,6856	-2589,6860	6,2	5,3
10	5	COO-	CO	2,469	90	-2589,6850	-2589,6873	6,6	4,5
11	5		His	2,593	90	-2589,6842	-2589,6858	7,1	5,5
12	4	COO-			90	-2589,6840	-2589,6852	7,2	5,8
13	4	COO-			90	-2589,6831	-2589,6851	7,8	5,9
14	5		COO-	2,414	180	-2589,6829	-2589,6832	7,9	7,1
15	4	NH2			90	-2589,6829	-2589,6832	7,9	7,1
16	4	COO-			90	-2589,6818	-2589,6835	8,6	6,9
17	5		COO-	2,414	180	-2589,6814	-2589,6863	8,9	5,1
18	5		COO-	2,366	90	-2589,6789	-2589,6849	10,4	6,0
19	5		NH2	2,449	90	-2589,6773	-2589,6819	11,4	7,9

**Table S2.** DFT Optimized **2-His** model coordinations.

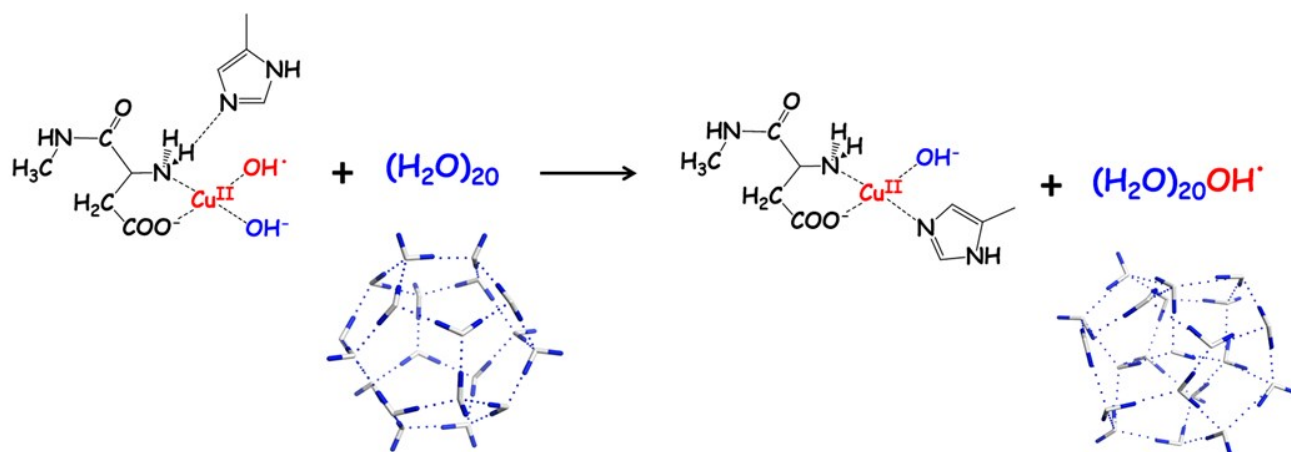
	Cu Coordination Number	Dissociated ligand	Apical Ligand	Apical Cu-L distance (Å)	N <sub>His</sub> -Cu-N <sub>His</sub> angle	HO-Cu-OH Angle	Total Energy (Hartree)		$\Delta E N\delta$ (kcal/mol)	$\Delta E N\epsilon$ (kcal/mol)
							N $\delta$	N $\epsilon$		
1	4	2His				90	-2855,3725	-2855,3727	0	0,0
2	4	NH2			90	90	-2855,3640	-2855,368	5,3	2,9
3	5	His	COO-			90	-2855,3608	-2855,3596	7,4	8,2
4	5		His		90	180	-2855,3604	-2855,3629	7,6	6,1
5	5		His	2,520	90	180	-2855,3586	-2855,3614	8,8	7,1
6	5	His	CO	2,496		90	-2855,3578	-2855,3679	9,2	3,0
7	4	His				90	-2855,3578	-2855,3574	9,2	9,6
8	4	His			90	90	-2855,3561	-2855,3591	10,3	8,5
9	5		His	2,555	90	90	-2855,3522	-2855,3587	12,8	8,8
10	5		NH2	2,521	90	90	-2855,3521	-2855,3601	12,8	7,9
11	5		COO-	2,567	180	180	-2855,3488	-2855,357	14,9	9,9
12	5		COO-	2,493	90	180	-2855,3470	-2855,3577	16,0	9,4



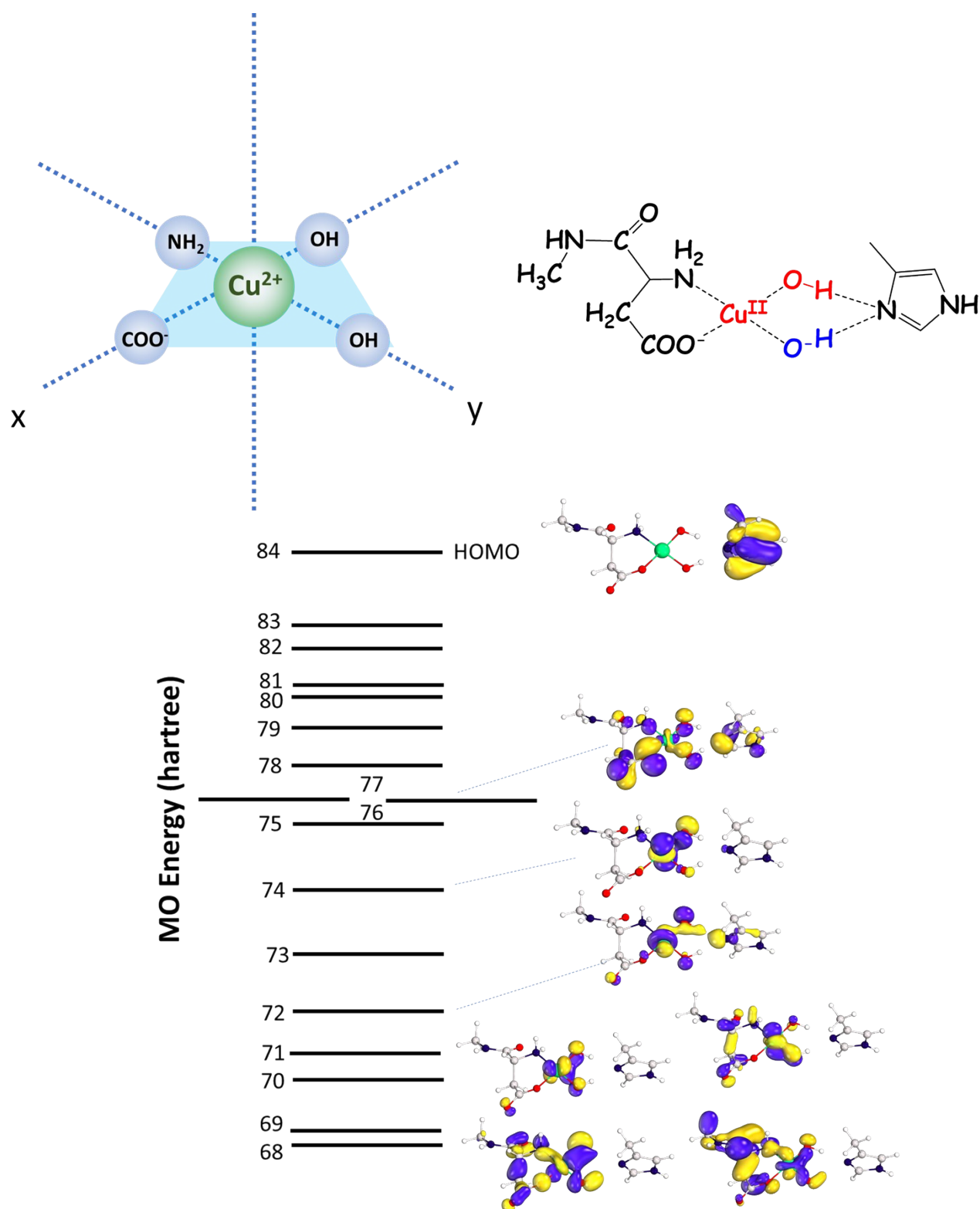
**Scheme S1a.** Energy (in hartree) history of the geometry optimization of the **1** oxyl form which evolves toward the  $\text{Cu(II)(OH)}_2\text{-A}\beta$  coordination. Similar results is also found for **2** and **3** (data not shown).



