Supplementary Information

Structural Determination of Fe^{2+}, Cu^{2+}, and Zn^{2+} Complexed with Glutathione by IRMPD Spectroscopy and Complimentary ab Initio Calculations

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Contents:

Table S1. Mass-to-charge ratios for the products monitored for FELIX experiments

$Fe(GSH-H)^+$ m/z		**$Cu(GSH-H)^+$ m/z**	**$Zn(GSH-H)^+$ m/z**	
361.9	214.6	368.9	369.9	220.6
344.8	212.6	367.9	367.9	212.6
343.9	208.6	366.9	361.4	205.6
325.9	206.6	351.9	351.9	204.6
317.9	204.6	341.8	349.9	203.6
315.9	198.6	338.4	332.4	202.6
309.8	197.6	332.4	323.9	194.6
300.8	196.6	330.8	306.8	193.6
299.8	189.6	325.8	305.8	184.7
298.8	188.6	324.9	300.7	180.6
297.8	187.6	323.9	298.7	175.6
286.8	186.6	322.9	296.8	155.6
283.8	184.6	321.9	295.8	151.6
282.8	173.5	308.8	294.8	140.7
272.7	172.6	306.8	289.8	139.7
271.8	171.6	305.9	287.8	138.7
270.7	170.6	304.8	280.7	137.6
269.7	169.6	294.8	279.7	129.8
258.7	160.6	293.8	278.7	123.8
255.8	157.6	278.8	277.8	
254.7	155.6	267.7	276.7	
253.7	147.6	266.7	268.7	
252.7	143.6	265.7	267.7	
251.7	141.6	260.7	266.7	
250.7	139.7	250.7	260.7	
243.7	129.7	242.7	259.7	
242.7		235.6	250.7	
240.7		230.7	248.7	
235.6		222.6	243.7	
233.6		192.6	240.7	
232.6		176.7	238.7	
230.6		152.8	232.7	
226.7		148.7	230.7	
225.7		129.8	223.6	
224.6			222.6	
215.6			221.6	

Table S2. Additional structures not listed in main text. Relative energies (E_{rel} in kJ/mol) are 298 K Gibbs energies at the level of theory indicated. Calculated energies, zero-point energies (ZPE), and 298 K thermal corrections (TC) are listed in Hartree. Frequencies scaled (sc) by 0.9896. Structures in bold are included in Tables in the main text and are usually the lowest energy structure of a particular binding motif. The designation of the metal binding site is followed by the amino acid orientation, which is represented by characterization of dihedral angles along the peptide backbone as cis (c, for angles between 0°– 45°), gauche (g, 45°–135°), or trans (t, 135°– 180°). Dihedrals are measured from the nitrogen atom of the N-terminus to the C-terminus ending with the carboxylic acid hydrogen.

Zn(GSH-H)$^+$	E_{rel}	B3LYP/				
Binding Motif	kJ/mol	6-311+G(d,p)	ZPE	TC	ZPE(sc)	TC(sc)
[N^1, CO$^\gamma$, S$^-$, CO^3]-ggttcgtgtt	**0.4**	**-3184.093343**	**0.286782**	**0.23597**	**0.283799**	**0.232792**
[N^1, CO^1, CO$^\gamma$, CO^{3-}]-gggtgttgt	**7.8**	**-3184.088693**	**0.284194**	**0.234144**	**0.281239**	**0.230995**
[CO^1, CO$^\gamma$, S$^-$, CO^3]-gggtcgtgtt	**6.4**	**-3184.090269**	**0.286024**	**0.235208**	**0.28305**	**0.232037**
[N^1, CO$^\gamma$, N^{2}H, S$^-$]-gggtgttttt	**0.0**	**-3184.0905**	**0.286507**	**0.232994**	**0.283527**	**0.229814**
[N^1, N^{2}H, S$^-$, CO^2]-gggtgttttt	2.3	-3184.088867	0.286277	0.232241	0.2833	0.229062
[N^1, CO^{1-}, N^2, SH, CO^3]-ggctgctgtt	20.5	-3184.08097	0.285745	0.231278	0.282773	0.228103
[CO^1, S$^-$, CO^2, CO^3]-gggtgttgtt	23.5	-3184.084269	0.285899	0.235718	0.282925	0.232549
[N^1, N^2, S$^-$, CO^3]-ggctgctgtt	26.3	-3184.080177	0.28618	0.232698	0.283204	0.229521
[N^1, CO^1, N^2, S–, CO^2]-ggctgttttt	32.6	-3184.076719	0.285333	0.231614	0.282366	0.228443
[N^1, N^2, S$^-$, N^3, CO^3]-ggctgctttt	38.1	-3184.076022	0.285362	0.232997	0.282394	0.229834
[N^1, N^2, S$^-$, CO^3]-ggcttctgtt	52.3	-3184.071583	0.285871	0.234007	0.282898	0.230834
[N^1, N^2, S$^-$, CO^3]-ggctgctgtt	54.7	-3184.070631	0.28594	0.233939	0.282967	0.230766
[N^1, CO^{1-}, N^2, SH, CO^3]-ggctgctgtt	54.8	-3184.069865	0.283321	0.233181	0.280374	0.230038
[N^1, N^2, S$^-$, N^3, CO^3]-ggctgctttt	57.7	-3184.068296	0.285166	0.232769	0.2822	0.229601

Cu(GSH-H)$^+$	E_{rel}	B3LYP/				
Binding Motif	kJ/mol	6-311+G(d,p)	ZPE	TC	ZPE(sc)	TC(sc)
[N^1, CO^1, N^{2}H, S$^-$]-ggttttttt	**0.0**	**-3045.229718**	**0.285842**	**0.230838**	**0.282869**	**0.227662**
[N^1, S$^-$, CO^2]-gggtcgtttt	**20.5**	**-3045.222133**	**0.285647**	**0.231069**	**0.282677**	**0.227895**
[N^1, CO$^\gamma$, S$^-$, CO^3]-ggttcgtgtt	**54.9**	**-3045.213215**	**0.286349**	**0.23526**	**0.283371**	**0.232087**

