

Supplementary Materials

S1 Table. Primers used in this study.

Targeted gene	Replicon	Primer	Sequence (5'- 3')
<i>merTPAGB₁</i>	pTP6	pTP6mer_Fw	CACTATAGGGCGAATTACGGTCTT TTCTGACGTTGG
		pTP6mer_Rv	AAGGGAACAAAAGCTGTTCTAGC CACTTTCGGTTCG
MCS	pBBR1	pBBR1MCS2_GA_Fw	CAGCTTTTGTTCCTTTAGTGAG
<i>cadA</i>	Chromosome	pBBR1MCS2_GA_Rv	AATTCGCCCTATAGTGAGTCGTAT
		CadA_Fw_CR1	GCACACTCACACATGGCAAG
<i>zniA</i>	Chromid	CadA_Rv_CR1	CTGAGACACAGGATGGTCCG
		ZniA_Fw_CR2	GCGCAGATCACCCAGGAATA
<i>ncca</i>	pMOL30	ZniA_Rv_CR2	ACTATGACCGTTCGACGCTG
		NccA_Fw_P30	AAGAGTGAATGGCCGGATGG
<i>cnrA</i>	pMOL28	NccA_Rv_P30	TCTCAATGGGTTGGGTGACG
		CnrA_Fw_P28	AAATGTCCCGATCACAGTCGG
<i>merG</i>	pTP6	CnrA_Rv_P28	GCCCACCACCGTTTCATTG
		MerG_Fw_PT6	TACAAATGCGGTATGGGCGT
		MerG_Rv_PT6	CGAGGTTGAACTGTGCATCG

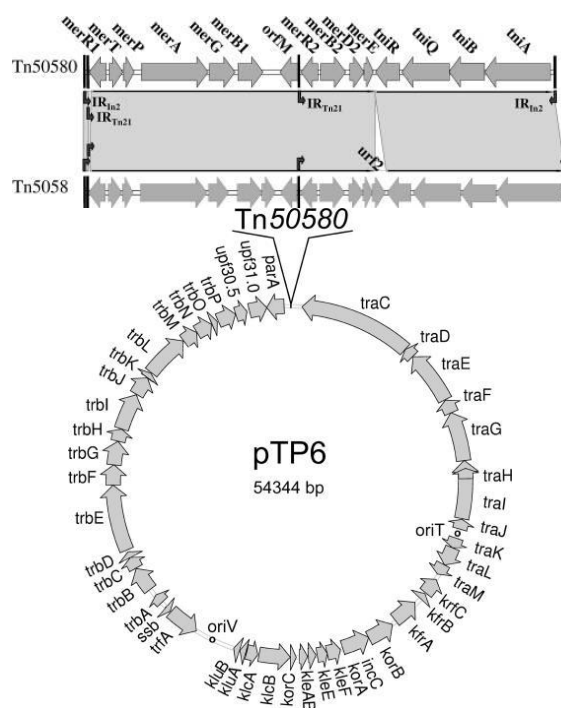


Fig. S1. Genetic map of plasmid pTP6. Figure adapted from Smalla et al., 2006 (Copyright © 2006, Appl Environ Microbiol. 2006 Nov; 72(11): 7253–7259).

