Electronic Supplementary Information

First-principles calculations on Ni,Fe-containing carbon monoxide dehydrogenases reveal key stereoelectronic features for binding and release of CO$_2$ to/from the C-cluster

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Table S1 Detailed information of the atoms included in the LM model of the active site. H atoms are not explicitly reported in the list. C, O and N are backbone carbonylic C and O, and amidic N, respectively. Atoms belonging also to the SM model are indicated with bold labels.

| Residue     | Atoms included in the model | Basis set | Atoms constrained at the X-ray position |
|-------------|-----------------------------|-----------|----------------------------------------|
| NiFeS<sub>4</sub> core | all | def-TZVP | none |
| Ala91       | C, O, C<sub>α</sub>         | def-SVP   | Ca (terminal atom), O (carbonyl oxygen atom forms an H-bond with the amidic hydrogen of Gly95 in a α-helix) |
| Gly92       | all                         | def-SVP   | O, H<sub>8</sub> (carbonyl oxygen forms an H-bond with the amidic hydrogen of His96 and the hydrogen atom on N<sub>ε</sub> of Gln332, whereas H<sub>8</sub> forms an H-bond with the carbonyl oxygen atom of Ala88 in a α-helix. H<sub>8</sub> has been added at 1.02 Å from N in the C-N-C<sub>α</sub> plane, and constrained at that position) |
| His93       | all                         | def-SVP   | O, H<sub>8</sub> (carbonyl oxygen forms an H-bond with the amidic hydrogen of Ala97, whereas H<sub>8</sub> forms an H-bond with the carbonyl oxygen atom of Gly89 in a α-helix. H<sub>8</sub> has been added at 1.02 Å from N in the C-N-C<sub>α</sub> plane, and constrained at that position) |
| Ser94       | N, C<sub>α</sub>            | def-SVP   | Ca (terminal atom), H<sub>8</sub> (H<sub>8</sub> forms an H-bond with the carbonyl oxygen atom of Ala90 in a α-helix. H<sub>8</sub> has been added at 1.02 Å from N in the C-N-C<sub>α</sub> plane, and constrained at that position) |
| His96       | side chain, C<sub>α</sub>   | def-SVP   | Ca (terminal atom) |
| Asp219      | side chain, C<sub>α</sub>   | def-SVP   | Ca (terminal atom) |
| Cys223      | side chain, C<sub>α</sub>   | def-SVP   | Cα, Cβ (terminal atoms) |
| Asp231      | side chain                  | def-SVP   | Cγ, Cβ (terminal atoms) |
| Hys261      | side chain, C<sub>α</sub>   | def-TZVP  | Ca (terminal atom) |
| Cys295      | side chain, C<sub>α</sub>   | def-TZVP  | Ca (terminal atom) |
| Glu299      | side chain, C<sub>α</sub>   | def-SVP   | Ca (terminal atom) |
| Arg303      | side chain terminated at C<sub>γ</sub> | def-SVP | Cγ, Cδ, Nε (three terminal atoms of the Arg303 sidechain are constrained to the X-ray positions to avoid unrealistic distortions at the boundary of the model) |
| Gln332      | side chain, C<sub>α</sub>   | def-SVP   | Ca (terminal atom) |
| Cys333      | side chain, C<sub>α</sub>   | def-SVP   | Ca (terminal atom) |
| Cys446      | side chain, C<sub>α</sub>   | def-TZVP  | Ca (terminal atom) |
| Cys476      | side chain, C<sub>α</sub>   | def-TZVP  | Ca (terminal atom) |
| Ser525      | side chain, C, O, C<sub>α</sub> | def-SVP | Ca (terminal atom) |
| Cys526      | all                         | def-TZVP (side chain); SVP (other atoms) | none |
| Val527      | N, C<sub>α</sub>            | def-SVP   | Ca (terminal atom) |
| Ile567      | side chain, C<sub>α</sub>   | def-SVP   | all C atoms (all carbon atoms are constrained to the X-ray positions to avoid an unrealistic large rotation of the sidechain during geometry optimization) |
| Trp570      | side chain, C<sub>α</sub>   | def-SVP   | Cα, Cβ, Cγ (three terminal atoms of the Trp570 sidechain are constrained to the X-ray positions to avoid unrealistic distortions at the boundary of the model) |
Table S2 Binding energies (in kcal/mol) of CO₂ to the terminal position of the Ni atom of the unbound form of the C-cluster for C_{red2} and C_{int} (ΔE tCO₂) and the OH-bound form of C-cluster for C_{red1} (ΔE CO₂-OH) calculated, using LM^{H+K+} model, with BP86, B3LYP, PBE0 and M06 functionals with and without D3 and D3(BJ) dispersion-corrections. The tCO₂ → μCO₂ isomerization reaction energies are also shown for C_{red2} and C_{int} (ΔE tCO₂ → μCO₂). In all calculations solvation effects are added using the conductor-like screening model (COSMO) with a polarizable continuum medium at ε = 4.

|         | ΔE tCO₂ | ΔE CO₂-OH | ΔE tCO₂ → μCO₂ |
|---------|---------|-----------|----------------|
|         | C_{red2} | C_{int}  | C_{red1}       | C_{red2} | C_{int} |
| BP86    | -24.5   | -8.8     | 5.3            | -10.9    | -10.8  |
| B3LYP   | -23.6   | -4.9     | 7.7            | -15.9    | -18.0  |
| PBE0    | -28.0   | -9.4     | 5.6            | -13.3    | -16.3  |
| M06     | -36.3   | -19.0    | 2.3            | -12.1    | -14.9  |
| BP86-D3 | -54.0   | -38.1    | -17.2          | -5.9     | -5.4   |
| B3LYP-D3| -48.5   | -29.4    | -10.8          | -11.3    | -13.2  |
| PBE0-D3 | -43.8   | -25.2    | -6.1           | -10.3    | -13.3  |
| M06-D3  | -43.8   | -25.9    | -2.8           | -11.2    | -14.1  |
| BP86-D3(BJ)| -55.4 | -39.5    | -16.3          | -5.5     | -5.4   |
| B3LYP-D3(BJ)| -53.1| -34.2    | -12.9          | -10.4    | -12.7  |
| PBE0-D3(BJ)| -45.0 | -26.5    | -6.3           | -10.2    | -13.1  |

Table S3 Reaction energies (ΔE), zero-point energies corrections (ΔZPE) and zero-point corrected energies (ΔE + ΔZPE) for CO₂ binding and tCO₂ → μCO₂ isomerization, in kcal/mol, in the C_{red2}, C_{int} and C_{red1} redox-states.

|         | C_{red2} | C_{int} | C_{red1} |
|---------|----------|---------|----------|
| CO₂ binding | ΔE_{LM}  | -24.5   | -8.8     | 5.3      |
|          | ΔZPE_{MM} | 2.50    | 2.98     | 2.03     |
|          | ΔE_{LM} + ΔZPE_{MM} | -22.0   | -5.8     | 7.4      |
| tCO₂ → μCO₂ isomerization | ΔE_{LM} | -10.9    | -10.8    | -        |
|          | ΔZPE_{MM} | -0.24   | 0.00     | -        |
|          | ΔE_{LM} + ΔZPE_{MM} | -11.1   | -10.8    | -        |
Scheme S1 Schematic representation of all possible non-equivalent spin coupling schemes for the C-cluster, in which two pairs of Fe atoms are coupled antiferromagnetically.
Table S4 Relative energies (in kcal/mol) for all possible spin coupling schemes of all species investigated in this work, optimized at the RI-BP86/def-TZVP level in COSMO with $\varepsilon=4$, using the SM model in the $C_{\text{red}1}$, $C_{\text{int}}$ and $C_{\text{red}2}$ redox states of the C-cluster. NBO atomic charges and Mulliken spin densities (in parenthesis) of selected atoms of the C-cluster and of the layers L1 and L2 (corresponding respectively to the blue and red layers of the BS coupling schemes shown in Scheme S1) are also indicated.

a) unbound forms of the C-cluster
| BS | ΔE | Ni   | Fe\textsubscript{u} | Fe\textsubscript{1} | Fe\textsubscript{2} | Fe\textsubscript{3} | L1   | L2   |
|----|----|------|------------------|------------------|------------------|------------------|------|------|
| C\textsubscript{red} |     |      |                  |                  |                  |                  |      |      |
| 1  | 12.74 | 0.56(0.40) | 0.88(-3.12) | 0.78(3.20) | 0.77(3.14) | 0.72(-3.03) | -1.13(7.03) | -0.87(-6.05) |
| 2  | 14.76 | 0.53(0.31) | 0.91(-3.19) | 0.78(3.24) | 0.68(-2.91) | 0.75(3.14) | -1.20(7.11) | -0.80(-6.10) |
| 3  | 0.00  | 0.54(0.27) | 0.96(3.40) | 0.73(3.12) | 0.71(-3.00) | 0.71(-3.02) | -0.77(7.31) | -1.23(-6.32) |
| 4  | 8.04  | 0.53(0.42) | 0.96(3.38) | 0.70(-3.01) | 0.74(3.09) | 0.75(3.16) | -1.09(-6.64) | -0.91(7.61) |
| 5  | 12.48 | 0.54(0.52) | 0.87(3.26) | 0.72(-3.06) | 0.76(3.12) | 0.67(-3.02) | -1.15(-6.54) | -0.85(7.51) |
| 6  | 9.25  | 0.50(0.39) | 0.96(-3.36) | 0.53(-2.44) | 0.74(3.06) | 0.74(3.11) | -0.86(-5.59) | -1.14(-6.60) |
| C\textsubscript{int} |     |      |                  |                  |                  |                  |      |      |
| 1  | 1.59  | 0.54(-0.16) | 0.91(-3.28) | 0.78(3.22) | 0.80(3.20) | 0.78(-3.20) | -1.50(7.06) | -1.50(-7.05) |
| 2  | 5.45  | 0.53(-0.18) | 0.92(-3.30) | 0.79(3.24) | 0.78(-3.17) | 0.79(3.23) | -1.63(7.14) | -1.37(-7.15) |
| 3  | 0.00  | 0.53(-0.01) | 0.91(3.27) | 0.74(3.11) | 0.76(-3.14) | 0.76(-3.16) | -1.39(6.67) | -1.61(-6.66) |
| 4  | 6.18  | 0.53(0.22) | 0.90(3.26) | 0.77(-3.21) | 0.79(-3.21) | 0.77(3.11) | -1.49(-7.06) | -1.51(7.04) |
| 5  | 10.29 | 0.52(0.36) | 0.84(3.18) | 0.77(-3.18) | 0.72(2.95) | 0.73(-3.15) | -1.50(-6.80) | -1.50(6.79) |
| 6  | 0.04  | 0.53(-0.01) | 0.91(-3.27) | 0.74(-3.11) | 0.76(3.14) | 0.76(3.16) | -1.39(-6.67) | -1.61(6.66) |
| C\textsubscript{red2} |     |      |                  |                  |                  |                  |      |      |
| 1  | 2.05  | 0.53(0.03) | 0.86(-3.19) | 0.82(3.31) | 0.84(3.30) | 0.74(-3.03) | -1.90(7.40) | -2.10(-6.43) |
| 2  | 4.67  | 0.48(0.11) | 0.90(-3.22) | 0.81(3.31) | 0.76(-2.99) | 0.81(3.28) | -1.96(7.21) | -2.04(-6.25) |
| 3  | 7.72  | 0.43(0.27) | 0.93(3.27) | 0.81(3.29) | 0.75(-3.04) | 0.76(-3.12) | -2.00(7.24) | -2.00(-6.30) |
| 4  | 12.46 | 0.47(0.47) | 0.90(3.35) | 0.78(-3.22) | 0.69(-2.77) | 0.67(2.97) | -1.90(-6.35) | -2.10(7.34) |
| 5  | 11.14 | 0.47(-0.33) | 0.92(3.34) | 0.80(-3.25) | 0.83(3.26) | 0.68(-2.78) | -2.04(-6.37) | -1.96(7.38) |
| 6  | 0.00  | 0.52(0.09) | 0.87(-3.21) | 0.70(-2.84) | 0.81(3.24) | 0.80(3.25) | -2.08(-5.90) | -1.92(6.87) |
### b) $\mu$CO$_2$-adducts of the C-cluster

