

Table S1. Determination of intracellular iron and manganese content of *B. japonicum* wild type, *fur*⁺, and *mur* strains. Data are expressed as average nmol metal per mg protein \pm standard deviation of triplicate samples.

Medium (μ M)		Cellular Iron Content		
		<i>Wild type</i>	<i>fur</i> ⁺	<i>mur</i>
Mn	Fe	nmol Fe/ mg protein	nmol Fe/ mg protein	nmol Fe/ mg protein
0	0	0.96 \pm 0.01	1.05 \pm 0.04	1.13 \pm 0.06
0	20	46 \pm 3	46 \pm 4	42 \pm 0.1
0	100	307 \pm 5	306 \pm 10	306 \pm 12
20	0	0.96 \pm 0.02	0.93 \pm 0.01	0.96 \pm 0.02
20	20	48 \pm 3	48 \pm 3	46 \pm 1
20	100	307 \pm 8	308 \pm 9	308 \pm 12
100	0	0.95 \pm 0.00	0.98 \pm 0.02	0.94 \pm 0.02
100	20	47 \pm 1	48 \pm 1	47 \pm 1
100	100	307 \pm 7	307 \pm 6	302 \pm 5

Medium (μ M)		Cellular Manganese Content		
		<i>Wild type</i>	<i>fur</i> ⁺	<i>mur</i>
Mn	Fe	nmol Mn/ mg protein	nmol Mn/ mg protein	nmol Mn/ mg protein
0	0	0.92 \pm 0.01	0.87 \pm 0.04	0.90 \pm 0.04
0	20	1.15 \pm 0.01	1.37 \pm 0.01	1.36 \pm 0.02
0	100	1.10 \pm 0.04	1.20 \pm 0.07	1.27 \pm 0.08
20	0	8.6 \pm 0.3	9.6 \pm 0.1	9.4 \pm 0.2
20	20	9.6 \pm 0.3	9.3 \pm 0.3	9.8 \pm 0.1
20	100	9.8 \pm 0.2	9.5 \pm 0.2	9.6 \pm 0.1
100	0	13.2 \pm 0.3	12.9 \pm 0.4	13.0 \pm 0.1
100	20	13.6 \pm 0.1	13.4 \pm 0.3	13.2 \pm 0.2
100	100	13.3 \pm 0.3	12.8 \pm 0.1	14.0 \pm 0.1

Table S2. Determination of intracellular iron and manganese content of *E. coli* wild type, *mur*⁺, and *fur* cells. Data are expressed as average nmol metal per mg protein \pm standard deviation of triplicate samples.

Medium (μ M)		Cellular Iron Content		
		<i>Wild type</i>	<i>mur</i> ⁺	<i>fur</i>
Mn	Fe	nmol Fe/ mg protein	nmol Fe/ mg protein	nmol Fe/ mg protein
0	0	2.02 \pm 0.02	2.22 \pm 0.06	2.02 \pm 0.08
0	20	214 \pm 4	216 \pm 7	238 \pm 1
0	100	1054 \pm 0	1150 \pm 9	1013 \pm 7
20	0	2.18 \pm 0.09	2.27 \pm 0.11	2.34 \pm 0.05
20	20	228 \pm 3	225 \pm 1	233 \pm 4
20	100	1268 \pm 52	1258 \pm 42	1226 \pm 42
100	0	2.09 \pm 0.01	2.07 \pm 0.05	2.09 \pm 0.14
100	20	274 \pm 2	267 \pm 5	272 \pm 12
100	100	1259 \pm 45	1246 \pm 4	1247 \pm 34

Medium (μ M)		Cellular Manganese Content		
		<i>Wild type</i>	<i>mur</i> ⁺	<i>fur</i>
Mn	Fe	nmol Mn/ mg protein	nmol Mn/ mg protein	nmol Mn/ mg protein
0	0	0.14 \pm 0.01	0.15 \pm 0.00	0.09 \pm 0.02
0	20	0.39 \pm 0.00	0.45 \pm 0.00	0.19 \pm 0.00
0	100	0.96 \pm 0.04	0.92 \pm 0.02	0.62 \pm 0.03
20	0	4.5 \pm 0.2	7.7 \pm 0.3	2.6 \pm 0.2
20	20	31 \pm 3	27 \pm 2	22 \pm 2
20	100	82 \pm 2	87 \pm 2	80 \pm 8
100	0	69 \pm 3	74 \pm 1	20 \pm 1
100	20	237 \pm 5	241 \pm 13	165 \pm 2
100	100	276 \pm 6	263 \pm 1	181 \pm 6

Fig S1. Metal binding residues of Mur and Fur are conserved.

Amino acid sequences and alignment of *E. coli* Fur (EcFur), *Magnetospirillum. gryphiswaldense* Fur (MgFur), and *B. japonicum* Mur (BjMur). Metal binding site 1 (bold) and metal binding site 2 (underlined) are shown as determined by crystal structure of *M. gryphiswaldense* Fur.

Fig S2. Western blot analysis of Mur or Fur expression in *B. japonicum* and *E. coli*.

