Electronic Supplementary Material (ESI) for MedChemComm.

Supplementary Materials

Probing substrate binding to the metal binding sites in metallo-β-lactamase L1 during catalysis

Mahesh Aitha,[1] David L. Tierney,[1*] Michael W. Crowder,[1*]

[†]*Department of Chemistry and Biochemistry, Miami University, 650 East High Street, Oxford, Ohio 45056, USA*

***Corresponding authors:** David L. Tierney and Michael W. Crowder

Phone: (513) 529-2813; **Fax:** (513) 529-5715; **e-mail: tiernedl@miamioh.edu;** crowdemw@MiamiOH.edu

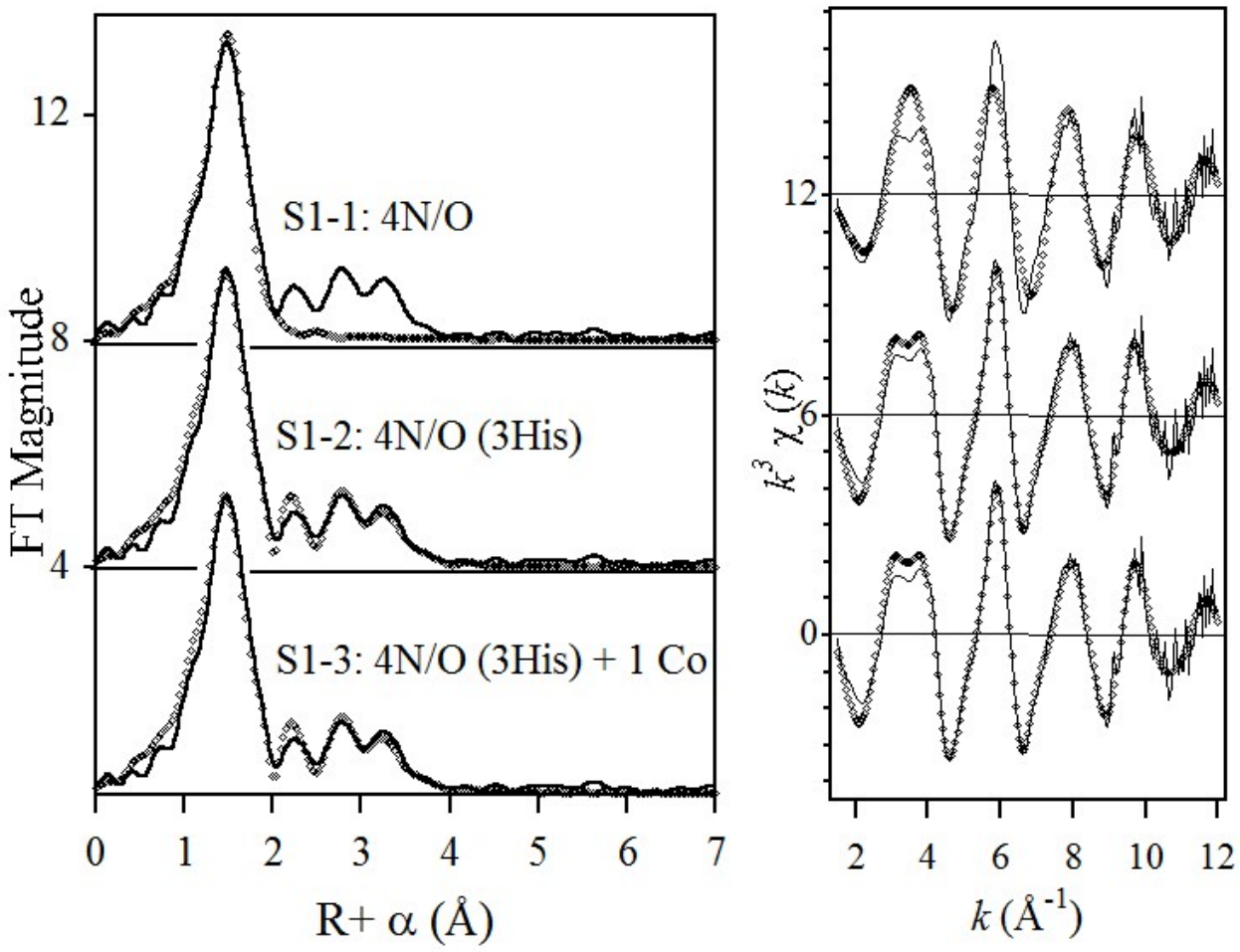

Figure S1. Fourier transforms (A) of k^3-weighted EXAFS (B) for Zn K-edge of ZnCo-L1 resting. The solid lines represent the data, the open symbols represent the various fits, corresponding to Table S1.

Table S1. Fitting results for Zn K-edge EXAFS of the resting ZnCo-L1 sample.[a]

Fit	Model	Zn-N/O	Zn-His[b]	Zn-Co	R_f[c]	R_u[c]
1-1	4 N/O	2.01 (4.4)			25	142
1-2	4 N/O (3His)	2.01 (6.1)	2.92 (5.9) 3.17 (4.7) 4.10 (17) 4.45 (24)		27	43
1-3	4N/O (2His) + Zn-Co	2.01 (6.1)	2.92 (6.1) 3.17 (5.0) 4.10 (19) 4.44 (24)	3.48 (28)	23	38

[a] Distances (Å) and disorder parameters (in parentheses, σ^2 (10^{-3} Å^2)) shown derive from integer or half-integer coordination number fits to filtered EXAFS data [Δk = 1.5-12 Å^{-1}; ΔR = 0.5 – 2.0 Å (fit 1), 0.3-4.0 Å (fits 2-3)].

[b] Imidazole multiple scattering paths represent combined paths, as described previously (see Materials and Methods).