| BS | $\Delta$E | Ni  | Fe$^6$ | Fe$^7$ | Fe$^8$ | Fe$^9$ | L1     | L2     | CO$_2$ |
|----|-----------|-----|--------|--------|--------|--------|--------|--------|--------|
| C$_{red1}$ |         |     |        |        |        |        |        |        |        |
| 0   |  1.56     | 0.52(0.03) | 0.76(2.13) | 0.73(-3.12) | 0.72(3.00) | 0.75(3.09) | -0.30(-5.53) | -0.90(6.50) | -0.81(0.00) |
| 1   |  7.00     | 0.55(-0.17) | 0.79(-2.25) | 0.75(3.15) | 0.73(3.01) | 0.76(-3.15) | -0.98(6.81) | -0.22(-5.81) | -0.80(0.01) |
| 2   |  8.44     | 0.56(-0.20) | 0.80(-2.25) | 0.79(3.22) | 0.77(-3.15) | 0.75(3.03) | -0.96(6.87) | -0.24(-5.83) | -0.80(0.00) |
| 3   |  0.00     | 0.51(0.15) | 1.16(3.60) | 0.72(3.10) | 0.75(-3.05) | 0.76(-3.12) | -0.24(7.51) | -0.91(-6.56) | -0.82(0.05) |
| 4   |  7.04     | 0.54(0.23) | 1.18(3.64) | 0.75(-3.14) | 0.73(-2.99) | 0.76(3.15) | -0.89(-6.82) | -0.28(7.73) | -0.82(0.07) |
| 5   |  9.07     | 0.54(0.23) | 1.21(3.68) | 0.78(-3.19) | 0.78(3.17) | 0.77(-3.10) | -0.90(-6.93) | -0.28(-7.80) | -0.82(0.06) |
| 6   |  1.56     | 0.52(-0.03) | 0.76(-2.13) | 0.73(-3.12) | 0.72(3.00) | 0.75(3.09) | -0.30(-5.53) | -0.90(6.50) | -0.81(0.00) |

### c) tCO$_2$-adducts of the C-cluster

| SM | BS | $\Delta$E | Ni  | Fe$^6$ | Fe$^7$ | L1     | L2     | CO$_2$ |
|----|----|-----------|-----|--------|--------|--------|--------|--------|
| C$_{int1}$ |     |  1.14     | 0.97(-3.32) | 0.66(-2.77) | 0.88(3.33) | 0.85(3.29) | -1.45(-6.22) | -1.57(7.26) | -0.98(-0.04) |
| 0   | 16.88 | 0.57(0.30) | 0.88(-2.89) | 0.76(3.14) | 0.81(3.16) | 0.75(-3.12) | -0.98(6.99) | -0.38(-5.97) | -0.64(0.00) |
| 1   | 10.61 | 0.55(0.23) | 0.91(-3.11) | 0.77(3.17) | 0.69(-2.91) | 0.78(3.18) | -1.02(7.14) | -0.44(-6.18) | -0.55(0.05) |
| 2   |  8.85 | 0.49(0.12) | 1.14(-3.52) | 0.84(3.36) | 0.73(-2.89) | 0.87(3.35) | -1.57(7.42) | -1.43(-6.34) | -1.00(-0.07) |
| 3   |  3.26 | 0.49(0.15) | 1.15(3.54) | 0.76(3.16) | 0.84(-3.20) | 0.75(-3.01) | -1.28(7.32) | -1.70(-6.43) | -1.02(0.09) |
| 4   |  6.51 | 0.52(0.21) | 1.17(3.57) | 0.77(3.16) | 0.77(-2.96) | 0.84(3.29) | -1.71(-6.59) | -1.27(7.52) | -1.01(0.06) |
| 5   |  6.35 | 0.49(0.11) | 1.16(3.58) | 0.79(-3.18) | 0.88(3.33) | 0.75(-2.98) | -1.71(-6.56) | -1.27(7.48) | -1.01(0.06) |
| 6   |  0.00 | 0.48(0.09) | 1.11(-3.45) | 0.66(-2.79) | 0.90(3.38) | 0.85(3.32) | -1.45(-6.22) | -1.57(7.26) | -0.98(-0.04) |

### C$_{red2}$

| SM | BS | $\Delta$E | Ni  | Fe$^6$ | Fe$^7$ | Fe$^8$ | Fe$^9$ | L1     | L2     | CO$_2$ |
|----|----|-----------|-----|--------|--------|--------|--------|--------|--------|--------|
| 0   |  1.56 | 0.52(-0.14) | 1.00(-3.36) | 0.71(-3.06) | 0.82(3.23) | 0.81(3.21) | -0.96(6.97) | -1.27(6.94) | -0.54(-0.02) |
| 1   |  5.58 | 0.56(0.02) | 0.95(-3.29) | 0.80(3.27) | 0.87(3.32) | 0.72(-2.94) | -1.62(7.34) | -1.51(-6.33) | -0.87(0.00) |
| 2   |  1.02 | 0.52(0.12) | 0.96(-3.28) | 0.83(3.31) | 0.77(-3.03) | 0.86(3.34) | -1.60(7.35) | -1.52(-6.34) | -0.88(-0.01) |
| 3   |  3.37 | 0.53(0.21) | 0.95(3.27) | 0.81(3.27) | 0.79(-3.10) | 0.74(-2.93) | -1.46(7.29) | -1.65(-6.29) | -0.89(-0.01) |
| 4   |  7.46 | 0.56(0.23) | 1.03(3.47) | 0.78(-3.13) | 0.76(-2.91) | 0.85(3.28) | -1.71(-6.47) | -1.37(7.45) | -0.92(0.01) |
| 5   |  6.66 | 0.52(0.25) | 1.02(3.45) | 0.75(-3.08) | 0.80(3.18) | 0.75(-3.01) | -1.76(-6.47) | -1.36(7.44) | -0.88(0.01) |
| 6   |  0.00 | 0.49(0.02) | 0.97(-3.32) | 0.66(-2.77) | 0.88(3.33) | 0.85(3.29) | -1.55(-6.15) | -1.55(7.12) | -0.89(0.00) |
### d) OH-adducts of the C-cluster

| BS | ΔE | Ni | Fe⁰ | Fe¹ | Fe² | Fe³ | L1  | L2  | OH   |
|----|----|----|-----|-----|-----|-----|-----|-----|------|
| C₀₅₁ |   | 1  | 9.46 | 0.64(0.65) | 1.17(-3.60) | 0.80(3.27) | 0.80(3.27) | 0.75(-3.09) | -1.40(7.35) -0.94(-16.16) -0.66(-0.18) |
|     |   | 2  | 11.01 | 0.60(0.08) | 1.09(-3.33) | 0.82(3.30) | 0.62(-2.65) | 0.81(3.27) | -1.39(7.26) -0.99(-6.08) -0.62(-0.15) |
|     |   | 3  | 0.00 | 0.58(0.31) | 1.16(3.59) | 0.71(3.06) | 0.75(-3.11) | 0.75(-3.10) | -0.94(7.35) -1.41(-6.53) -0.65(-0.17) |
|     |   | 4  | 7.18 | 0.60(0.61) | 1.17(3.60) | 0.75(-3.16) | 0.80(-3.24) | 0.74(3.10) | -1.33(-7.06) -1.00(7.86) -0.66(-0.16) |
|     |   | 5  | 10.38 | 0.63(0.63) | 1.14(3.53) | 0.80(-3.26) | 0.78(3.18) | 0.82(-3.27) | -1.34(-7.07) -0.99(7.89) -0.67(-0.15) |
|     |   | 6  | 5.02 | 0.64(0.68) | 1.13(-3.50) | 0.70(-2.98) | 0.76(3.13) | 0.80(3.21) | -0.94(-5.73) -1.40(6.88) -0.66(-0.16) |

### e) CO₂-OH adducts of the C-cluster

| BS | ΔE | Ni | Fe⁰ | Fe¹ | Fe² | Fe³ | L1  | L2  | CO₂ | OH  |
|----|----|----|-----|-----|-----|-----|-----|-----|-----|-----|
| C₀₅₂ |   | 1  | 4.96 | 0.53(0.07) | 1.06(-3.42) | 0.81(3.28) | 0.84(3.33) | 0.71(-2.90) | -2.06(7.33) -2.18(-6.27) -0.76(-0.08) |
|     |   | 2  | 6.79 | 0.56(0.37) | 1.09(-3.48) | 0.83(3.32) | 0.76(-3.05) | 0.82(3.30) | -2.10(7.26) -2.15(-6.17) -0.75(-0.10) |
|     |   | 3  | 6.96 | 0.55(0.42) | 1.05(3.40) | 0.80(3.21) | 0.74(-3.00) | 0.76(-3.11) | -2.13(7.25) -2.11(-6.33) -0.76(0.08) |
|     |   | 4  | 7.00 | 0.56(0.47) | 1.08(3.44) | 0.78(-3.19) | 0.77(-3.10) | 0.83(3.29) | -2.19(-6.71) -2.06(7.61) -0.75(-0.10) |
|     |   | 5  | 10.05 | 0.60(0.70) | 1.08(3.48) | 0.80(-3.20) | 0.79(3.15) | 0.78(-3.15) | -2.11(-6.67) -2.14(7.58) -0.76(0.09) |
|     |   | 6  | 0.00 | 0.54(0.36) | 1.06(-3.37) | 0.71(-2.91) | 0.81(3.26) | 0.79(3.22) | -2.19(-5.77) -2.08(6.86) -0.74(-0.10) |
Table S5 Energies (in Hartree) and relative energies (in kcal/mol) for \( S = 0 \), \( S = 1 \) and \( S = 2 \) spin states of the more relevant species in the \( C \text{int} \) redox state discussed in this work, optimized at the RI-BP86/def-TZVP-SVP level in COSMO with \( \varepsilon = 4 \), using the \( \text{LM}^{\text{H},\text{K}^+} \) and the \( \text{LM}^{\text{H}^0,\text{K}^+} \) models.

| Structure                     | \( S = 0 \)       | \( S = 1 \)       | \( S = 2 \)       |
|-------------------------------|-------------------|-------------------|-------------------|
| \( \text{C}_{\text{int}}\text{LM}^{\text{H},\text{K}^+} \) | -15589.90172 (0.0) | -15589.893000 (5.5) | -15589.88408 (11.1) |
| \( \text{C}_{\text{int}}\text{tCO}_2\text{LM}^{\text{H},\text{K}^+} \) | -15778.59541 (0.6) | -15778.59640 (0.0) | converges to \( \text{C}_{\text{int}}\text{μCO}_2\text{LM}^{\text{H},\text{K}^+} \) |
| \( \text{C}_{\text{int}}\text{μCO}_2\text{LM}^{\text{H},\text{K}^+} \) | -15778.61256 (0.0) | -15778.61087 (1.1) | -15778.59111 (13.5) |
| \( \text{C}_{\text{int}}\text{OH}\text{LM}^{\text{H},\text{K}^+} \) | -15665.84848 (0.0) | -15665.83984 (5.4) | -15665.82156 (16.9) |
| \( \text{C}_{\text{int}}\text{CO}_2\text{OH}\text{LM}^{\text{H},\text{K}^+} \) | -15854.51395 (0.0) | -15854.50489 (5.7) | -15854.49818 (9.9) |
| \( \text{C}_{\text{int}}\text{LM}^{\text{H}^0,\text{K}^+} \) | -15589.34172 (0.0) | -15589.34068 (0.7) | -15589.32323 (11.6) |
| \( \text{C}_{\text{int}}\text{tCO}_2\text{LM}^{\text{H}^0,\text{K}^+} \) | -15778.01578 (0.0) | -15778.01442 (1.1) | -15778.0095 (12.1) |
| \( \text{C}_{\text{int}}\text{μCO}_2\text{H}_2\text{O}\text{LM}^{\text{H}^0,\text{K}^+} \) | -15854.51737 (0.0) | -15854.50512 (7.7) | -15854.4952 (13.9) |
| \( \text{C}_{\text{int}}\text{μCO}_2\text{H}_2\text{O}\text{LM}^{\text{H}^0,\text{K}^+} \) | -15854.52845 (0.0) | -15854.51245 (10.0) | -15854.5073 (13.3) |