**Scheme S1b.** Protonation of **1**  $\text{Cu(II)(OH)}_2\text{-A}\beta$  form. In the first step the proton source is an isolated acetic acid molecule which is then deprotonated to acetate anion. The total energy difference among products and reactant is +19.8 kcal/mol. In the second step is computed the energy difference for the replacement of water molecule with the non-coordination histidine ligand, finding an energy gain of 11.4 kcal/mol.

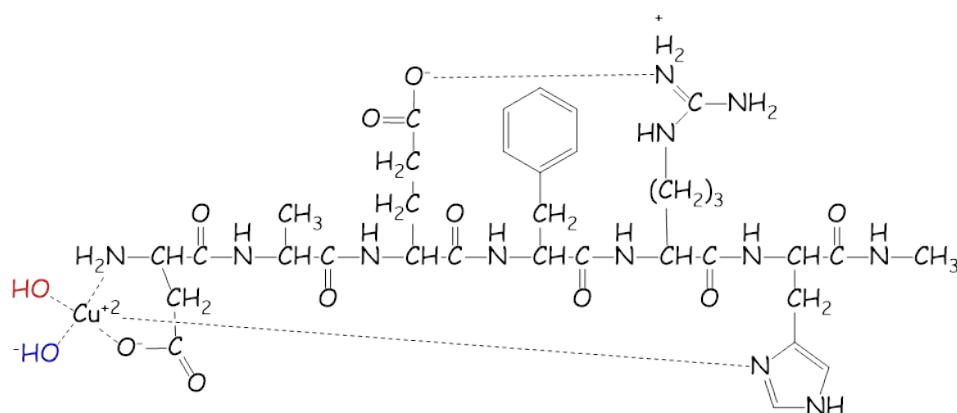


**Scheme S1c.** OH dissociation from the Cu(II) coordination sphere from **1** Cu(II)(OH)<sub>2</sub><sup>-</sup>Aβ form. Water is modelled as an isolated (H<sub>2</sub>O)<sub>20</sub> dodecahedral cluster while solvate OH is the OH(H<sub>2</sub>O)<sub>20</sub> one reported in Figure S4 of this document (see below). Upon OH dissociation, the Cu(II) coordination sphere is saturated with a non-coordinating His side chain. This process is thermodynamically unfavorable by 25.5 kcal/mol.



**Scheme S2.** Occupied frontier Molecular Orbital (MO) diagram for the Cu(II) 1-His most stable four-coordinated model with Cu(II) coordination in almost square planar geometry (OON dihedral 2.9 degree, the four possible Cu-L-L-L dihedrals all close to 179 degree). The 84 MO is the HOMO. MO Isosurfaces (0.05 a.u) for selected MO are reported for the most significant MO with Cu-L bonding character.

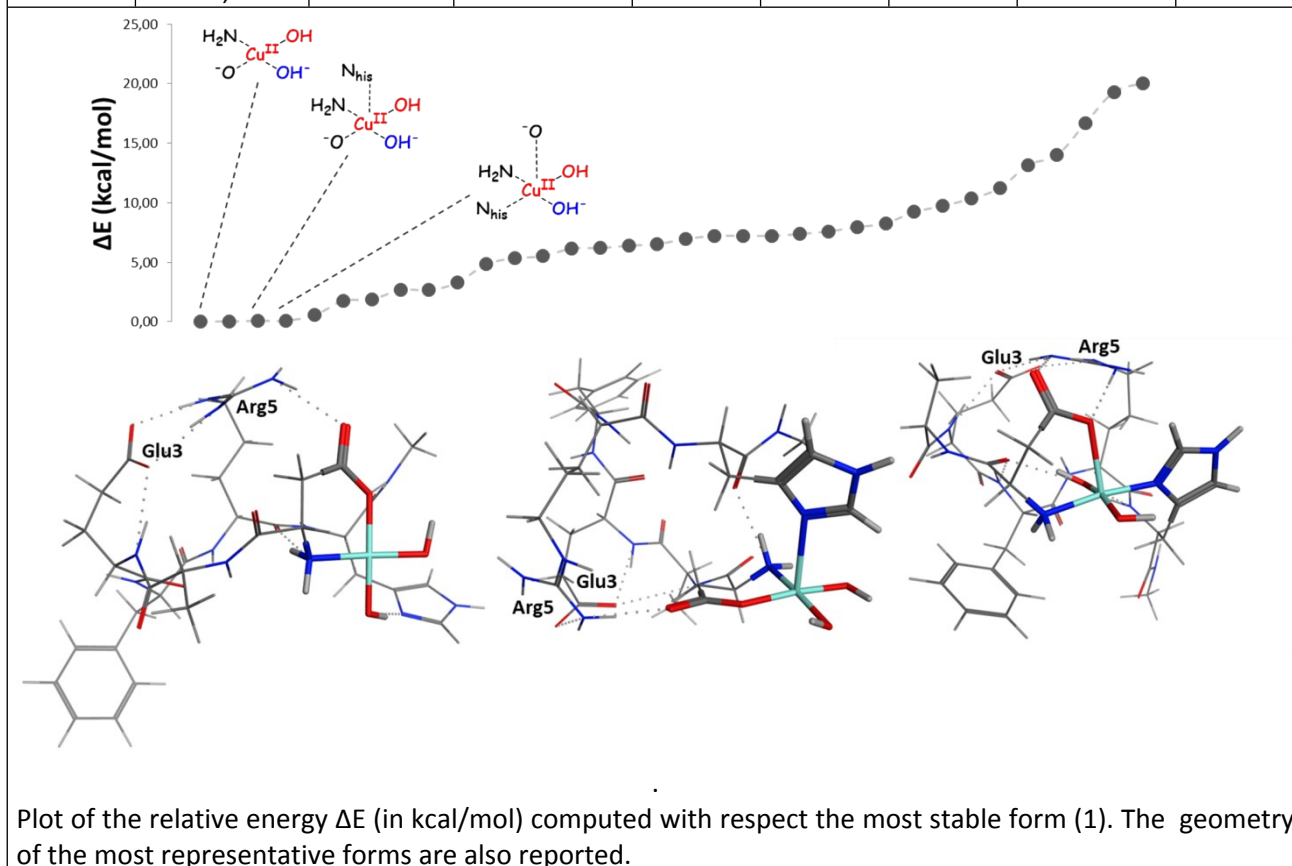
# 1 DAEFRH 6



**Table S3a.** DFT Optimized Cu(II)(OH)<sub>2</sub><sup>-</sup>·Aβ(1-6) models. Aβ(1-6) is the DAEFRH capped peptide. The total net charge of this model is zero. E and ΔE are the total energy and the total energy difference computed with respect to the most stable isomer (1).

	E (hartree)	ΔE (kcal/mol)	Cu Coordination Number	Apical Ligand	Apical Cu-L distance (Å)	HO-Cu- OH Angle	Dissociated ligand	Arg5 salt bridge?
1	-4528,3818	0.00	4			90	His	√
2	-4528,3817	0.04	4			90	His	√
3	-4528,3817	0.08	5	His	2.680	90		√
4	-4528,3816	0.10	5	COO <sup>-</sup>	2.348	180		√
5	-4528,3809	0.6	5	His	2.546	90		√
6	-4528,3790	1.8	4			90	His	√
7	-4528,3788	1.9	5	His	2.540	90		√
8	-4528,3775	2.7	4			180	NH <sub>2</sub>	√
9	-4528,3776	2.7	4			90	NH <sub>2</sub>	√
10	-4528,3766	3.3	5	CO		90	His	√
11	-4528,3741	4.8	4			90	His	√
12	-4528,3733	5.3	4	His	2.675	90		√
13	-4528,3730	5.5	5	His	2.299	90		√
14	-4528,3720	6.2	5	COO <sup>-</sup>	2.688	180		√
15	-4528,3719	6.2	5	CO	2.574	90		√
16	-4528,3716	6.4	4			180	COO <sup>-</sup>	√
17	-4528,3714	6.5	4			90	His	√
18	-4528,3707	7.0	4	COO <sup>-</sup>	2.908	180	COO <sup>-</sup>	√
19	-4528,3703	7.2	4			90	His	
20	-4528,3703	7.2	5	CO	2.601	90	NH <sub>2</sub>	√
21	-4528,3703	7.2	5	CO	2.509	90		
22	-4528,3700	7.4	5	His	2.486	90		√
23	-4528,3697	7.6	5	His	2.744	90		
24	-4528,3691	8.0	5	CO		90		