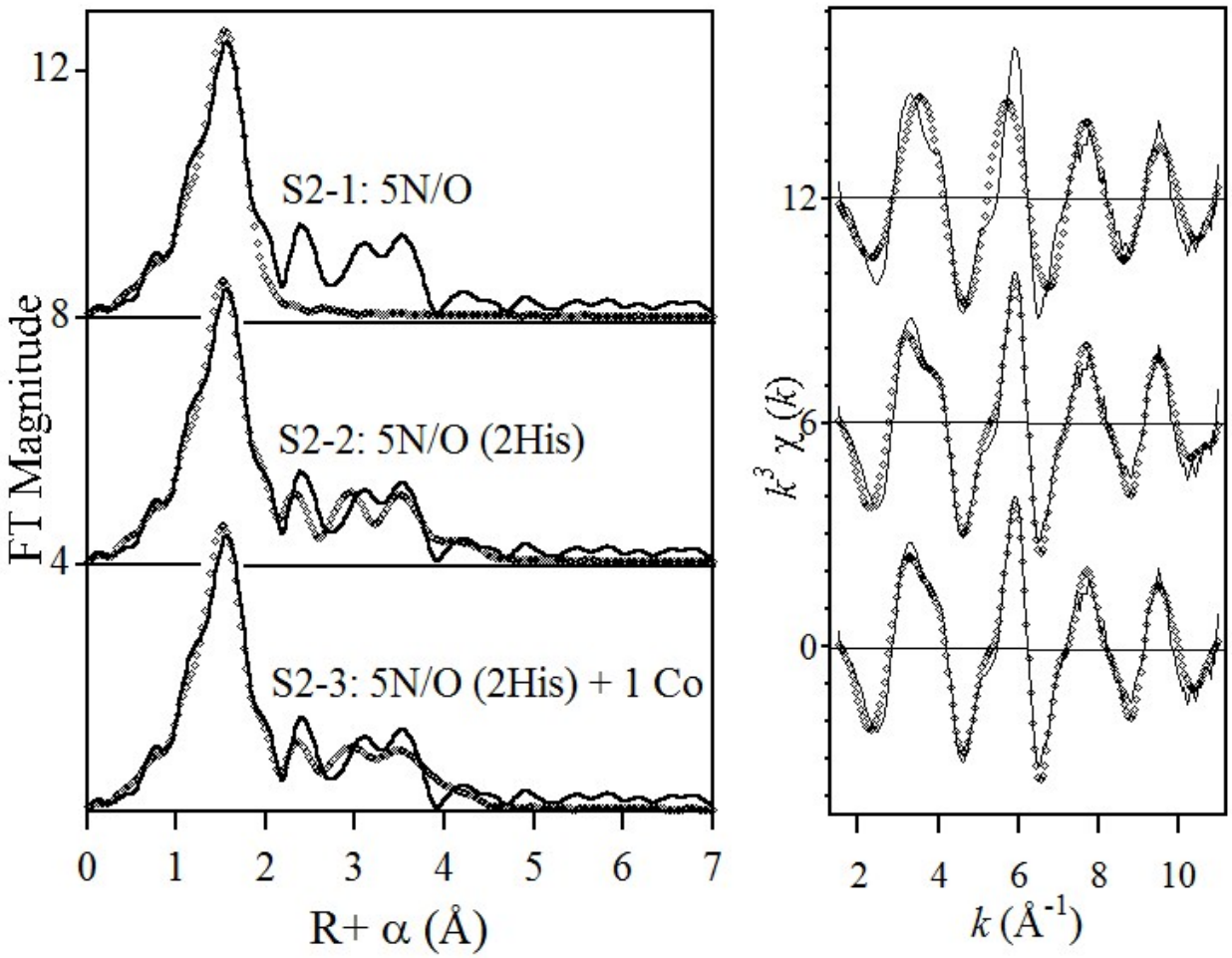

Figure S2. Fourier transforms (A) of k^3-weighted EXAFS (B) for Co K-edge of ZnCo-L1 resting. The solid lines represent the data, the open symbols represent the various fits, corresponding to Table S2.

Table S2. Fitting results for Co K-edge EXAFS of the resting ZnCo-L1 sample.[a]

Fit	Model	Co-N/O	Co-His[b]	Co-C	Co-Zn	R_f[c]	R_u[c]
2-1	5 N/O	2.08 (6.8)				41	247
2-2	5 N/O (2His)	2.08 (6.6)	2.99 (12) 3.28 (1.0) 4.13 (32) 4.61 (11)			61	46
2-3	5N/O(2His) + Co-Zn	2.08 (6.6)	3.01 (12) 3.26 (1.0) 4.24 (8.6) 4.61 (12)		3.50 (13)	47	39
2-4	5N/O(2His) +1C + Co-Zn	2.08 (6.6)	3.01 (12) 3.26 (1.0) 4.24 (8.6) 4.61 (12)	2.41 (10)	3.50 (13)	41	35

[a] Distances (Å) and disorder parameters (in parentheses, σ^2 (10^{-3} Å^2)) shown derive from integer or half-integer coordination number fits to filtered EXAFS data [Δk = 1.5-11 Å^{-1}; ΔR = 0.5 – 2.2 Å (fit 1), 0.3-4.0 Å (fits 2-3)].

[b] Imidazole multiple scattering paths represent combined paths, as described previously (see Materials and Methods).

