

Supplementary Information

Supplementary Table S1. Properties of the characterized cluster A-I solute binding proteins.

Protein	Organism	Ligand	PDB codes	UniProtKB code	References	KO	Sites	Kd	Binding site 1 st coordination sphere	Flexible loop	Gram
ZnuA	<i>Acinetobacter baumannii</i>	Zn ²⁺	Not found	B0VNJ6	[1,2]	Zn[2]	nd	nd	nd	nd	-
ZnuA (YcdH)	<i>Bacillus subtilis</i>	Mn ²⁺	2O1E	O34966	Unpublished [3]	Zn[4]	nd	nd	His ₂ (His51 and His128)/ Glu ₁ (Glu267)	Length: 16 a.a. 5 histidines	+
ZnuA	<i>Brucella abortus</i> biovar 1	Zn ²⁺	Not found	Q576K1	[5]	Zn[5]	nd	nd	nd	nd	-
ZnuA2	<i>Candidatus Liberibacter asiaticus</i> str. psy62	Mn ²⁺ Mn ²⁺ (mut) Cd ²⁺ Zn ²⁺ Apo Apo (mut)	4UD0, 4CL2 5ZHA, 5Z2K 6IXI 5AFS 4UDN 5Z35, 5Z2J	C6XF58	[6-9]	nd	1	Zn:430uM Mn:370uM [10]	His ₂ (His39, His106) Asp ₁ (Asp247) Glu ₁ (172) H-bond: asp and arginine	No loop.	-
ZnuA	<i>Campylobacter jejuni</i>	Zn ²⁺	Not found	Q0PBZ4	[11]	Zn[11]	nd	nd	nd	Length: 14 a.a. 8 histidines	-
YtgA	<i>Chlamydia trachomatis</i>	Fe ³⁺	6NSI	Q9S529	[12]	nd	1	nd	His ₃ (His75, His141, His207) Asp ₁ (299)	No major loop structure The loop connecting alpha 9 and B6 on the surface of the C-terminal domain dwells directly above the Fe(III).	-
ZnuA	<i>Escherichia coli</i>	Zn ²⁺ Co ²⁺ Apo	2OSV, 2OGW, 2PS0 2PRS 2PS9 2PS3	P39172	[13] [14] [15]	Zn[16]	2	Zn: < 20 nM [15]	His ₃ (His78, His161, His225) Glu ₁ (Glu77).	Length: 21a.a 6 histidines	-
ZnuA (PZP1)	<i>Haemophilus influenzae</i> Rd KW20 (serotype d)	Zn ²⁺	Not found	P44526	[17]	Zn[17]	nd	nd	nd	Length: 50 a.a. 25 histidines[18]	-
Lmo1671	<i>Listeria monocytogenes</i> EGD-e	unknown	Not found	Q8Y6L3	none	nd	nd	nd	nd	Loop with histidines[19]	+
MntA	<i>Listeria monocytogenes</i> EGD-e	Cd ²⁺ Mn ²⁺ Apo	5JPD 5I4K 5HX7	Q8Y653	Unpublished[20]	nd	nd	nd	His ₂ (His67, His140) Glu ₁ (206) Asp ₁ (281)	No loop	+
ZinA	<i>Listeria monocytogenes</i>	Zn ²⁺	Not found	Q8YAH3	[21]	Zn[21]	nd	nd	nd	nd	+
MntC	<i>Neisseria gonorrhoeae</i>	Zn ²⁺ Mn ²⁺	Not found	B4RIV9	[22]	Zn, Mn[22]	1	Zn ²⁺ : 104 ±5 nM Mn ²⁺ : 100 ±8 nM[22]	nd	Histidine rich loop[22]	-
AztC	<i>Paracoccus denitrificans</i>	Zn ²⁺ Zn ²⁺ (mut) Apo	5W57 5KZJ 5W56	A1B2F3	[23]	Zn[24]	1	Zn: 0.3 ± 0.1 nM[24]	His ₃ (His61, His138, His204) Asp ₁ (279)	Length: 15 a.a 3 histidines	-
ZnuA	<i>Paracoccus denitrificans</i>	Zn ²⁺	Not found	A1B9L0	[24]	Zn[24]	5	Main binding site Zn: 1nM [24]	nd	Length: 38 a.a. 15 Histidines	-