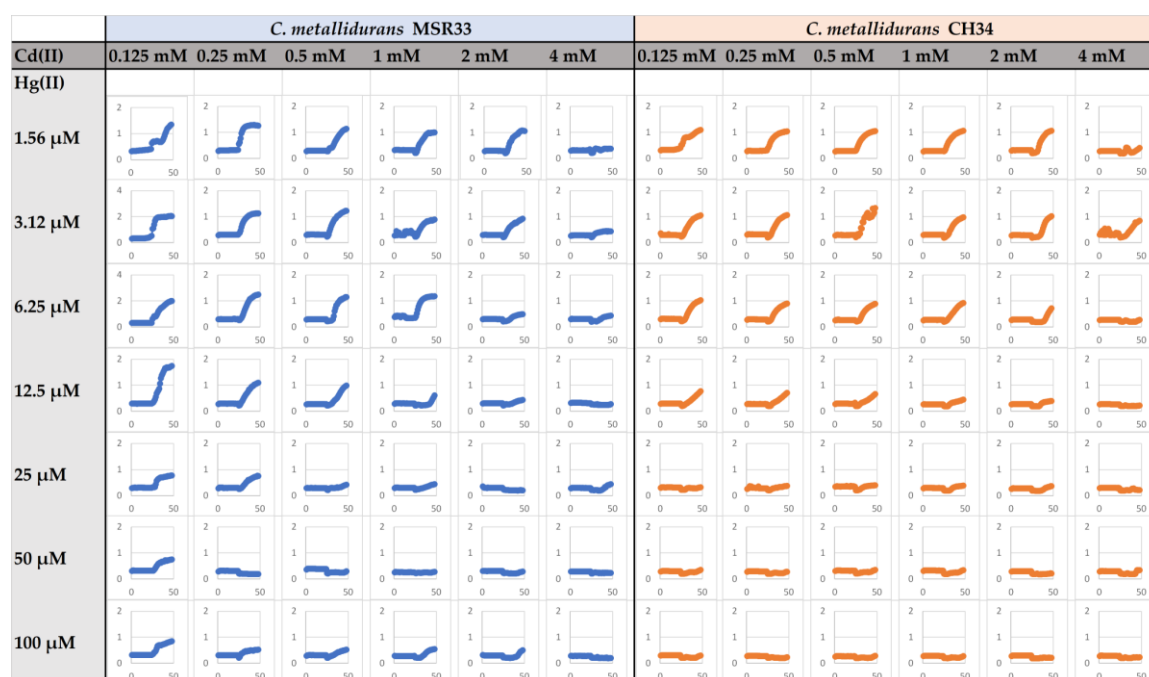
S2 Table. Transcriptional changes in MSR33 vs CH34 under equal, non-selective conditions.

	No genes	%Total
Total genes	6099	100
Pval >0.05	5462	89.56
Pval <0.05	637	10.44
Overexpressed (log ₂ >1)	87	0.014
Repressed (log ₂ <-1)	15	0.002
Highly overexpressed (log ₂ >2)	27	0.004
Highly repressed (log ₂ <-2)	1	0.0001

S3 Table. Sequence similarity of *mer* gene products present in plasmid pTP6.

Gene	Protein (aa)	Function	Organism (Reference)	%ID (aa)
<i>merR1</i>	MerR (144)	activator/repressor of <i>mer</i> operon	<i>C. metallidurans</i> CH34 (YP_145639.1)	95% (144)
<i>merT</i>	MerT (116)	mercuric ion transport protein	<i>C. metallidurans</i> CH34 (YP_145638.1)	87% (116)
<i>merP</i>	MerP (91)	periplasmic mercuric-ion binding protein	<i>C. metallidurans</i> CH34 (YP_145637.1)	77% (91)
<i>merA</i>	MerA (569)	mercuric-ion reductase Fad flavoprotein	<i>C. metallidurans</i> CH34 (YP_145636.1)	71% (561)
<i>merG</i> *	MerG (217)	organomercurial transporter	<i>C. testosterone</i> JL40 (KGGH30768.1)	100% (190)
<i>merB-1</i> *	MerB (212)	organomercurial lyase	<i>B. cepacia</i> 2a (YP_006965881.1)	100% (212)
<i>merR2</i>	MerR (144)	activator/repressor of <i>mer</i> operon	<i>C. metallidurans</i> CH34 (YP_145639.1)	93% (144)
<i>merB-2</i> *	MerB (212)	organomercurial lyase	<i>B. cepacia</i> 2a (YP_006965883.1)	100% (212)
<i>merD</i>	MerD (121)	secondary regulatory protein	<i>C. metallidurans</i> CH34 (YP_145635.1)	92% (121)
<i>merE</i>	MerE (78)	membrane mercuric resistance protein	<i>C. metallidurans</i> CH34 (YP_145634.1)	79% (78)

