Supplementary Material:

Dissecting the metal-selectivity of MerR monovalent metal ion sensors in *Salmonella*

María M. Ibáñez, Sebastián Cerminati, Susana K. Checa, and Fernando C. Soncini#

Instituto de Biología Molecular y Celular de Rosario, Departamento de Microbiología, Facultad de Ciencias Bioquímicas y Farmacéuticas, Universidad Nacional de Rosario, Consejo Nacional de Investigaciones Científicas y Técnicas. Ocampo y Esmeralda, 2000-Rosario, Argentina.

Table S1. Bacterial strains and plasmids used in this study

Strain	Relevant genotype	Reference or source
PB4406	LB5010 Δ*golS golB*::*kan*	Laboratory stock
PB5295	LB5010 Δ*cueR*::*kan*	Laboratory stock
14028s	wild type	ATCC
PB5257	Δ*golS*	(1)
PB7116	*golS golB*::*lacZY*$^{+}$*-kan*	This study
PB5040	*golS*$_{L}$ *golB*::*lacZY*$^{+}$*-kan*	This study
PB8261	*golS*$_{sL}$ *golB*::*lacZY*$^{+}$*-kan*	This study
PB7173	*golS*$_{C111S}$ *golB*::*lacZY*$^{+}$*-kan*	This study
PB7169	*golS*$_{A113P}$ *golB*::*lacZY*$^{+}$*-kan*	This study
PB7680	*golS*$_{A116D}$ *golB*::*lacZY*$^{+}$*-kan*	This study
PB8538	*golS*$_{L117S}$ *golB*::*lacZY*$^{+}$*-kan*	This study
PB7171	*golS*$_{P118A}$ *golB*::*lacZY*$^{+}$*-kan*	This study
PB7967	*golS golB*::*lacZY*$^{+}$*-kan* Δ*cueR-copA*::*cat*	This study
PB8274	*golS*$_{L}$ *golB*::*lacZY*$^{+}$*-kan* Δ*cueR-copA*::*cat*	This study
PB8275	*golS*$_{sL}$ *golB*::*lacZY*$^{+}$*-kan* Δ*cueR-copA*::*cat*	This study
PB7968	*golS*$_{A113P}$ *golB*::*lacZY*$^{+}$*-kan* Δ*cueR-copA*::*cat*	This study
PB7979	*golS*$_{A116D}$ *golB*::*lacZY*$^{+}$*-kan* Δ*cueR-copA*::*cat*	This study
PB9707	*golS*$_{L117S}$ *golB*::*lacZY*$^{+}$*-kan* Δ*cueR-copA*::*cat*	This study
PB7969	*golS*$_{P118A}$ *golB*::*lacZY*$^{+}$*-kan* Δ*cueR-copA*::*cat*	This study
PB7608	*golS*$_{A113S}$ *golB*::*lacZY*$^{+}$*-kan*	This study
PB7610	*golS*$_{A113H}$ *golB*::*lacZY*$^{+}$*-kan*	This study
PB10441	*golS*$_{A113T}$ *golB*::*lacZY*$^{+}$*-kan*	This study
PB7980	*golS*$_{A113S}$ *golB*::*lacZY*$^{+}$*-kan* Δ*cueR-copA*::*cat*	This study
PB7981	*golS*$_{A113H}$ *golB*::*lacZY*$^{+}$*-kan* Δ*cueR-copA*::*cat*	This study

PB10448	*golS*$_{A113T}$ *golB*::*lacZY*$^{+}$*-kan* Δ*cueR-copA*::*cat*	This study
PB8534	*golS*$_{A113P-A116D}$ *golB*::*lacZY*$^{+}$*-kan*	This study
PB8536	*golS*$_{A113P-L117S}$ *golB*::*lacZY*$^{+}$*-kan*	This study
PB7175	*golS*$_{A113P-P118A}$ *golB*::*lacZY*$^{+}$*-kan*	This study
PB7867	*golS*$_{A116D-L117S}$ *golB*::*lacZY*$^{+}$*-kan*	This study
PB7675	*golS*$_{A116D-L117S-P118A}$ *golB*::*lacZY*$^{+}$*-kan*	This study
PB8257	*golS*$_{A113P-A116D-P118A}$ *golB*::*lacZY*$^{+}$*-kan*	This study
PB8259	*golS*$_{A113P-L117S-P118A}$ *golB*::*lacZY*$^{+}$*-kan*	This study
PB7673	*golS*$_{C111S-A113P}$ *golB*::*lacZY*$^{+}$*-kan*	This study
PB7606	*golS*$_{C111S-P118A}$ *golB*::*lacZY*$^{+}$*-kan*	This study
PB7869	*golS*$_{C111S-A113P-P118A}$ *golB*::*lacZY*$^{+}$*-kan*	This study
PB9388	*cueR-cat*	This study
PB7325	*cueR*$_{L}$*-cat*	This study
PB10330	*cueR*$_{P113A}$*-cat*	This study
PB10851	*cueR*$_{A118P}$*-cat*	This study
PB10332	*cueR*$_{P113A-A118P}$*-cat*	This study
PB10370	*cueR*$_{L}$*-cat golS*::*kan*	This study
PB10371	*cueR*$_{P113A}$*-cat golS*::*kan*	This study
PB10852	*cueR*$_{A118P}$*-cat golS*::*kan*	This study
PB10373	*cueR*$_{P113A-A118P}$*-cat golS*::*kan*	This study
PB5292	*copA-lacZY-cat*	(2)
PB7414	*cueR*$_{L}$ *copA-lacZY-cat*	This study