ZnuA	<i>P. aeruginosa</i> PAO1: PA5498	Zn ²⁺	Not found	Q9HT75	[18]	nd	2	Zn: 22.6 ± 6.4 nM[18]	His ₃ (His60, His140 and His204)	Length: 15 a.a. 5 Histidines	-

								Water molecule Based on homology[18]		
ZnuA	<i>Salmonella enterica</i> subsp. <i>enterica</i> serovar Typhimurium	Zn ⁺² Zn ⁺² /PH Apo (mut)	2XQV, 2XY4 4BBP 2XH8	Q8ZNV8	[25,26]	Zn[27]	2	1 st site: <20nM 2 nd site: uM range[28]	His3(His140, His147, His211) Glu1(Glu59)	Length: 21a.a 3 histidines
SitA	<i>Salmonella enterica</i> subsp. <i>enterica</i> serovar Typhimurium str. LT2	Apo	5JG7	Q7CPX8	Unpublished[29]	nd	nd	nd	nd	Length: 11 a.a No histidines
MntC	<i>Staphylococcus aureus</i>	Fe ⁺³ /co structure with mAB 305-78-7 Zn ⁺² Mn ⁺² Apo/anta gonistic fab fragment	5HDQ 4NNO 4K3V 4NNP	A0A0H3JT S7	[30-32]	Mn[33]	1	Mn ⁺² : ~ 4 nM[32]	Mn ⁺² /Fe ⁺³ His2(His50, His123) Asp:(Asp264) Glu1(Glu189) Zn ⁺² : His2(His67, His140) Asp:(Asp281) Glu1(Glu206)	No major loop structure Peptides 28-51 and 37-51 include a flexible loop with one of the four Mn+2 coordinating residues :H50 residues covered by peptides 111- 125 and 111-126. They include another flexible loop and another Mn2+- coordinating residue, H123
SitA	<i>Staphylococcus pseudintermedius</i>	Zn ⁺² Mn ⁺²	4OXQ 4OXR	NCBI WP_014614 644.1	[34]	nd	1	Mn ⁺² and Zn ⁺² : Low nM range.[34]	His2(His64, His137) Glu1(Glu203) Asp:(Asp278)	No Loop
SitA	<i>Sinorhizobium meliloti</i>	Mn ⁺² Fe ⁺²	Not found	F7XAF4	[35]	Mn, Fe(II)[35]	nd	nd	nd	nd
Lmb	<i>Streptococcus agalactiae</i> NGBS572	Zn ⁺² Zn ⁺² (mut)	3HJT 4HOF	Q9ZHG8	[36],[37]	Zn[38]	nd	nd	His2(His66, His142 and His206). Glu1(Glu281).	Length: 12a.a No histidines Residue range: 124-135
AdcA	<i>Streptococcus agalactiae</i> A909 serotype Ia	Zn ⁺²	Not found	Q8E128	[38]	Zn[38]	nd	nd	3 Histidines 1 Glutamate Predicted from sequence homology. [39]	Length: 11 a.a. 6 Histidines
PsaA	<i>Streptococcus pneumoniae</i>	Apo Mn ⁺² Zn ⁺² Cd ⁺²	3ZK8 3ZK7 3ZK9 3ZKA 3ZTT 1PSZ 4UTO 4UTP	P0A4G2	[39-41]	Mn[42]	nd	231 nM Zn, 3.3 nM Mn[43]	His2(His67, His139) Glu205 Asp280	7 a.a No histidines Residue range: 129-135
AdcA	<i>Streptococcus pneumoniae</i>	Zn ⁺²	Not found	Q8CWN2	[44,45]	Zn[45]	2	Zn ⁺² : High Affinity site: 4±0.1 nM Low Affinity Site: 228±88 nM[46]	His2(His63, His140, His204) Glu1(Glu279) Based on homology.[46]	Histidine Rich Loop is present.