*Genes not present in *C. metallidurans* CH34

S4 Table: Effect of various mixtures of Hg²⁺ and Cd²⁺ on *C. metallidurans* strains MSR33 and CH34 growth.**S5 Table.** Plasmid copy number (PCN) for *C. metallidurans* strains MSR33 and CH34

Replicon (gene)	<i>C. metallidurans</i> MSR33				<i>C. metallidurans</i> CH34				Average PCN
	Ct	SD	Log (DNA)	PCN	Ct	SD	log (DNA)	PCN	
Chromosome (<i>cadA</i>)	23.38	0.05	10.18	1.00	23.47	0.21	10.18	1.00	1.00
Chromid (<i>zniA</i>)	20.90	0.19	10.24	1.16	20.18	0.07	10.24	1.14	1.15
pMOL30 (<i>nccA</i>)	21.71	0.29	10.26	1.21	19.75	0.20	10.24	1.14	1.18
pMOL28 (<i>cnrA</i>)	27.80	0.24	10.25	1.18	24.62	0.07	10.23	1.12	1.15
pTP6 (<i>merG</i>)	20.67	0.08	10.50	1.81	35.28	0.60	9.48	0.17	0.99

S6 Table: *mer* gene occurrence on replicons of strains CH34 and MSR33 (individual genes of the *merRT*, *merRTPA*, *merRDE*, *merRTPA* or *merRTPADE* loci are indicated)

Replicon	<i>mer</i> genes present in each replicon						PCN*
CHR1	<i>R</i>	<i>T</i>	<i>P</i>	<i>A</i>			1.00
pMOL28	<i>R</i>	<i>T</i>	<i>P</i>	<i>A</i>	<i>D</i>	<i>E</i>	1.15
pMOL30 – cluster 1	<i>R</i>	<i>T</i>	<i>P</i>	<i>A</i>	<i>D</i>	<i>E</i>	1.18
pMOL30 – cluster 2 (CH34)	<i>R</i>	<i>T</i>					1.18
	4.51	4.51	3.33	3.33	2.33	2.33	
pTP6 – cluster 1	<i>R</i>	<i>T</i>	<i>P</i>	<i>A</i>			1.8
pTP6 – cluster 2 (MSR33)	<i>R</i>				<i>D</i>	<i>E</i>	1.8
	8.11	6.31	5.13	5.13	4.13	4.13	
Gene unit increase	+2	+1	+1	+1	+1	+1	
Gene content increase	80%	40%	54%	54%	77%	77%	

* PCN values were taken from S5 Table.

S7 Table: Expression changes under non-selective conditions of *C. metallidurans* MSR33 against *C. metallidurans* CH34, represented in a colour coded table.

Movement related genes	Transcriptional regulators	Transporters
Recombinase activity	Catalytic function	Miscellaneous

*Highly over expressed (log₂ ratio > +1) or repressed (log₂ ratio < -1) genes are shown as bold text.