Table S6 Energies (in Hartree) of all structures investigated in this work, optimized at the RI-BP86/def-TZVP-SVP level in COSMO with \( \varepsilon = 4 \), using the a) SM, b) \( \text{LM}^{\text{H},\text{K}^+} \), c) \( \text{LM}^{\text{H}^0,\text{K}^+} \) and d) \( \text{LM}^{\text{H}^+,\text{K}^0} \) DFT models.

a) SM model

| \( \text{C}_{\text{red}1} \) | charge | multiplicity | BS state | E          | \( \Delta E \) |
|----------------------------|--------|-------------|----------|------------|--------------|
|                            | -2     | 2           | BS-1     | -10850.70496516 | 12.74        |
|                            |        |             | BS-2     | -10850.70174919 | 14.76        |
|                            |        |             | BS-3     | -10850.72526804 | 0.00         |
|                            |        |             | BS-4     | -10850.71245146 | 8.04         |
|                            |        |             | BS-5     | -10850.70538738 | 12.48        |
|                            |        |             | BS-6     | -10850.71053395 | 9.25         |

| \( \text{C}_{\text{int}} \) | charge | multiplicity | BS state | E          | \( \Delta E \) |
|----------------------------|--------|-------------|----------|------------|--------------|
|                            | -3     | 1           | BS-1     | -10850.74059174 | 5.19         |
|                            |        |             | BS-2     | -10850.74016559 | 5.45         |
|                            |        |             | BS-3     | -10850.74885795 | 0.00         |
|                            |        |             | BS-4     | -10850.73901312 | 6.18         |
|                            |        |             | BS-5     | -10850.73245202 | 10.29        |
|                            |        |             | BS-6     | -10850.74879664 | 0.04         |

| \( \text{C}_{\text{red}2} \) | charge | multiplicity | BS state | E          | \( \Delta E \) |
|----------------------------|--------|-------------|----------|------------|--------------|
|                            | -4     | 2           | BS-1     | -10850.71198545 | 2.05         |
|                            |        |             | BS-2     | -10850.70781231 | 4.67         |
|                            |        |             | BS-3     | -10850.70294196 | 7.72         |
|                            |        |             | BS-4     | -10850.69538835 | 12.46        |
|                            |        |             | BS-5     | -10850.69750347 | 11.14        |
|       |       |       | BS-1     | BS-2     | BS-3     | BS-4     | BS-5     | BS-6    |
|-------|-------|-------|----------|----------|----------|----------|----------|---------|
| $C_{\text{red1}-\text{OH}}$ | -3    | 2     |          |          |          |          |          |         |
|       |       |       | -10926.60243105 | -10926.59997131 | -10926.61751139 | -10926.60607055 | -10926.60096684 | -10926.60950723 |
| $C_{\text{int}-\text{OH}}$ | -4    | 1     |          |          |          |          |          |         |
|       |       |       | -10926.58941386 | -10926.58485503 | -10926.59367579 | -10926.58944438 | -10926.58506162 | -10926.58914696 |
| $C_{\text{red2}-\text{OH}}$ | -5    | 2     |          |          |          |          |          |         |
|       |       |       | -10926.51815717 | -10926.51524727 | -10926.51496907 | -10926.51490133 | -10926.51004464 | -10926.52606314 |
| $C_{\text{red1}-\mu\text{CO}_2}$ | -2    | 2     |          |          |          |          |          |         |
|       |       |       | -11039.38354099 | -11039.38123894 | -11039.39469143 | -11039.38347495 | -11039.38023284 | -11039.39220625 |
| $C_{\text{int}-\mu\text{CO}_2}$ | -3    | 1     |          |          |          |          |          |         |
|       |       |       | -11039.41941177 | -11039.42126966 | -11039.42621751 | -11039.42232471 | -11039.42112889 | -11039.42617502 |
| $C_{\text{red2}-\mu\text{CO}_2}$ | -4    | 2     |          |          |          |          |          |         |
|       |       |       | -11039.40492823 | -11039.40697637 | -11039.40312525 | -11039.3979460 | -11039.39819926 | -11039.40832457 |
| $C_{\text{red1}-\text{tCO}_2}$ | -2    | 2     |          |          |          |          |          |         |
|       |       |       | -11039.37021021 | -11039.38020187 | -10850.71524969 | -10926.60243105 | -10926.59997131 | -10926.61751139 |
|       |       |       |         |         |          |          |          | 0.00    |
|       |       |       |         |         |          |          |          | 9.46    |
|       |       |       |         |         |          |          |          | 11.01   |
|       |       |       |         |         |          |          |          | 0.00    |
|       |       |       |         |         |          |          |          | 7.18    |
|       |       |       |         |         |          |          |          | 10.38   |
|       |       |       |         |         |          |          |          | 5.02    |
|       |       |       |         |         |          |          |          | 2.67    |
|       |       |       |         |         |          |          |          | 5.54    |
|       |       |       |         |         |          |          |          | 0.00    |
|       |       |       |         |         |          |          |          | 9.46    |
|       |       |       |         |         |          |          |          | 5.41    |
|       |       |       |         |         |          |          |          | 0.00    |
|       |       |       |         |         |          |          |          | 9.07    |
|       |       |       |         |         |          |          |          | 1.56    |
|       |       |       |         |         |          |          |          | 2.13    |
|       |       |       |         |         |          |          |          | 3.10    |
|       |       |       |         |         |          |          |          | 0.00    |
|       |       |       |         |         |          |          |          | 2.44    |
|       |       |       |         |         |          |          |          | 3.19    |
|       |       |       |         |         |          |          |          | 0.03    |
|       |       |       |         |         |          |          |          | 2.13    |
|       |       |       |         |         |          |          |          | 0.85    |
|       |       |       |         |         |          |          |          | 3.26    |
|       |       |       |         |         |          |          |          | 6.51    |
|       |       |       |         |         |          |          |          | 6.35    |
|       |       |       |         |         |          |          |          | 0.00    |
|       |       |       |         |         |          |          |          | 16.88   |
|       |       |       |         |         |          |          |          | 10.61   |
|       | BS-3         | BS-4         | BS-5         | BS-6         |
|-------|--------------|--------------|--------------|--------------|
| \(C_{\text{int}} - \text{tCO}_2\) | -11039.39710996 | -11039.38291466 | -11039.38610109 | -11039.38717377 |
|       | 0.00         | 8.91         | 6.91         | 6.24         |

|       | BS-1         | BS-2         | BS-3         | BS-4         |
|-------|--------------|--------------|--------------|--------------|
| \(C_{\text{red}} - \text{tCO}_2\) | -11039.42633709 | -11039.42908675 | -11039.43304589 | -11039.42633693 |
|       | 4.21         | 2.48         | 0.00         | 4.21         |

|       | BS-1         | BS-2         | BS-3         | BS-4         |
|-------|--------------|--------------|--------------|--------------|
| \(C_{\text{red}} - \text{CO}_2 - \text{OH}\) | -11115.3006146 | -11115.4144398 | -11115.3050474 | -11115.29738888 |
|       | 3.13         | 1.02         | 3.37         | 7.46         |

|       | BS-1         | BS-2         | BS-3         | BS-4         |
|-------|--------------|--------------|--------------|--------------|
| \(C_{\text{int}} - \text{CO}_2 - \text{OH}\) | -11115.29223557 | -11115.29295667 | -11115.3050474 | -11115.29619362 |
|       | 2.48         | 2.03         | 0.25         | 0.00         |

|       | BS-1         | BS-2         | BS-3         | BS-4         |
|-------|--------------|--------------|--------------|--------------|
| \(C_{\text{red}} - \text{CO}_2 - \text{OH}\) | -11115.23661335 | -11115.23538578 | -11115.23643082 | -11115.23696922 |
|       | 0.22         | 0.99         | 0.34         | 0.00         |
### b) \( \text{LM}^{\text{He,K}+} \text{model} \)

| Charge \( \text{C}_{\text{red1}} \) | Multiplicity | BS State | \( E \) | \( \Delta E \) |
|----------------|-------------|----------|--------|--------|
| -2             | 2           | BS-3     | -15589.84905936 | 0.00  |
|                 |             | BS-6     | -15589.84031942 | 5.48  |
| -3             | 1           | BS-3     | -15589.90165629 | 0.04  |
|                 |             | BS-6     | -15589.90172268 | 0.00  |
| -4             | 2           | BS-3     | -15589.95557115 | 1.81  |
|                 |             | BS-6     | -15589.89845975 | 0.00  |
| -3             | 2           | BS-3     | -15665.81630659 | 0.00  |
|                 |             | BS-6     | -15665.80491832 | 7.15  |
| -4             | 1           | BS-3     | -15665.8484392* | 0.00  |
|                 |             | BS-6     | -15665.8483119* | 0.08  |
| -5             | 2           | BS-3     | -15665.81935415*| 1.90  |
|                 |             | BS-6     | -15665.82237958*| 0.00  |
| -2             | 2           | BS-3     | -15778.54194728 | 0.00  |
|                 |             | BS-6     | -15778.53406558 | 4.95  |
| -3             | 1           | BS-3     | -15778.61255969 | 0.00  |
|                 |             | BS-6     | -15778.61247282 | 0.05  |
| -4             | 2           | BS-3     | -15778.63091730 | 2.23  |
|                 |             | BS-6     | -15778.63445048 | 0.00  |
| -2             | 2           | BS-3     | unstable        |       |
|                 |             | BS-6     | unstable        |       |
| -3             | 1           | BS-3     | -15778.59541465 | 0.00  |
|                 |             | BS-6     | unstable        |       |
| -4             | 2           | BS-3     | -15778.61713245 | 0.00  |
|                 |             | BS-6     | -15778.61628150 | 0.53  |
| -3             | 2           | BS-3     | -15854.48744800 | 0.00  |
|                 |             | BS-6     | -15854.47423537 | 8.29  |
| -4             | 1           | BS-3     | -15854.51296608 | 0.62  |
|                 |             | BS-6     | -15854.51395426 | 0.00  |
| -5             | 2           | BS-3     | -15854.49549139 | 0.00  |
|                 |             | BS-6     | -15854.49160506 | 1.87  |
### c) LM$^{H_{0},K_{+}}$ model