15 µg of *B. japonicum* whole cell lysates or 5 µg of *E. coli* whole cell lysates were loaded per lane of a 15% SDS-PAGE gel and analyzed using either anti-Mur antibodies (Mur) or anti-Fur antibodies (Fur). (A) Metal dependent expression of Fur in Wt *E. coli*, Mur in ecMur, Fur in bjFur, or Mur in Wt *B. japonicum*. Cells were grown in the presence (+) or absence (-) of 20 µM MnCl₂ and FeCl₃. (B) Fur and Mur expression in Wt *E. coli*, ecMur (*mur*⁺), JW0669 (*fur*⁻) Wt *B. japonicum*, bjFur (*fur*⁺), and GEM4 (*mur*⁻) strains.

Fig S3. Metal dependent gene expression of *mnoP* and *fiu* in *B. japonicum* and *E. coli* cells.

(A) Analysis of *mnoP* mRNA by qPCR in *B. japonicum* wild type (Wt) or *mur* cells grown in the presence or absence of 20 µM MnCl₂ or 20 µM FeCl₃. (B) Analysis of *fiu* mRNA by qPCR in *B. japonicum* wild type (Wt) or *mur* cells grown as described above. (C) Analysis of *fiu* mRNA by qPCR in *E. coli* parent strain (Wt) or *fur* cells grown as described above. (D) Analysis of *mnoP* mRNA by qPCR in *E. coli* strain expressing Mur (*mur*⁺) or *fur* cells grown as describe above. The data are expressed as the relative starting quantity (SQ) of *mntH* mRNA normalized to the housekeeping gene *gapA*, and are presented as the average of triplicate samples with the error bars representing the standard deviation.

EcFur	-----MTDNNTALKKAGLKVTLPRLKILEVLQEPDNHHVSAEDLYKRLIDMGEE	49
MgFur	-----MVSRIEQRCIDKGMKMTDQRRVIAQVLSDS-ADHPDVEEVYRRATAKDPR	49
BjMur	MTALKPSSASKASGIEARCAATGMRMTEQRRVIARVLAEA-VDHPDVEELYRRCVAVDDK	59
EcFur	IGLATVYRVLNQFDDAGIVTRHNFEGGKSVFELTQQHH <u>HD</u> HLICLDCGKVI <u>EF</u> SDDSI <u>EA</u>	109
MgFur	ISIATVYRTVRLFEEESILERHDFGDGRARY <u>EE</u> APSE <u>HD</u> HLIDVNSARVI <u>EF</u> TSPET <u>EA</u>	109
BjMur	ISISTVYRTVKLFEDAGIERHDFREGRARY <u>ET</u> MRDS <u>HD</u> HLINLRDGKVI <u>EF</u> TSEET <u>EK</u>	119
EcFur	RQREIAAKHGIRLTN <u>HS</u> LYLYGHCAEGDCREDEHAHEGK	148
MgFur	LQREIARKHGFRLV <u>GH</u> RLELYGVPLTSGGDSDDK-----	143
BjMur	LQAEIARKLGYKLVD <u>H</u> RLELYCVPLDDDKPTS-----	151