[N^1, CO^1, N^2, S^-, CO^2]-gggttttttt	**52.6**	**-3045.209712**	**0.285341**	**0.230849**	**0.282374**	**0.227678**
[N^1, CO^γ, S^-]-ggttttttt	25.4	-3045.219712	0.285498	0.23051	0.282529	0.227337
[CO^1, CO^γ, S^-, CO^3]-gggtcgtgtt	55.8	-3045.210295	0.284871	0.23264	0.281908	0.229475
[CO^1, S^-, CO^2, CO^3]-gggtgttgtt	59.3	-3045.20866	0.284585	0.23235	0.281626	0.229188
[N^1, CO^1, CO^γ, CO_2^-]-gggtgttgt	73.0	-3045.204958	0.284035	0.233835	0.281081	0.230688
[N^1, CO^1, S^-, CO^3]-gggttctgtt	86.2	-3045.198843	0.2853	0.232781	0.282333	0.229612
[N^1, CO^1, S^-, CO^3]-gggttctgtt	86.5	-3045.199334	0.28545	0.233379	0.282481	0.23021
[N^1, CO^{1-}, SH, N^3, CO^3]-gggttgtttt	98.4	-3045.193545	0.28299	0.232091	0.280047	0.228951
[N^1, CO^1, S^-, N^3, CO^3]-gggttctttt	98.9	-3045.193209	0.284638	0.23197	0.281678	0.228807
[N^1, CO^1, S^-, N^3, CO^3]-gggttctttt	105.1	-3045.190896	0.284618	0.232032	0.281658	0.228869
[N^1, CO^1, N^2, S^-, N^3, CO^3]-gggttctttt	105.1	-3045.190896	0.284618	0.232032	0.281658	0.228869
[N^1, CO^1, N^3]-ggctgctttt	116.5	-3045.185047	0.282081	0.230484	0.279147	0.227351
[N^1, CO^{1-}, SH, N^3]-ggctgctttt	120.1	-3045.183513	0.282069	0.230344	0.279135	0.227211

Fe(GSH-H)$^+$-Triplet Binding Motif	E_{rel} kJ/mol	B3LYP/ 6-311+G(d,p)	ZPE	TC	ZPE(sc)	TC(sc)
[N^1, CO^γ, S^-, NH, CO^3]-ggttcgtgtt	**0.0**	**-2668.441313**	**0.287347**	**0.237129**	**0.284359**	**0.233952**
[N^1, CO^1, N^2, S^-, CO^2]-gggttttttt	**3.0**	**-2668.437018**	**0.286477**	**0.233971**	**0.283498**	**0.230797**
[CO^1, CO^γ, S^-, CO^3]-gggtcgtgtt	**15.4**	**-2668.435036**	**0.28656**	**0.236692**	**0.28358**	**0.233522**
[N^1, CO^1, CO^γ, CO^{3-}]-gggtgttgt	**16.3**	**-2668.432841**	**0.284843**	**0.234837**	**0.28188**	**0.231686**
[N^1, CO^1, N^2, SH, CO^3]-ggctgctgtt	22.9	-2668.428562	0.283956	0.233057	0.281002	0.229913
[N^1, CO^{1-}, SH, N^3, CO^3]-gggttgtttt	26.8	-2668.428204	0.284121	0.23418	0.281166	0.231034
[N^1, CO^1, N^2, S^-, N^3, CO^3]-gggttctttt	39.0	-2668.424043	0.285694	0.234688	0.282722	0.231521
[N^1, CO^{1-}, N^2, N^3]-ggctgctttt	38.6	-2668.421736	0.283043	0.232202	0.280099	0.229064
[N^1, CO^1, N^2, S^-, N^3, CO^3]-gggttctttt	46.7	-2668.421043	0.285657	0.23464	0.282686	0.231473

[N^1, CO^1, N^2, S^-, N^3, CO^3]-gggttctttt	46.7	-2668.421043	0.285657	0.234639	0.282686	0.231473
[N^1, CO^{1-}, N^2, N^3]-ggctgctttt	41.6	-2668.420544	0.283077	0.232174	0.280133	0.229036
[N^1, CO^1, N^2, S^-, CO^3]-gggttctgtt	52.3	-2668.41926	0.286206	0.234989	0.283229	0.231817
[N^1, N^2, S^-, N^3H, CO^3]-ggctgctgtt	69.2	-2668.411784	0.285707	0.233948	0.282736	0.230783
[CO^1, S^-, CO^2, CO^3]-gggtgttgtt	73.4	-2668.410353	0.28562	0.234107	0.28265	0.23094

Fe(GSH-H)$^+$-Quintet Binding Motif	E_{rel} kJ/mol	B3LYP/ 6-311+G(d,p)	ZPE	TC	ZPE(sc)	TC(sc)
[CO^1, S^-, CO^2, CO^3]-gggtgttgtt	**34.8**	**-2668.462488**	**0.285587**	**0.233548**	**0.282617**	**0.230381**
[CO^1, CO^γ, S^-, CO^3]-gggtcgtgtt	**12.4**	**-2668.470776**	**0.285808**	**0.233329**	**0.282835**	**0.230159**
[N^1, CO^1, N^2, S^-, CO^2]-ggctttttt	**37.7**	**-2668.458361**	**0.285407**	**0.230525**	**0.282439**	**0.227354**
[N^1, N^2H, S^-, CO^2]-gggtgttttt	**34.1**	**-2668.459567**	**0.285997**	**0.230385**	**0.283022**	**0.227209**
[N^1, CO^1, CO^γ, CO^{3-}]-gggtgttgt	0.0	-2668.474078	0.283848	0.231874	0.280896	0.228727
[N^1, CO^1, N^2H, S^-]-ggttgttttt	44.7	-2668.456068	0.286103	0.23093	0.283128	0.227754
[CO^1, S^-, CO^2, CO^3]-gggtgttgtt	58.6	-2668.45244	0.284988	0.232563	0.282024	0.229398
[N^1, CO^1, N^2, CO^3]-ggctgctgtt	61.9	-2668.448902	0.282646	0.23025	0.279706	0.227113
[N^1, CO^{1-}, N^2, N^3, CO^3]-gggttctttt	63.5	-2668.445588	0.281325	0.227552	0.2784	0.224423
[N^1, CO^{1-}, N^3, CO^3]-gggttctttt	67.0	-2668.444195	0.281376	0.227488	0.27845	0.224358
[N^1, N^2, S^-, N^3, CO^3]-gggtgctttt	68.5	-2668.446749	0.284611	0.230648	0.281651	0.227485
[N^1, CO^1, N^2, S^-, CO^3]-gggtctgtt	69.5	-2668.448241	0.285313	0.232544	0.282346	0.229377
[N^1, N^2, S^-, CO^3]-gggttctgtt	69.8	-2668.448573	0.285343	0.232978	0.282376	0.229813
[N^1, CO^1, N^2H, S^-]-ggttgttttt	70.5	-2668.44605	0.285805	0.230716	0.282833	0.227542