25	-4528,3686	8.3	4			90	His	
26	-4528,3671	9.2	5	COO-	2.357	90		
27	-4528,3663	9.7	5	His	2.608	90		
28	-4528,3653	10.4	4			90	His	√
29	-4528,3639	11.2	5	NH2	2.332	90		
30	-4528,3608	13.2	5	His	2.489	90		√
31	-4528,3595	14.0	4			90	COO-	√
32	-4528,3552	16.7	4			180		
33	-4528,3510	19.3	4			180	NH2	√
34	-4528,3499	20.0	5	Glu3	2.351	90	NH2	√

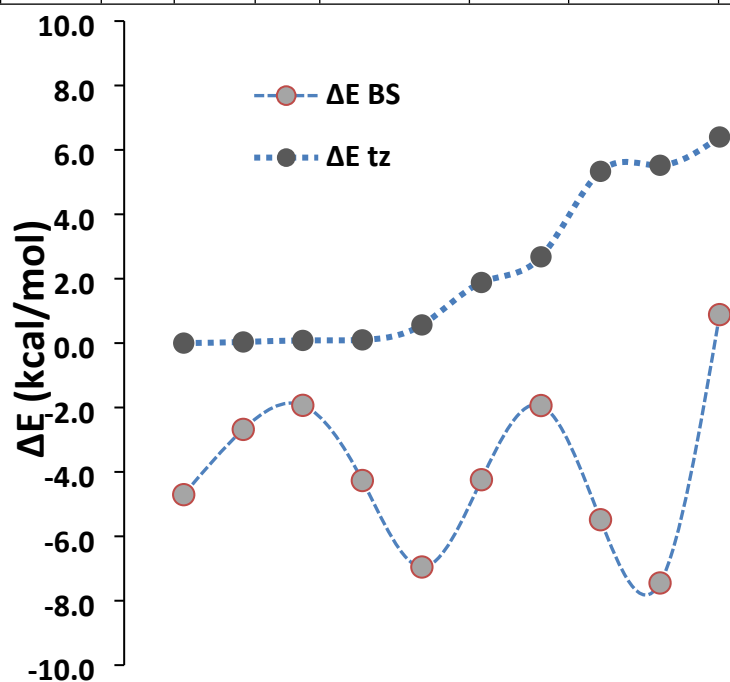


**Table S3b.** DFT Optimized  $\text{Cu(II)(OH)}_2 \cdot \text{A}\beta(1-6)\text{-Tyr}$  models with Tyr modelled as 4-methylphenol.  $\text{A}\beta(1-6)$  is the DAEFRH peptide. The label of the structure is equal to that of Table S3a. The total net charge of this model is zero. The energy values  $E$  in hartree, the  $\Delta E$  values are in kcal/mol.  $\Delta E(\text{Tyr})$  is the 4-methylphenol binding energy,  $E(S=1)$  and  $E(S=0 \text{ BS})$  are the total energy for the triplet state and Broken-Symmetry singlet state after the H abstraction; the corresponding  $\Delta E(S=1)$  and  $\Delta E(S=0 \text{ BS})$  are the energy differences upon H abstraction computed between the  $S=0$  reactant and the  $S=1$  or  $S=0 \text{ BS}$  products.

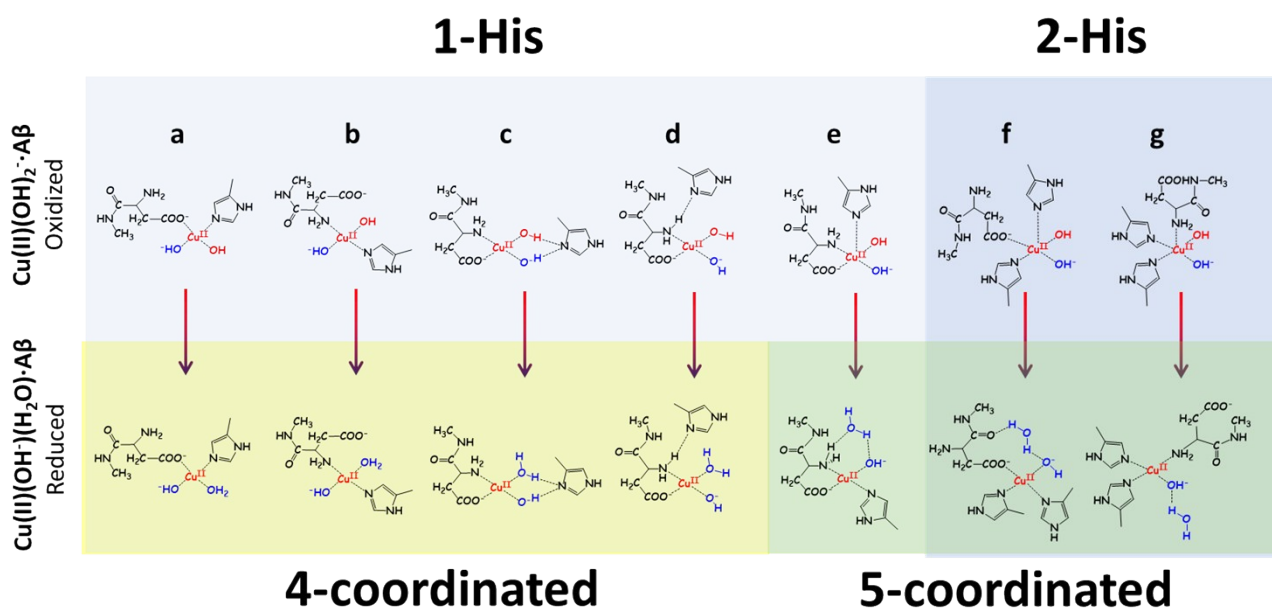
n	CN	apic	Cu-apic	oh	Diss		$E(S=0)$	$\Delta E(\text{Tyr})$	$E(S=1)$	$E(S=0 \text{ BS})$	$\Delta E S=1$	$\Delta E S=0 \text{ BS}$
1	4			90	His	2SB	-4875,3295	-11,5	-4875,3368	-4875,337	-4,6	-4,7
2	4			90	His	SB	-4875,3199	-5,6	-4875,3236	-4875,3242	-2,3	-2,7
3	5	His	2,680	90		SB	-4875,3206	-6,1	-4875,3227	-4875,3237	-1,3	-1,9



4	5	COO-	2,348	180		2SB	-4875,3188	-4,9	-4875,3247	-4875,3256	-3,7	-4,3
5	5	His	2,546	90		SB	-4875,3207	-6,6	-4875,3311	-4875,3317	-6,5	-7,0
7	5	His	2,540	90		SB	-4875,3167	-5,4	-4875,3235	-4875,3234	-4,3	-4,2
8	4			180	NH2	2SB	-4875,3142	-4,6	-4875,3172	-4875,3173	-1,9	-1,9
12	4	His	2,675	90		SB	-4875,3122	-6,0	-4875,3205	-4875,3209	-5,2	-5,5
13	5	His	2,299	90		SB	-4875,3118	-6,0	-4875,3226	-4875,3237	-6,8	-7,4
16	4			180	COO-	SB	-4875,3106	-6,1	-4875,3091	-4875,3092	0,9	0,9



Plot of the (1) relative energy  $\Delta E$  (in kcal/mol) computed with respect the most stable form ( $\Delta E$  tz) and; (2) energy differences upon H abstraction computed between the S=0 reactant and the S=0 BS products ( $\Delta E$  BS)



