

Supporting Information for Park et al.

Figure S1. Fet3p expression levels in cells harvested at different times. A, YPAD; B, MM. Percentage expression levels relative to that at 7 hr are given below the anti-Fet3p blots. Pma1p, a plasma membrane ATPase, was used as loading control, but its expression level was not constant with harvesting time.

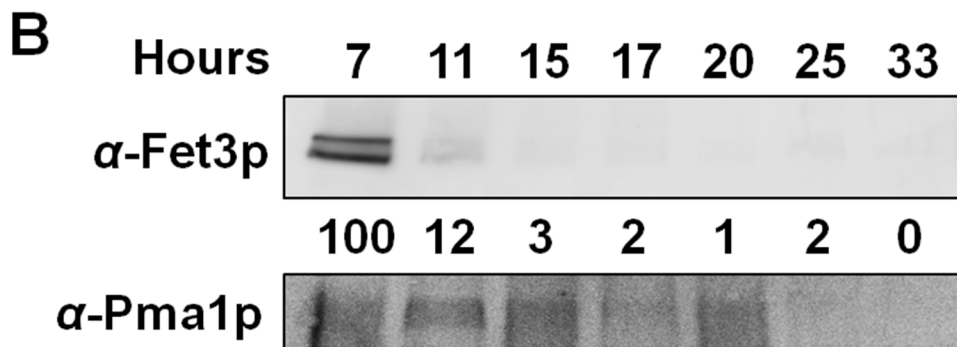
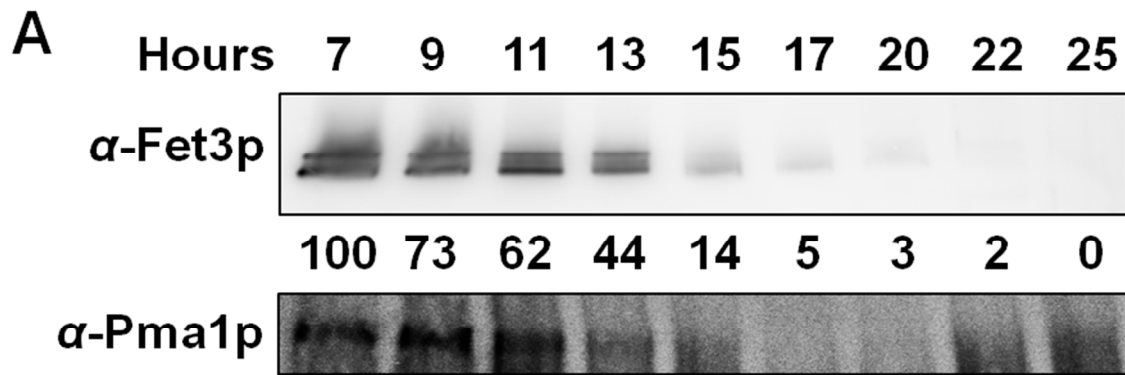


Figure S2. Chronological profile of Zn concentrations in cells grown in batch culture. These were the same samples as were used in Figures 1 and 2. Squares, YPAD; circles, MM. *Inset*, same as YPAD but showing spike between 6 – 20 hr. Plotted Zn concentrations are the average of two independent experiments. Error bars indicate standard deviations.