Table S3. Relative Enthalpies at 0 K (Gibbs Energies at 298 K) of $Zn(GSH-H)^+$ in kJ/mol

Structure	B3LYP// B3LYP	B3LYP// MP2	B3LYP-GD3BJ// B3LYP-GD3BJ	ωB97XD// B3LYP	ωB97XD// MP2	MP2(full)// B3LYP	MP2(full)// MP2
[N^1, CO^γ, S^-, CO^3]-ggttcgtgtt	0.0 (0.0)	0.0 (0.7)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)
[N^1, CO^1, CO^γ, CO^{3-}]-gggtgttgt	2.3 (4.3)	2.4 (7.1)	1.8 (4.9)	4.2 (6.2)	3.9 (7.8)	0.4 (2.4)	1.7 (5.7)
[CO^1, CO^γ, S^-, CO^3]-gggtcgtgtt	5.1 (5.1)	6.4 (7.4)	9.0 (9.4)	7.5 (7.5)	8.2 (8.4)	16.4 (16.4)	17.1 (17.4)
[N^1, CO^γ, N^2H, S^-]-gggtgttttt	7.3 (0.1)	5.8 (0.0)	23.6 (16.4)	22.2 (15.1)	21.4 (14.8)	18.4 (11.3)	20.0 (13.4)

Table S4. Relative Enthalpies at 0 K (Gibbs Energies at 298 K) of $Cu(GSH-H)^+$ in kJ/mol

Structure	B3LYP// B3LYP	B3LYP// MP2	B3LYP-GD3BJ// B3LYP-GD3BJ	ωB97XD// B3LYP	ωB97XD// MP2	MP2(full)// B3LYP	MP2(full)// MP2
[N^1, CO^1, N^2H, S^-]-ggttttttt	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	47.1 (36.8)	39.4 (29.9)
[N^1, S^-, CO^2]-gggtcgtttt	21.3 (22.4)	3.7 (2.8)	18.1 (19.8)	22.3 (23.4)	6.9 (6.0)	33.9 (24.7)	30.7 (20.2)
[N^1, CO^γ, S^-, CO^3]-ggttcgtgtt	46.6 (56.9)	27.3 (36.8)	19.4 (30.2)	22.6 (32.9)	4.8 (14.3)	0.0 (0.0)	0.0 (0.0)
[N^1, CO^1, N^2, S^-, CO^2]-gggttttttt	49.9 (51.3)	30.3 (31.5)	44.7 (47.8)	45.8 (47.1)	25.1 (26.3)	30.2 (21.2)	28.1 (19.8)

Table S5. Relative Enthalpies at 0 K (Gibbs Energies at 298 K) of quintet $Fe(GSH-H)^+$ in kJ/mol

Structure	B3LYP// B3LYP	B3LYP// MP2	B3LYP-GD3BJ// B3LYP-GD3BJ	ωB97XD// B3LYP	ωB97XD// MP2	MP2(full)// B3LYP	MP2(full)// MP2
$[CO^1, S^-, CO^2, CO^3]$-gggtgttgtt	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)
$[CO^1, CO^\gamma, S^-, CO^3]$-gggtcgtgtt	9.9 (8.7)	12.0 (12.0)	19.5 (16.7)	11.2 (10.0)	8.4 (8.4)	28.6 (27.4)	13.8 (13.9)
$[N^1, CO^1, N^2, S^-, CO^2]$-ggctttttt	40.6 (33.2)	19.3 (10.6)	60.4 (50.2)	34.9 (27.4)	33.0 (24.3)	47.6 (40.1)	20.0 (11.3)
$[N^1, N^2H, S^-, CO^2]$-gggtgttttt	41.3 (31.9)	19.0 (7.6)	62.9 (52.1)	39.4 (30.0)	37.0 (25.6)	64.1 (54.7)	31.8 (20.5)

Table S6. Experimental vibrations for $Zn(GSH-H)^+$ and corresponding calculated motions at the MP2/6-311+G(d,p)

Label	Wavenumber (cm^{-1})	Corresponding Motion
ν^1	1763	CO^1 stretch
ν^2	1688	CO^3 stretch
ν^3	1620	CO^γ stretch
ν^4	1543	CN^2H in-plane bend
ν^5	1426	CO^3H in-plane bend
ν^6	1284	CH_2 twists
ν^7	1231	N^1H_2 twist and CN^2H and CN^3H in-plane bends
ν^8	1141	CO^1H in-plane bend
ν^9	1073	N^1C stretch and CC backbone stretch

Table S7. Experimental vibrations for $Cu(GSH-H)^+$ and corresponding calculated motions at the MP2/6-311+G(d,p)

Label	Wavenumber (cm^{-1})	Corresponding Motion
ν'^1	1770	CO^3 stretch
ν'^2	1685	CO^γ stretch
ν'^3	1587	N^1H_2 scissor
ν'^4	1505	CN^3H in-plane bend
ν'^5	1423	CO^3H in-plane bend
ν'^7	1246	CO^3H in-plane bend and CH_2 wag
ν'^8	1159	CO^3H in-plane bend
ν'^9	1102	N^1H_2 wag

Table S8. Experimental vibrations for $Fe(GSH-H)^+$ and corresponding calculated motions at the MP2/6-311+G(d,p)

Label	Wavenumber (cm^{-1})	Corresponding Motion
ν''^2	1685	CO^γ stretch
ν''^3	1602	N^1H_2 scissor
ν''^4	1517	CN^3H in-plane bend
ν''^5	1425	CO^3H in-plane bend and nearby CH_2 wag
ν''^6	1263	CO^1H in-plane bend and nearby CH bend
ν''^7	1219	Not well assigned
ν''^8	1162	CO^3H in-plane bend
ν''^9	1040	Not well assigned

