

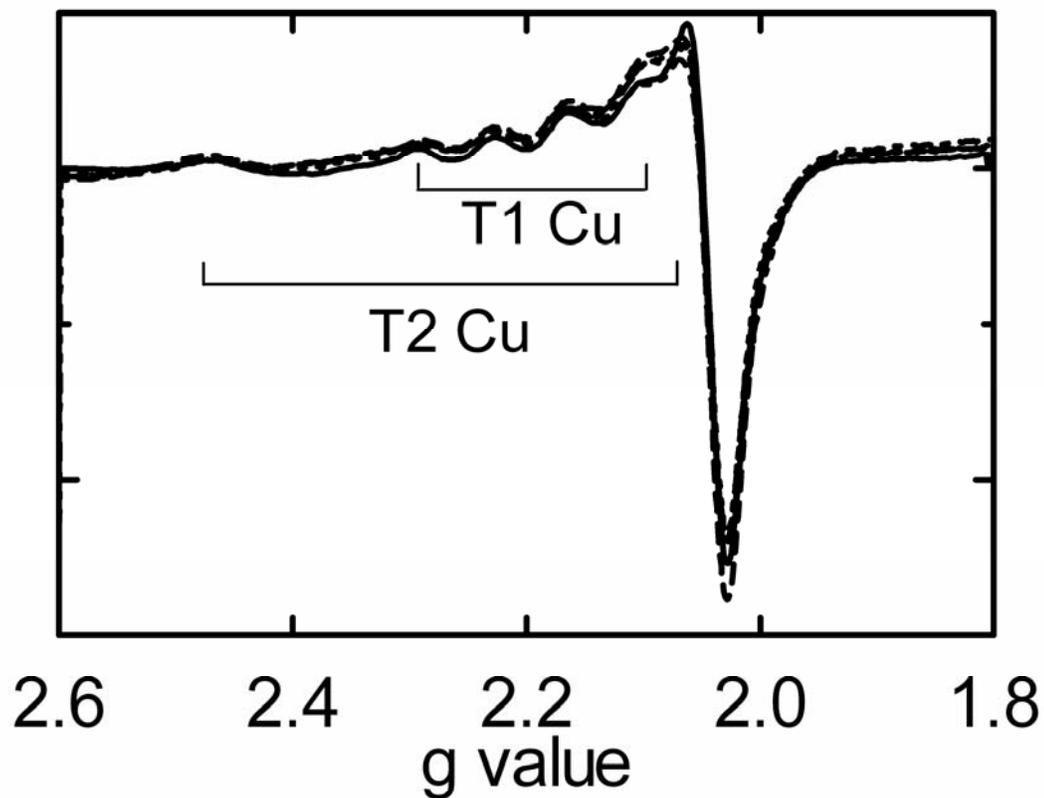
## SUPPORTING INFORMATION

**Table S1. Mutant Fet3p-supported  $^{59}\text{Fe}$ -Uptake**

Fet3p species	$\text{Fe}^{59}$ Uptake (% of WT)	
	0 mM Citrate	20 mM Citrate
WT	100	100
D283A	100	3
E185A	0	0
D409A	100	7
E185A/D409A	0	0

Iron uptake phenotypes for each mutant were examined for citrate sensitivity relative to yeast expressing the wild type ferroxidase by monitoring cellular  $^{59}\text{Fe}$  incorporation. All of the mutant enzymes were present at the yeast plasma membrane as viewed by the positive localization of a partner Ftr1:GFP fusion.

**FIGURE S1**



**FIGURE S1. EPR spectra of Fet3 wild-type and mutant proteins.** EPR spectra at 77 K for Fet3p WT (—), E185A (---), D283A (···), D409A (-·-), E185A/D409A (· · -). The  $^{63/65}\text{Cu}$  splitting in  $g_{\parallel}$  is indicated for the T1 and T2  $\text{Cu}^{\text{II}}$  sites. EPR spectra were recorded under the following conditions: 9.41 GHz microwave frequency, 10 mW microwave power, 20 G modulation amplitude, 100 kHz modulation frequency, 327 ms time constant. At least 6 scans were averaged to obtain the spectra presented.