|       | charge | multiplicity | BS state | E          | ΔE    |
|-------|--------|--------------|----------|------------|-------|
| $C_{\text{red1}}$ | -3     | 2            | BS-3     | -15589.32185204 | 0.00  |
| C       |         |              | BS-6     | -15589.31888675 | 1.86  |
| $C_{\text{int}}$ | -4     | 1            | BS-3     | -15589.34172213 | 0.00  |
| C       |         |              | BS-6     | -15589.34135562 | 0.23  |
| $C_{\text{red2}}$ | -5     | 2            | BS-3     | -15589.3135096 | 1.34  |
| C       |         |              | BS-6     | -15589.31565012 | 0.00  |
| $C_{\text{red1}-\text{OH}}$ | -4     | 2            | BS-3     | -15665.2654694 | 0.00  |
| C       |         |              | BS-6     | -15665.2537088 | 7.38  |
| $C_{\text{int}-\text{OH}}$ | -5     | 1            | BS-3     | -15665.25490477 | 0.00  |
| C       |         |              | BS-6     | -15665.2549083 | 0.00  |
| $C_{\text{red2}-\text{OH}}$ | -6     | 2            | BS-3     | -15665.19272796 | 2.55  |
| C       |         |              | BS-6     | -15665.19678426 | 0.00  |
| $C_{\text{red1}-\mu\text{CO}_2}$ | -3    | 2            | BS-3     | -15777.98333022 | 0.00  |
| C       |         |              | BS-6     | -15777.96786353 | 9.71  |
| $C_{\text{int}-\mu\text{CO}_2}$ | -4    | 1            | BS-3     | -15778.02882304 | 0.00  |
| C       |         |              | BS-6     | -15778.02814652 | 0.42  |
| $C_{\text{red2}-\mu\text{CO}_2}$ | -5    | 2            | BS-3     | -15778.02313132 | 2.08  |
| C       |         |              | BS-6     | -15778.02644899 | 0.00  |
| $C_{\text{red1}-\text{tCO}_2}$ | -3    | 2            | BS-3     | -15777.98061617 | 0.00  |
| C       |         |              | BS-6     | -15777.96916736 | 7.18  |
| $C_{\text{int}-\text{tCO}_2}$ | -4    | 1            | BS-3     | -15778.0267356 | 1.95  |
| C       |         |              | BS-6     | -15778.01578077 | 0.00  |
| $C_{\text{red2}-\text{tCO}_2}$ | -5    | 2            | BS-3     | -15778.0053658 | 0.00  |
| C       |         |              | BS-6     | -15777.99836502 | 1.36  |
| $C_{\text{red1}-\text{CO}_2-\text{OH}}$ | -4   | 2            | BS-3     | -15853.90997976 | 0.00  |
| C       |         |              | BS-6     | -15853.89785817 | 7.61  |
| $C_{\text{int}-\text{CO}_2-\text{OH}}$ | -5  | 1            | BS-3     | -15853.90673860 | 0.00  |
| C       |         |              | BS-6     | -15853.90480643 | 1.21  |
| $C_{\text{red2}-\text{CO}_2-\text{OH}}$ | -6  | 2            | BS-3     | -15853.86445580 | 0.33  |
| C       |         |              | BS-6     | -15853.86498952 | 0.00  |
### d) LM$^{H_2O_0}$ model

| Charge | Multiplicity | BS state | Energy | ΔE   |
|--------|--------------|----------|--------|------|
| $C_{red1}$ | -3 | 2 | BS-3 | -15589.31671105 | 0.00 |
|          |     |     | BS-6 | -15589.31085238 | 3.68 |
| $C_{int}$ | -4 | 1 | BS-3 | -15589.33900594 | 0.00 |
|          |     |     | BS-6 | -15589.33877147 | 0.15 |
| $C_{red2}$ | -5 | 2 | BS-3 | -15589.30186535 | 2.91 |
|          |     |     | BS-6 | -15589.30650957 | 0.00 |
| $C_{red1}$-OH | -4 | 2 | BS-3 | unstable |   |
|          |     |     | BS-6 | unstable |   |
| $C_{int}$-OH | -5 | 1 | BS-3 | unstable |   |
|          |     |     | BS-6 | unstable |   |
| $C_{red2}$-OH | -6 | 2 | BS-3 | unstable |   |
|          |     |     | BS-6 | unstable |   |
| $C_{red1}$-μ$CO_2$ | -3 | 2 | BS-3 | -15777.00042472 | 0.00 |
|          |     |     | BS-6 | -15777.98944869 | 6.89 |
| $C_{int}$-μ$CO_2$ | -4 | 1 | BS-3 | -15778.04647603 | 0.00 |
|          |     |     | BS-6 | -15778.04613268 | 0.22 |
| $C_{red2}$-μ$CO_2$ | -5 | 2 | BS-3 | -15778.03384183 | 2.64 |
|          |     |     | BS-6 | -15778.03986665 | 0.00 |
| $C_{red1}$-t$CO_2$ | -3 | 2 | BS-3 | unstable |   |
|          |     |     | BS-6 | unstable |   |
| $C_{int}$-t$CO_2$ | -4 | 1 | BS-3 | unstable |   |
|          |     |     | BS-6 | unstable |   |
| $C_{red2}$-t$CO_2$ | -5 | 2 | BS-3 | unstable |   |
|          |     |     | BS-6 | unstable |   |
| $C_{red1}$-CO$_2$-OH | -4 | 2 | BS-3 | 15853.90394311 | 0.00 |
|          |     |     | BS-6 | 15853.89130811 | 7.93 |
| $C_{int}$-CO$_2$-OH | -5 | 1 | BS-3 | 15853.89552282 | 0.00 |
|          |     |     | BS-6 | 15853.89553072 | 0.00 |
| $C_{red2}$-CO$_2$-OH | -6 | 2 | BS-3 | 15853.84325325 | 1.38 |
|          |     |     | BS-6 | 15853.84545353 | 0.00 |

* During geometry optimization, a proton is transferred from His93 to the OH ligand to form a H$_2$O molecule that dissociates from the C-cluster.

* During geometry optimization, a proton is transferred from Lys563 to the OH ligand to form a H$_2$O molecule.

* During geometry optimization, a proton is transferred from Lys563 to the OH ligand to form a H$_2$O molecule that dissociates from the C-cluster.
Table S7 Selected distances (Å) and angles (°) for small and large models (SM, LM<sup>H+,K+</sup>, LM<sup>H+,K+</sup>, LM<sup>H+,K0</sup>) of the μCO<sub>2</sub>-bound C-cluster in the C<sub>red2</sub>, C<sub>int</sub> and C<sub>red1</sub> redox states (in their most stable BS state), optimized at the RI-BP86/def-TZVP-SVP level in COSMO with ε=4, and for the 3B52 and 4UDX x-ray structures.

|                  | X-ray 3B52 | X-ray 4UDX | SM C<sub>red2</sub> C<sub>int</sub> C<sub>red1</sub> | LM<sup>H+,K+</sup> C<sub>red2</sub> C<sub>int</sub> C<sub>red1</sub> | LM<sup>H+,K+</sup> C<sub>red2</sub> C<sub>int</sub> C<sub>red1</sub> | LM<sup>H+,K0</sup> C<sub>red2</sub> C<sub>int</sub> C<sub>red1</sub> |
|------------------|------------|------------|---------------------------|-----------------------------|-----------------------------|-----------------------------|
| Ni-Fe<sub>u</sub>| 2.76       | 2.83       | 2.78 2.73 2.73            | 2.65 2.67 2.63              | 2.62 2.66 2.60              | 2.70 2.73 2.60              |
| Ni-S<sub>Cys526</sub>| 2.10 | 2.11       | 2.24 2.24 2.24            | 2.23 2.22 2.23              | 2.23 2.23 2.23              | 2.24 2.24 2.23              |
| Ni-S<sub>4</sub>| 3.59       | 3.73       | 3.40 3.28 3.19            | 3.59 3.53 3.27              | 3.62 3.56 3.27              | 3.61 3.57 3.48              |
| S<sub>2</sub>-Ni-S<sub>Cys526</sub>| 168.3 | 166.2      | 176.5 175.0 173.0         | 168.6 169.1 169.0           | 168.7 168.4 168.8           | 167.0 166.8 165.6           |
| S<sub>1</sub>-Ni-C| 171.3      | 174.9      | 169.4 167.4 165.0         | 175.1 173.2 167.2           | 175.4 173.7 167.3           | 175.4 174.7 172.5           |
| Fe<sub>7</sub>-S<sub>4</sub>| 2.19       | 2.33       | 2.30 2.26 2.23            | 2.26 2.25 2.22              | 2.27 2.26 2.23              | 2.26 2.25 2.21              |
| Fe<sub>7</sub>-S<sub>Cys295</sub>| 2.34 | 2.29       | 2.42 2.41 2.36            | 2.34 2.31 2.31              | 2.35 2.31 2.32              | 2.36 2.33 2.31              |
| Fe<sub>7</sub>-N<sub>His261</sub>| 2.03 | 2.15       | 2.24 2.25 2.22            | 2.16 2.16 2.21              | 2.16 2.15 2.21              | 2.17 2.15 2.15              |
| Fe<sub>7</sub>-S<sub>Cys526</sub>| 3.33 | 3.34       | 2.79 2.58 2.52            | 3.48 3.38 2.65              | 3.44 3.40 2.65              | 3.62 3.59 3.52              |
| Ni-Fe<sub>u</sub>-S<sub>4</sub>| 92.3      | 92.1       | 83.4 81.5 79.4            | 93.7 91.1 84.2              | 95.3 92.2 85.0              | 93.1 91.1 92.2              |
| Ni-C| 1.96       | 1.81       | 1.91 1.91 1.94            | 1.86 1.88 1.89              | 1.89 1.91 1.93              | 1.87 1.88 1.90              |
| Fe<sub>7</sub>-O<sub>1</sub>| 2.05       | 2.03       | 2.10 2.12 2.13            | 2.14 2.11 2.15              | 2.09 2.07 2.12              | 2.08 2.07 2.07              |
| C-O<sub>1</sub>| 1.25       | 1.32       | 1.30 1.29 1.29            | 1.32 1.32 1.31              | 1.34 1.33 1.31              | 1.30 1.30 1.29              |
| C-O<sub>2</sub>| 1.26       | 1.30       | 1.25 1.24 1.23            | 1.26 1.24 1.24              | 1.22 1.22 1.21              | 1.26 1.25 1.24              |
| O<sub>1</sub>-C-O<sub>2</sub>| 132.6      | 117.2      | 124.8 126.7 128.9         | 122.2 123.8 126.3           | 123.5 125.7 129.3           | 123.5 125.0 127.4           |
| Ni-Fe<sub>u</sub>-O<sub>1</sub>| 69.5       | 64.6       | 64.9 65.6 65.9            | 68.1 68.0 68.4              | 68.7 68.3 69.1              | 67.2 66.7 68.7              |
| Ni-C-O<sub>1</sub>| 119.4      | 119.8      | 112.1 1115.1113.1         | 116.1 115.0 114.8           | 111.8 111.6 111.9           | 115.5 114.4 112.4           |
| Ni-C-O<sub>2</sub>| 108.0      | 124.5      | 123.1 121.7 119.8         | 121.8 121.2 118.9           | 124.7 122.6 118.9           | 121.0 120.6 120.2           |
| O<sub>1</sub>-N<sub>His563</sub>| 2.64       | 2.72       | - - -                     | 2.82 2.86 2.86              | 2.74 2.79 2.82              | 3.18 3.21 3.21              |
| O<sub>1</sub>-H<sub>His563</sub>| - - -      | - - -      | 1.81 1.82                 | 1.67 1.74 1.77              | - - -                      | - - -                      |
| O<sub>2</sub>-εN<sub>His93</sub>| 2.88       | 2.70       | - - -                     | 2.56 2.64 2.70              | 3.52 3.37 3.25              | 2.54 2.60 2.66              |
| O<sub>2</sub>-εH<sub>His93</sub>| - - -      | - - -      | 1.47 1.58 1.66            | - - -                      | 1.42 1.52 1.60              | - - -                      |
| O<sub>1</sub>-εN<sub>His93</sub>| 3.93       | 3.92       | - - -                     | 3.78 3.86 3.82              | 5.29 5.05 4.75              | 3.76 3.83 3.87              |
| O<sub>1</sub>-εH<sub>His93</sub>| - - -      | - - -      | 2.88 2.98 2.98            | - - -                      | 2.84 2.93 3.00              | - - -                      |
Figure S1 Superimposition of the optimized geometry of a) C_{red2-μCO2}, b) C_{red2-tCO2} and c) C_{red2-CO2-OH} optimized using the LM^{H+,K+} (red), LM^{H0,K+} (blue) and LM^{H+,K0} (white) models of the active site.
Table S8 RMSD values calculated for the structures of the $\mu$CO$_2$-bound metallic cluster, the [NiFe$_4$S$_4$] core and the CO$_2$ ligand extracted by the theoretical geometries, compared to the 3B52 and 4UDX crystal structures.