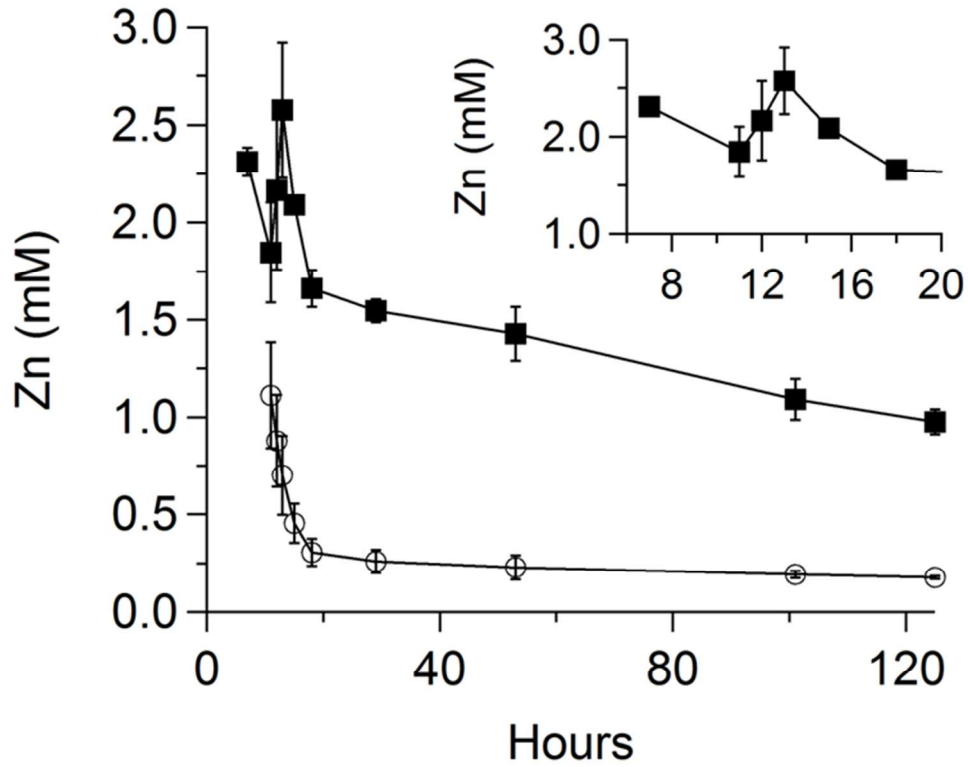


Figure S3. 100 K Mössbauer spectrum of 5-day-grown cells on MM. The field was 0.04 T applied parallel to the radiation.

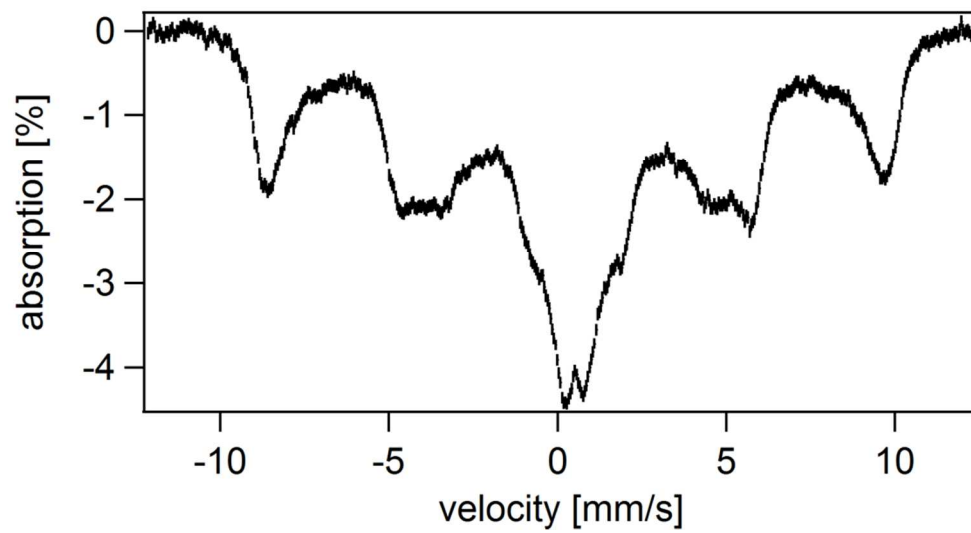


Figure S4. Mössbauer spectrum of mitochondria isolated from 5-day MM-grown cells (OD 2.2) at 0.04 T and 5 K. Blue line indicates Fe^{III} nanoparticle doublet. Red line is a composite simulation that includes contributions from Fe^{III} nanoparticles, the CD and $[\text{Fe}_2\text{S}_2]^{2+}$ clusters.