Rmet_code	Gene name	Function	Log ratio
Rmet_0063	<i>Rmet_0063</i>	putative allophanate hydrolase subunit 2	-1.67650883
Rmet_0065	<i>Rmet_0065</i>	conserved hypothetical protein	-1.23932973
Rmet_0066	<i>Rmet_0066</i>	conserved hypothetical protein	-1.37945687
Rmet_0067	<i>pcp1</i>	pyrrolidone-carboxylate peptidase	-1.05550107
Rmet_0114	<i>bioA</i>	7,8-diaminopelargonic acid synthase, PLP-dependent	-1.42445765
Rmet_0115	<i>bioF</i>	8-amino-7-oxononanoate synthase	-2.28376218*
Rmet_0116	<i>bioD</i>	dethiobiotin synthetase	-1.45848096
		biotin synthase slightly down RPM proteomics BL (0,72)	
Rmet_0117	<i>bioB</i>	down also RWV BL (0.68)	-1.4034115
Rmet_0118	<i>ycdW</i>	2-ketoacid reductase	-1.55585535
Rmet_0120	<i>Rmet_0120</i>	predicted Fe-S protein	-1.23051237
Rmet_0121	<i>Rmet_0121</i>	beta-lactamase-like protein	-1.3041744
Rmet_0122	<i>Rmet_0122</i>	putative acetyltransferase	-1.23552293
Rmet_0297	<i>Rmet_0297</i>	probable nucleoside triphosphate hydrolase domain	1.12141351
Rmet_0310	<i>Rmet_0310</i>	putative intracellular protease/amidase/DJ-1/Pfpl family	1.11868
Rmet_0410	<i>rplM</i>	50S ribosomal protein L13	1.22429

Rmet_0484	<i>tnpB</i>	IstB-like ATP binding protein ISRme4	2.58535001*
		molybdate transporter subunit ; membrane component	
Rmet_0570	<i>modB</i>	of ABC superfamily	2.66830376*
Rmet_0599	<i>Rmet_0599</i>	cytochrome c oxidase, subunit II	1.91275581
Rmet_0653	<i>pilA</i>	Flp/Fap pilin component; Putative pilus subunit protein	2.04791129
		phosphonate/organophosphate ester transporter subunit;	
Rmet_0775	<i>phnC</i>	ATP-binding component of ABC superfamily	1.11565916
Rmet_0777	<i>bcr</i>	drug resisttransporter Bcr/CflA subfamily	2.00829538*
Rmet_0865	<i>Rmet_0865</i>	conserved hypothetical protein	1.03606223
Rmet_0887	<i>Rmet_0887</i>	putative membrane protein	1.13689678
Rmet_0942	<i>Rmet_0942</i>	putative ADP-ribose pyrophosphatase	1.45577081
Rmet_0986	<i>Rmet_0986</i>	putative ATPase, AAA family	2.01441667*
Rmet_1220	<i>boxA</i>	benzoyl-CoA oxygenase component A	1.3183696
Rmet_1690	<i>flhD1</i>	DNA-binding transcriptional dual regulator with FlhC	2.94450715*
Rmet_1767	<i>Rmet_1767</i>	transcriptional regulator, LysR family	2.0439383*
Rmet_1841	<i>Rmet_1841</i>	4-hydroxybenzoyl-CoA thioesterase	2.1924016*
Rmet_1960	<i>phaY</i>	D-(-)-3-hydroxybutyrate oligomer hydrolase	2.19656841*
Rmet_1980	<i>ansB</i>	periplasmic L-asparaginase II	1.74419116
Rmet_2072	<i>Rmet_2072</i>	major facilitator superfamily MFS_1	1.06419117
Rmet_2171	<i>Rmet_2171</i>	conserved hypothetical protein	1.35381313
		Tyr recombinase activity site-specific recombination	
Rmet_2172	<i>Int</i>	Tyr recombinase activity	2.82265137*
Rmet_2177	<i>ppx</i>	exopolyphosphatase	1.10508189
		phosphate transporter subunit; ATP-binding component	
Rmet_2182	<i>pstB</i>	of ABC superfamily	1.00939883
		phosphate transporter subunit; membrane component of	
Rmet_2183	<i>pstA</i>	ABC superfamily	1.63382487
Rmet_2185	<i>pstS</i>	phosphate ABC transporter periplasmic-binding protein	1.01072
Rmet_2382	<i>tnpA</i>	transposase IS1088	5.17170573*
Rmet_2382	<i>tnpA</i>	transposase IS1088	4.72423726*
Rmet_2535	<i>bug</i>	extra-cytoplasmic Solute Receptor	1.31844478
Rmet_2737	<i>Rmet_2737</i>	hypothetical protein	1.03944721
Rmet_2932	<i>Rmet_2932</i>	lactoylglutathione lyase-like protein	1.27848603
		phosphonate/organophosphate ester transporter subunit;	
Rmet_2995	<i>ptxA</i>	ATP-binding component of ABC superfamily	1.50871938
Rmet_3036	<i>Rmet_3036</i>	conserved hypothetical protein	2.0934961*
Rmet_3126	<i>ddlB</i>	D-alanine:D-alanine ligase	2.27184325*
Rmet_3313	<i>rpsS</i>	30S ribosomal subunit protein S19	1.58336143
Rmet_3358	<i>Rmet_3358</i>	conserved hypothetical protein	1.54868879
Rmet_3359	<i>Rmet_3359</i>	thiamine biosynthesis protein	1.43945338
Rmet_3361	<i>Rmet_3361</i>	hypothetical protein; membrane protein	1.11203697