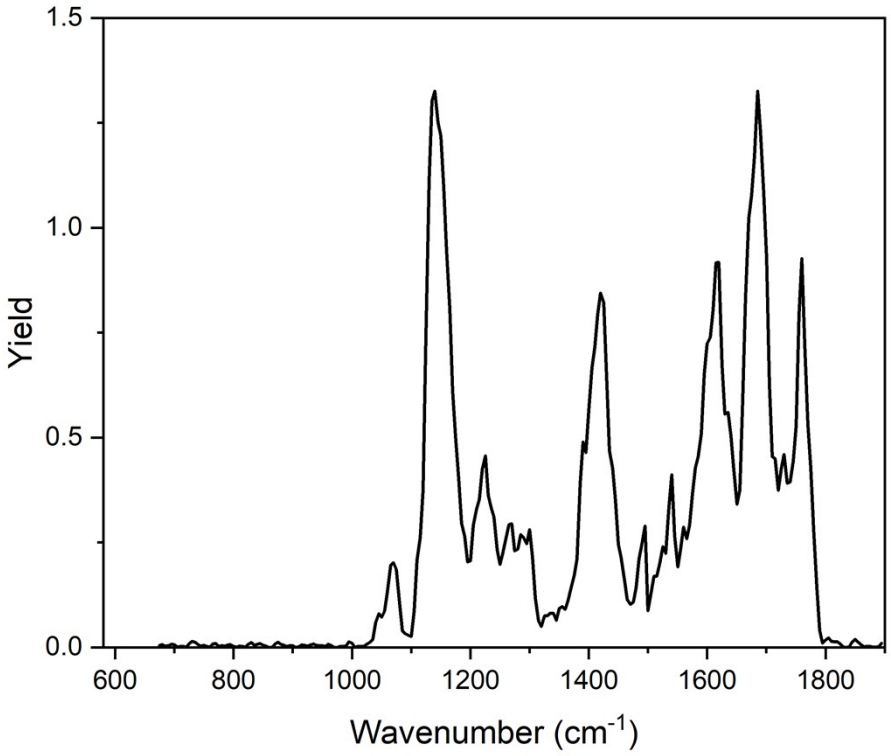
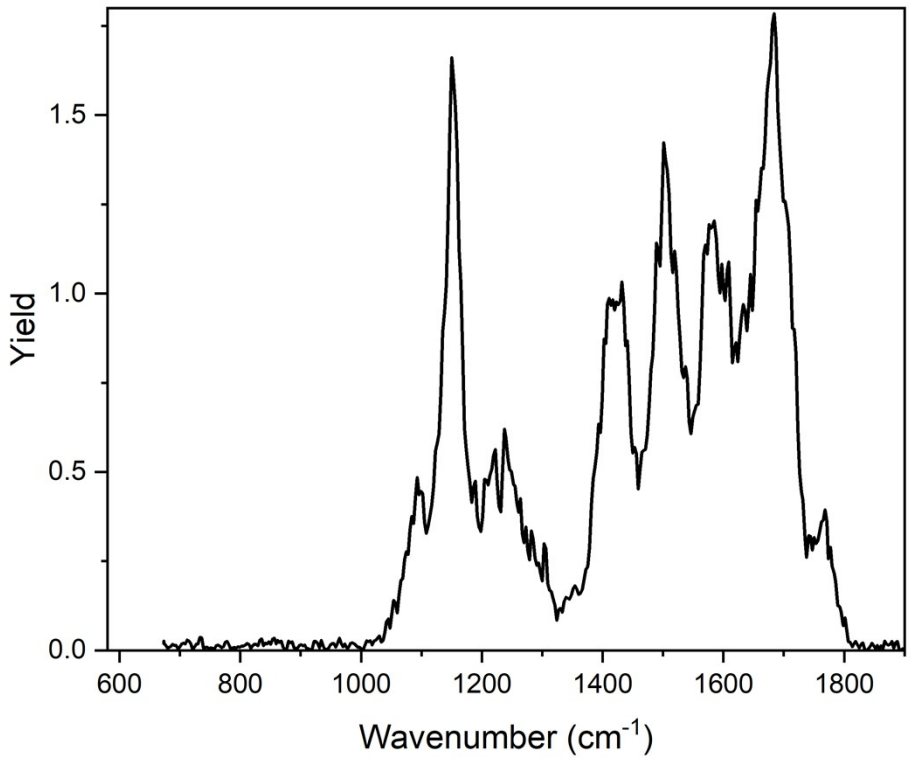


Figure S1. $Zn(GSH-H)^+$ full IRMPD experimental spectrum.

Figure S2. $Cu(GSH-H)^+$ full IRMPD experimental spectrum.



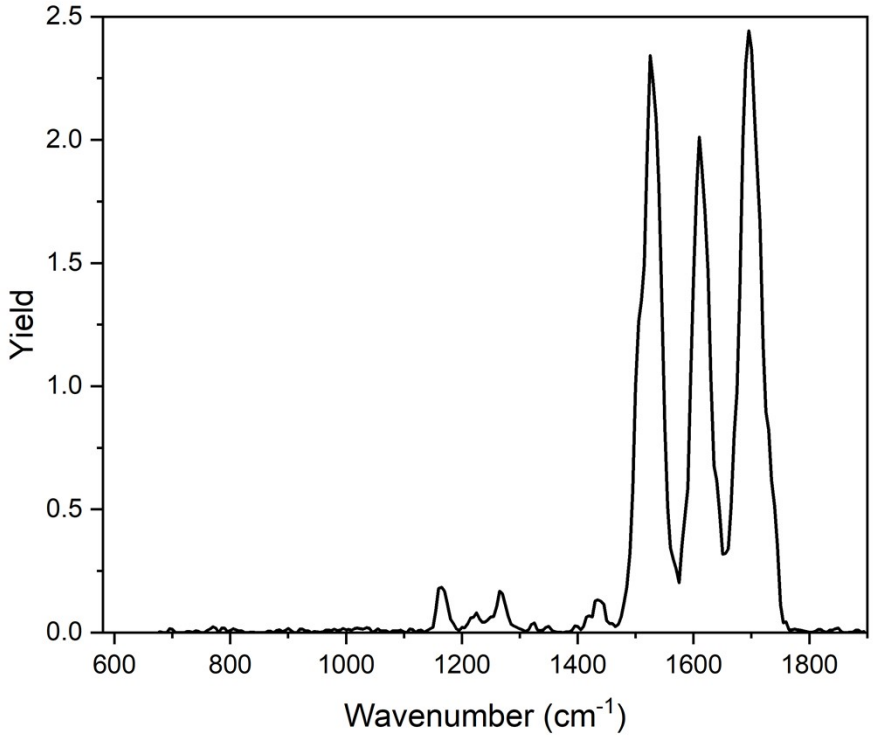


Figure S3. $Fe(GSH\text{-}H)^+$ full IRMPD experimental spectrum.