Plasmid

pKD3	ori$_{\gamma}$ Amp^{R} FRT Cm^{R} FRT	(3)
pCP20	λ-cI857-ts P$_{r}$-*flp*, Amp^{R} Cm^{R}	(3)
pKD46	oriR$_{pSC101}$ts P$_{araB}$*exo-bet-gam* Amp^{R}	(3)
pKG136	ori$_{R6K}$ FRT lacZY^{+} t$_{his}$ Km^{R}	J. M. Slauch
pUH21-2*lacI*^{q}	ori$_{pMB1}$ Ap^{r} *lacI*^{q}	(4)
pMC1871	pBR322, Tc^{R}, *lacI*	Amersham Pharmacia
pPB1225 (p*PcopA*)	pMC1871::P*copA*	(2)
pPB1205 (pGolS)	pUH::*golS*	(1)
pPB1353 (pGolS$_{P118A}$)	pUH:: *golS*$_{P118A}$	This study

Table S2. Oligonucleotides used in this study.

Primer name	Sequence (5`-3`)	5'- restriction site	Purpose
golS-F	ATGAGGAGGAGCGTCATGAACATCG	-	γ Red insertion of *golS* mutants
RvP1-*golB*-R	GTGAACTCCTTTTGTGTGGGAACTG	-	γ Red insertion of *golS* mutants
golB-P1-F	CACTGGCAAGGTCCAGACTGGCAACAGTTCCCACACAAAAGGAGTTCACT GTGTAGGCTGGAGCTGCTTCG	-	γ Red insertion of *golS* mutants
golB-P2-R	TGGCTAGCGTATCGCGACCGGCCTGTCGCCAGACCGATCGCCATTGACGAC ATATGAATATCCTCCTTA	-	γ Red insertion of *golS* mutants
$golS_L$-R	GGTTGTCCAAGCGTATGCAGAATGGGGCAATCGGCGCTGTCATCGCCAGG GCAGGCATTCGCCAGCGCTTTGAGGG	-	Mutagenesis of *golS*
gol_{SL}-*R*	GCAATCTGCGCTGTCGTCGCCGGGGCAGCA	-	Mutagenesis of *golS*
$golS_{C111S}$-R	CGCCGGCGCAGGAGTGAATCAGCG	-	Mutagenesis of *golS*
$golS_{A113P}$-R	CGCCGGGGCAGCAGTGAATCAG	-	Mutagenesis of *golS*
$golS_{A116D}$-R	GGCAATCTGGCAGGTCGTCGCCGGCGCAGCA	-	Mutagenesis of *golS*
$golS_{L117S}$-R	GCAATCTGGGCTCGCGTCGCCCGCGCAGCA	-	Mutagenesis of *golS*
$golS_{P118A}$-R	GCAATCTGCCAGCGCGTCGC	-	Mutagenesis of *golS*
$golS_{A113P\text{-}A116D}$-R	GCAATCTGGCAGGTCGTCGCCGGGGCAGCA	-	Mutagenesis of *golS*
$golS_{A113P\text{-}L117S}$-R	GCAATCTGGGCTCGCGTCGCCGGGGCAGCA	-	Mutagenesis of *golS*
$golS_{A113P\text{-}P118A}$-R	GCAATCTGCCAGCGCGTCGCCGGGGCAGCAGTG	-	Mutagenesis of *golS*
$golS_{A113P\text{-}A116D\text{-}P118A}$-R	GCAATCTGCCAGGTCGTCGCCGGGGCAGCA	-	Mutagenesis of *golS*
$golS_{A113P\text{-}L117S\text{-}P118A}$-R	GCAATCTGCGCTCGCGTCGCCGGGGCAGCA	-	Mutagenesis of *golS*
$golS_{A116D\text{-}L117S}$-R	GAATGGGGCAATCTGGGCTGTCGTCGCCGGCGC	-	Mutagenesis of *golS*
$golS_{A116D\text{-}L117S\text{-}P118A}$-R	GCAGAATGGGGCAATCGGCGCTGTCGTCGCCGGCGCAGCAGT	-	Mutagenesis of *golS*
$golS_{A113S}$-R	GCAGCGCGTCGCCACTGCAGCAGTGAAT	-	Mutagenesis of *golS*
$golS_{A113H}$-R	GCAGCGCGTCGCCGTGGCAGCAGTGAAT	-	Mutagenesis of *golS*
