Supplementary Information

EXAFS Deconvolution

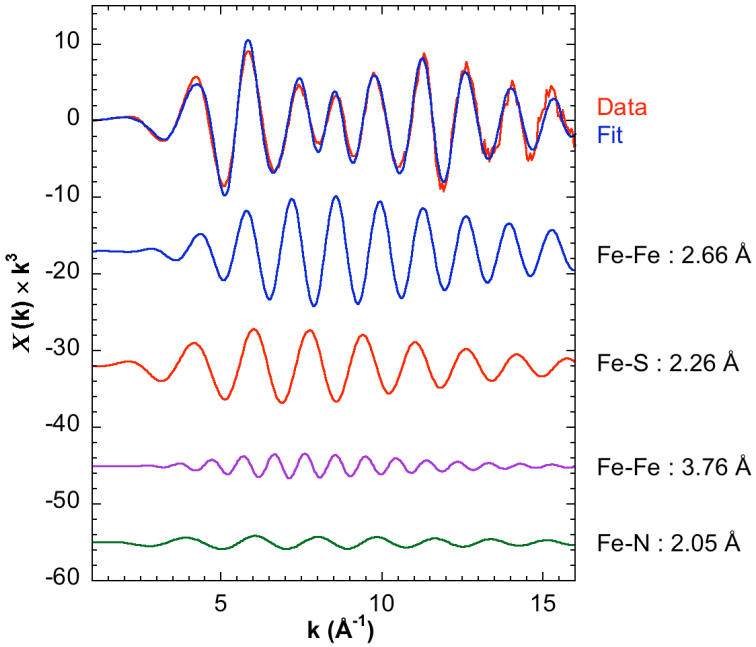

Figure S1. Deconvolution of the EXAFS fit of figure 4a into individual scatterer-backscatterer contributions. The data is the Fe K-edge EXAFS of NifX:NifB-co and the fit is for the 6Fe model. In this fit the interstitial atom X has been modeled as N. The spectra of the individual contributions have been offset.

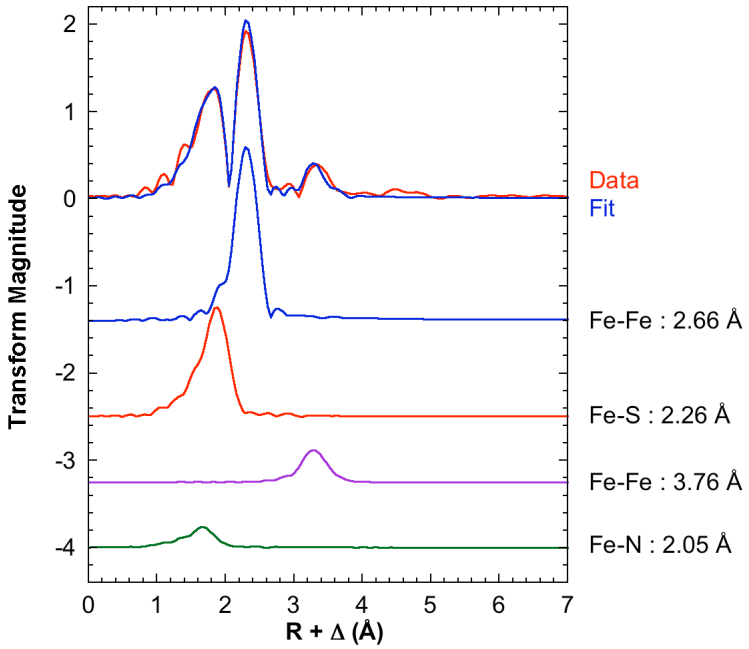

Figure S2. Fourier transforms of the individual EXAFS contributions in figure S1. Note these data are not phase corrected so the peaks are negatively offset from the fitted distance.

The deconvolution of the fit of the NifX:NifB-co EXAFS to the 6Fe model of figure 4 is shown in figure S1 with the corresponding Fourier transforms in figure S2. These data show the individual contributions from each scatterer–backscatterer pair. It is clear that each fitted component, including the interstitial atom X (here modeled as N), has at least 6% intensity. The impact of each component towards the overall fit can readily be assessed from figures S1 and S2.

NRVS Analysis using the 8Fe Model

Figure S3 and Table S1 illustrate the problems fitting the NRVS data to the 8Fe model; figure S3 presents example simulations while Table S1 augments the fitted Urey-Bradley force constants of Table 2 for the 6Fe and 7Fe models with an additional highlighted column giving an example optimized fit using the 8Fe model.