Figure S1

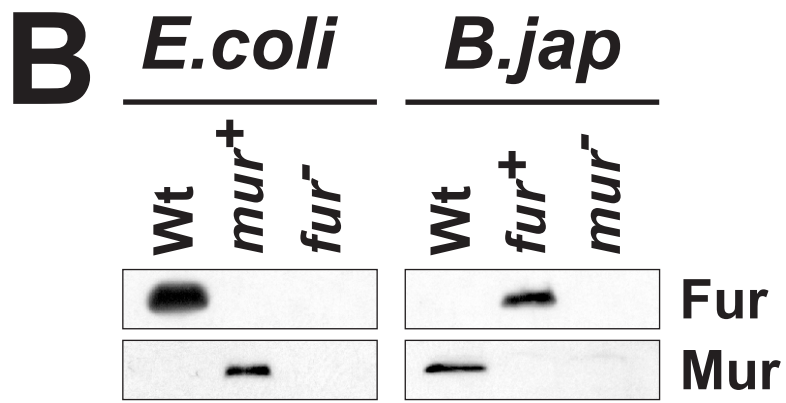
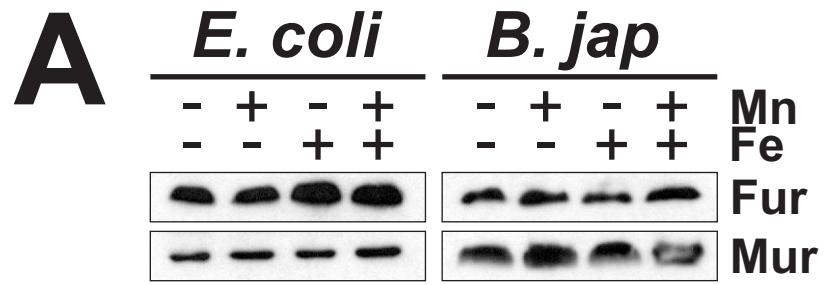
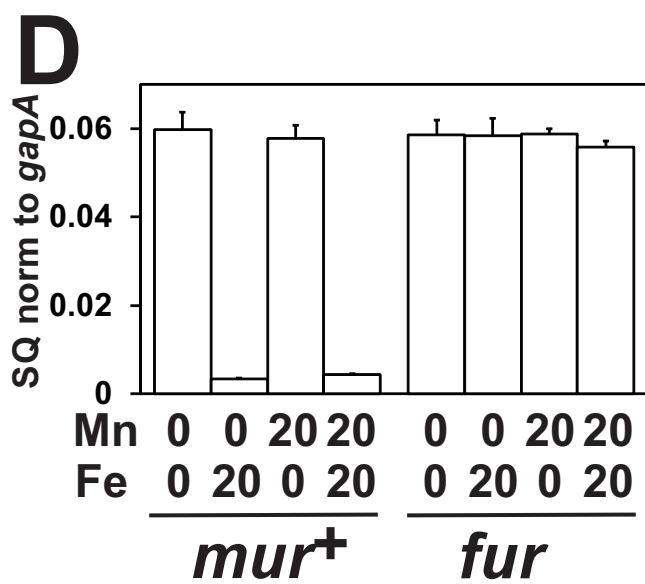
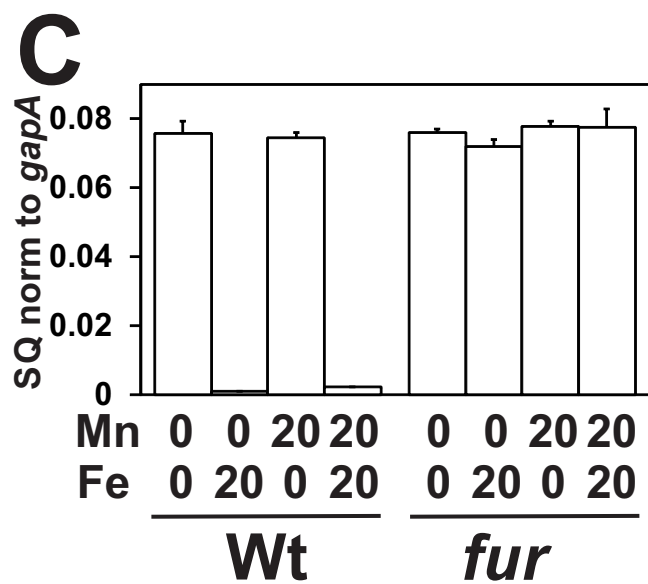
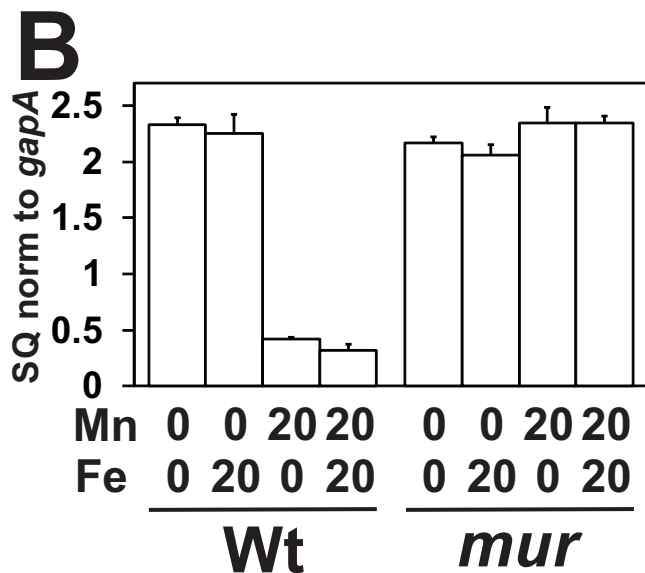
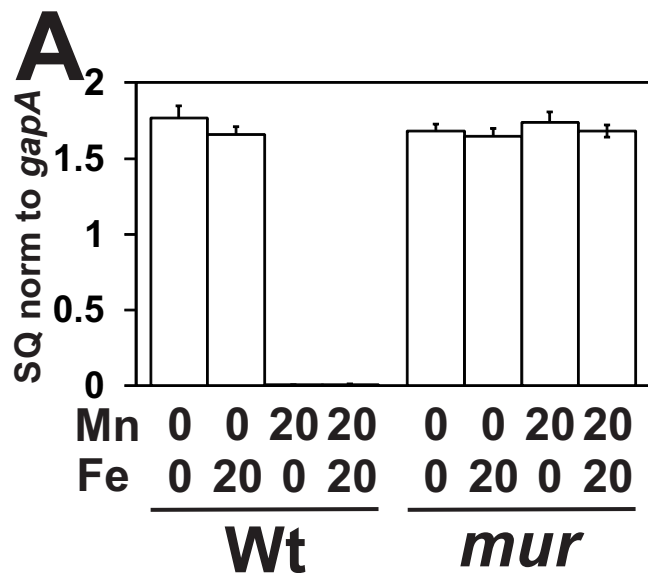


Figure S2



FigureS3