Rmet_3378	<i>Rmet_3378</i>	GCN5-related N-acetyltransferase	1.42687412
Rmet_3533	<i>phhA</i>	phenylalanine-4-hydroxylase (PAH) (Phe-4-monooxygenase)	1.17501249
Rmet_3534	<i>phhB</i>	pterin-4-alpha-carbinolamine dehydratase (transcriptional co-activator)	1.42952206
Rmet_3549	<i>tctC</i>	periplasmic tricarboxylate binding receptor (TctC) component of transporter, tripartite tricarboxylate	-1.2197541
Rmet_3550	<i>tctB</i>	transport (TTT) family	-1.22615821
Rmet_3589	<i>acrB</i>	multidrug efflux system protein	1.1724
Rmet_3628	<i>Rmet_3628</i>	hypothetical protein	1.20980184
Rmet_3681	<i>cheW</i>	purine-binding chemotaxis protein	1.04754265
Rmet_3693	<i>cheB1</i>	chemotaxis response regulator protein-glutamate methylesterase 1	2.3968041*
Rmet_3731	<i>flgN</i>	flagella synthesis protein FlgN	1.09027262
Rmet_3732	<i>flgM</i>	negative regulator of flagellin synthesis (anti-sigma-28 factor, FlgM)	1.2232021
Rmet_3742	<i>flgJ</i>	peptidoglycan hydrolase (muramidase)	1.15496048
Rmet_3744	<i>flgL</i>	flagellar hook-filament junction protein	1.07665334
Rmet_3826	<i>Rmet_3826</i>	alkaline phosphatase	1.46719238
Rmet_4115	<i>Rmet_4115</i>	conserved hypothetical protein	2.29657966*
Rmet_4161	<i>pelG</i>	conserved hypothetical protein	-1.05049624
Rmet_4299	<i>dctA</i>	C4-dicarboxylate transport protein 2	4.21730516*
Rmet_4332	<i>tauC</i>	ABC transporter, permease protein; possible sulfonate/taurine transporter inner membrane component	1.25074713
Rmet_4403	<i>Shc</i>	squalene cyclase	1.24741055
Rmet_4404	<i>shcR</i>	transcriptional regulator, TetR family	1.21796966
Rmet_4623	<i>Rmet_4623</i>	hypothetical protein	1.10951785
Rmet_4664	<i>Rmet_4664</i>	phosphoryl transfer system, HPr	1.09003277
Rmet_4726	<i>Rmet_4726</i>	diguanylate cyclase/phosphodiesterase	1.52401544
Rmet_4727	<i>Rmet_4727</i>	methyl-accepting chemotaxis sensory transducer	1.29962115
Rmet_4834	<i>ompP</i>	putative outer membrane pore protein (gram-negative type)	1.27816628
Rmet_4839	<i>Rmet_4839</i>	putative transcriptional regulator, LacI family	1.57630345
Rmet_4993	<i>Rmet_4993</i>	membrane efflux protein (major facilitator superfamily MFS_1)	1.4303943
Rmet_5075	<i>musE1</i>	putative NAD(P)H-dependent FMN reductase SsuE	-1.0976
Rmet_5235	<i>Rmet_5235</i>	conserved hypothetical protein; TPR domain protein	1.10058015
Rmet_5238	<i>Rmet_5238</i>	conserved hypothetical protein	1.02270936
Rmet_5239	<i>Rmet_5239</i>	hypothetical protein (putative dioxygenase)	1.03401905
Rmet_5252	<i>fliC2</i>	<i>fliC2</i> flagellin	1.84863