|        | SM    | LM$^{H+,K^+}$ | LM$^{H^0,K^+}$ | LM$^{H^+,K^0}$ |
|--------|-------|---------------|----------------|----------------|
|        | C$_{\text{red2}}$ | C$_{\text{int}}$ | C$_{\text{red1}}$ | C$_{\text{red2}}$ | C$_{\text{int}}$ | C$_{\text{red1}}$ | C$_{\text{red2}}$ | C$_{\text{int}}$ | C$_{\text{red1}}$ | C$_{\text{red2}}$ | C$_{\text{int}}$ | C$_{\text{red1}}$ | C$_{\text{red2}}$ | C$_{\text{int}}$ | C$_{\text{red1}}$ |
| 3B52   |       |               |                |                |                |                |                |                |                |                |                |                |                |                |                |                |
| [NiFe$_4$S$_4$]-CO$_2$ | 0.219 | 0.220 | 0.230 | 0.140 | 0.131 | 0.167 | 0.151 | 0.132 | 0.161 | 0.132 | 0.118 | 0.108 |          |          |          |          |
| [NiFe$_4$S$_4$] | 0.239 | 0.247 | 0.260 | 0.145 | 0.138 | 0.181 | 0.153 | 0.140 | 0.176 | 0.145 | 0.129 | 0.117 |          |          |          |          |
| CO$_2$ | 0.047 | 0.037 | 0.027 | 0.062 | 0.054 | 0.041 | 0.061 | 0.050 | 0.035 | 0.053 | 0.045 | 0.034 |          |          |          |          |
| 4UDX   |       |               |                |                |                |                |                |                |                |                |                |                |          |          |          |          |
| [NiFe$_4$S$_4$]-CO$_2$ | 0.215 | 0.223 | 0.239 | 0.125 | 0.117 | 0.174 | 0.134 | 0.119 | 0.169 | 0.119 | 0.113 | 0.118 |          |          |          |          |
| [NiFe$_4$S$_4$] | 0.217 | 0.233 | 0.253 | 0.127 | 0.120 | 0.175 | 0.134 | 0.122 | 0.169 | 0.119 | 0.109 | 0.113 |          |          |          |          |
| CO$_2$ | 0.061 | 0.072 | 0.084 | 0.048 | 0.058 | 0.071 | 0.061 | 0.071 | 0.089 | 0.052 | 0.062 | 0.076 |          |          |          |          |

Figure S2 RMSD values and superimposition of the optimized geometry of a) $C_{\text{red2}}$-$\mu$CO$_2$-$LM^{H^+,K^+}$, b) $C_{\text{red2}}$-$\mu$CO$_2$-$LM^{H^0,K^+}$ and c) $C_{\text{red2}}$-$\mu$CO$_2$-$LM^{H^+,K^0}$ (white) with the 4UDX X-ray structure (blue). The indicated RMSD values have been calculated on the [NiFe$_4$S$_4$] cluster, the CO$_2$ ligand and the side chains and C$\alpha$ atoms of the His93 and Lys563 residues, extracted from the theoretical and experimental geometries. For the sake of clarity, only the hydrogens of the N atom of Lys563 and of the $\varepsilon$N atom of His93 are shown.
Table S9 RMSD values calculated among the structures extracted by the LM\textsuperscript{H+,K+}, LM\textsuperscript{H0,K+} and LM\textsuperscript{H+,K0} theoretical geometries of the [NiFe\textsubscript{4}S\textsubscript{4}] core and bounded ligands, [NiFe\textsubscript{4}S\textsubscript{4}] core, bounded ligands and the side chains and \textit{C}\textalpha{} atoms of the His93 and Lys563 residues, and all (non-hydrogen) atoms of the models.

|                | LM\textsuperscript{H+,K+}-LM\textsuperscript{H0,K+} | LM\textsuperscript{H+,K+}-LM\textsuperscript{H+,K0} | LM\textsuperscript{H0,K+}-LM\textsuperscript{H+,K0} | LM\textsuperscript{H+,K+}-LM\textsuperscript{H+,K0} |
|----------------|------------------------------------------------------|------------------------------------------------------|------------------------------------------------------|------------------------------------------------------|
| \textit{C}\textsubscript{red2}-\textit{μCO}\textsubscript{2} | | | | |
| [NiFe\textsubscript{4}S\textsubscript{4}]-CO\textsubscript{2} | 0.039 | 0.056 | 0.070 | |
| [NiFe\textsubscript{4}S\textsubscript{4}]-CO-His93-Lys563 | 0.547 | 0.321 | 0.504 | |
| all (non-hydrogen) atoms | 0.377 | 0.382 | 0.506 | |
| \textit{C}\textsubscript{red2}-tCO\textsubscript{2} | | | | |
| [NiFe\textsubscript{4}S\textsubscript{4}]-CO\textsubscript{2} | 0.583 | - | - | |
| [NiFe\textsubscript{4}S\textsubscript{4}]-CO-His93-Lys563 | 0.780 | - | - | |
| all (non-hydrogen) atoms | 0.392 | - | - | |
| \textit{C}\textsubscript{red2}-CO\textsubscript{2}-OH | | | | |
| [NiFe\textsubscript{4}S\textsubscript{4}]-CO\textsubscript{2}-OH | 0.169 | 0.176 | 0.136 | |
| [NiFe\textsubscript{4}S\textsubscript{4}]-CO\textsubscript{2}-OH-His93-Lys563 | 0.444 | 0.357 | 0.575 | |
| all (non-hydrogen) atoms | 0.325 | 0.366 | 0.506 | |

Table S10 Computed NBO charges and, in parenthesis, Mulliken spin population for the CO\textsubscript{2} molecule bound to the C-cluster in \textit{μCO}, \textit{tCO} and CO\textsubscript{2}-OH calculated using the SM, LM\textsuperscript{H+,K+}, LM\textsuperscript{H0,K+} and LM\textsuperscript{H+,K0} models of the active site.

a) SM model

|                | C   | O1  | O2  | CO\textsubscript{2} |
|----------------|-----|-----|-----|---------------------|
| \textit{C}\textsubscript{red1}-\textit{μCO}\textsubscript{2} | 0.55(0.00) | -0.73(0.05) | -0.63(0.00) | -0.82(0.05) |
| \textit{C}\textsubscript{red1}-tCO\textsubscript{2} | 0.63(-0.02) | -0.63(0.00) | -0.62(0.00) | -0.63(-0.02) |
| \textit{C}\textsubscript{red1}-CO\textsubscript{2}-OH | 0.61(-0.02) | -0.68(0.00) | -0.64(0.00) | -0.71(-0.02) |
| \textit{C}\textsubscript{int}-\textit{μCO}\textsubscript{2} | 0.53(0.00) | -0.76(0.05) | -0.68(0.00) | -0.91(0.05) |
| \textit{C}\textsubscript{int}-tCO\textsubscript{2} | 0.59(-0.01) | -0.68(0.00) | -0.68(0.00) | -0.78(-0.01) |
| \textit{C}\textsubscript{int}-CO\textsubscript{2}-OH | 0.56(0.02) | -0.73(0.00) | -0.70(0.01) | -0.86(0.04) |
| \textit{C}\textsubscript{red2}-\textit{μCO}\textsubscript{2} | 0.52(0.00) | -0.79(-0.04) | -0.72(0.00) | -0.98(-0.04) |
| \textit{C}\textsubscript{red2}-tCO\textsubscript{2} | 0.56(0.00) | -0.72(0.00) | -0.73(0.00) | -0.89(0.00) |
| \textit{C}\textsubscript{red2}-CO\textsubscript{2}-OH | 0.54(0.03) | -0.76(0.00) | -0.75(0.01) | -0.97(0.04) |
b) $\text{LM}^{\text{H}^+,\text{K}^+}$ model

|        | C  | O1   | O2    | CO₂    |
|--------|----|------|-------|--------|
| $C_{\text{red1}-\mu CO₂}$ | 0.55(0.00) | -0.82(0.04) | -0.64(0.00) | -0.91(0.04) |
| $C_{\text{red1}-tCO₂}$ | - | - | - | - |
| $C_{\text{red1}-CO₂-OH}$ | 0.64(-0.03) | -0.73(0.00) | -0.58(0.00) | -0.67(-0.03) |
| $C_{\text{int}-\mu CO₂}$ | 0.54(0.00) | -0.84(-0.02) | -0.68(0.00) | -0.98(-0.02) |
| $C_{\text{int}-tCO₂}$ | 0.57(-0.01) | -0.82(0.00) | -0.65(-0.01) | -0.92(-0.02) |
| $C_{\text{int}-CO₂-OH}$ | 0.60(0.02) | -0.77(0.00) | -0.63(0.02) | -0.80(0.04) |
| $C_{\text{red2}-\mu CO₂}$ | 0.53(0.00) | -0.85(-0.02) | -0.71(0.00) | -1.03(-0.02) |
| $C_{\text{red2}-tCO₂}$ | 0.53(0.00) | -0.84(0.00) | -0.69(0.00) | -1.00(0.00) |
| $C_{\text{red2}-CO₂-OH}$ | 0.55(0.01) | -0.80(0.00) | -0.68(0.00) | -0.94(0.01) |

c) $\text{LM}^{\text{H},\text{K}^+}$ model

|        | C  | O1   | O2    | CO₂    |
|--------|----|------|-------|--------|
| $C_{\text{red1}-\mu CO₂}$ | 0.56(0.00) | -0.82(0.04) | -0.53(0.00) | -0.79(0.04) |
| $C_{\text{red1}-tCO₂}$ | 0.73(-0.02) | -0.56(0.00) | -0.54(0.00) | -0.37(-0.02) |
| $C_{\text{red1}-CO₂-OH}$ | 0.67(-0.02) | -0.59(0.00) | -0.58(0.00) | -0.49(-0.02) |
| $C_{\text{int}-\mu CO₂}$ | 0.54(0.00) | -0.85(0.03) | -0.57(0.00) | -0.88(0.03) |
| $C_{\text{int}-tCO₂}$ | 0.66(0.01) | -0.62(0.01) | -0.60(0.00) | -0.56(0.02) |
| $C_{\text{int}-CO₂-OH}$ | 0.64(-0.03) | -0.62(-0.02) | -0.63(-0.01) | -0.61(-0.06) |
| $C_{\text{red2}-\mu CO₂}$ | 0.53(0.00) | -0.87(-0.03) | -0.61(0.00) | -0.95(-0.03) |
| $C_{\text{red2}-tCO₂}$ | 0.61(0.00) | -0.68(0.00) | -0.66(0.00) | -0.73(0.00) |
| $C_{\text{red2}-CO₂-OH}$ | 0.61(0.02) | -0.67(0.00) | -0.66(0.01) | -0.71(0.04) |

d) $\text{LM}^{\text{H}^+,\text{K}₀}$ model

|        | C  | O1   | O2    | CO₂    |
|--------|----|------|-------|--------|
| $C_{\text{red1}-\mu CO₂}$ | 0.56(0.00) | -0.75(0.03) | -0.65(0.00) | -0.84(0.03) |
| $C_{\text{red1}-tCO₂}$ | - | - | - | - |
| $C_{\text{red1}-CO₂-OH}$ | 0.63(-0.01) | -0.77(0.00) | -0.60(0.00) | -0.69(-0.01) |
| $C_{\text{int}-\mu CO₂}$ | 0.56(0.00) | -0.78(0.03) | -0.69(0.00) | -0.91(0.03) |
| $C_{\text{int}-tCO₂}$ | - | - | - | - |
| $C_{\text{int}-CO₂-OH}$ | 0.60(0.02) | -0.77(0.00) | -0.65(0.00) | -0.81(0.02) |
| $C_{\text{red2}-\mu CO₂}$ | 0.54(0.00) | -0.79(-0.02) | -0.71(0.00) | -0.96(-0.02) |
| $C_{\text{red2}-tCO₂}$ | - | - | - | - |
| $C_{\text{red2}-CO₂-OH}$ | 0.58(0.02) | -0.78(0.00) | -0.67(0.00) | -0.87(0.02) |
Table S11 NBO atomic charges and Mulliken spin densities (in parenthesis) of selected atoms of the C-cluster and of the layers L1 and L2 (corresponding respectively to the blue and red layers of the BS coupling schemes shown in Scheme S1) for the unbound form of the C-cluster and μCO2 and tCO2 adducts in the C_red1, C_int and C_red2 redox states (in the most stable spin coupling scheme) optimized using the SM, LM^H+K+, LM^B0,K+ and LM^H+K0 DFT models.