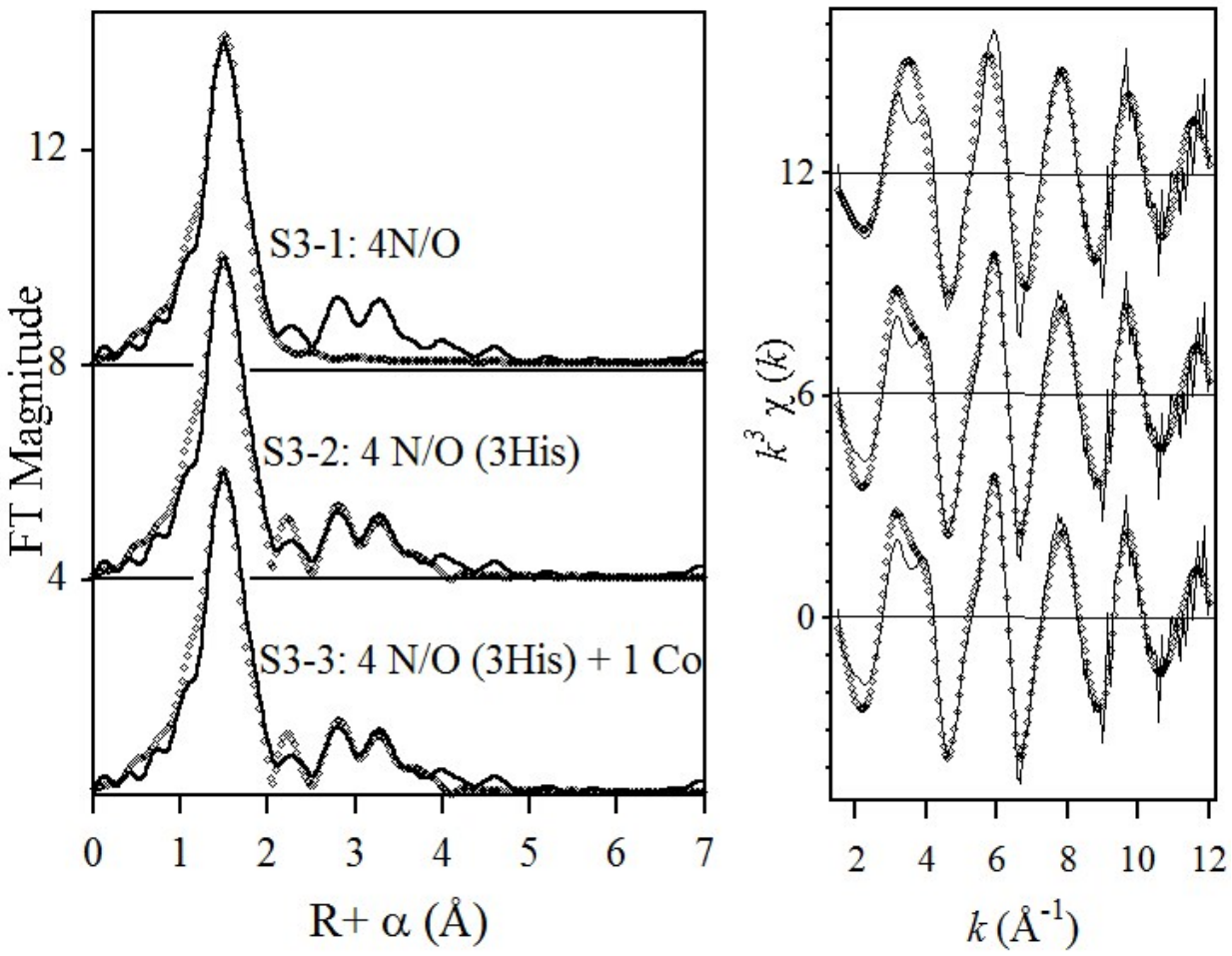

Figure S3. Fourier transforms (A) of k^3-weighted EXAFS (B) for Zn K-edge of the ZnCo-L1/chromacef sample quenched at 10 ms. The solid lines represent the data, the open symbols represent the various fits, corresponding to Table S3.

Table S3. Fitting results for Zn K-edge EXAFS spectra of the ZnCo-L1/chromacef sample quenched at 10 ms.[a]

Fit	Model	Zn-N/O	Zn-His[b]	Zn-Co	R_f[c]	R_u[c]
3-1	4 N/O	2.02 (4.8)			27	135
3-2	4 N/O (3His)	2.02 (5.0)	2.89 (7.5) 3.13 (4.2) 4.12 (22) 4.39 (19)		49	68
3-3	4N/O(2His) + Zn-Co	2.02 (5.0)	2.90 (7.6) 3.13 (4.3) 4.17 (23) 4.41 (17)	3.82 (16)	43	59

[a] Distances (Å) and disorder parameters (in parentheses, σ^2 (10^{-3} Å^2)) shown derive from integer or half-integer coordination number fits to filtered EXAFS data [Δk = 1.5-12 Å^{-1}; ΔR = 0.5 – 2.0 Å (fit 1), 0.3-4.0 Å (fits 2-3)].

[b] Imidazole multiple scattering paths represent combined paths, as described previously (see Materials and Methods).

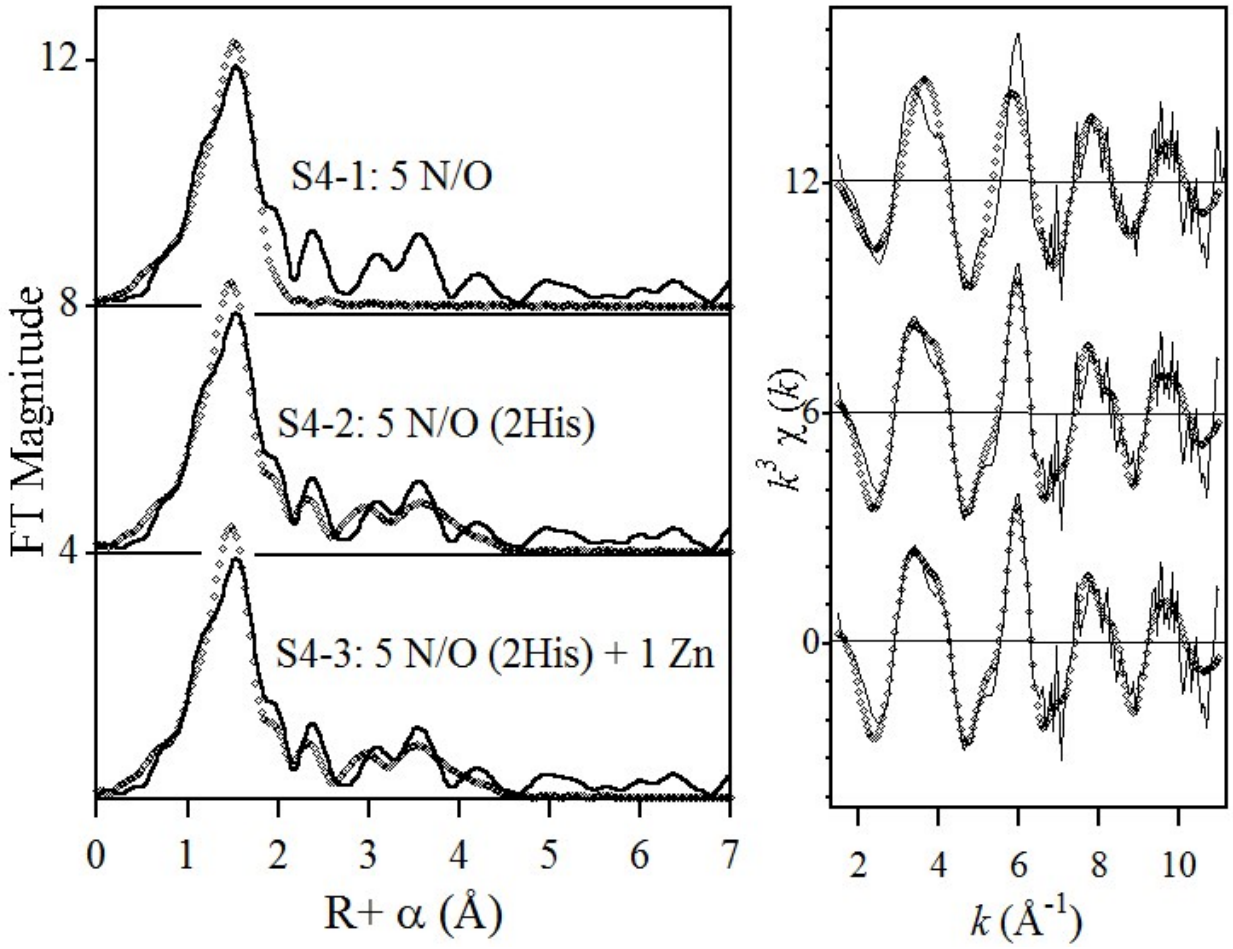

Figure S4. Fourier transforms (A) of k^3-weighted EXAFS (B) for Co K-edge of the ZnCo-L1/chromacef sample quenched at 10 ms. The solid lines represent the data, the open symbols represent the various fits, corresponding to Table S4.