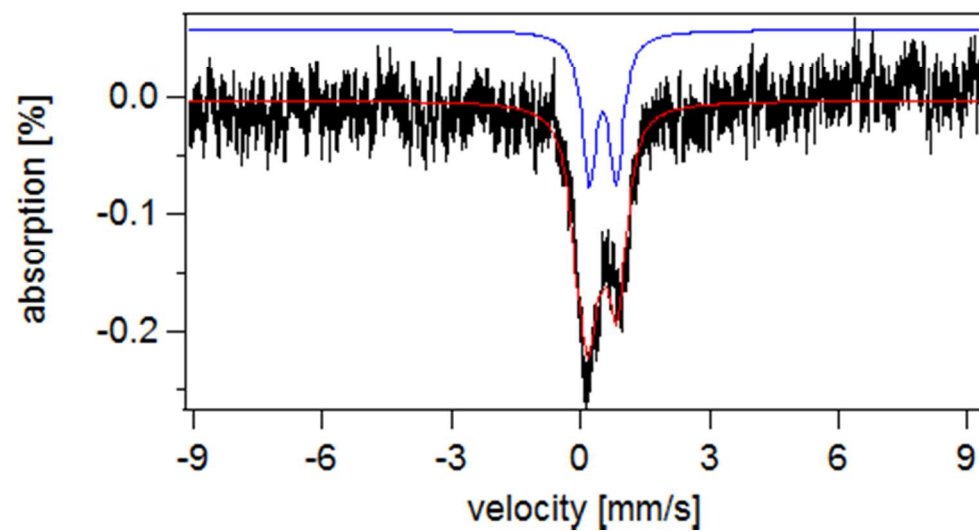


Figure S5. Loading controls for Western blots. A, matched with Figure 7A; B, matched with Figure 7B and 7C.

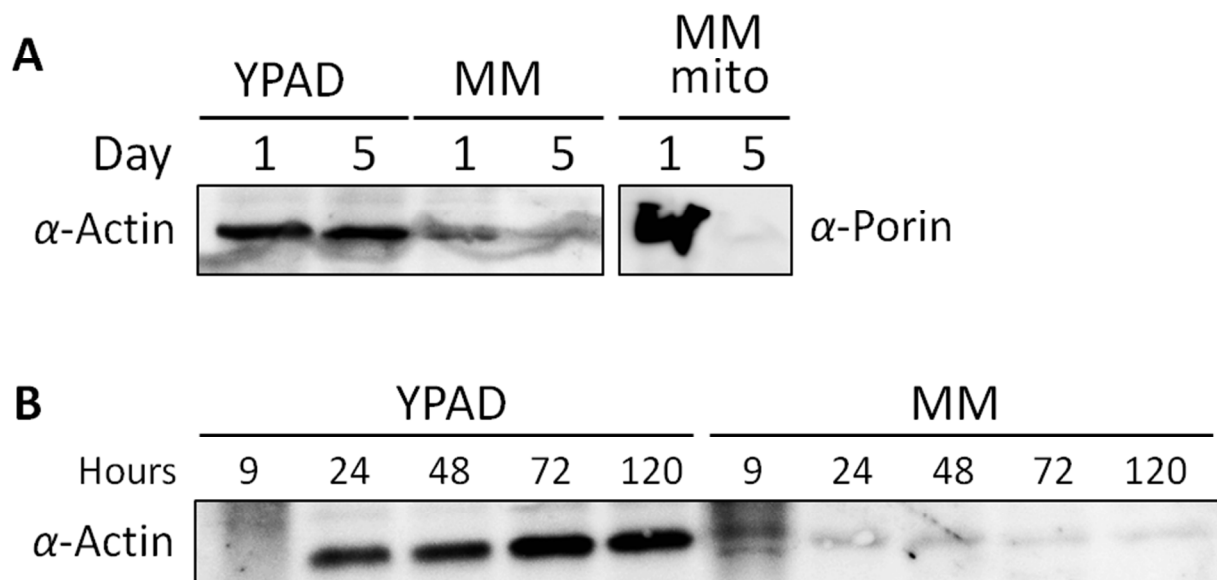


Figure S6. Set of Ordinary Differential Equations that describe version V of the model.