AdcAll (Lmb)	<i>Streptococcus pneumoniae</i>	Zn ⁺²	3CX3	Q8DQ09	[19]	Zn [45]	nd	nd	His2(His71, His147, His211) Glu1(Glu286)	Length: 13 a.a No histidines Residue range: 129-142
MtsA	<i>Streptococcus pyogenes</i>	Fe ⁺³	3HH8	P0A4G4	[46]	Mn, Fe[47,48]	1	4.3 μM Fe(II)	His2(His68, His140)	No loop

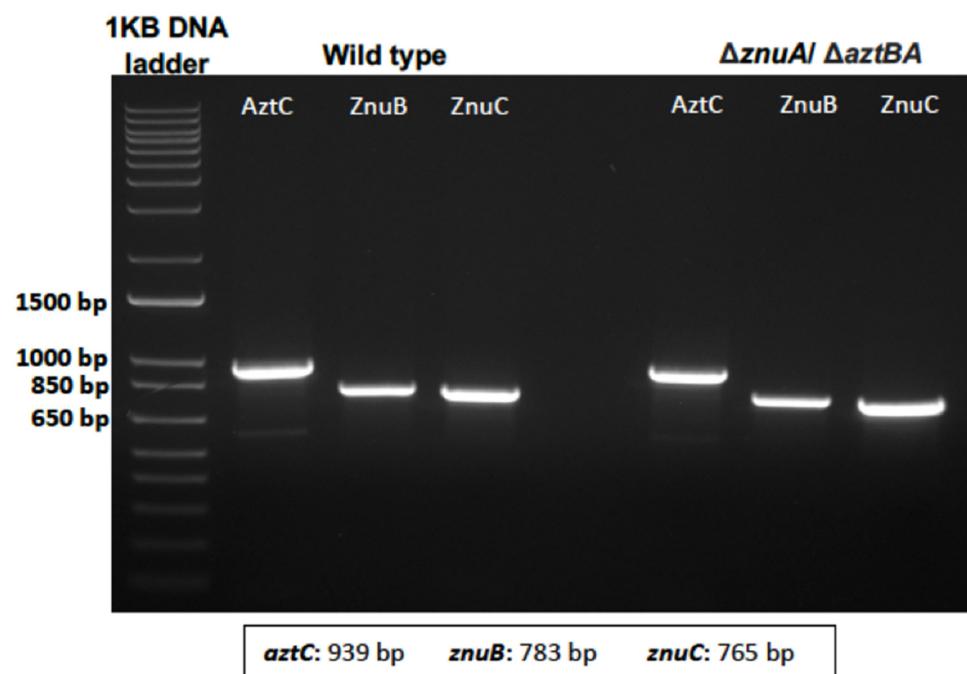
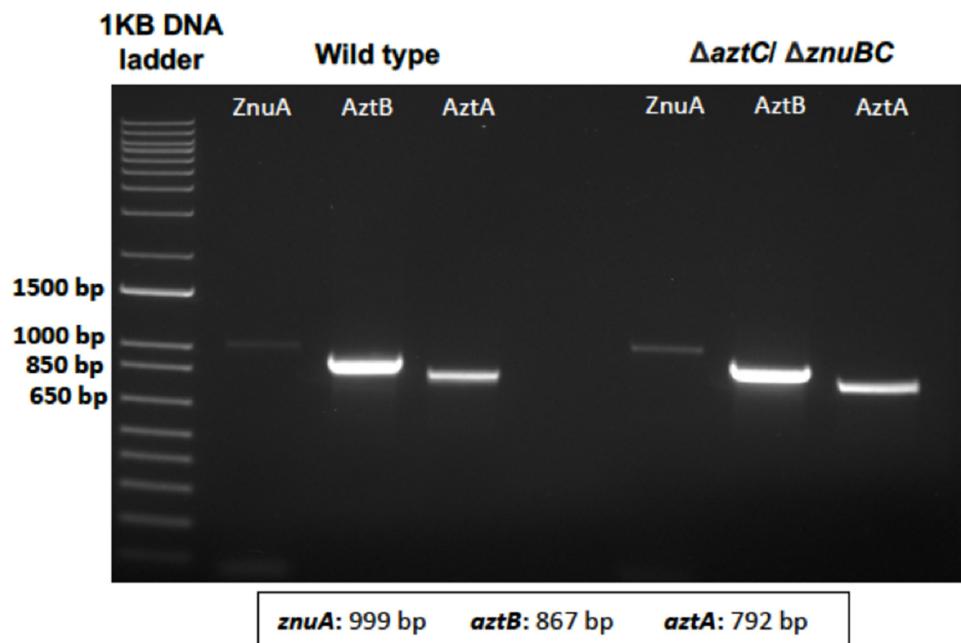
								50 μM Mn	Glu:(Glu206) Asp:(Asp281)		
Lmb (Lbp)	<i>Streptococcus pyogenes</i> Serotype M1	Zn ⁺²	3GI1	Q99XV3	[49]	Zn[50],[51]]	1	nd	His ₃ (His66, His142, His206) Glu ₁ (Glu281)	Length: 15 a.a. No histidine Residue range: 123-137	+
TroA	<i>Streptococcus suis</i>	Zn ⁺²	3MFQ	A4VY63	[52]	Mn[53]	1	434nM Zn 254nM Mn[53]	His ₃ (His76, His139, His205) Asp:(Asp289)	Length: 4a.a Residue range: 126-129	+
ZnuA	<i>Synechocystis</i> sp. PCC 6803 substr. Kazusa	Zn ⁺² Zn ⁺² (mut) Apo (mut)	1PQ4 2OV3 2OV1	P73085	[54,55]	nd	4	7 nM, 9 μM Zn[55]	His ₃ (His83, His179, His243) Water molecule	Length: 34 a.a 8 histidines	-
MntC	<i>Synechocystis</i> sp. PCC 6803	Mn ⁺² Mn ⁺² (mut) Zn ⁺² , Mn ⁺²	1XVL 4IRM 3UJP	Q79EF9	[56,57]	Mn[58]	nd	nd	His ₂ (His89, His154) Glu ₁ (Glu220) Asp:(Asp295)	No loop	-
TroA	<i>Treponema pallidum</i>	Zn ⁺² Apo	1TOA 1K0F	P96116	[59,60]	nd	1	23nM Zn 7.1nM Mn[61],[62]	His ₃ (His68, His133, His 199) Asp:(Asp279)	4a.a No histidines	-
VC_2081 (ZnuA)	<i>Vibrio cholerae</i>	Zn ²⁺	Not found	Q9KQB9	[63]	Zn[63]	nd	nd	nd	nd	-
VC_2552 (ZrgA)	<i>Vibrio cholerae</i>	Zn ²⁺	Not found	Q9KP27	[63]	Zn[63]	nd	nd	nd	nd	-
YfeA	<i>Yersinia pestis</i>	Zn ⁺² Fe ⁺³ Mn ⁺² Apo	5UXS 5UXU 5UY0 6Q1D 5UYG 5UYD 5UYA 5UYW 5UYV 5UY4 5UYE 5UYB 5UY5 5UYH 5UYF 5UYC 6Q1C	Q56952	[64-65]	Fe[66]	2	17.8 nM Mn, 6.6 nM Zn[67] Iron nd	His ₂ (His76, His141) Glu ₁ (207(holo), Glu256(apo)) Asp:(Asp282)	Length: 21 a.a. Residue range: 119-140	-