Table S4. Fitting results for Co K-edge EXAFS spectra of the ZnCo-L1/chromacef sample quenched at 10 ms.[a]

Fit	Model	Co-N/O	Co-His[b]	Co-Zn	R_f[c]	R_u[c]
4-1	5 N/O	2.05 (8.0)			85	227
4-2	5 N/O (2His)	2.06 (7.9)	3.28 (5.6) 3.36 (1.0) 4.30 (9.8) 4.61 (12)		67	121
4-3	5N/O(2His) + Co-Zn	2.06 (7.9)	3.28 (5.0) 3.35 (1.0) 4.29 (15) 4.61 (14)	3.81 (13)	50	107

[a] Distances (Å) and disorder parameters (in parentheses, σ^2 (10^{-3} Å^2)) shown derive from integer or half-integer coordination number fits to filtered EXAFS data [Δk = 1.5-11 Å^{-1}; ΔR = 0.5 – 2.2 Å (fit 1), 0.3-4.0 Å (fits 2-3)].

[b] Imidazole multiple scattering paths represent combined paths, as described previously (see Materials and Methods).

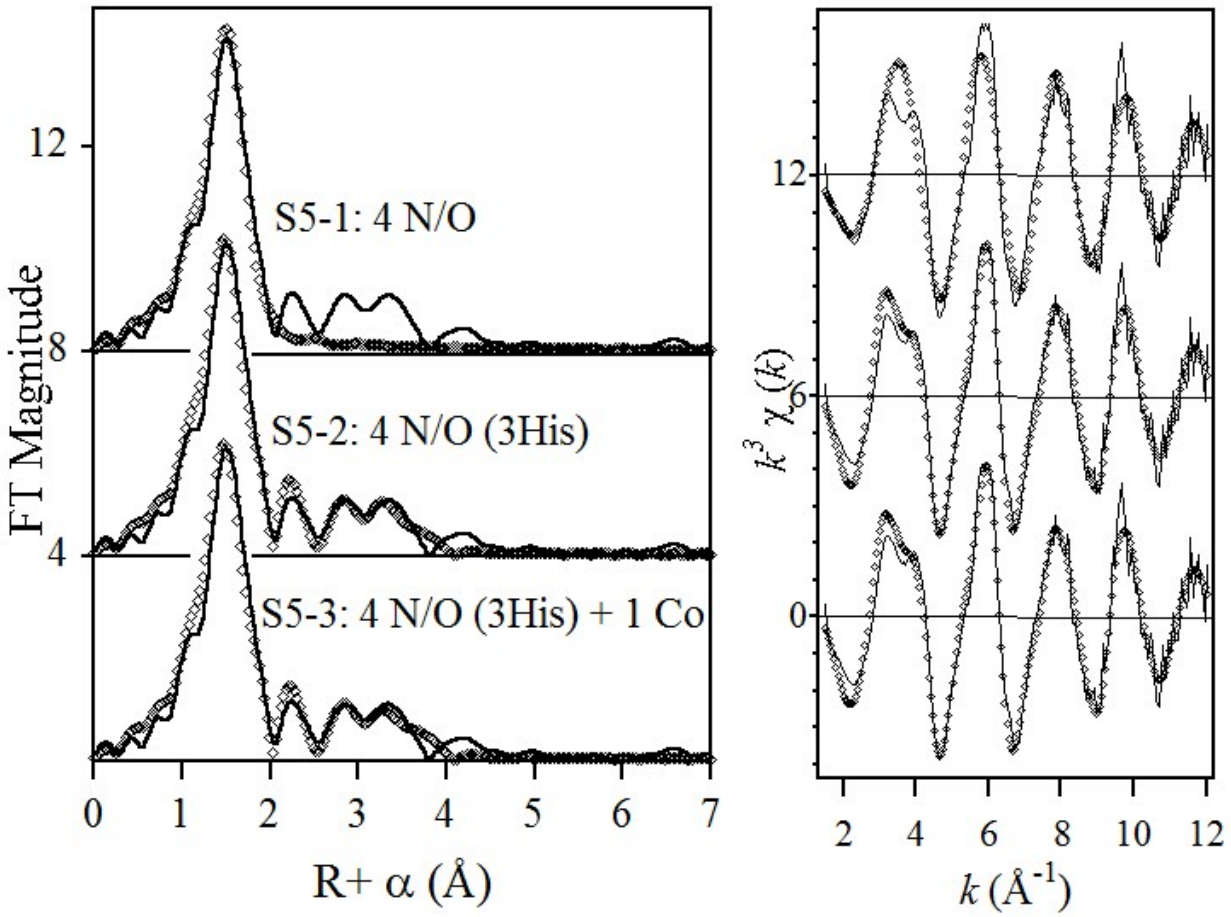

Figure S5. Fourier transforms (A) of k^3-weighted EXAFS (B) for Zn K-edge of the ZnCo-L1/chromacef sample quenched at 50 ms. The solid lines represent the data, the open symbols represent the various fits, corresponding to Table S5.

Table S5. Fitting results for Zn K-edge EXAFS spectra of the ZnCo-L1/chromacef sample quenched at 50 ms.[a]

Fit	Model	Zn-N/O	Zn-His[b]	Zn-Co	R_f[c]	R_u[c]
5-1	4 N/O	2.01 (4.7)			22	120
5-2	4 N/O (3His)	2.01 (4.8)	2.89 (6.8) 3.15 (5.9) 4.16 (21) 4.38 (17)		30	50
5-3	4N/O(2His) + Zn-Co	2.01 (4.8)	2.90 (6.9) 3.15 (6.2) 4.16 (16) 4.38 (17)	3.70 (16)	25	45

[a] Distances (Å) and disorder parameters (in parentheses, σ^2 (10^{-3} Å^2)) shown derive from integer or half-integer coordination number fits to filtered EXAFS data [Δk = 1.5-12 Å^{-1}; ΔR = 0.5 – 2.0 Å (fit 1), 0.3-4.0 Å (fits 2-3)].