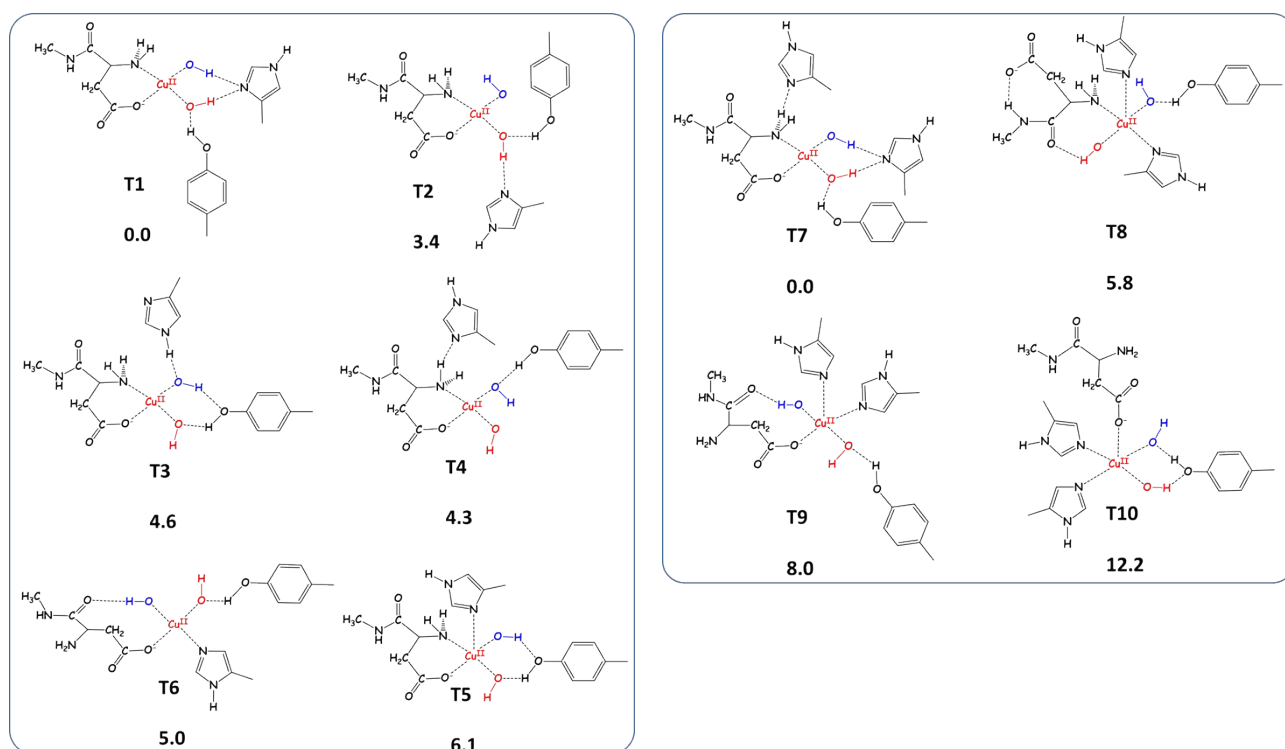
**Table S4.** DFT computed values of the relative standard half reaction reduction potential ( $\Delta\Delta E^\circ$  in Volt) for the 7 models reported in the scheme above (a-g) with respect to that computed to the  $(\text{H}_2\text{O})_{20}\text{OH}/(\text{H}_2\text{O})_{21}$  half reaction.  $\Delta E^\circ = 2.73 - \Delta\Delta E^\circ$  is the estimated  $\Delta E^\circ$  for the Cu(II) coordinated OH radical considering the experimental value of the OH/ $\text{H}_2\text{O}$  potential equal to 2.73 V. The Natural bond orbital copper atomic charges in the oxidized and reduced form ( $q(\text{Cu})_{\text{ox}}$  and  $q(\text{Cu})_{\text{red}}$ ) and are reported in electrons.

	$\Delta\Delta E^\circ$ (V)	$\Delta E^\circ = 2.73 - \Delta\Delta E^\circ$ (V)	$q(\text{Cu})_{\text{ox}}$	$q(\text{Cu})_{\text{red}}$
<b>a</b>	-1.61	1.12	1,31	1,20
<b>b</b>	-1.53	1.20	1,32	1,21
<b>c</b>	-1.42	1.31	1,31	1,21
<b>d</b>	-1.30	1.43	1,30	1,19
<b>e</b>	-1.00	1.73	1,33	1,21
<b>f</b>	-0.88	1.85	1,34	1,20
<b>g</b>	-0.87	1.86	1,37	1,23

**Potential energy surface scans.** The copper population are computed according to the Natural bond order approach. The effects of ZPE and thermal and entropic contributions on the purely electronic total energy values to compute free energies were investigated by means of evaluation of the approximated roto-translational partition function of each molecular species, at  $T = 298$  K and  $P = 1$  bar.

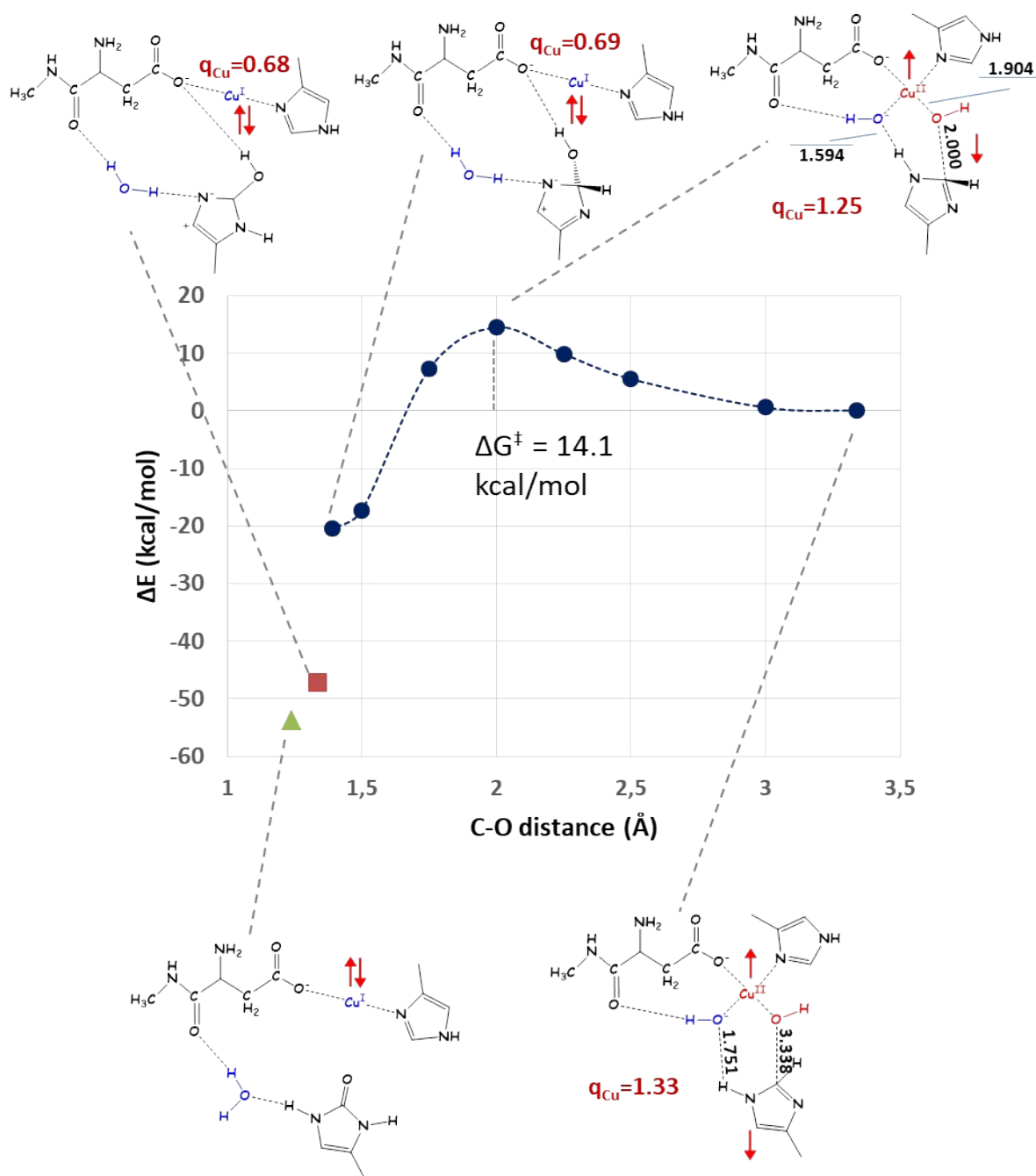
**Table S5.** Tyr side chain adducts (T1-T10 according to the panel below). Singlet ground state total energy  $E(S=0)$ , triplet and singlet broken symmetry total energy  $E(S=1)$  and  $E(S=0$  BS) in Hartree.  $\Delta E$  is the relative energy of 1-His and 2-His adducts computed against the most stable form.  $\Delta E$  ( $S=1$ ) and  $\Delta E$  ( $S=0$  BS) are the energy differences between  $S=0$  Tyr adduct and  $S=1$  of  $S=0$  BS form after H abstraction. All  $\Delta E$  in kcal/mol.