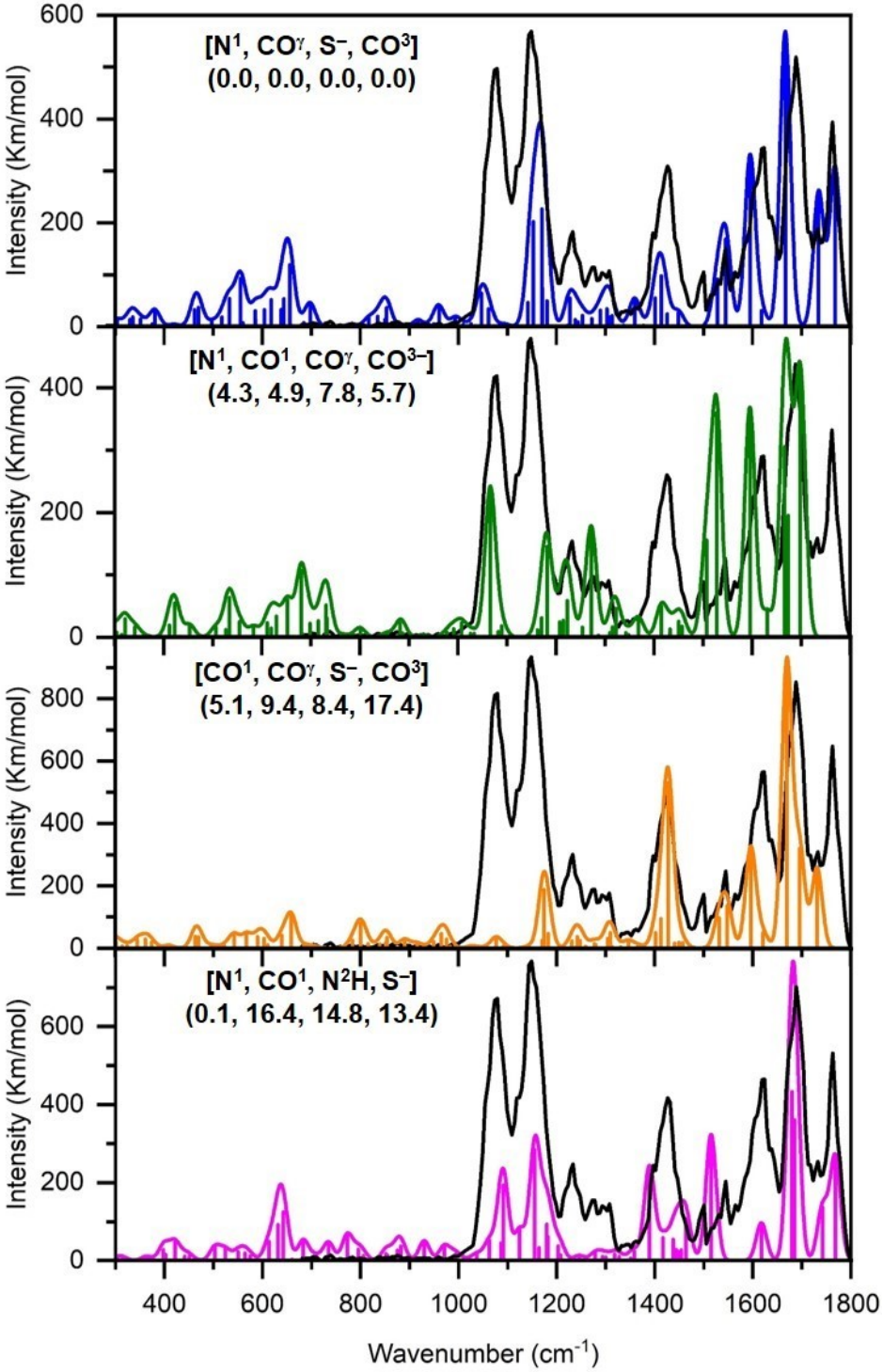


Figure S4. Comparison of the $Zn(GSH-H)^+$ experimental IRMPD spectrum (solid black line) with spectra calculated at the MP2/6-311+G(d,p) level of theory for low-lying conformers. Relative 298 K Gibbs energies (kJ/mol) are given at the B3LYP//B3LYP, B3LYP-GD3BJ//B3LYP-GD3BJ, ωB97XD//MP2, and MP2(full)//MP2 levels, respectively, using the 6-311+G(2d,2p) basis set.