[b] Imidazole multiple scattering paths represent combined paths, as described previously (see Materials and Methods).

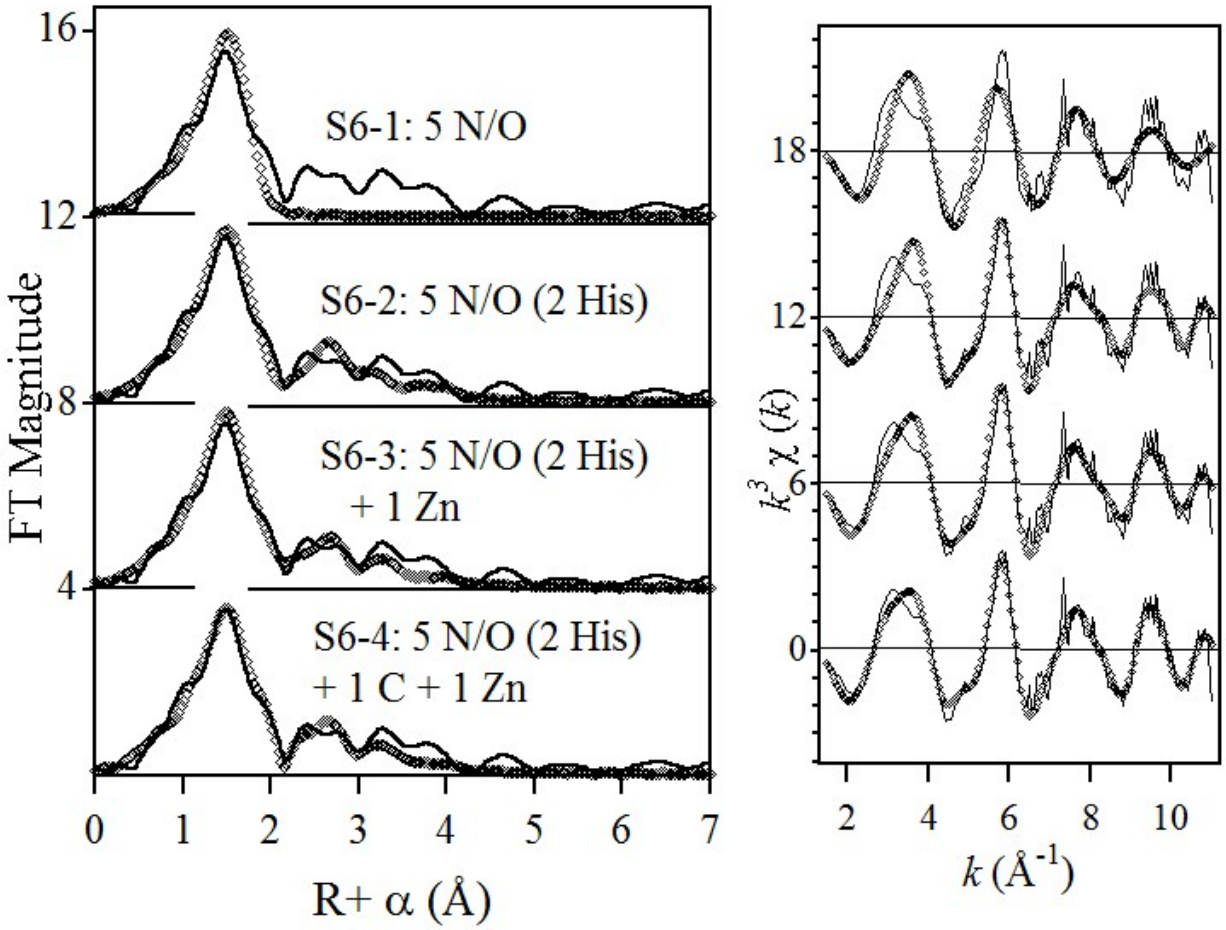

Figure S6. Fourier transforms (A) of k^3-weighted EXAFS (B) for Co K-edge of the ZnCo-L1/chromacef sample quenched at 50 ms. The solid lines represent the data, the open symbols represent the various fits, corresponding to Table S6.

Table S6. Fitting results for Co K-edge EXAFS spectra of the ZnCo-L1/chromacef sample quenched at 50 ms.[a]

Fit	Model	Co-N/O	Co-His[b]	Co-Zn	Co-C	R_f[c]	R_u[c]
6-1	5 N/O	2.09 (8.0)				98	257
6-2	5 N/O (2His)	2.09 (8.5)	3.02 (6.7) 3.31 (5.3) 4.13 (1.0) 4.24 (9.2)			163	141
6-3	5N/O(2His) + Co-Zn	2.10 (8.2)	3.01 (11) 3.31 (1.0) 4.10 (1.0) 4.21 (14)	3.68 (6.4)		123	113
6-4	5N/O(2His) + 1C + Co-Zn	2.10 (7.9)	3.01 (8.6) 3.31 (1.2) 4.10 (1.0) 4.21 (13)	3.68 (7.5)	2.41 (5.7)	103	97

[a] Distances (Å) and disorder parameters (in parentheses, σ^2 (10^{-3} Å^2)) shown derive from integer or half-integer coordination number fits to filtered EXAFS data [Δk = 1.5-11 Å^{-1}; ΔR = 0.5 – 2.2 Å (fit 1), 0.3-4.0 Å (fits 2-4)].

[b] Imidazole multiple scattering paths represent combined paths, as described previously (see Materials and Methods).