$golS_{A113T}$-R	CAATCTGGCAGCGCGTCGCCCGTGCAGCA	-	Mutagenesis of *golS*
$golS_{C111S\text{-}A113P}$-R	GCAGCGCGTCGCCGGGGCAACTGTGAATCA	-	Mutagenesis of *golS*
$golS_{C111S\text{-}P118A}$-R	GCAATCTGCCAGCGCGTCGCCGGCGCAACTGTGAAT	-	Mutagenesis of *golS*
$golS_{C111S\text{-}A113P\text{-}P118A}$-R	GCAATCTGCCAGCGCGTCGCCGGGGCAGCTGTGAAT	-	Mutagenesis of *golS*
golS-ORF-F	GAGGATCCATATGAACATCGGTAAAGC	*Bam*HI	Cloned *golS*
golS-ORF-R	ACCCAAGCTTACAGACGCTTTGCCAG	*Hind*III	Cloned *golS*
cueR-F	GAGGATCCATATGAATATTAGCG	*BamH*I	γ Red insertion of *cueR* mutants
RvP1-*cueR*-R	CCGCCGGTTTATGCTTGATGCCGCGTTAGT	-	γ Red insertion of *cueR* mutants
cueR-P1-F	CCGTCCGTTCACTAACGCGGCATCAAGCATAAACCGGCGGCTGTGTAGGCT GGAGCTGCTTCG	-	γ Red insertion of *cueR* mutants
cueR-P2-R	GCCTTAACGTTGCTGGCGGCCTGGCTTTGGTGGCGCTGGCCATATGAATAT CCTCCTTA	-	γ Red insertion of *cueR* mutants
$cueR_L$-R	CAGCCGGAAAGATTATCAATAATCGGGCAGTCTGGCAGCGCGTCGCCGGC GCAGCAGTGAATCAGCGCCAACAGTT	-	Mutagenesis of *cueR*
$cueR_{P113A}$-R	GCTGTCATCGCCGGCACAACTCTCCGCCA	-	Mutagenesis of *cueR*
$cueR_{A118P}$-R	CAATAATCGGGCAGTCGGGGCTGTCATCGCCGGGA	-	Mutagenesis of *cueR*
$cueR_{P113A\text{-}A118P}$-R	CGGGCAGTCGGGGCTGTCATCGCCGGCACAACTCTCCGCCA	-	Mutagenesis of *cueR*
cueR-copA-P1-F	CAACGTGGCTTTTGCGCCTTGTGATGACAGCAGCCGGAAACATATGAATAT CCTCCTTA	-	γ Red deletion of *cueR-copA*
cueR-copA-P2-R	AACGCTATATCAGGCAGCATCAGCTGCCTGATATATTATTTGTAGGCTGGA GCTGCTTCG	-	γ Red deletion of *cueR-copA*
PgolB-F	GACCCGGGACGTATCCAGAACATGC	*Xma*I	Electrophoretic gel mobility shift assays
PgolB-R	TCCCCCGGGGCAGCCGCCGCAGGTC	*Xma*I	Electrophoretic gel mobility shift assays

Supplementary References

1. **Checa SK, Espariz M, Pérez Audero ME, Botta PE, Spinelli SV, Soncini FC.** 2007. Bacterial sensing of and resistance to gold salts. Mol Microbiol **63:**1307-1318.
2. **Pérez Audero ME, Podoroska BM, Ibáñez MM, Cauerhff A, Checa SK, Soncini FC.** 2010. Target transcription binding sites differentiate two groups of MerR-monovalent metal ion sensors. Mol Microbiol **78:**853-865.
3. **Datsenko KA, Wanner BL.** 2000. One-step inactivation of chromosomal genes in *Escherichia coli* K-12 using PCR products. Proc Natl Acad Sci U S A **97:**6640-6645.
4. **Soncini FC, García Véscovi E, Groisman EA.** 1995. Transcriptional autoregulation of the *Salmonella typhimurium phoPQ* operon. J Bacteriol **177:**4364-4371.