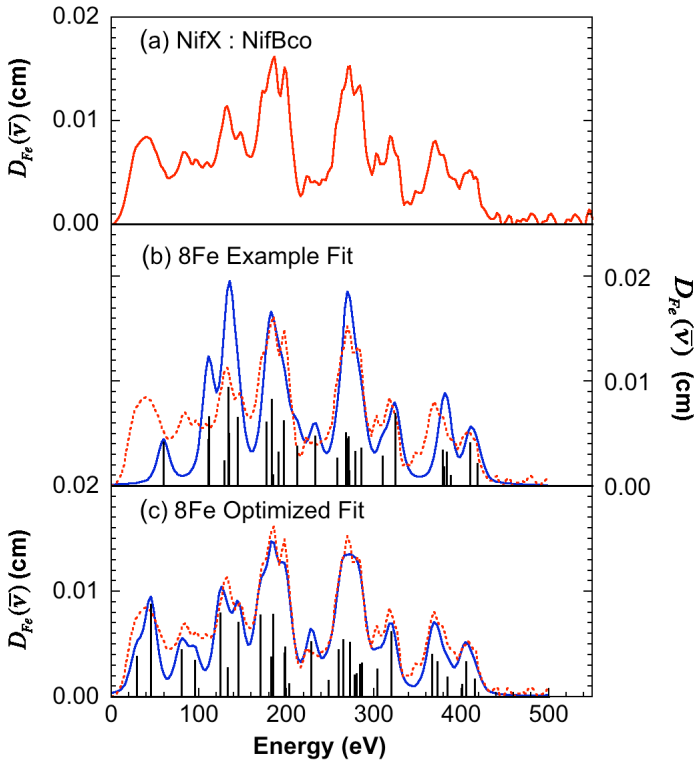

Figure S3. NRVS data for NifX:NifB-co and example Urey-Bradley force field simulations using the 8Fe model. (a) Data from NifX:NifB-co (b) Simulation using the 8Fe model with the (reasonable) force constants from the 7Fe model simulation in Figure 7c and Table 2. (c) Visually good simulation using the 8Fe model but with the unreasonable force constants generated by the optimization. Both simulations are overlaid by the NRVS spectrum as a broken line. The sticks in the simulated spectra show the calculated normal modes. The simulated spectra were obtained by broadening the stick spectrum with 7 cm^{-1} Gaussian lineshapes. Force constants are given in Table S1.

Mode		6Fe model	7Fe model (8Fe Example)	8Fe model (8Fe optimized)
Stretching (mdyn / Å)	Fe-S^U	1.136	1.200	1.199
	Fe-S	1.065	0.960	0.841
	Fe-N	0.265	0.300	0.088
	Fe-Fe	0.225	0.155	0.165
	Fe^T-S		0.960	1.28
Bending (mdyn Å /rad^2)	S-Fe-S	0.130	0.140	-0.651
	S-Fe-S^U	0.455	0.388	0.389
	S-Fe-N	0.179	0.140	0.433
	S^U-Fe-N	0.453	0.400	0.153
	Fe-N-Fe	0.056	0.250	0.067
	Fe-S-Fe	0.536	0.450	-0.057
	Fe-S^U-Fe	0.556	0.450	1.514
	S-Fe^T-S		0.120	0.224
	Fe-S-Fe^T		0.120	0.324
Nonbonding (mdyn / Å)	S—S	0.00	0.00	-0.021
	S—N	0.270	0.250	0.248
	S—S^U	0.047	0.118	0.19
	S^U—N	0.280	0.224	0.137
stretch-stretch stretch-bend (mdyn / rad)	FeS^U/FeS^U	0.114	0.074	-0.02
	FeS/FeS	0.124	0.070	0.075
	FeN/FeN	0.265	0.136	0.159
	Fe^TS/Fe^TS		0.113	0.35

Table S1. NRVS force constants used to simulate the NifB-co spectrum including the 8Fe optimized fit (highlighted). The 7Fe model parameters were used in the 8Fe example simulation in Figure S3b. Optimization using VIBRATZ generates the simulation in Figure S3c with the 8Fe model optimized parameters in the highlighted right-hand column. Note the unreasonable and negative values for the bending modes. S^U = trigonal face bridging S (3 coordinate S atoms in 7Fe or 8Fe models). Fe^T = terminal Fe.

Taking the 7Fe fit as a starting point generates the example simulation in Figure S3b. This simulation is a good starting point as the force constants have reasonable and appropriate

values. However, while the Fe-S stretching region is fit quite well, the bending modes below 160 cm^{-1} are not well reproduced. Unconstrained optimization of the simulation by VIBRATZ generates the fit in Figure S3c with force constants given in the highlighted column in Table S1. This fit is visually very good, however, some of the bending mode force constants are either negative, such as the S-Fe-S bend, or unreasonably large, like the Fe-S^U-Fe bend. Despite extensive efforts, it proved not possible to constrain these force constants to reasonable values and generate a good simulation of the NRVS data with the 8Fe model. Hence we conclude that the 8Fe model in figure 5 does not fit the NRVS data.