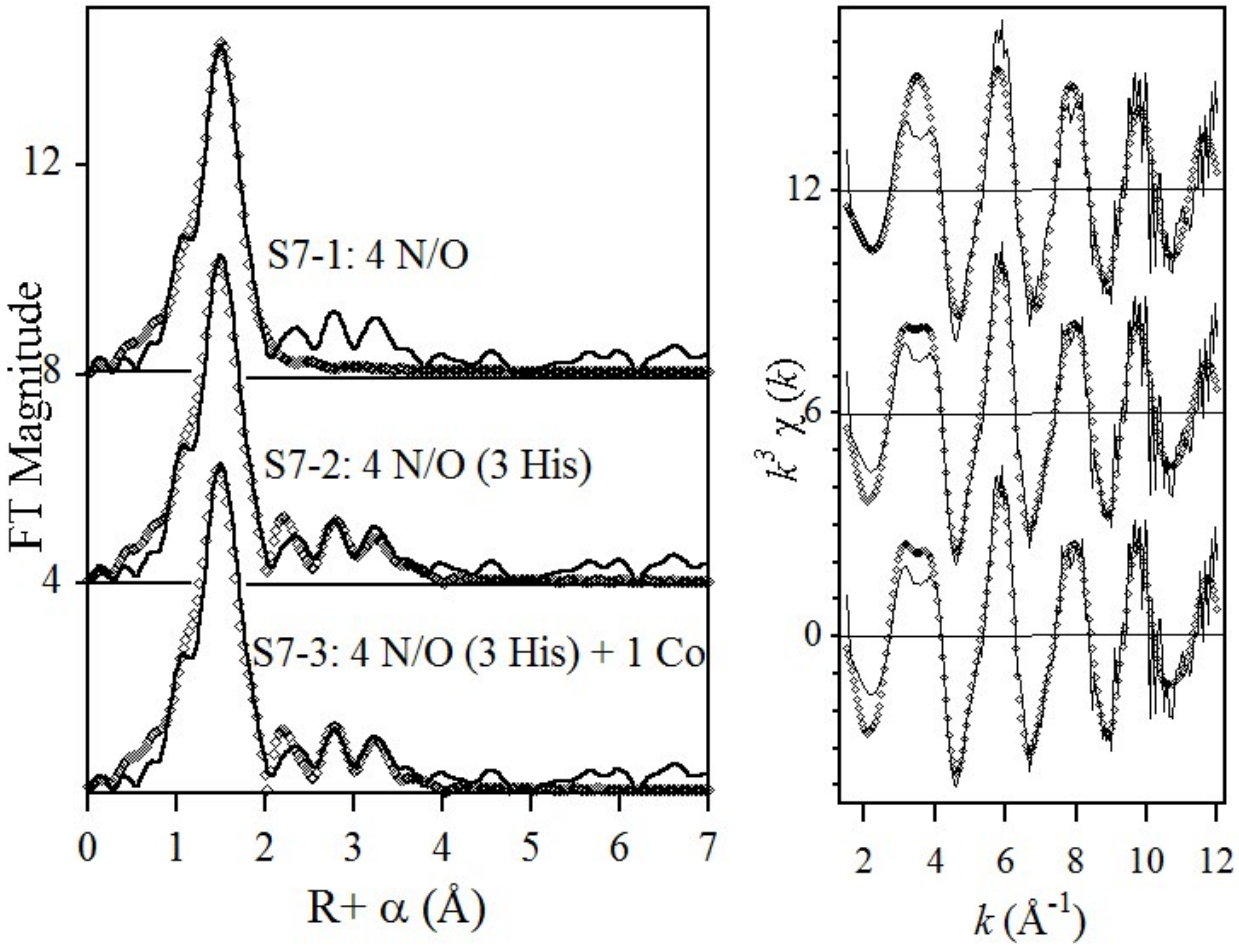

Figure S7. Fourier transforms (A) of k^3-weighted EXAFS (B) for Zn K-edge of the ZnCo-L1/chromacef sample quenched at 100 ms. The solid lines represent the data, the open symbols represent the various fits, corresponding to Table S7.

Table S7. Fitting results for Zn K-edge EXAFS spectra of the ZnCo-L1/chromacef sample quenched at 100 ms.[a]

Fit	Model	Zn-N/O	Zn-His[b]	Zn-Co	R_f[c]	R_u[c]
7-1	4 N/O	2.01 (4.6)			18	147
7-2	4 N/O (3His)	2.01 (4.8)	2.93 (9.0) 3.16 (6.6) 4.10 (22) 4.43 (24)		53	106
7-3	4N/O(2His) + Zn-Co	2.01 (4.8)	2.92 (7.4) 3.14 (4.4) 4.12 (22) 4.44 (24)	3.59 (14)	45	97

[a] Distances (Å) and disorder parameters (in parentheses, σ^2 (10^{-3} Å^2)) shown derive from integer or half-integer coordination number fits to filtered EXAFS data [Δk = 1.5-12 Å^{-1}; ΔR = 0.5 – 2.0 Å (fit 1), 0.3-4.0 Å (fits 2-3)].

[b] Imidazole multiple scattering paths represent combined paths, as described previously (see Materials and Methods).

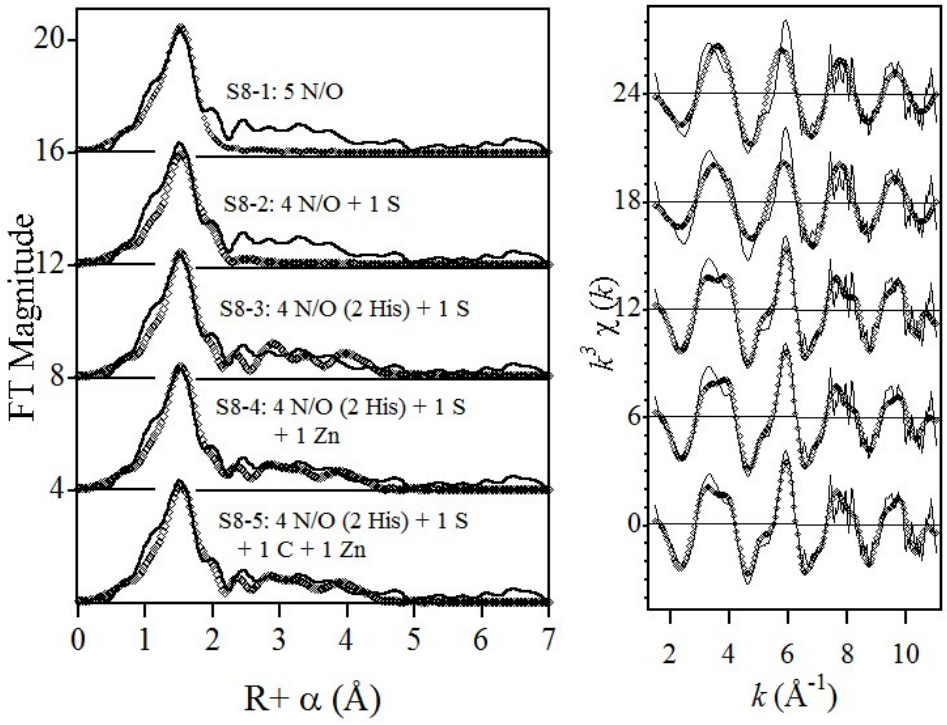

Figure S8. Fourier transforms (A) of k^3-weighted EXAFS (B) for Co K-edge of the ZnCo-L1/chromacef sample quenched at 100 ms. The solid lines represent the data, the open symbols represent the various fits, corresponding to Table S8.