ZnuA	<i>Yersinia pestis</i>	Zn ²⁺	Not found	Q8ZEU2	[67,68]	Zn[67]	nd	nd	His ₃ (His62, His153, His217) Water molecule Based on homology modeling.[69]	Histidine Rich loop present.	-

Supplementary Table S2. Strains used in this study.

Strain/plasmid	Genotype/ description	Source
PD1222	<i>P. denitrificans</i> Wild-type, Rm ^r	A gift from Dr. Stephen Sprio UT Dallas
	PD1222 mutant, Δ <i>aztC</i> , Rif ^r	[24]
	PD1222 mutant, Δ <i>znuA</i> , Rif ^r	[24]
	PD1222 mutant, Δ <i>aztC</i> , Δ <i>znuA</i> Rif ^r	[24]
	PD1222 mutant, ΔD-loop <i>aztC</i> , Δ <i>znuA</i> , Rif ^r	This work
	PD1222 mutant, ΔZ-loop <i>aztC</i> , Δ <i>znuA</i> , Rif ^r	This work
	PD1222 mutant, ΔD-loop <i>znuA</i> , Δ <i>aztC</i> , Rif ^r	This work
	PD1222 mutant, Δ <i>aztC</i> Δ <i>znuBC</i> , Rif ^r	This work
	PD1222 mutant, Δ <i>znuA</i> Δ <i>aztBA</i> , Rif ^r	This work
S17-1	<i>E. coli</i> , with derivative RP4 plasmid integrated, for incorporation of plasmid constructs into <i>P. denitrificans</i> .	A gift from Dr. Stephen Sprio UT Dallas
PRK2013	<i>E. coli</i> with pRK2013 plasmid, which contains transfer system RK2 for mobilization of non-self-transmissible plasmids	A gift from Dr. Stephen Sprio UT Dallas
Plasmids		
pK18mobsacB	Gene replacement vector, sacB, Km ^r	
	pK18mobsacB, with upstream and downstream 600bp flanking sequences of D-loop from <i>aztC</i> , Km ^r	This work
	pK18mobsacB, with upstream and downstream 600bp flanking sequences of Z-loop from <i>aztC</i> , Km ^r	This work
	pK18mobsacB, with upstream and downstream 600bp flanking sequences of loop from <i>znuA</i> , Km ^r	This work
	pK18mobsacB, with upstream and downstream 600bp flanking sequences of <i>znuBC</i> , Km ^r	This work
	pK18mobsacB, with upstream and downstream 600bp flanking sequences of <i>aztBA</i> , Km ^r	This work

Supplementary Table S3. Primers used in this study.