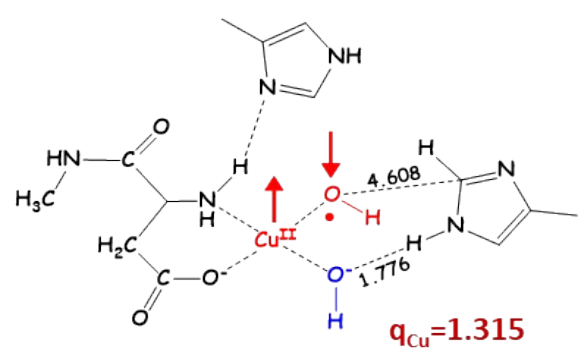
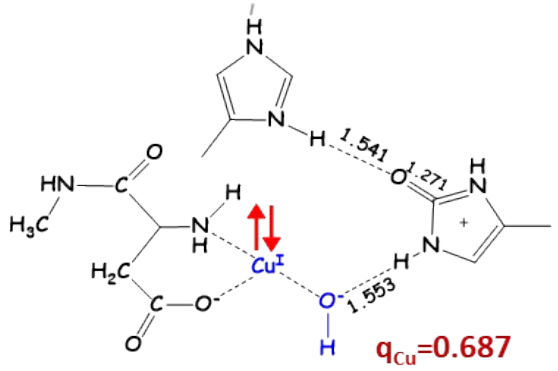
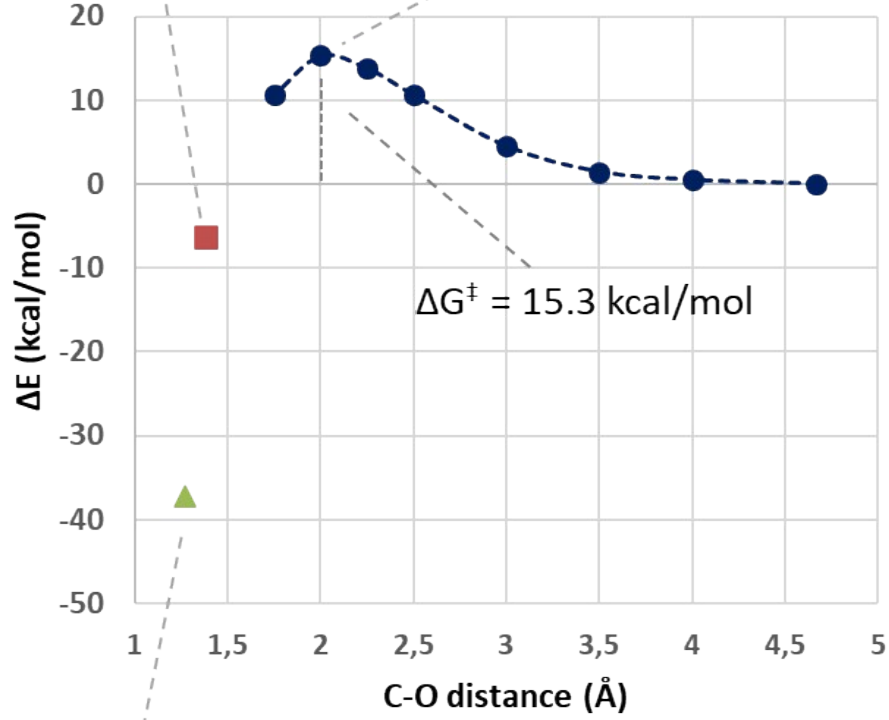
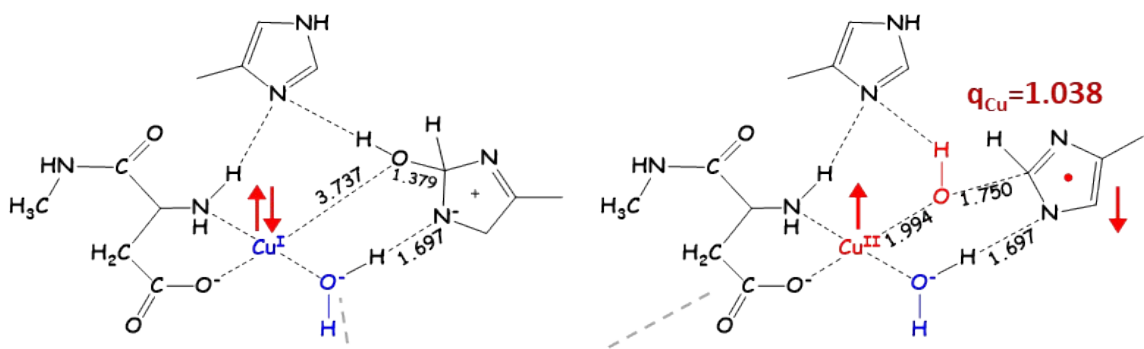
	$E(S=0)$	$\Delta E$	$E(S=1)$	$E(S=0$ BS)	$\Delta E$ ( $S=1$ )	$\Delta E$ ( $S=0$ BS)
<b>1-His</b>						
T1	-2936,6475	0	-2936,6425	-2936,6436	3,1	2,4
T2	-2936,6421	3,4	-2936,6407	-2936,6406	0,9	0,9
T3	-2936,6406	4,3	-2936,6345	-2936,6358	3,8	3,0
T4	-2936,6401	4,6	-2936,6406	-2936,6407	-0,3	-0,4
T5	-2936,6395	5,0	-2936,6283	-2936,6359	7,0	2,3
T6	-2936,6378	6,1	-2936,6383	-2936,6383	-0,3	-0,3
<b>2-His</b>						
T7	-3123,6567	0	-3123,6481	-3123,6503	5,0	4,0
T8	-3123,6474	5,8	-3123,6451	-3123,6500	1,4	0,8
T9	-3123,6439	8,0	-3123,6366	-3123,6435	4,6	0,3
T10	-3123,6373	10,2	-3123,6412	-3123,6435	-2,5	-3,9

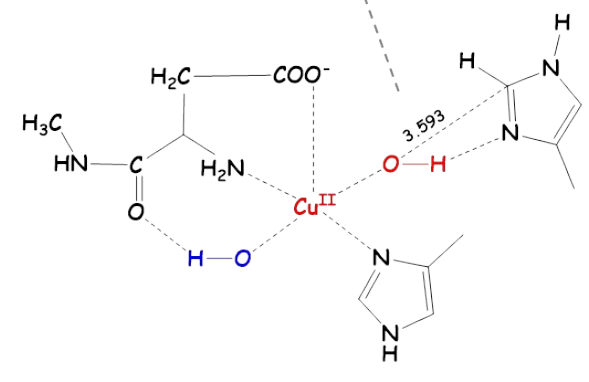
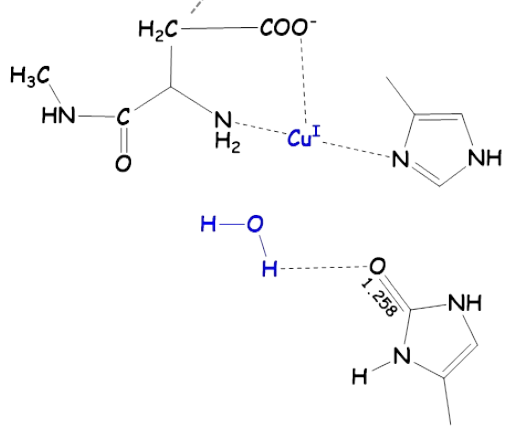
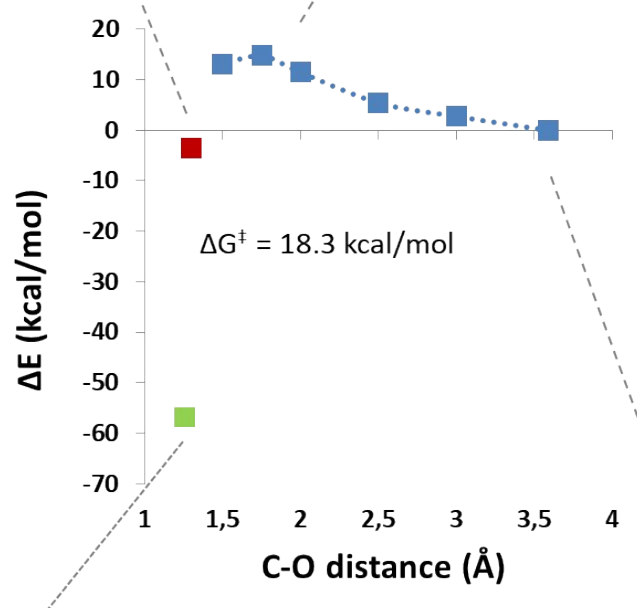
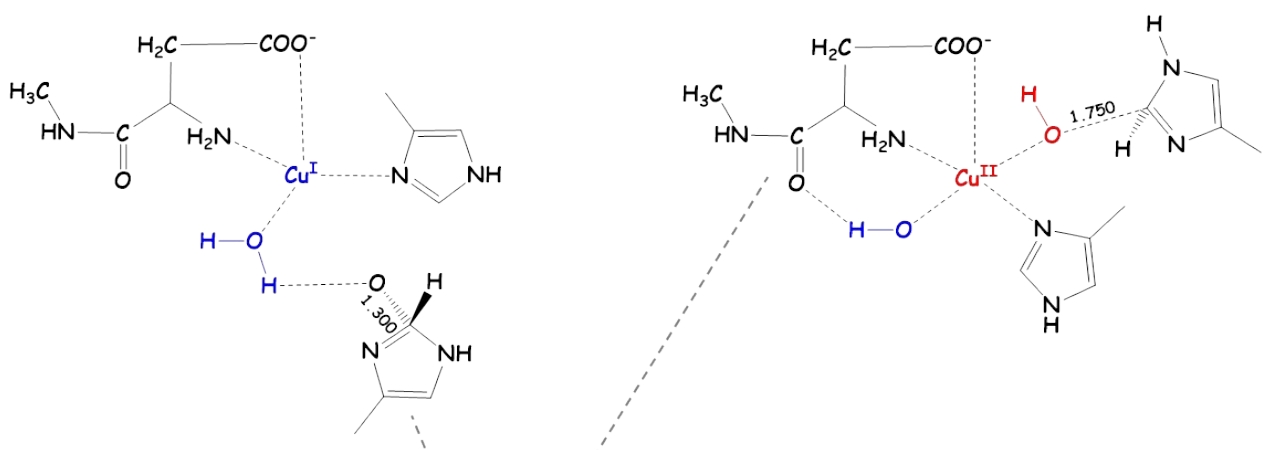