Table S8. Fitting results for Co K-edge EXAFS spectra of the ZnCo-L1/chromacef sample quenched at 100 ms.[a]

Fit	Model	Co-N/O	Co-S	Co-His[b]	Co-Zn	Co-C	R_f[c]	R_u[c]
8-1	5 N/O	2.07 (7.4)					115	232
8-2	4 N/O +1 S	2.10 (5.3)	2.25 (8.3)				95	201
8-3	4 N/O(2His) + 1 S	2.07 (5.7)	2.28 (17)	3.06 (15) 3.26 (1.0) 4.16 (3.0) 4.64 (8.5)			77	132
8-4	4 N/O(2His) + 1 S + Co-Zn	2.07 (27)	2.27 (16)	3.05 (13) 3.24 (1.0) 4.12 (7.8) 4.64 (11)	3.57 (12)		71	122
8-5	4N/O(2His)+ 1S +1C + Co-Zn	2.07 (7.5)	2.28 (13)	3.05 (12) 3.24 (1.0) 4.12 (6.7) 4.64 (10)	3.58 (13)	2.40 (4.9)	52	110
8-6	5N/O(2His)+ +1C + Co-Zn	2.07 (7.5)		3.05 (10) 3.24 (1.0) 4.12 (6.5) 4.63 (11)	3.58 (9.5)	2.40 (5.1)	68	125

[a] Distances (Å) and disorder parameters (in parentheses, σ^2 (10^{-3} Å^2)) shown derive from integer or half-integer coordination number fits to filtered EXAFS data [Δk = 1.5-11 Å^{-1}; ΔR = 0.5 – 2.5 Å (fits 1-2), 0.3-4.0 Å (fits 3-5)].
[b] Imidazole multiple scattering paths represent combined paths, as described previously (see Materials and Methods).

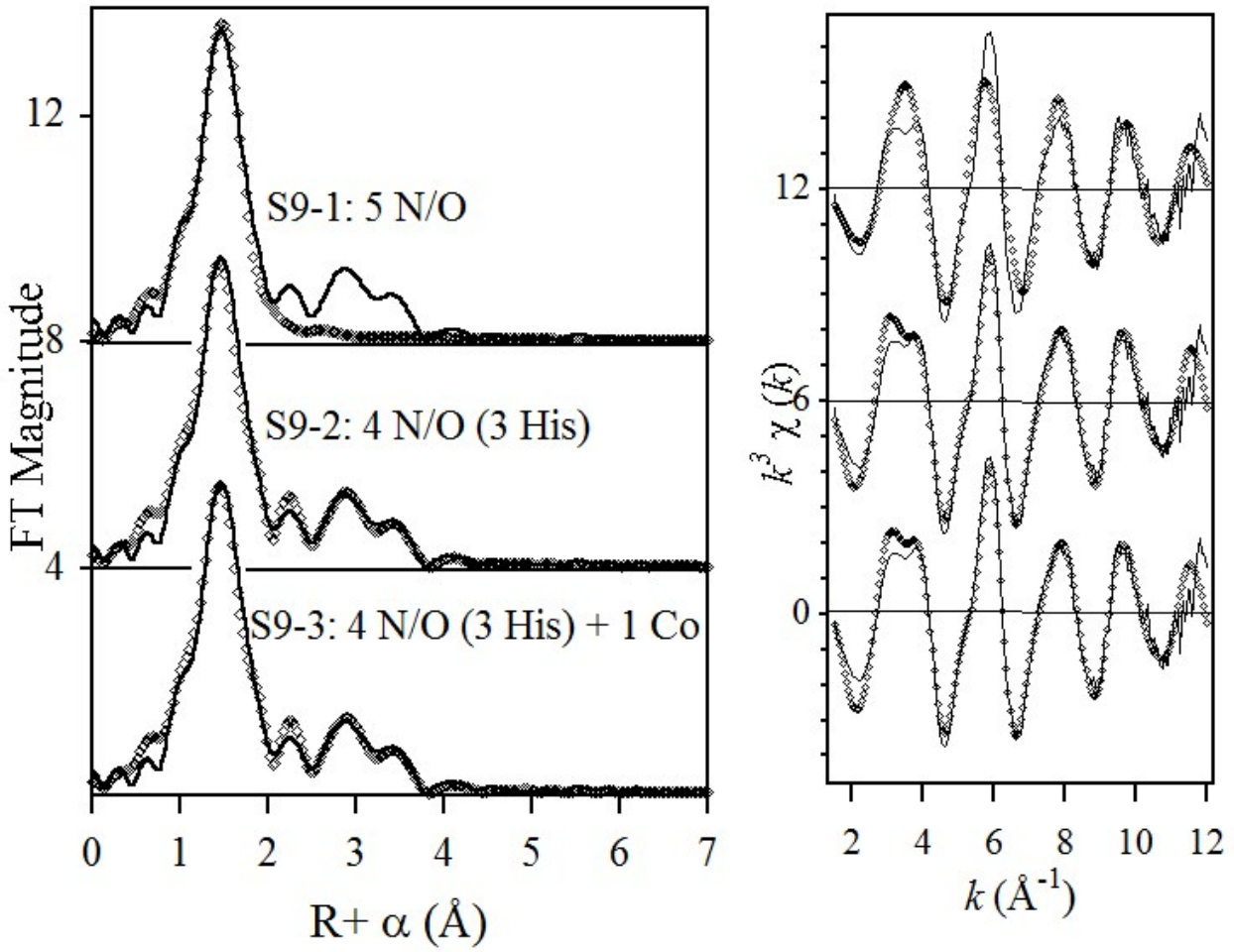

Figure S9. Fourier transforms (A) of k^3-weighted EXAFS (B) for Zn K-edge of the ZnCo-L1/product complex. The solid lines represent the data, the open symbols represent the various fits, corresponding to Table S9.