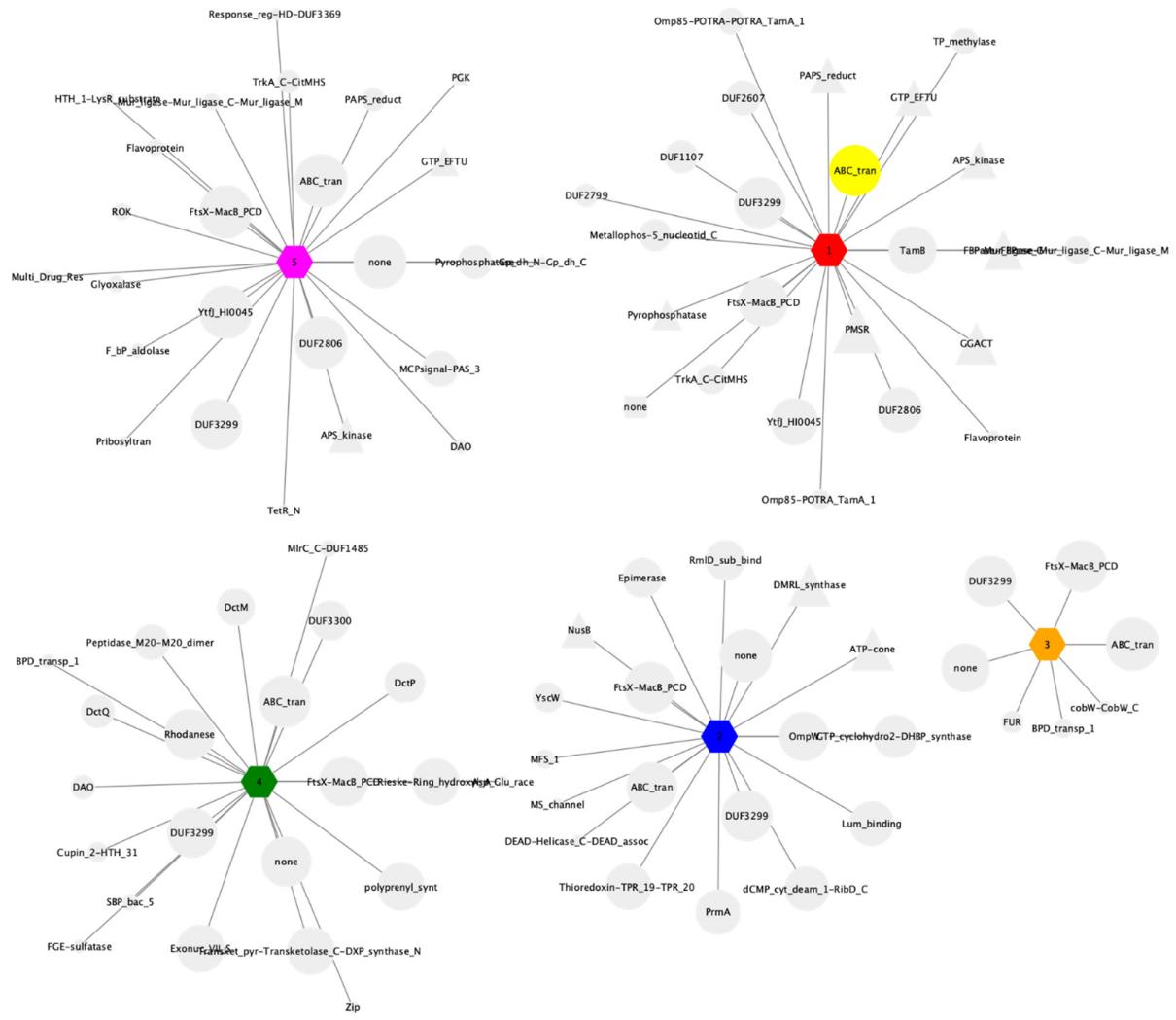
Primer	Primer sequence 5' to 3'
UP600aztAB_FWD	gaaacagctatgacatgattacgaacctcgggcggatccgg
UP600aztAB_REV	cgtcgccgaaagagccagtcttcatcaactggatagccgactc
DWN600aztAB_FWD	caaaatatcgagtccgctatccaggatgaaagactggcttc
DWN600aztAB_REV	gaggatccccgggtaccggatcgaaacacggtgccggcttg
UP600znuBC_FWD	cagctatgacatgattacggggttatataccatccagacgcgc
UP600znuBC_REV	caccgaggccggcctgtcacccatcgagcggcggaaa
DWN600znuBC_FWD	tttccggcgcctccgatacgttagtacaggccggctcggt
DWN600znuBC_REV	atccccgggtacccgagctcgacacggaaatcggtcacc
UP600Loop_znuA_FWD	ctatgacatgattacggccatggcgcgtatgcgtt
UP600Loop_znuA_REV	catgaggatcggtgcggcatagtccggcgtatgcgt
DWN600Loop_znuA_FWD	catcgccggactatgccggcaccatccatgcct
DWN600Loop_znuA_REV	cgggtaccgagctcgtaaaaggcgg
UP600D-Loop_aztC_FWD	gctatgacatgattacgcggcgggtctggtaacg
UP600D-Loop_aztC_REV	tggggatcatgcgcggccgcggccgtcc
DWN600D-Loop_aztC_FWD	gagccggcgccggcgccgcgtatccccatgcc
DWN600D-Loop_aztC_REV	cgggtaccgagctcggtcctttaggggg
UP600Z-Loop_aztC_FWD	ctatgacatgattacgcaggcgaaaccgtggacg
UP600 Z-Loop_aztC_REV	cacatggccgcagggctgcggcgacaggaaatgcac
DWN600Z-Loop_aztC_FWD	ttccctgcgcgcaggccgtcggtcgatgt
DWN600Z-Loop_aztC_REV	cgggtaccgagctcgacagatgccgtcggtact
znuA_RT-PCR_FWD	actatcatatgatccgacccttc
znuA_RT-PCR_REV	actatggtagccatcgaggcccagg
znuB_RT-PCR_FWD	ttatttcctgc aaaccact
znuB_RT-PCR_REV	atgcttgacgtttctgt
znuC_RT-PCR_FWD	tcaatcggtggcg
znuC_RT-PCR_REV	gtgacgcccgtatgt
aztA_RT-PCR_FWD	atgcccagaaccgtccg
aztA_RT-PCR_REV	tcacgcacccctgtac
aztB_RT-PCR_FWD	atggtcgacgggtcg
aztB_RT-PCR_REV	tcaggccgtccgtat
aztC_RT-PCR_FWD	atgaaagactggcttc
aztC_RT-PCR_REV	gaaatctggcagcggc



Supplementary Figure S1. RT-PCR of full-length ABC transporter genes from WT and hybrid transporter strains.