### a) SM model

| Redox state | BS state | Ni  | Fe₄  | Fe₁  | Fe₂  | Fe₃  | L1    | L2    | CO₂   |
|-------------|----------|-----|------|------|------|------|-------|-------|-------|
| C_red1      | 3        | 0.54(0.27) | 0.96(3.40) | 0.73(3.12) | 0.71(-3.00) | 0.71(-3.02) | -0.77(7.31) | -1.23(-6.32) | -      |
| C_red1-μCO₂ | 3        | 0.51(0.15) | 1.16(3.60) | 0.72(3.10) | 0.75(-3.05) | 0.76(-3.12) | -0.24(7.51) | -0.91(-6.56) | -0.82(0.05) |
| C_red1-tCO₂ | 3        | 0.53(0.35) | 1.04(3.50) | 0.71(3.08) | 0.76(-3.10) | 0.77(-3.11) | -0.48(7.62) | -0.89(-6.63) | -0.63(-0.02) |
| C_int       | 3        | 0.53(0.01) | 0.91(3.27) | 0.74(3.11) | 0.76(-3.14) | 0.76(-3.16) | -1.39(6.67) | -1.61(-6.66) | -      |
| C_int-μCO₂  | 3        | 0.51(0.09) | 1.17(3.58) | 0.68(2.93) | 0.85(-3.28) | 0.80(-3.23) | -0.83(6.93) | -1.26(-6.98) | -0.91(0.05) |
| C_int-tCO₂  | 3        | 0.52(0.14) | 1.00(3.36) | 0.71(3.06) | 0.82(-3.23) | 0.81(-3.21) | -0.96(6.97) | -1.27(-6.94) | -0.78(-0.01) |
| C_red2      | 6        | 0.52(0.09) | 0.87(-3.21) | 0.70(-2.84) | 0.81(3.24) | 0.80(3.25) | -2.08(-5.90) | -1.92(6.87) | -      |
| C_red2-μCO₂ | 6        | 0.48(0.09) | 1.11(-3.45) | 0.66(-2.79) | 0.90(3.38) | 0.85(3.32) | -1.45(-6.22) | -1.57(7.26) | -0.98(-0.04) |
| C_red2-tCO₂ | 6        | 0.49(0.02) | 0.97(-3.32) | 0.66(-2.77) | 0.88(3.33) | 0.85(3.29) | -1.55(-6.15) | -1.55(7.12) | -0.89(0.00) |

### b) LM^H+K+ model

| Redox state | BS state | Ni  | Fe₄  | Fe₁  | Fe₂  | Fe₃  | L1    | L2    | CO₂   |
|-------------|----------|-----|------|------|------|------|-------|-------|-------|
| C_red1      | 3        | 0.52(0.22) | 0.96(3.37) | 0.71(3.08) | 0.67(-2.91) | 0.70(-3.00) | -0.82(7.14) | -1.04(-6.16) | -      |
| C_red1-μCO₂ | 3        | 0.54(0.17) | 1.14(3.55) | 0.69(3.04) | 0.73(-3.02) | 0.74(-3.08) | -0.17(7.45) | -0.73(-6.51) | -0.91(0.04) |
| C_red1-tCO₂ | 3        | 0.53(0.17) | 1.09(3.45) | 0.69(3.04) | 0.74(-3.02) | 0.75(-3.08) | -0.24(7.51) | -0.91(-6.56) | -0.82(0.05) |
| C_int       | 6        | 0.50(0.00) | 0.88(-3.17) | 0.75(-3.15) | 0.73(3.07) | 0.77(3.19) | -1.40(-6.62) | -1.37(6.63) | -      |
| C_int-μCO₂  | 6        | 0.51(-0.02) | 1.06(-3.37) | 0.70(-3.06) | 0.81(3.22) | 0.77(3.17) | -0.74(-6.90) | -1.05(-6.92) | -0.98(-0.02) |
| C_int-tCO₂  | 6        | 0.50(0.06) | 1.02(3.40) | 0.66(2.92) | 0.80(-3.19) | 0.78(-3.17) | -0.79(6.83) | -1.06(-6.80) | -0.92(-0.02) |
| C_red2      | 6        | 0.47(0.05) | 0.86(-3.17) | 0.68(-2.85) | 0.81(3.23) | 0.79(3.25) | -1.95(-5.98) | -1.64(6.88) | -      |
| C_red2-μCO₂ | 6        | 0.51(-0.05) | 1.04(-3.35) | 0.63(-2.68) | 0.86(3.32) | 0.80(3.24) | -1.26(-5.99) | -1.36(7.08) | -1.03(-0.02) |
| C_red2-tCO₂ | 3        | 0.49(-0.15) | 1.00(3.36) | 0.74(3.14) | 0.79(-3.15) | 0.67(-2.83) | -1.17(7.17) | -1.50(-6.21) | -1.00(0.00) |
c) \( \text{LM}^{10, K^+} \) model

| Redox state | BS state | Ni    | Fe_α  | Fe_1  | Fe_2  | Fe_3  | L1      | L2      | CO_2     |
|-------------|----------|-------|-------|-------|-------|-------|---------|---------|----------|
| C_{red1}    | 3        | 0.51(0.31) | 0.96(3.40) | 0.70(3.05) | 0.69(-2.97) | 0.72(-3.05) | -0.83(7.27) | -1.12(-6.30) | -         |
| C_{red1}μCO_2 | 3       | 0.54(0.22) | 1.15(3.56) | 0.69(3.05) | 0.76(-3.10) | 0.75(-3.05) | -0.28(7.50) | -0.84(-6.56) | -0.79(0.04) |
| C_{red1}tCO_2 | 3      | 0.51(0.29) | 1.01(3.45) | 0.66(2.94) | 0.71(-2.95) | 0.71(-2.98) | -0.61(7.20) | -1.04(-3.23) | -0.37(-0.02) |
| C_{int}     | 3        | 0.49(0.08) | 0.86(3.17) | 0.72(3.10) | 0.75(-3.10) | 0.76(-3.18) | -1.38(6.63) | -1.45(-6.66) | -         |
| C_{int}μCO_2 | 3       | 0.50(0.00) | 1.05(3.35) | 0.72(3.10) | 0.78(-3.16) | 0.82(-3.22) | -0.82(6.88) | -1.16(-6.91) | -0.88(0.03) |
| C_{int}tCO_2 | 6       | 0.53(-0.16) | 0.95(-3.29) | 0.67(-2.96) | 0.78(3.15)  | 0.77(3.14)  | -1.07(-6.71) | -1.34(-6.71) | -0.56(0.02) |
| C_{red2}    | 6        | 0.46(-0.02) | 0.85(-3.17) | 0.66(-2.79) | 0.83(3.26)  | 0.77(3.22)  | -1.92(-6.00) | -1.73(6.89)  | -         |
| C_{red2}μCO_2 | 6    | 0.50(0.05) | 1.05(-3.36) | 0.63(-2.69) | 0.81(3.26)  | 0.87(3.33)  | -1.36(-6.05) | -1.47(7.09)  | -0.95(-0.03) |
| C_{red2}tCO_2 | 3      | 0.54(0.29) | 0.94(3.27)  | 0.75(3.14)  | 0.79(-3.13) | 0.68(-2.86) | -1.40(7.11)  | -1.77(-6.20) | -0.73(0.00) |

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d) \( \text{LM}^{11+, K^0} \) model

| Redox state | BS state | Ni    | Fe_α  | Fe_1  | Fe_2  | Fe_3  | L1      | L2      | CO_2     |
|-------------|----------|-------|-------|-------|-------|-------|---------|---------|----------|
| C_{red1}    | 3        | 0.53(0.24) | 0.95(3.34) | 0.71(3.08) | 0.67(-2.91) | 0.68(-2.96) | -0.83(7.10) | -1.16(-6.13) | -         |
| C_{red1}μCO_2 | 3       | 0.54(0.24) | 1.14(3.55) | 0.69(3.05) | 0.76(-3.06) | 0.70(-3.01) | -0.28(7.46) | -0.80(-6.49) | -0.84(0.03) |
| C_{red1}tCO_2 | -     | -     | -     | -     | -     | -     | -       | -       | -         |
| C_{int}     | 3        | 0.52(0.00) | 0.86(3.15) | 0.73(3.14) | 0.73(-3.05) | 0.77(-3.16) | -1.40(6.55) | -1.54(-6.58) | -         |
| C_{int}μCO_2 | 3       | 0.51(0.00) | 1.08(3.41) | 0.71(3.08) | 0.83(-3.24) | 0.77(-3.17) | -0.79(6.91) | -1.14(-6.94) | -0.91(0.03) |
| C_{int}tCO_2 | -      | -     | -     | -     | -     | -     | -       | -       | -         |
| C_{red2}    | 6        | 0.47(-0.03) | 0.87(-3.17) | 0.69(-2.93) | 0.81(3.23)  | 0.79(3.25)  | -1.87(-6.17) | -1.77(6.90)  | -         |
| C_{red2}μCO_2 | 6     | 0.51(0.03) | 1.06(-3.36) | 0.63(-2.70) | 0.88(3.34)  | 0.80(3.25)  | -1.34(-6.11) | -1.46(7.12)  | -0.96(-0.02) |
| C_{red2}tCO_2 | -      | -     | -     | -     | -     | -     | -       | -       | -         |
Table S12 Energies (in eV) of LUMO and HOMO orbitals of selected species of the C-cluster optimized using the LM$^{H+,K+}$ and the LM$^{H0,K+}$ models of the active site.