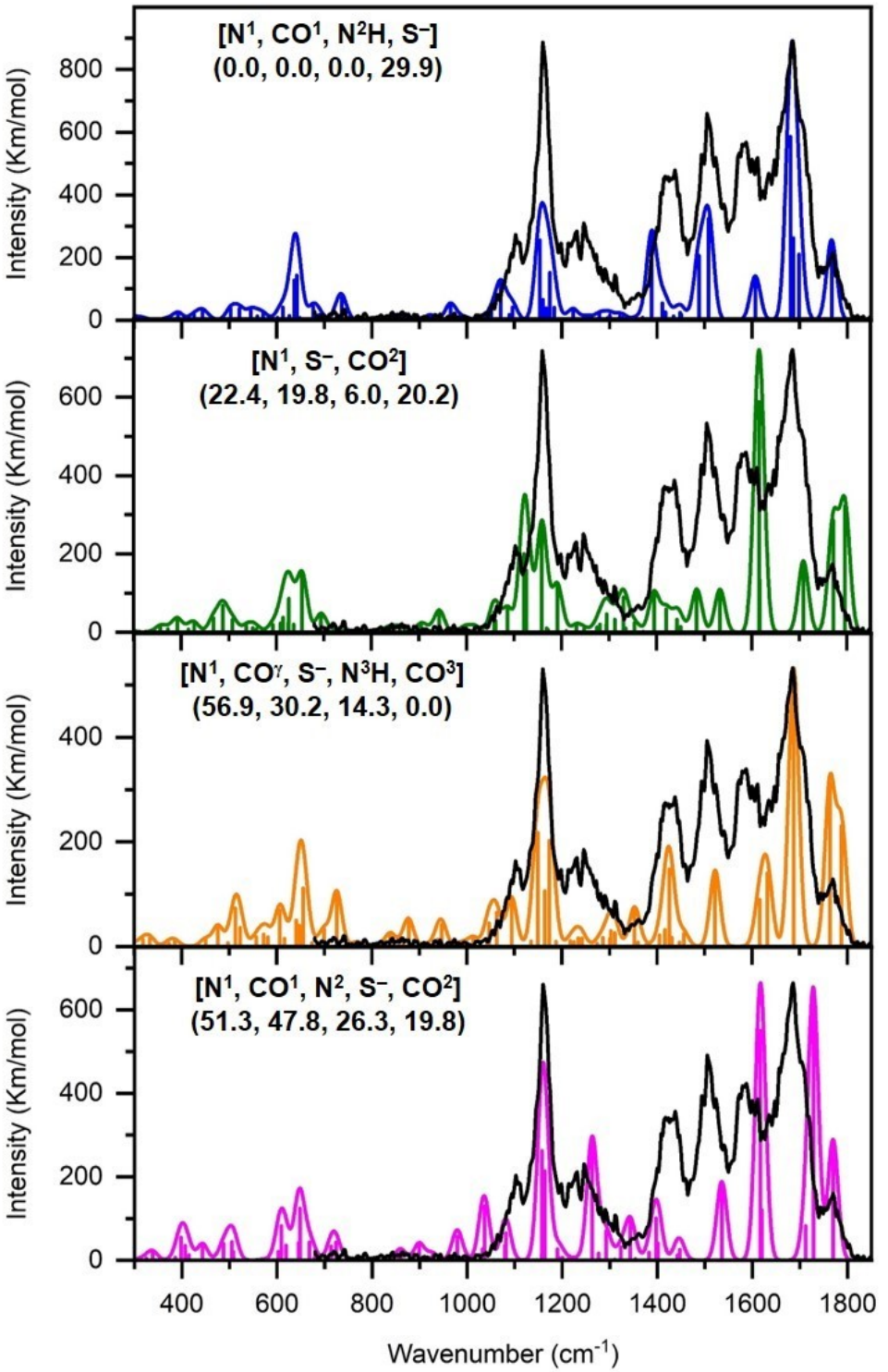


Figure S5. Comparison of the $Cu(GSH-H)^+$ experimental IRMPD spectrum (solid black line) with spectra calculated at the MP2/6-311+G(d,p) level of theory for low-lying conformers. Relative 298 K Gibbs energies (kJ/mol) are given at the B3LYP//B3LYP, B3LYP-GD3BJ//B3LYP-GD3BJ, ωB97XD//MP2, and MP2(full)//MP2 levels, respectively, using the 6-311+G(2d,2p) basis set.

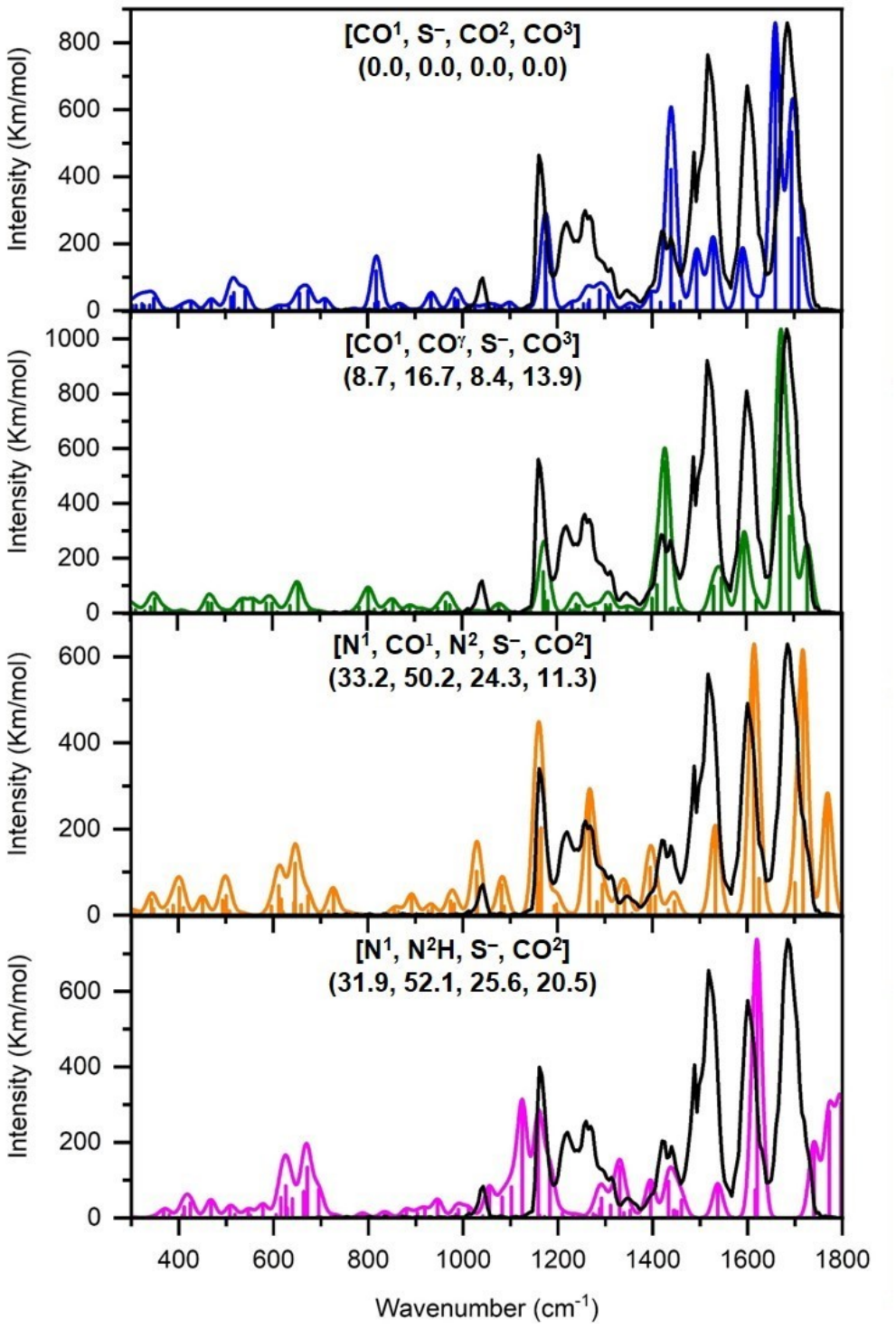


Figure S6. Comparison of the $Fe(GSH-H)^+$ experimental IRMPD spectrum (solid black line) with spectra calculated at the MP2/6-311+G(d,p) level of theory for low-lying conformers. Relative 298 K Gibbs energies (kJ/mol) are given at the B3LYP//B3LYP, B3LYP-GD3BJ//B3LYP-GD3BJ, ωB97XD//MP2, and MP2(full)//MP2 levels, respectively, using the 6-311+G(2d,2p) basis set.