	1	10	20	30	40	50		
V.cholerae VC2552/25-242	S.DHQRHQHBAHVHGQVELNIAQ..DGHDLTLEITAPGADVQHEAHPQDA						QI	
C.jejuni ZnuA/29-296	TSSNLIVSVSIAPAOAFFIKKIAANTLDVNVL	U	PPNSNELNSEPKPSTMKMEKSDYPTFQ				KVF	
Synechocystis ZnuA/46-334	ADAMDTVISIDPQYVLEKTTGCDLVRVSVL	V	PPNSNELNSEPKPSTMKMEKSDYPTFQ				QPF	
L.monocytogenes Lmo1671/33-315	SDQLTVTTFVPPYVYEFTKRNVYCDKKA	DVHS	PPNSDASPEEDTQKMMKTDSDS	AYFVY	QIGM		GE	
S.agalactiae AdcA/29-311	DSKLKVTTTFVPPYVYEFTKRNVYCDKKA	DVHS	GACTEPDPEEDSTKNIAAJQDSNAF	YVYMODNM			TW	
S.pneumoniae AdcA/26-307	DGKLNIVTTTFVPPYVYEFTKRNVYCDKKA	DVHS	GACTEPDPEEDSTKNIAAJQDSNAF	YVYMODNM			TW	
B.subtilis ZnuA/34-315	GDKLKVTTFVPPYVYEFTKRNVYCDKKA	DVHS	PSSVEPDEDTPKDIANJODADIF	FVYNESEYM			TW	
L.monocytogenes ZnuA/32-312	KDKLKVTTFVPPYVYEFTKRNVYCDN	ASIASLEM	DACTEPDPEEDSAKDIJKIEA	YDADV	FVYNSHTLB		TW	
S.pneumoniae AdcA/35-310	GKGMKIVTSFVPPYVYEFTKRNVYCDLN	DRVM	OSSAGIISSEDVSNVAAIYDADIF	FVYNSHTLB			SW	
S.agalactiae Lmb/30-305	TQGMSVTSFVPPYVYEFTKRNVYCDLN	DRVM	OSAGIISSEDVSNVAAIYDADIF	FVYNSHTLB			AH	
S.pyogenes Lmb/30-305	TQGMSVTSFVPPYVYEFTKRNVYCDLN	DRVM	OSAGIISSEDVSNVAAIYDADIF	FVYNSHTLB			AH	
S.aureus MntC/30-308	NGRLKLVTTNSILYDMARNVGGNNDVIH	S	PVCQDPPEYEVKPKD	KKRTDADVLY	YGLNLTG		NG	
L.monocytogenes MntA/30-308	DGKLNVATYSILADIVKRNVYCDLN	DRVM	PVCQDPPEYEVKPKD	KKRTDADVLY	YGLNLTG		NG	
S.pyogenes MtsA/31-308	SDKLKVATNSIADMTKAIAGDKIDLH	S	PVCQDPPEYEVKPKD	KKRTDADVLY	YGLNLTG		NG	
S.pneumoniae Psaa/30-307	GQRLKVVATNSIADMTKAIAGDKIDLH	S	PVCQDPPEYEVKPKD	KKRTDADVLY	YGLNLTG		NG	
Ca.L.asiaticus ZnuA2/21-293	TQKVVVSSFSI	GDI	EAGNDSSSYQVTS	TAIKIQNADL	LCNGLH		ET	
Synechocystis MntC/52-322	EERKKVTTFTVLA	M	RIGAEIIGYEE	TPDIVSAODADIL	LYNMN		RW	