|                  | LM$^{H+,K+}$ |        | LM$^{H0,K+}$ |        |
|------------------|---------------|--------|---------------|--------|
|                  | LUMO   | HOMO   | LUMO   | HOMO   |
| $C_{\text{red}1}$ | -2.086 | -2.548 | -1.421 | -1.915 |
| $C_{\text{red}1}$-$\mu CO_2$ | -2.476 | -3.049 | -1.782 | -2.579 |
| $C_{\text{red}1}$-$t CO_2$ | -      | -     | -1.509 | -2.004 |
| $C_{\text{red}1}$-$OH$     | -0.998 | -1.518 | -0.162 | -0.678 |
| $C_{\text{red}1}$-$CO_2$-$OH$ | -1.258 | -1.933 | -0.451 | -1.070 |
| $C_{\text{red}1}$-$CO_2$     | -0.553 | -0.670 | 0.070  | -0.082 |
| $C_{\text{int}}$-$\mu CO_2$ | -1.238 | -1.247 | -0.565 | -0.594 |
| $C_{\text{red}2}$-$t CO_2$ | -1.225 | -1.251 | -0.231 | -0.233 |
| $C_{\text{red}2}$     | 0.669   | 0.669 | 1.283  | 1.281  |
| $C_{\text{red}2}$-$\mu CO_2$ | 0.229  | 0.122 | 0.921  | 0.781  |
| $C_{\text{red}2}$-$t CO_2$ | 0.182  | 0.115 | 1.199  | 1.019  |

Table S13 Energy differences (in eV) between reduced and oxidized species ($E_{\text{red}} - E_{\text{ox}}$) calculated for unbound, $\mu CO_2$ and $t CO_2$ forms of the C-cluster optimized using the LM$^{H+,K+}$, LM$^{H0,K+}$ and LM$^{H+,K0}$ models of the active site. Such values are expected to follow the same trend as the experimental reduction potentials.

|                  | SM       | LM$^{H+,K+}$ | LM$^{H0,K+}$ | LM$^{H+,K0}$ |
|------------------|----------|--------------|--------------|--------------|
| $C_{\text{red}1}$-$C_{\text{int}}$ | -0.64    | -1.43        | -0.54        | -0.61        |
| $C_{\text{red}1}$-$\mu CO_2$-$C_{\text{int}}$-$\mu CO_2$ | -0.86    | -1.92        | -1.24        | -1.24        |
| $C_{\text{red}1}$-$t CO_2$-$C_{\text{int}}$-$t CO_2$ | -0.98    | -1.92        | -0.96        | -2.24        |
| $C_{\text{int}}$-$C_{\text{red}2}$ | +0.91    | +0.09        | +0.71        | +0.88        |
| $C_{\text{int}}$-$\mu CO_2$-$C_{\text{red}2}$-$\mu CO_2$ | +0.49    | -0.60        | +0.06        | +0.33        |
| $C_{\text{int}}$-$t CO_2$-$C_{\text{red}2}$-$t CO_2$ | +0.46    | -0.59        | +0.41        | -2.24        |
Table S14 Selected distances (Å) and angles (°) for small and large models (SM, LM^{H+,K+}, LM^{H0,K+}, LM^{H+,K0}) of the tCO2-bound C-cluster in the C_{red2}, C_{int} and C_{red1} redox states (in their most stable BS state), optimized at the RI-BP86/def-TZVP-SVP level in COSMO with ε=4.

|          | SM       | LM^{H+,K+} | LM^{H0,K+} | LM^{H+,K0} |
|----------|----------|------------|------------|------------|
|          | C_{red2} | C_{int}    | C_{red1}   | C_{red2}   | C_{int}    | C_{red1}   | C_{red2}   | C_{int}    | C_{red1}   |
| Ni-Fe_u  | 2.97     | 3.01       | 2.69       | 2.65       | 2.66       | -          | 2.90       | 2.86       | 2.69       |
| Ni-S{Cys526} | 2.24     | 2.24       | 2.22       | 2.23       | 2.22       | -          | 2.24       | 2.22       | 2.22       |
| Ni-S{4}  | 3.57     | 3.49       | 3.33       | 3.69       | 3.55       | -          | 3.72       | 3.60       | 3.26       |
| S2-Ni-{S{Cys526}} | 171.9  | 165.4      | 165.7      | 166.6      | 167.2      | -          | 148.7      | 149.6      | 156.6      |
| S1-Ni-C  | 151.7    | 147.6      | 142.3      | 158.1      | 159.4      | -          | 117.0      | 117.6      | 108.9      |
| Fe_{u}-{S{4}} | 2.26     | 2.36       | 2.33       | 2.24       | 2.23       | -          | 2.25       | 2.24       | 2.21       |
| Fe_{u}-{S{Cys295}} | 2.39     | 2.25       | 2.20       | 2.34       | 2.31       | -          | 2.39       | 2.36       | 2.32       |
| Fe_{u}-N{His261} | 2.14     | 2.13       | 2.11       | 2.19       | 2.19       | -          | 2.18       | 2.18       | 2.18       |
| Fe_{u}-{S{Cys526}} | 2.34     | 2.33       | 2.37       | 2.65       | 2.55       | -          | 2.40       | 2.39       | 2.40       |
| Ni-Fe_{u}-S{4} | 85.0     | 81.8       | 85.1       | 97.6       | 92.5       | -          | 91.7       | 89.1       | 82.9       |
| Ni-C     | 1.95     | 1.99       | 2.04       | 1.89       | 1.90       | -          | 1.98       | 2.02       | 2.18       |
| Fe_{u}-O{1} | 4.12     | 4.12       | 3.90       | 3.19       | 3.05       | -          | 4.62       | 4.56       | 4.46       |
| C-O{1}   | 1.25     | 1.24       | 1.23       | 1.32       | 1.31       | -          | 1.24       | 1.23       | 1.21       |
| C-O{2}   | 1.26     | 1.25       | 1.23       | 1.24       | 1.24       | -          | 1.24       | 1.23       | 1.21       |
| O1-C-O{2} | 130.3    | 134.0      | 138.9      | 122.4      | 125.6      | -          | 134.5      | 140.0      | 147.7      |
| Ni-C-O{1} | 120.2    | 119.0      | 118.5      | 120.6      | 120.6      | -          | 115.5      | 114.0      | 107.9      |
| Ni-C-O{2} | 109.4    | 107.0      | 102.6      | 117.0      | 113.8      | -          | 110.0      | 106.0      | 104.4      |
| O1-N{lys563} | -        | -          | -          | 2.78       | 2.80       | -          | 4.07       | 4.08       | 4.05       |
| O1-H{lys563} | -        | -          | -          | 1.72       | 1.74       | -          | 3.20       | 3.29       | 3.24       |
| O2-r{N{His93}} | -        | -          | -          | 3.03       | 3.07       | -          | 5.54       | 5.59       | 5.59       |
| O2-r{H{His93}} | -        | -          | -          | 2.37       | 2.40       | -          | -          | -          | -          |
| O1-e{N{His93}} | -        | -          | -          | 2.62       | 2.69       | -          | 3.27       | 3.31       | 3.28       |
| O1-e{H{His93}} | -        | -          | -          | 1.52       | 1.62       | -          | -          | -          | -          |
Table S15 Selected distances (Å) and angles (°) for small and large models (SM, LM^{H+,K+}, LM^{H0,K+}, LM^{H+,K0}) of the CO2-OH-bound C-cluster in the C_red2, C_int and C_red1 redox states (in their most stable BS state), optimized at the RI-BP86/def-TZVP-SVP level in COSMO with ε=4, and for the 2YIV x-ray structure.

|                  | X-ray        | SM            | LM^{H+,K+}    | LM^{H0,K+}    | LM^{H+,K0}    |
|------------------|--------------|---------------|---------------|---------------|---------------|
|                  | 2YIV C_red2  | C_int C_red1  | C_red2 C_int | C_red1 C_red2| C_int C_red1  |
| Ni-Fe_u          | 3.10         | 3.34 3.28 2.98| 3.04 3.03 2.80| 2.79 3.04 2.72| 2.90 2.92 2.76|
| Ni-S_{Cys526}    | 2.09         | 2.25 2.25 2.25| 2.26 2.23 2.23| 2.25 2.26 2.25| 2.21 2.22 2.22|
| Ni-S_{4}         | 3.99         | 3.52 3.41 2.94| 3.70 3.61 3.25| 3.60 3.70 3.05| 3.62 3.52 3.15|
| S2-Ni-S_{Cys526} | 161.0        | 153.3 153.9 165.2| 157.4 149.7 156.6| 153.6 151.6 153.2| 147.8 144.6 152.1|
| S1-Ni-C          | 147.8        | 141.3 137.6 118.6| 141.0 140.3 128.1| 133.5 141.0 112.8| 138.7 134.9 122.5|
| Fe_u-S_{4}       | 2.32         | 2.39 2.33 2.28| 2.32 2.30 2.25| 2.32 2.32 2.26| 2.34 2.32 2.26|
| Fe_u-S_{Cys295}  | 2.30         | 2.51 2.46 2.45| 2.38 2.36 2.37| 2.39 2.38 2.38| 2.45 2.48 2.45|
| Fe_u-N_{His261}  | 2.05         | 2.34 2.34 2.28| 2.25 2.28 2.27| 2.20 2.25 2.26| 2.25 2.26 2.23|
| Fe_u-S_{Cys526}  | 3.44         | 2.54 2.52 2.49| 2.71 2.65 2.58| 2.78 2.71 2.54| 2.77 2.62 2.55|
| Ni-Fe_{4},S_{4}  | 93.7         | 73.8 72.5 66.6| 86.3 84.2 79.4| 89.1 86.3 75.0| 86.8 83.6 77.1|
| Ni-C             | 1.95         | 1.93 1.94 1.99| 1.92 1.92 1.95| 1.94 1.92 2.05| 1.91 1.92 1.96|
| Fe_{4}-O_{1}     | 4.06         | 4.04 4.00 3.96| 4.00 3.99 3.93| 3.98 4.00 4.03| 3.91 3.93 3.89|
| C-O_{1}          | 1.34         | 1.27 1.26 1.25| 1.30 1.28 1.26| 1.25 1.30 1.23| 1.28 1.27 1.26|
| C-O_{2}          | 1.24         | 1.26 1.25 1.24| 1.24 1.24 1.23| 1.24 1.24 1.22| 1.24 1.24 1.23|
| O_{1}-C-O_{2}    | 121.1        | 127.7 130.6 134.5| 125.8 130.4 135.0| 133.1 125.8 142.0| 127.6 129.5 133.4|
| Ni-Fe_{4},O_{1}  | 43.0         | 43.6 44.0 43.8| 43.8 44.3 44.9| 43.8 43.8 41.9| 45.5 45.4 45.7|
| Ni-C-O_{1}       | 113.0        | 122.3 121.2 113.9| 116.3 120.5 118.0| 118.0 117.9 108.9| 120.6 121.0 118.2|
| Ni-C-O_{2}       | 125.9        | 110.0 108.2 111.6| 117.9 109.1 107.0| 109.0 116.3 109.1| 111.9 109.4 108.4|
| Fe_{4}-O_{OH}    | 1.96         | 1.94 1.92 1.90| 2.03 2.00 1.97| 2.17 2.03 1.99| 1.93 1.93 1.92|
| O_{1}-O_{OH}     | 2.51         | 2.91 2.88 2.87| 2.66 2.69 2.72| 2.62 2.66 2.93| 2.71 2.73 2.74|
| O_{1}-O_{HOH}    | -            | 1.93 1.90 1.89| 1.66 1.70 1.73| 1.60 1.66 1.95| 1.73 1.75 1.76|
| Fe_{4},O_{OH}-H_{OHH} | -   | 108.8 110.3 112.2| 110.9 111.1 110.9| 111.4 110.9 114.3| 112.1 1112.8 113.3|
| S_{4},Fe_{4},O_{OH} | 148.4       | 140.0 140.7 134.8| 143.7 140.9 134.2| 147.4 143.7 136.3| 157.8 150.2 141.9|
| O_{1}-N_{Lys563} | 3.94         | - - -     | 3.97 4.08 4.22| 4.23 3.97 4.45| 4.24 4.21 4.22|
| O_{1}-H_{Lys563} | -            | - - -     | 3.15 3.26 3.39| 3.11 3.15 3.63| - - -     |
| O_{2},eN_{His93} | 4.39         | - - -     | 4.14 4.42 4.63| 5.10 4.14 5.35| 4.25 4.36 4.59|
| O_{2},eH_{His93} | -            | - - -     | 3.18 3.48 3.70| - - -     | 3.32 3.44 3.68|
| O_{1},eN_{His93} | 2.85         | - - -     | 2.59 2.68 2.77| 3.54 2.59 3.26| 2.64 2.68 2.80|
| O_{1},eH_{His93} | -            | - - -     | 1.49 1.62 1.75| - - -     | 1.56 1.62 1.78|
| O_{OH}^{+}N_{Lys563} | -   | - - - | 2.55 2.60 2.63| 2.63 2.55 2.61| 3.38 3.28 3.14|
| O_{OH}^{+}H_{Lys563} | -  | - - - | 1.40 1.47 1.53| 1.08 1.40 1.50| - - -     |
Scheme S2 Schematic representation of the a) $C_{\text{red2}}/C_{\text{int}}-\text{CO}_2-\text{OH} - \text{LM}^{\text{Hi},K^+}$, b) $C_{\text{red2}}-\text{CO}_2-\text{OH} - \text{LM}^{\text{Hi},K^+}$, c) $C_{\text{int}}-\text{CO}_2-\text{OH} - \text{LM}^{\text{Hi},K^+}$ and d) $C_{\text{red2}}/C_{\text{int}}-\text{CO}_2-\text{OH} - \text{LM}^{\text{Hi},K^0}$ formation process from the naked C-cluster, CO$_2$ and H$_2$O. The proton that is transferred from His93 and Lys563 to the OH-bound C-cluster to form a H$_2$O-adduct, is depicted in red and blue, respectively.