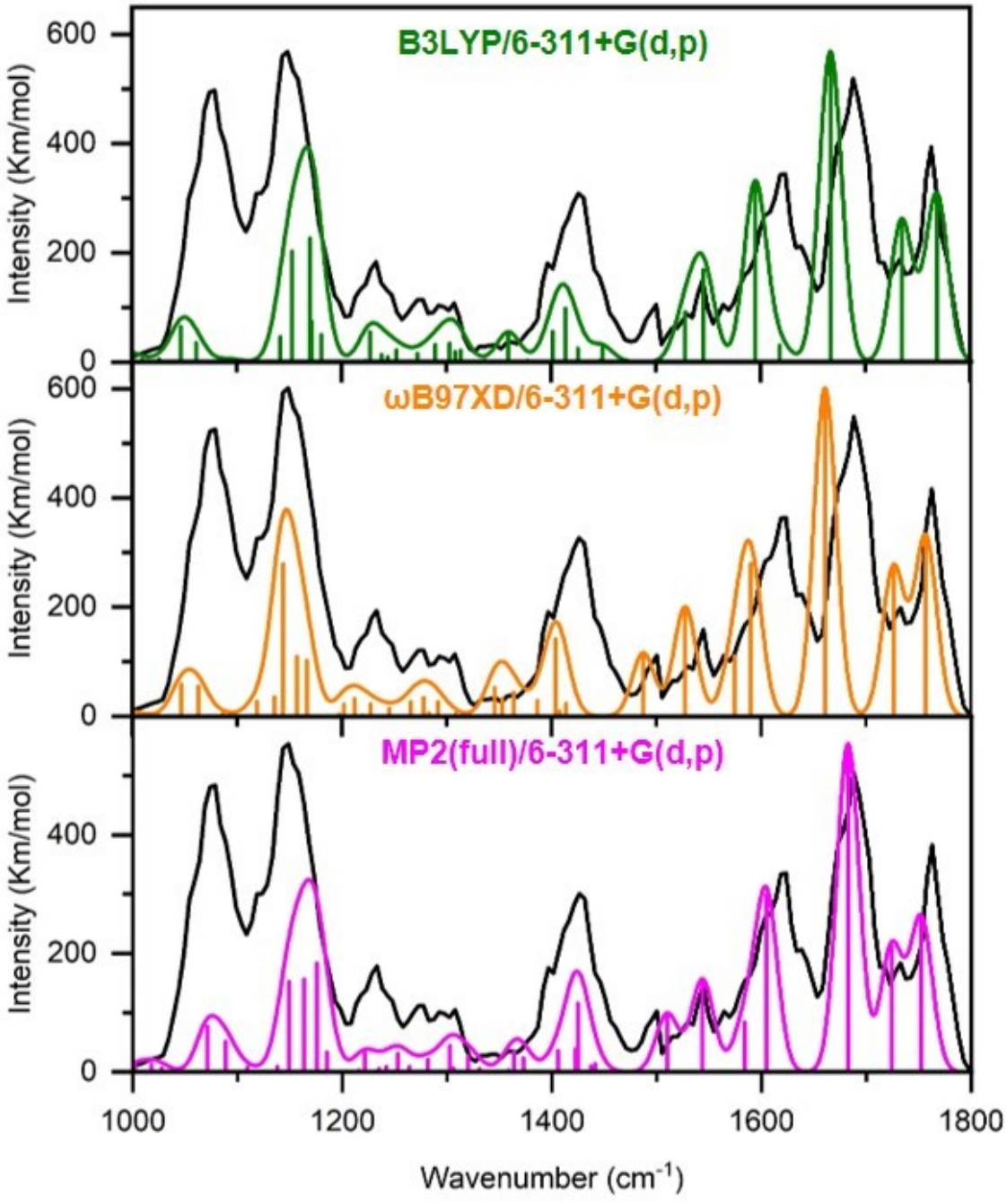


Figure S7. Comparison of the $Zn(GSH\text{-}H)^+$ experimental IRMPD spectrum (solid black line) with spectra calculated at several levels of theory for the [N^1, CO^γ, S^-, CO^3] conformer.