Y.pestis Yfa/39-309	AKKEKVTTFTVLA	I	KPGAEIHDYOPTPD	IVSAODADIL	LWN		RW	
S.enterica Sita/27-297	KEEKKVTTFTVLA	I	KPGAEIHDYOPTPD	IVSAODADIL	LWN		RW	
S.meliloti Sita/29-299	ANELKAVTTFTVLA	I	KPGAEIHDYOPTPD	IVSAODADIL	LWN		RW	
Rhizobium/Agrobacterium TroA/24-294	QEKPKVTTFTVLA	I	KPGAEIHDYOPTPD	IVSAODADIL	LWN		RW	
N.gonorrhoeae MntC/19-302	AAPLDVTTSFSLGDVYAKQVIGGERVA	QVQSLV	KANODDTAYHNTSCD	KKIRSAK1V	LGGL		AA	
P.denitrificans AztC/24-306	GDPLDVTTSFSLGDVYAKQVIGGERVA	QVQSLV	GPDSDDTIVYEDPRPAAIA	LAGADV	TLN		GF	
Rhizobium/Agrobacterium ZnuA/26-324	AaelkvV	ASFSIADFAKNVYGNRNVETT	LLV	GPDDADAIVYEDPRPAAVA	VSKADYV	LGFLF		GF
C.trachomatis YtgA/38-320	DESYI	V1CDVSR	EDRMAMSQ	UFCNGL	LGFLF		AS	
S.suis TroA/40-316	S.KPRAVVTTISFLND	MVYV	PAGBDPHS	YENVKD	EDRHS		AS	
T.pallidum TroA/32-306	G.KPLVTTIGM1ADAVKNI	ADQDVHKL	PAGBDPHS	YVAKSS	LSK1OKADIV		AS	
A.baumannii ZnuA/17-274	GWPSGLVTVTHPIYI	TAKEI	GQPSGIDVQ	LPKPHRK	INDASIV		AS	
P.denitrificans Psaa/22-323	AEVPRVTTIPVGA	MVQOQ	QAGQDPH	SYQLRPSQARS	LODADL		PW	
B.abortus ZnuA/24-332	GEREGVTVISIKPLH	S	QAGQDPH	SYQLRPSQARS	LODADL		PW	
P.aeruginosa ZnuA/23-304	RAEVS	VIISIKPLQVAAI	QDGQGQPDV	LLV	PPQGASED	YVYSLKPS	AA	
V.cholerae VC2081/21-294	ASAIEV	TIISIKPLQVAAI	TYELMLGTGTPD	V	PPQGASED	YQYALRPSD	YR	
H.influenzae ZnuA/20-335	MANAD	VIASRKLGF	IVSIALGVTG	QVLLV	PPQGASED	YQYALRPSD	YR	
Y.pestis ZnuA/25-317	LASAA	VVTSIRPLGF	IAAAIADGVLPTEV	LL	PPQGASED	YDYNKLS	YR	
E.coli ZnuA/23-307	AADAA	VVASLKPVGFI	IASAIADGVTE	LL	PPQGASED	YDYSLRPSD	YR	
S.enterica ZnuA/42-330	AADAA	VVASLKPVGFI	IASAIADGVTD	LL	PPQGASED	YDYSLRPSD	YR	

	60	70	80	90	100	110	
V.cholerae VC2552/25-242	LEALETIIHHPE	..	KLFIAQSDKAQCEK	REVIKHTLG	GEYQHSHAY	GEDEEH	EHQHGEGEHDHK
C.jejuni ZnuA/29-296	TDFKQNFS	KL	QVVMNQK	RLNQH			DT..HEHSH..E..