Table S16 NBO atomic charges and Mulliken spin densities (in parenthesis) of selected atoms of the C-cluster and of the layers L1 and L2 (corresponding respectively to the blue and red layers of the BS coupling schemes shown in Scheme S1) for the OH-bound form of the C-cluster and CO$_2$-OH adducts in the $C_{\text{red2}}$, $C_{\text{int}}$ and $C_{\text{red1}}$ redox states (in the most stable spin coupling scheme) optimized using the SM, LM$^{\text{Hi},K^+}$, LM$^{\text{Hi},K^+}$ and LM$^{\text{Hi},K^0}$ DFT models.

a) SM model

| Redox state | BS state | Ni  | Fe1  | Fe2  | Fe3  | L1            | L2            | CO$_2$         | OH            |
|-------------|----------|-----|------|------|------|---------------|---------------|----------------|---------------|
| $C_{\text{red2}}$-OH  | 3        | 0.58(0.31) | 1.16(3.59) | 0.71(3.06) | 0.75(-3.11) | -0.94(7.35) | -1.41(-6.53) | -0.65(-0.17) |
| $C_{\text{red2}}$-CO$_2$-OH | 3        | 0.57(0.15) | 1.24(3.65) | 0.73(3.11) | 0.73(-3.01) | -0.45(7.42) | -1.20(-6.63) | -0.71(-0.01) |
| $C_{\text{int}}$-OH | 3        | 0.56(0.03) | 1.09(3.45) | 0.72(3.05) | 0.80(-3.21) | -1.55(6.67) | -1.73(-6.77) | -0.73(0.10)  |
| $C_{\text{int}}$-CO$_2$-OH | 6        | 0.56(-0.03) | 1.25(-3.64) | 0.70(-2.98) | 0.82(3.25) | -0.98(-6.94) | -1.48(7.09) | -0.86(0.04) |
| $C_{\text{red1}}$-OH | 6        | 0.54(0.36) | 1.06(-3.37) | 0.71(-2.91) | 0.81(3.26) | -2.19(-5.77) | -2.08(6.86) | -0.74(-0.10) |
| $C_{\text{red2}}$-CO$_2$-OH | 6        | 0.55(0.10) | 1.19(-3.51) | 0.67(-2.79) | 0.90(3.38) | -1.53(-6.27) | -1.77(7.36) | -0.97(0.04) |

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### b) $\text{LM}^{\text{H}+,\text{K}+}$ model

| Redox state | BS state | Ni    | Fe0   | Fe1   | Fe2   | Fe3   | L1     | L2     | CO2    | OH     |
|-------------|----------|-------|-------|-------|-------|-------|--------|--------|--------|--------|
| C_rred=OH   | 3        | 0.53(0.12) | 1.12(3.52) | 0.73(3.13) | 0.70(-2.99) | 0.73(-3.07) | -0.83(7.26) | -1.21(-6.34) | -     | -0.72(0.05) |
| C_rred=CO2-OH | 3   | 0.57(0.27) | 1.22(3.62) | 0.68(3.02) | 0.74(-3.05) | 0.77(-3.12) | -0.40(7.48) | -0.99(-6.58) | -0.67(-0.03) | -0.64(0.11) |
| C_rred=OH   | 3        | 0.49(0.04) | 0.87(3.17) | 0.75(3.13) | 0.74(-3.07) | 0.78(-3.18) | -1.40(-6.63) | -1.48(-6.63) | -     | -0.55(-0.02) |
| C_rred=CO2-OH | 6   | 0.57(+0.12) | 1.18(-3.53) | 0.65(-2.92) | 0.82(3.23) | 0.81(3.22) | -0.87(-6.93) | -1.27(-6.98) | -0.80(0.04) | -0.65(-0.08) |
| C_rred=OH   | 6        | 0.43(-0.03) | 0.87(-3.19) | 0.67(-2.82) | 0.83(3.26) | 0.80(3.25) | -1.94(-6.05) | -1.75(-6.92) | -     | -0.56(0.02) |
| C_rred=CO2-OH | 3   | 0.56(0.19) | 1.15(3.47) | 0.74(3.15) | 0.82(-3.20) | 0.72(-2.94) | -1.16(-7.30) | -1.68(-6.45) | -0.94(0.01) | -0.64(0.06) |

### c) $\text{LM}^{\text{H}0,\text{K}+}$ model

| Redox state | BS state | Ni    | Fe0   | Fe1   | Fe2   | Fe3   | L1     | L2     | CO2    | OH     |
|-------------|----------|-------|-------|-------|-------|-------|--------|--------|--------|--------|
| C_rred=OH   | 3        | 0.56(0.13) | 1.13(3.51) | 0.73(3.13) | 0.70(-2.98) | 0.72(-3.05) | -0.80(7.24) | -1.29(-6.31) | -     | -0.56(0.03) |
| C_rred=CO2-OH | 3   | 0.57(0.28) | 1.20(3.60) | 0.68(3.00) | 0.74(-3.04) | 0.75(-3.09) | -0.51(7.40) | -1.16(-6.52) | -0.49(-0.02) | -0.62(0.10) |
| C_rred=OH   | 6        | 0.55(0.05) | 1.06(-3.38) | 0.72(-3.06) | 0.76(3.12) | 0.76(3.14) | -1.40(-6.57) | -1.62(6.62) | -     | -0.55(-0.02) |
| C_rred=CO2-OH | 3   | 0.55(0.15) | 1.17(3.52) | 0.66(2.92) | 0.81(-3.20) | 0.81(-3.21) | -1.00(6.90) | -1.45(-6.91) | -0.61(-0.06) | -0.63(0.07) |
| C_rred=OH   | 6        | 0.54(0.12) | 1.02(-3.31) | 0.71(-2.98) | 0.80(3.22) | 0.78(3.21) | -1.77(-6.17) | -1.84(6.84) | -     | -0.54(0.00) |
| C_rred=CO2-OH | 6   | 0.52(-0.02) | 1.12(-3.46) | 0.63(-2.74) | 0.85(3.29) | 0.82(3.27) | -1.39(-6.31) | -1.61(7.05) | -0.71(0.04) | -0.57(-0.03) |

### d) $\text{LM}^{\text{H}+,\text{K}0}$ model

| Redox state | BS state | Ni    | Fe0   | Fe1   | Fe2   | Fe3   | L1     | L2     | CO2    | OH     |
|-------------|----------|-------|-------|-------|-------|-------|--------|--------|--------|--------|
| C_rred=OH   | 3        | 0.56(0.13) | 1.13(3.51) | 0.73(3.13) | 0.70(-2.98) | 0.72(-3.05) | -0.80(7.24) | -1.29(-6.31) | -     | -0.56(0.03) |
| C_rred=CO2-OH | 3   | 0.58(0.29) | 1.23(3.61) | 0.72(3.11) | 0.76(-3.12) | 0.81(-3.21) | -0.55(7.59) | -1.04(-6.80) | -0.69(-0.01) | -0.61(0.17) |
| C_rred=OH   | 6        | 0.55(0.05) | 1.06(-3.38) | 0.72(-3.06) | 0.76(3.12) | 0.76(3.14) | -1.40(-6.57) | -1.62(6.62) | -     | -0.55(-0.02) |
| C_rred=CO2-OH | 6   | 0.58(-0.16) | 1.23(-3.57) | 0.68(-2.96) | 0.82(3.26) | 0.86(3.32) | -1.01(-7.00) | -0.70(7.16) | -0.81(0.02) | -0.63(-0.14) |
| C_rred=OH   | 6        | 0.54(0.12) | 1.02(-3.31) | 0.71(-2.98) | 0.80(3.22) | 0.78(3.21) | -1.77(-6.17) | -1.84(6.84) | -     | -0.54(0.00) |
| C_rred=CO2-OH | 6   | 0.56(-0.05) | 1.22(-3.54) | 0.65(-2.78) | 0.84(3.30) | 0.88(3.37) | -1.32(-6.51) | -0.85(7.16) | -0.87(0.02) | -0.63(-0.12) |
| Element | Mass     | Standard Deviation | Percent of Total |
|---------|----------|--------------------|------------------|
| Ni      | 54.7224059 | 7.276434 | 47.9303434 |
| Fe      | 54.6607500 | 10.2740919 | 49.6756594 |
| Fe      | 53.4327793 |                | 49.6756594 |
| H       | 53.3715548 | 15.1901962 | 45.9651564 |
| S       | 53.3300379 | 12.276434 | 47.9303434 |
| N       | 49.7098285 | 17.9873034 | 47.9303434 |
| H       | 54.6486700 | 17.9873034 | 47.9303434 |
| C       | 56.0621870 | 17.9873034 | 47.9303434 |
| H       | 55.2345670 | 17.9873034 | 47.9303434 |
| O       | 55.8927850 | 17.9873034 | 47.9303434 |
| C       | 55.2536890 | 17.9873034 | 47.9303434 |
| H       | 54.7890120 | 17.9873034 | 47.9303434 |
| S       | 55.2267890 | 17.9873034 | 47.9303434 |
| N       | 50.0493750 | 17.9873034 | 47.9303434 |
| H       | 53.7245670 | 17.9873034 | 47.9303434 |
| C       | 53.8627850 | 17.9873034 | 47.9303434 |
| C       | 53.2267890 | 17.9873034 | 47.9303434 |
| O       | 52.8972850 | 17.9873034 | 47.9303434 |
| C       | 52.4637890 | 17.9873034 | 47.9303434 |
| H       | 51.9245670 | 17.9873034 | 47.9303434 |
| S       | 51.3857890 | 17.9873034 | 47.9303434 |
| C       | 51.0923456 | 17.9873034 | 47.9303434 |
| H       | 50.5923456 | 17.9873034 | 47.9303434 |
| C       | 50.0923456 | 17.9873034 | 47.9303434 |
| O       | 49.5923456 | 17.9873034 | 47.9303434 |

**Table continued...**