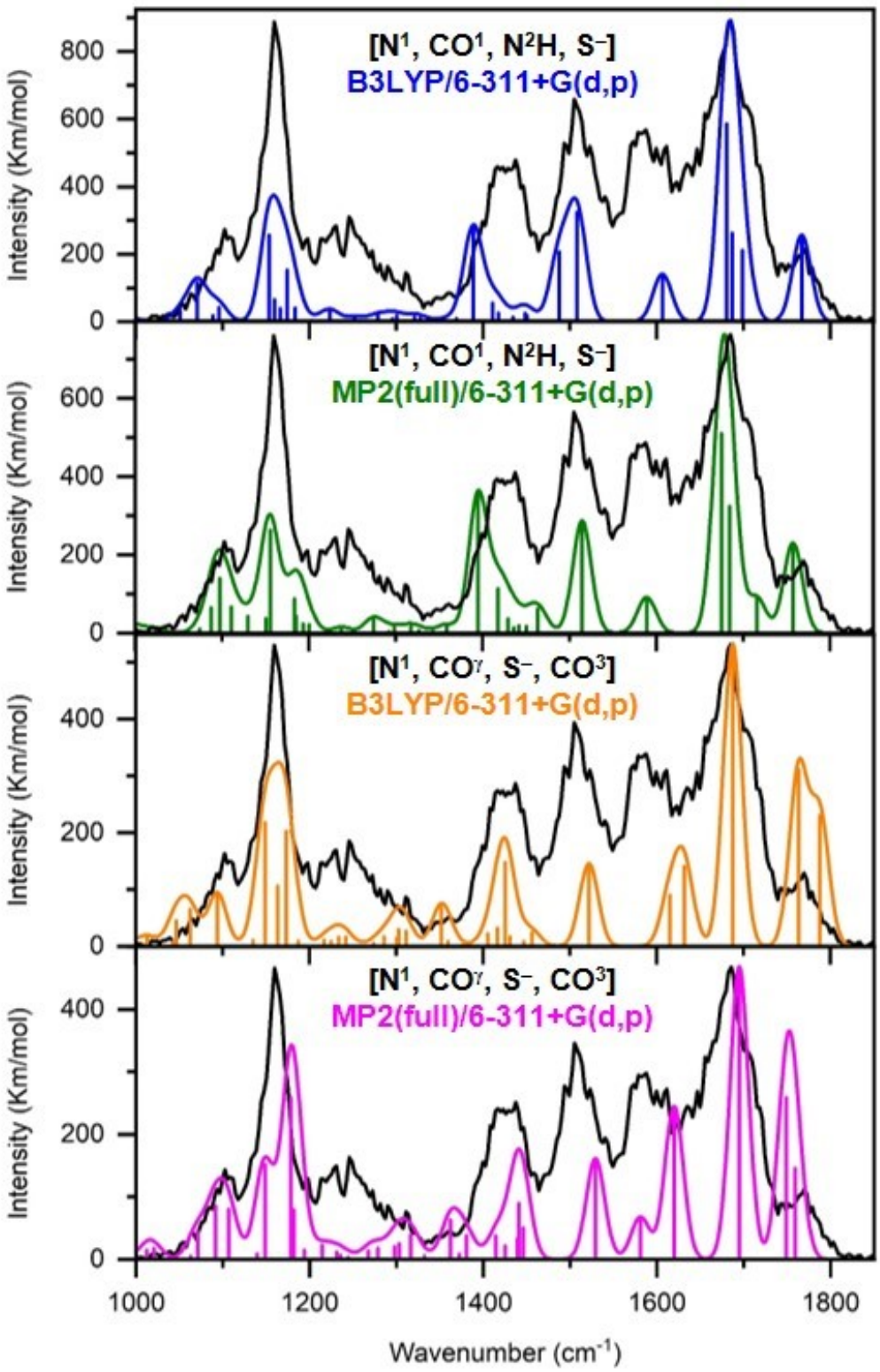


Figure S8. Comparison of the Cu(GSH-H)$^+$ experimental IRMPD spectrum (solid black line) with spectra calculated at several levels of theory.

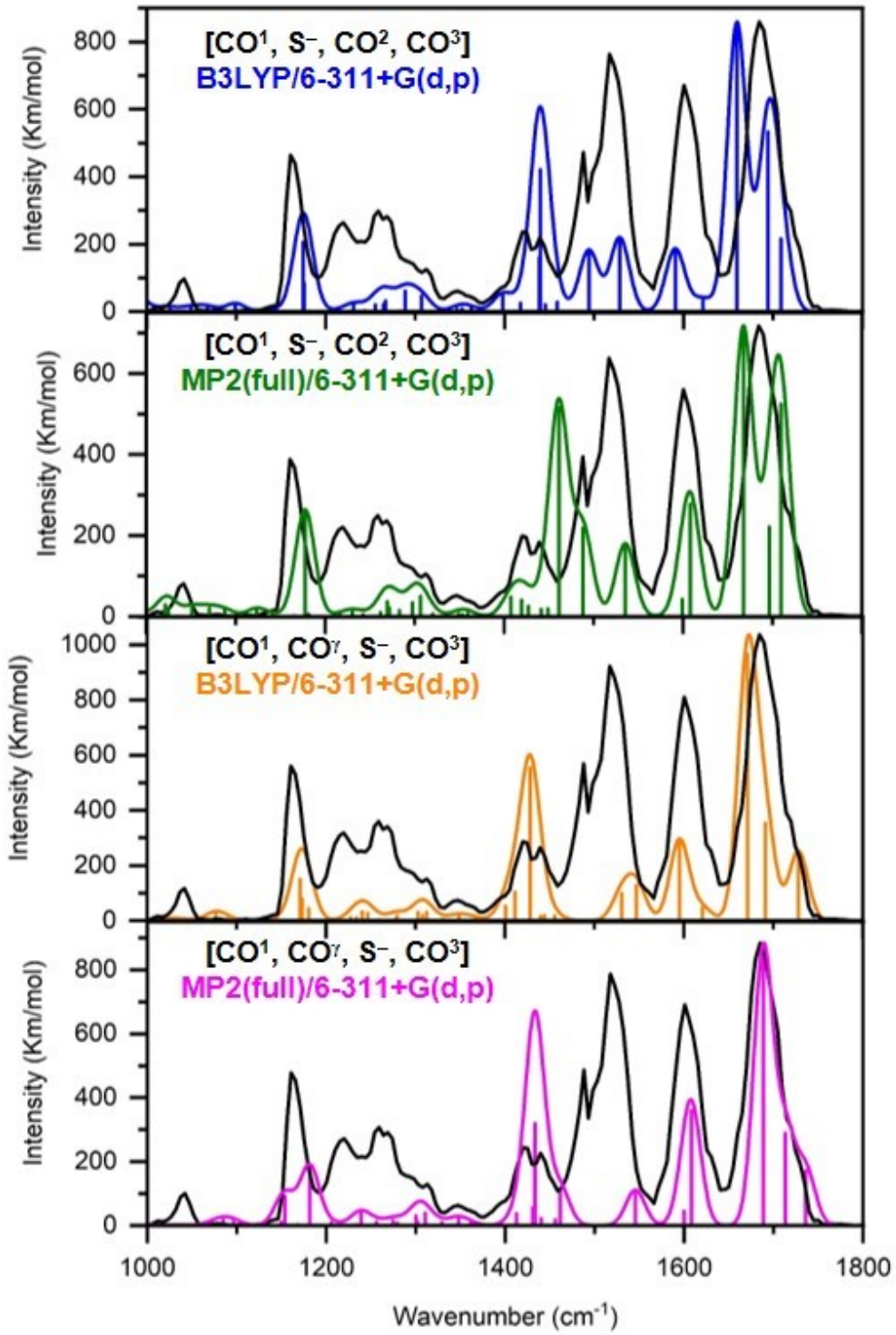


Figure S9. Comparison of the $Fe(GSH\text{-}H)^+$ experimental IRMPD spectrum (solid black line) with spectra calculated at several levels of theory.