**Scheme S3.** 1-His (on left) and 2-His (on right) Tyrosine model structures. Energy differences calculated with respect to the most stable in kcal/mol.

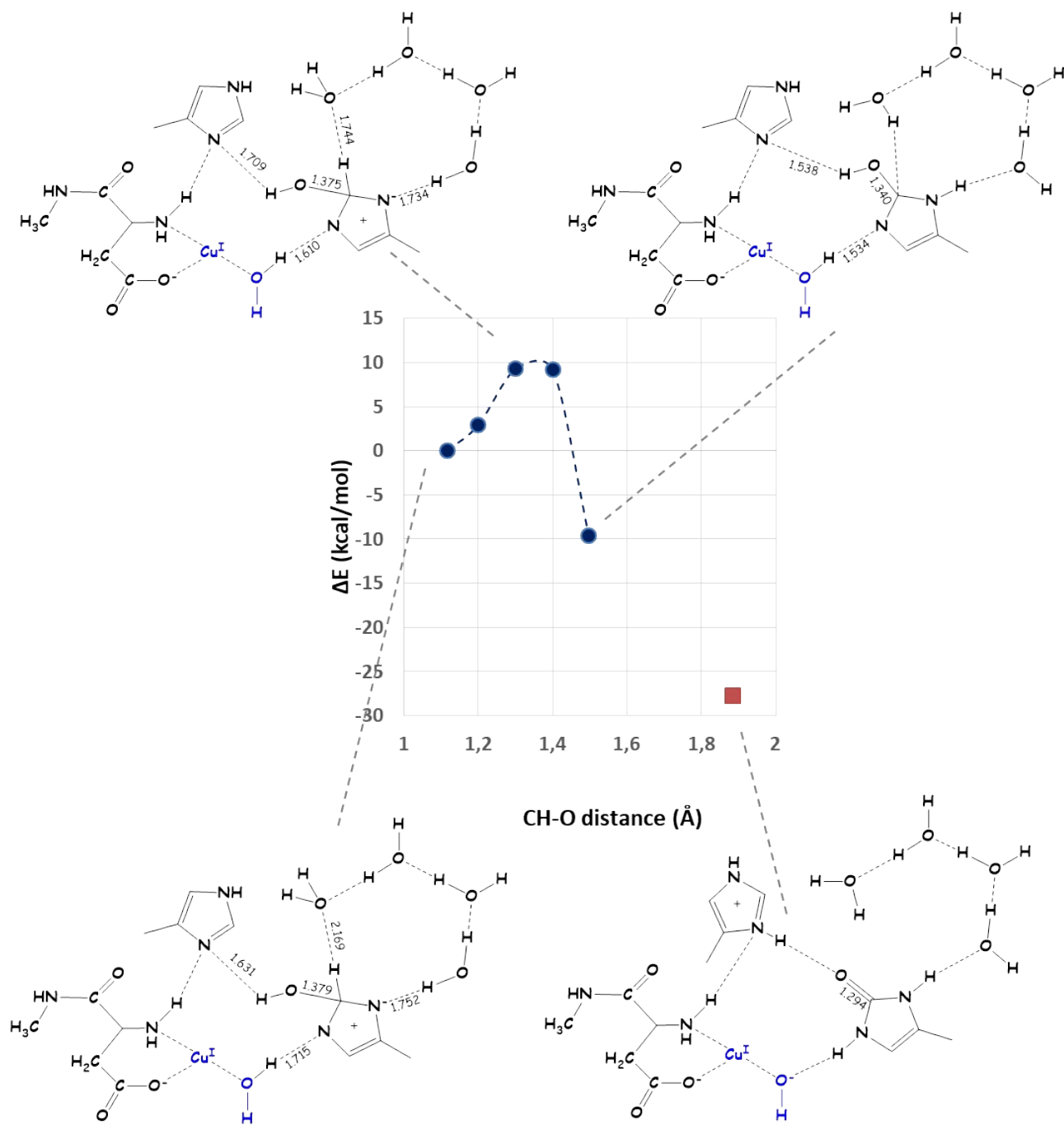
**Figure S1.** S=0 Potential energy surface scans computed along the His oxidation pathway.



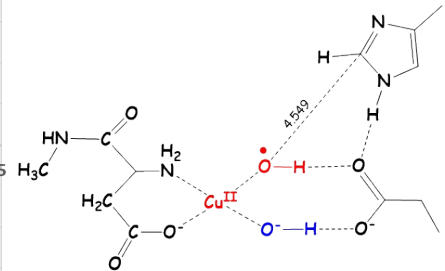
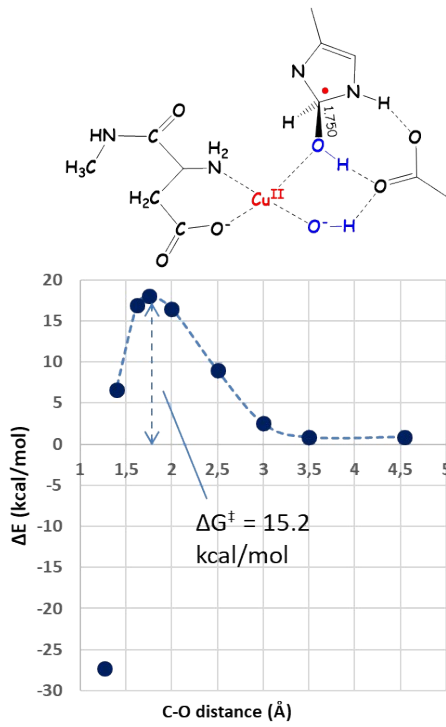
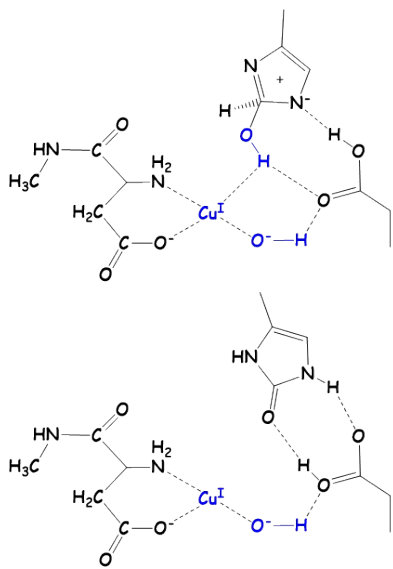