$$\frac{d[Fe_C]}{dt} = R_{HI} \left(\frac{1}{1 + \left(\frac{[Fe_C]}{K_C} \right)^{cs}} \right) + R_{LO} - k_M \left(\frac{1}{1 + \left(\frac{[Fe_M]}{K_M} \right)^{ms}} \right) [Fe_C] - (k_V + \alpha)[Fe_C]$$

$$\frac{d[Fe_V]}{dt} = k_V[Fe_C] - k_P[Fe_V]^{px} - \alpha[Fe_V]$$

$$\frac{d[Fe_M]}{dt} = k_M \left(\frac{1}{1 + \left(\frac{[Fe_M]}{K_M} \right)^{ms}} \right) [Fe_C] - \alpha[Fe_M]$$

$$\frac{d[Fe_P]}{dt} = k_P[Fe_V]^{px} - \alpha[Fe_P]$$

Table S1. Isomer shift (δ , mm/s), quadrupole splitting (ΔE_Q , mm/s), line width (Γ , mm/s) and percentage of Fe species as determined from Mössbauer spectra. For HS Fe^{III}, A_{iso} , D, E/D, η values were also listed. Parameters were averaged from all spectra presented in this paper, but some parameters (δ , ΔE_Q , and Γ) were listed separately if they differed significantly from the averaged numbers. *Percentages of signals with broad range of A values (Fe aggregates) was estimated based on the intensity left after subtracting all assignable signals from original spectra (See Figure S2). Bold-style parameters were fixed when simulated.

Spectrum (Total [Fe])	Non-heme HS Fe ^{III}	Non-heme HS Fe ^{II}	HS Fe ^{II} heme	Central doublet (LS heme Fe ^{II} and [Fe ₄ S ₄] ²⁺)	Fe ^{III} nano- particles (and broad feature*)
A_{iso} (kG)	232 ± 2	-	-	-	-
δ (mm/s)	0.55	1.39 ± 0.05	0.91 ± 0.05	0.45	0.53
ΔE_Q (mm/s)	0.42	3.02 ± 0.10	2.31 ± 0.09	1.15	0.50
Γ (mm/s)	0.34	0.70 ± 0.11	0.32 ± 0.11	0.60	0.46
D (cm ⁻¹)	0.15	-	-	-	-
E/D	0.21	-	-	-	-
η	1.3	-	-	-	-
YPAD/OD 2.1 Fig. 3A (700 μ M)	79%	8%	3%	9%	-
YPAD/OD 3.2 Fig. 3B (1000 μ M)	67%	7%	-	3%	19%
Fig. 3B – 3A Fig. 3C (~ 300 μ M)	37%	-	-	-	63%
YPAD/OD 8.0 Fig. 3D (1500 μ M)	66%	5%	-	9%	19%
Fig. 2D – 2B Fig. 3E (~ 500 μ M)	53%	-	-	29%	18%
MM/OD 0.2 Fig. 4A (430 μ M)	76%	3%	3%	15%	-

MM/OD 1.2 Fig. 4B (620 μM)	73%	8%	-	13%	4%
MM/OD 1.8 Fig. 4C (1500 μM)	66%	12%	-	9%	11%
Fig. 4C – 4B×2 Fig. 4D (~ 300 μM)	32%	39%	-	-	29%
MM/OD 1.8 /5 days Fig. 4E (8100 μM)	72%	-	-	-	12%
MM/OD 1.7 /5 days/6 T Fig. 4F (4900 μM)	72%	-	-	-	Unknown parameters 15%
400Fe/OD 2.2 /5 days Fig. 6A (28.4 mM)	9%	-	-	-	0.54/0.65/0.80 53% (< 30%)
Fig. 6A – nanoparticle and HS Fe ^{III} Fig. 6B (~11 mM)	-	-	-	-	-
Same as Fig. 6A, but at 6 T Fig. 6C (28.4 mM)	9%	-	-	-	Unknown parameters > 90%
Fig. 6C – HS Fe ^{III} Fig. 6D (~25 mM)	-	-	-	-	Unknown parameters > 90%
MM/OD 1.8 /5 days mitochondria Fig. S4 (220 μM)	-	-	-	15% [Fe ₂ S ₂] ²⁺ , 30%	0.54/0.60/0.40 40%