Rmet_5279	<i>Rmet_5279</i>	transcriptional regulator, LysR family	1.24140191
Rmet_5334	<i>Rmet_5334</i>	MgtC/SapB family transporter	1.31906819
Rmet_5426	<i>Rmet_5426</i>	conserved hypothetical protein	2.78212006*
Rmet_5559	<i>Rmet_5559</i>	hypothetical protein	2.00853447*
Rmet_5560	<i>Rmet_5560</i>	hypothetical protein	1.76768503
Rmet_5620	<i>Rmet_5620</i>	conserved hypothetical protein	1.04143379
Rmet_5642	<i>fliD3</i>	flagellar hook-associated protein 2	1.23738918
Rmet_5655	<i>Rmet_5655</i>	conserved hypothetical protein	1.73791832
Rmet_5855	<i>wcaF</i>	glycose-acyl transferase	1.04033538
Rmet_5930	<i>fusA2</i>	elongation factor G 2 (EF-G 2)	1.07594255
Rmet_5957	<i>tnpA</i>	<i>orf4</i> , <i>ISRme19</i>	1.54296414
Rmet_6006	<i>Int</i>	Tyr recombinase activity site-specific recombination	1.30660657
Rmet_6171	<i>merR</i>	MerR from Tn4380, regulatory protein involved in Hg(II) resistance	1.23849795
Rmet_6196	<i>chrP</i>	permease of the major facilitator superfamily MFS_1	2.14766347*
Rmet_6205	<i>cnrY</i>	CnrY, antisigma factor, regulatory protein, involved in Co(II), Ni(II) response	5.4418141*
Rmet_6206	<i>cnrX</i>	CnrX, antisigma factor, regulatory protein, involved in Co(II), Ni(II) response	5.03229334*
Rmet_6207	<i>cnrH</i>	CnrH, sigma factor, involved in Co(II), Ni(II) response	3.78953232
Rmet_6208	<i>cnrC</i>	CnrC, outer membrane protein, three components cation proton antiporter efflux system, involved in Co(II), Ni(II) resistance	4.60736056*
Rmet_6209	<i>cnrB</i>	CnrB, membrane fusion protein, three components cation proton antiporter efflux system, involved in Co(II), Ni(II) resistance	5.9867383*
Rmet_6210	<i>cnrA</i>	CnrA, inner membrane efflux pump, three components cation proton antiporter efflux system, involved in Co(II), Ni(II) resistance	5.81023103*
Rmet_6211	<i>cnrT</i>	CnrT, Cation Diffusion Facilitator, involved in Co(II), Ni(II) resistance	5.3174816*
Rmet_6252	<i>int</i>	tyrosine-based site-specific recombinase activity	1.26521262
Rmet_6255	<i>Rmet_6255</i>	TetR family transcriptional regulator-like protein	1.18017164
Rmet_6262	<i>orf112</i>	Alpha/beta hydrolase	1.22794711
Rmet_6268	<i>Rmet_6268</i>	DNA/RNA non-specific endonuclease	1.65810803
Rmet_6300	<i>trbG</i>	mating pair formation	1.19131289