Synechocystis ZnuA/46-334	LERLKAANA	NM	KLIDSAGQITPL	EME			K..DHHSHEEEG
L.monocytogenes Lmo1671/33-315	VDRAKRTI	LANEH	..VTFVPTA	EKLDDP			KDPD..AAE
S.agalactiae AdcA/29-311	VPPVAKRS	VSKR	..VTTI	IKGTGDM	LLT		KGVE..EEG
S.pneumoniae AdcA/26-307	VPPVLLD	IKRK	..VTKT	IKATGDM	LLT		PGGE..EEE
B.subtilis ZnuA/34-315	L..M..monocytogenes	ZnuA/32-312	VPSAERS	MGQOH	..AVFVN	ASKGID	DM
S.pneumoniae AdcA/35-310	VPSVLFV	LSRKR	..DVI	IDA	SAG	GESE..EEH	
S.agalactiae Lmb/30-305	AGSPVNL	IKRSK	..IVK	VLEAS	SGMT	EE..FED	
S.pyogenes Lmb/30-305	ARDLDPN	IKRSK	..VWD	VEASRPL	TD..	RVPG..LED	
S.aureus MntC/30-308	ADP	IKRSK	..VWD	VEASRPL	TD..	RVKG..LED	
L.monocytogenes MntA/30-308	FEDMLR	AGKSK	..KD	KVVA	ADDKPL	RVKG..LED	
S.pyogenes MtsA/31-308	FDMLR	DKGRED	DNV	LGSKGV	KPMY	EEGG..	
S.pneumoniae Psaa/30-307	FTL	LVKNAOKTKN	..KD	VFAV	SDGIDV	YI	EG..
Ca.L.asiaticus ZnuA/21-293	FTL	LVKNAOKTKN	..KD	VFAV	SDGIDV	YI	GA..
Synechocystis MntC/52-322	FEQFLG	ENV	..K	YTF	TDGINDIC	CS	GO..N..
Y.pestis Yfa/39-309	FEFPFF	IK..DV	..PSV	VLE	EGIEPI	IP	EDTS..
S.enterica Sita/27-297	FAFVYQH	IK..DV	..PSAV	VTAGIT	PLP	R	DGP..
S.meliloti Sita/29-299	FEFPFF	QNF	..P	PEV	VSTG	VPMG	ECP..
Rhizobium/Agrobacterium TroA/24-294	FEFPFF	QNF	..P	PGV	VSEGV	PEPMG	ECP..
N.gonorrhoeae MntC/19-302	DIQR	AVKQSKV	..P	PDV	TV	SDGIEPMA	GGA..
P.denitrificans AztC/24-306	FEFPFF	QNF	..P	SYA	ATFKGI	QPLK	E..EG..
Rhizobium/Agrobacterium ZnuA/26-324	LTL	LLIAASG	TDAA	AVATL	TDG	IEPME	.PGG..
C.trachomatis YtgA/38-320	LRLB	LLIDT	SGTKA	PVW	ELTKGV	KPLRS	DEPAGAHAP
S.suis TroA/40-316	MVEA	LEKRTGV	..P	KVVD	DLQRL	PS	HAPEEEEA
T.pallidum TroA/32-306	MGEVF	FSKLRGSR	..LV	VAV	SETIP	V	KN..
A.baumannii ZnuA/17-274	LNK	LLSLNNK	..K	KAL	LLDSGLS	ITP	
P.denitrificans ZnuA/22-323	LEDAAT	SUSAQS	..EMIAL	LLDLPATH	RRBD		ORNT..
B.abortus ZnuA/24-332	LDP	IDT	..LGE	KVVA	QDAKGLT	KLK	YAGG..
P.aeruginosa ZnuA/23-304	LPRV	LAGRQGT	..S	VAV	QDLPGMH	LRK	EHEHEH..EH
V.cholerae VC2081/21-294	MSK	LLLEGRT	..S	AL	TLSQVP	NLA	FREGGPFE..AHDHG
H.influenzae ZnuA/20-335	LDRK	PISOI	..K	V	ADLADV	KPL	Y..EEHAG..
Y.pestis ZnuA/25-317	LSKPL	TVAEN	..K	KQJ	IALSQL	PSVTP	FVN..FEE..EEHAG..
E.coli ZnuA/23-307	MOKPV	SKLPGA	..K	KQV	TAQLEDV	KPL	LRE..YG..
S.enterica ZnuA/42-330	MESV	SRV	..P	KQV	TAQLADV	KPL	MKSDEHDEAEGESGH..



Supplementary Figure S3. Genome neighborhood network where the hub node represents each group indicated in Figure 5. The gray spoke nodes indicate the prevalence of Pfam [69] protein family genes within 10 genes of *zrgA* in at least 20% of genomes.

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