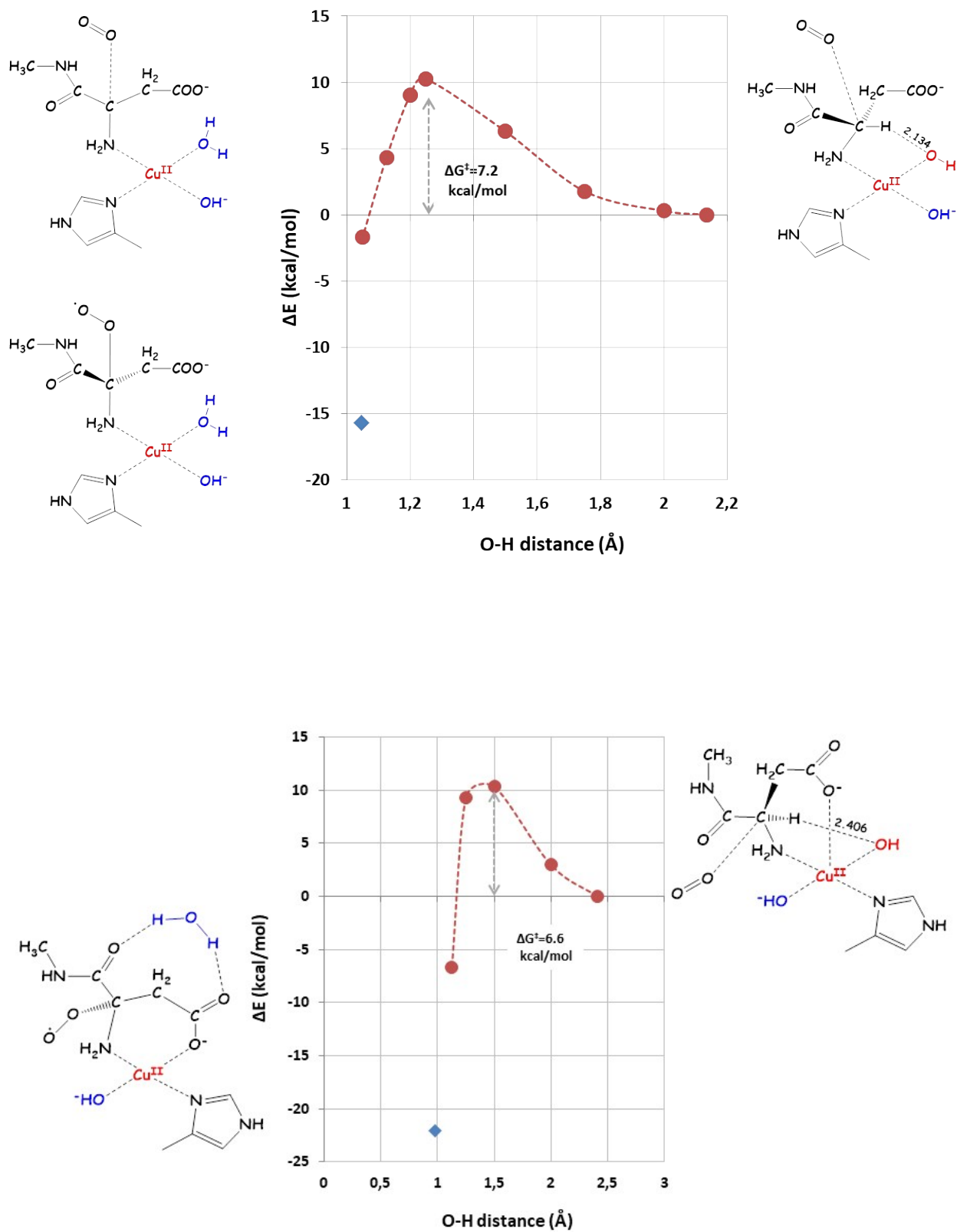
**Figure S2.** His oxidation Proton transfer. Potential energy surface scans computed along the His oxidation pathway. Characterization of the proton transfer assisted rearrangement of the CHOH group that belong to the zwitterionic form of the 2-oxo imidazole.

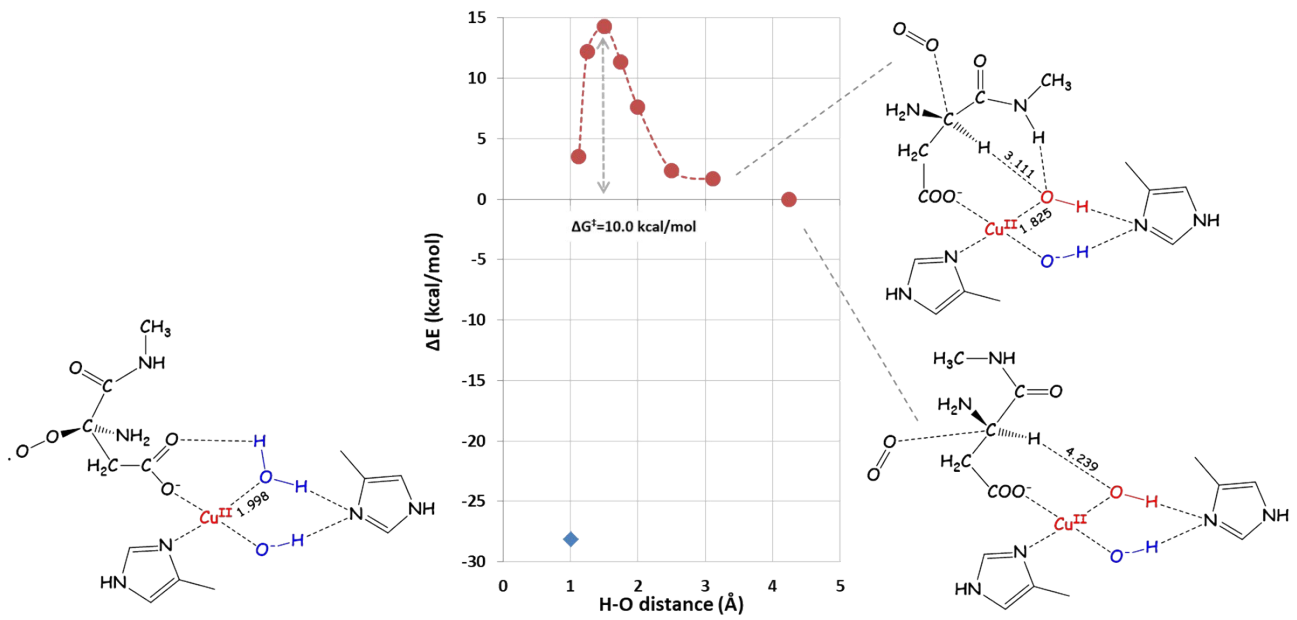
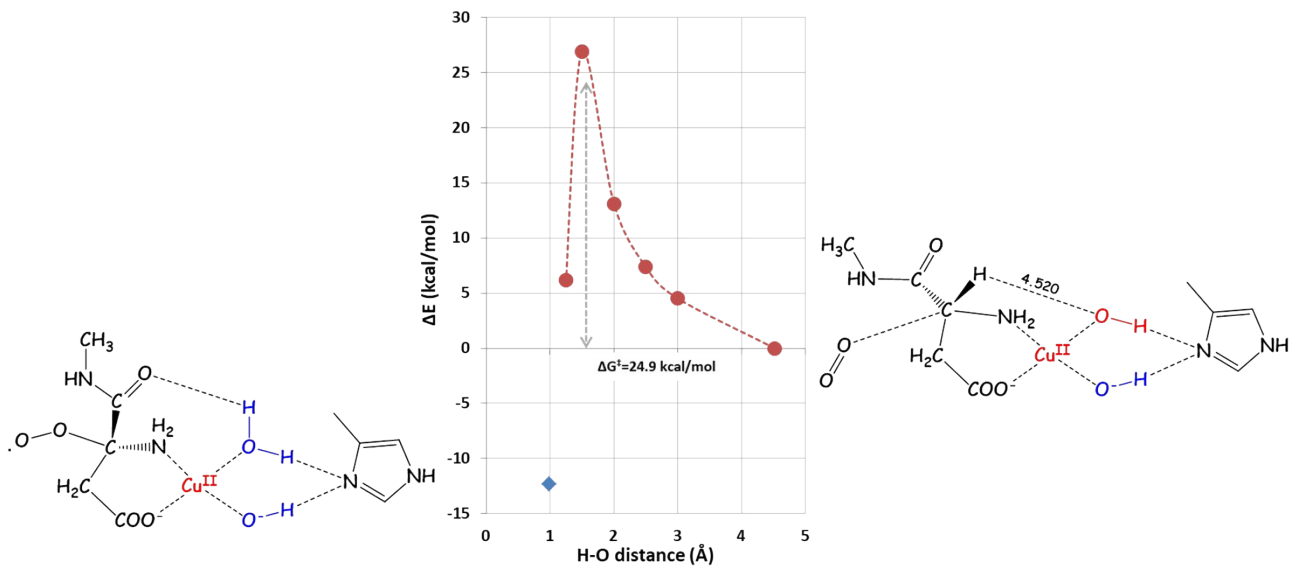