Table S9. Fitting results for Zn K-edge EXAFS spectra of the ZnCo-L1/product complex.[a]

Fit	Model	Zn-N/O	Zn-His[b]	Zn-Co	R_f[c]	R_u[c]
9-1	4 N/O	2.00 (6.0)			62	255
9-2	4 N/O (3His)	2.00 (5.9)	2.90 (2.5) 3.14 (6.4) 4.01 (20) 4.44 (14)		69	105
9-3	4N/O(2His) + Zn-Co	2.00 (5.9)	2.90 (2.4) 3.14 (1.0) 4.01 (20) 4.45 (14)	3.59 (11)	45	93

[a] Distances (Å) and disorder parameters (in parentheses, σ^2 (10^{-3} Å^2)) shown derive from integer or half-integer coordination number fits to filtered EXAFS data [Δk = 1.5-12 Å^{-1}; ΔR = 0.5 – 2.0 Å (fit 1), 0.3-4.0 Å (fits 2-3)].

[b] Imidazole multiple scattering paths represent combined paths, as described previously (see Materials and Methods).

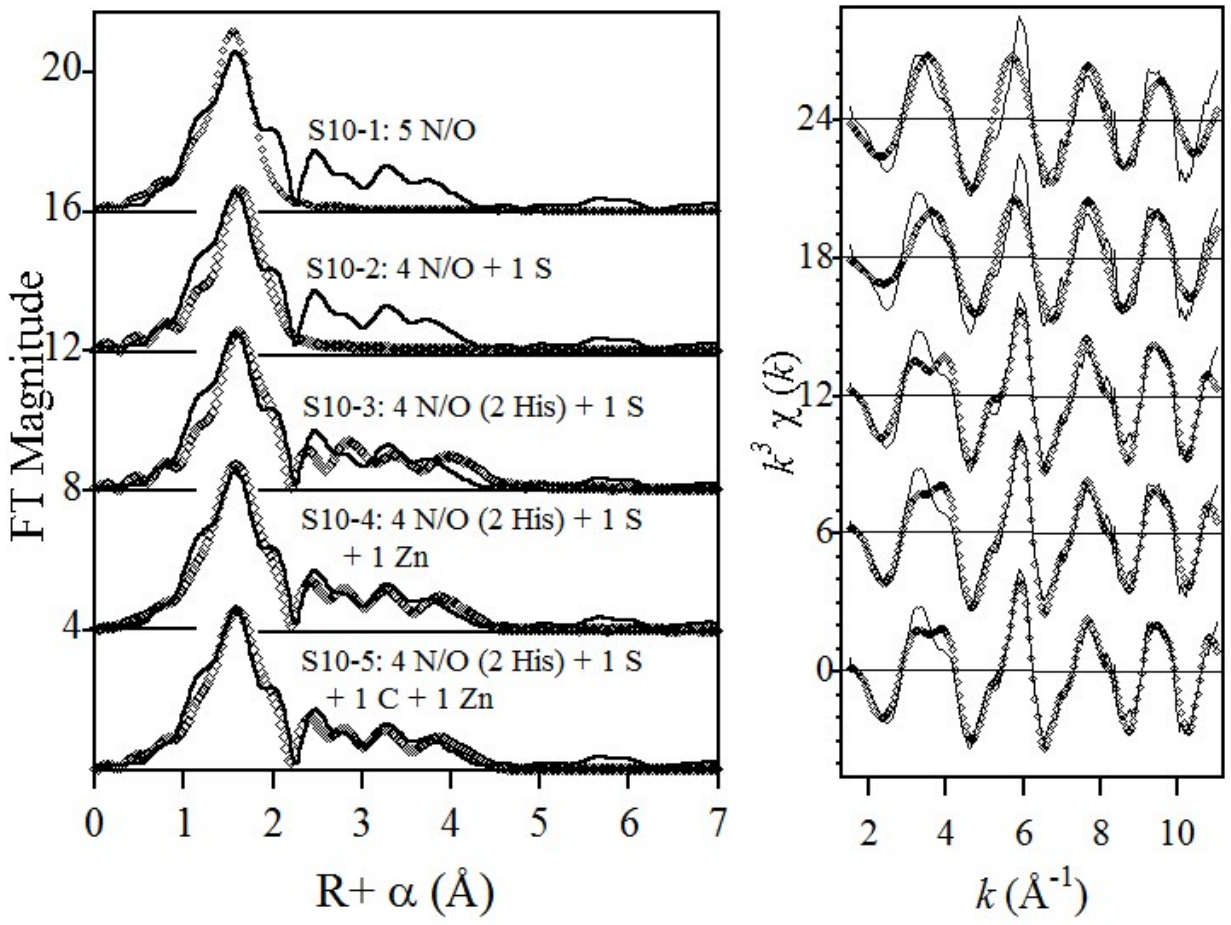

Figure S10. Fourier transforms (A) of k^3-weighted EXAFS (B) for Co K-edge of the ZnCo-L1/product complex. The solid lines represent the data, the open symbols represent the various fits, corresponding to Table S10.

Table S10. Fitting results for Co K-edge EXAFS spectra of the ZnCo-L1/product complex.[a]

Fit	Model	Co-N/O	Co-S	Co-His[b]	Co-Zn	Co-C	R_f[c]	R_u[c]
10-1	5 N/O	2.09 (7.1)					178	291
10-2	4 N/O +1 S	2.08 (6.8)	2.30 (3.1)				110	220
10-3	4 N/O(2His) + 1 S	2.08 (4.8)	2.29 (5.3)	3.00 (1.0) 3.18 (1.9) 4.10 (3.5) 4.58 (6.9)			238	242
10-4	4 N/O(2His) + 1S+ Co-Zn	2.08 (4.8)	2.29 (5.3)	3.00 (1.1) 3.18 (1.9) 4.10 (3.5) 4.58 (7.1)	3.57 (9.3)		174	222
10-5	4N/O(2His) +1S +1C + Co-Zn	2.08 (4.8)	2.29 (5.3)	3.00 (1.0) 3.18 (1.5) 4.10 (3.1) 4.58 (6.9)	3.57 (9.1)	2.47 (0.9)	102	178

[a] Distances (Å) and disorder parameters (in parentheses, σ^2 (10^{-3} Å^2)) shown derive from integer or half-integer coordination number fits to filtered EXAFS data [Δk = 1.5-11 Å^{-1}; ΔR = 0.5 – 2.5 Å (fits 1-2), 0.3-4.0 Å (fits 3-5)].
[b] Imidazole multiple scattering paths represent combined paths, as described previously (see Materials and Methods).