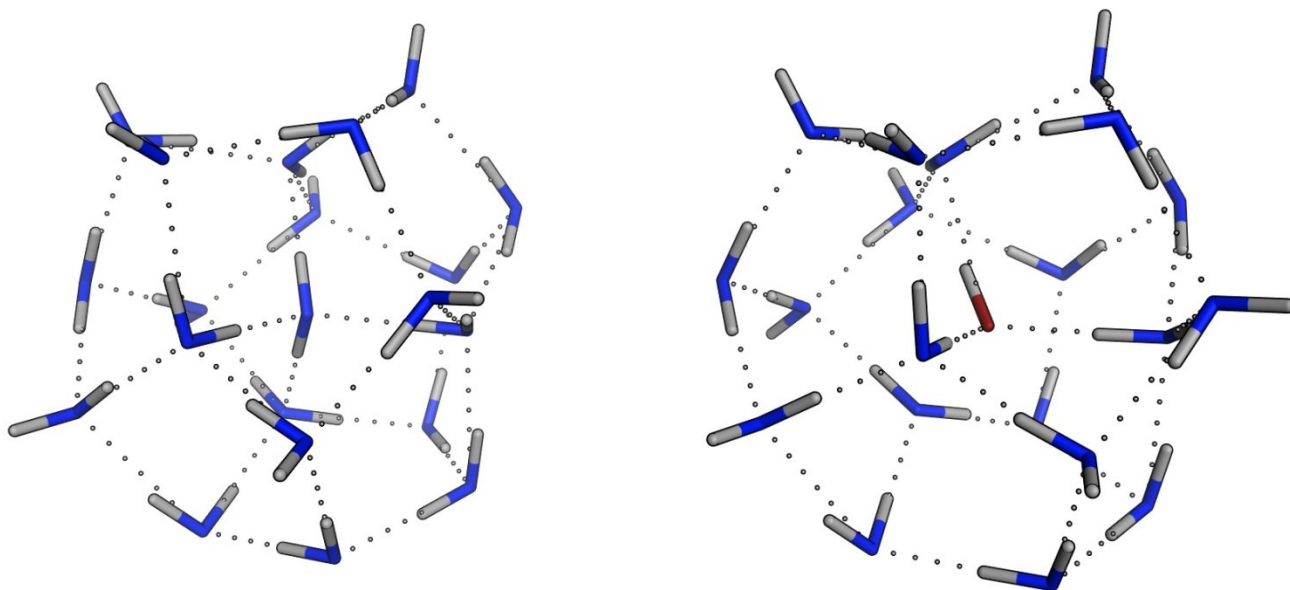




**Figure S3.** Potential energy surface scans computed along the C $\alpha$ -H hydrogen abstraction / O<sub>2</sub> addition Asp1 oxidation reaction coordinate.





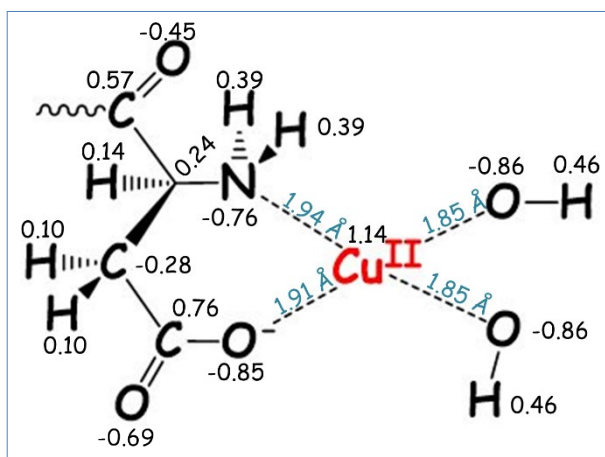


**Figure S4.** Structures of the most stable  $(\text{H}_2\text{O})_{21}$  and  $(\text{OH})(\text{H}_2\text{O})_{20}$  forms.  $(\text{H}_2\text{O})_{21}$  is characterized by a single water molecule in the center of an  $(\text{H}_2\text{O})_{20}$  distorted dodecahedral cage, in agreement with Cui et al. (*Theoretical Characterization of the  $(\text{H}_2\text{O})_{21}$  Cluster: Application of an  $n$ -body Decomposition Procedure*; J. Phys. Chem. B 2006, 110, 38, 18872-18878) findings.  $(\text{OH})(\text{H}_2\text{O})_{20}$  most stable form derived from the most stable  $(\text{H}_2\text{O})_{21}$  by removing one H atom from the internal water molecules (in the figure below in red).

**Table S6.**  $(\text{H}_2\text{O})_{21}$  and  $(\text{H}_2\text{O})_{20}\text{OH}$ . E(BP86) total energy and relative energy with respect to the most stable form ( $\Delta E$ ) in kcal/mol.  $\mu$  is the chemical potential such that the free energy G is computed as  $G = E(\text{BP86}) + \mu/2626,754767$ . G and  $\mu$  in hartree.  $\Delta E^\circ$  and  $\Delta\Delta E^\circ$  are the half-reaction and relative half-reaction potentials in Volt, considering the OH radical reduction potentials are  $\text{OH} + e^- + \text{H}^+ \rightarrow \text{H}_2\text{O}$  equal to 2.730 V vs SHE.

$(\text{H}_2\text{O})_{21}$							
	E(BP86)	$\Delta E$	$\mu$	G			
1	-1606,2516	0.5	1123,47	-1605,82394			
2	-1606,2517	0	1124,07	-1605,82373			
$(\text{H}_2\text{O})_{20}\text{OH}$							
	E(BP86)	$\Delta E$	$\mu$	G	$\Delta G$	$\Delta E^\circ$	$\Delta\Delta E^\circ$
1	-1605,5504	0	1085,35	-1605,13718	-0,69	18,68	-1,42
2	-1605,5485	1,19					
3	-1605,5485	1,19					
4	-1605,5476	1,74					
5	-1605,5397	6,70					
6	-1605,5314	11,90					
7	-1605,5309	12,23					
8	-1605,5287	13,59					
9	-1605,5287	13,60					
10	-1605,5263	15,10					
11	-1605,5244	16,30					
12	-1605,5233	16,99					
13	-1605,5208	18,54					
14	-1605,5207	18,62					
15	-1605,5174	20,70					
16	-1605,5109	24,77					
17	-1605,5086	26,23					
18	-1605,5086	26,23					

**Scheme S4.** Ligands parameters used for the MD simulations. The MP2/6-31G\* atomic charges for CHARMM36m force field parametrization of the Cu(II) (OH)<sub>2</sub>-Aβ four-coordinated model at the equilibrium distances. A conventional bond constant of 200000 kJ mol<sup>-1</sup> nm<sup>-2</sup> has been used for all coordination bonds between Cu(II) and its ligands. An equilibrium angle of 90 degrees have been used between Cu(II) and its ligands with a conventional angle constant of 10000 kJ mol<sup>-1</sup> rad<sup>-2</sup>. interactions between atoms in the coordination complex were turned off. TIP3P atom types have been used for OH radicals. CHARMM36m bond and Lennard-Jones parameters were used for Asp1



**Table S7.** Average Centre-of-mass distances (in nm) among hAβ(1-16) residues involved in salt-bridge interactions (positively charged Arg5 and Lys16 with Asp1 with negatively charged Glu3, Asp7 and Glu11, see inset below) along with the number of frames with centre-of-mass distances less than 0.4 nm and the corresponding percentage, obtained from the MD simulation sampled every 100 ps.



	lys16-glu11	lys16-glu3	lys16-asp7	lys16-asp1	arg5-glu11	arg5-glu3	arg5-asp7	arg5-asp1
Average	1,292 ± 0,447	2,203 ± 0,839	1,883 ± 0,604	2,246 ± 0,870	1,479 ± 0,551	0,798 ± 0,351	0,951 ± 0,334	0,898 ± 0,440
Frames <0,4 nm	2914	2473	1053	2693	1410	6504	4955	9427
%	5,8	4,9	2,1	5,4	2,8	13,0	9,9	18,9