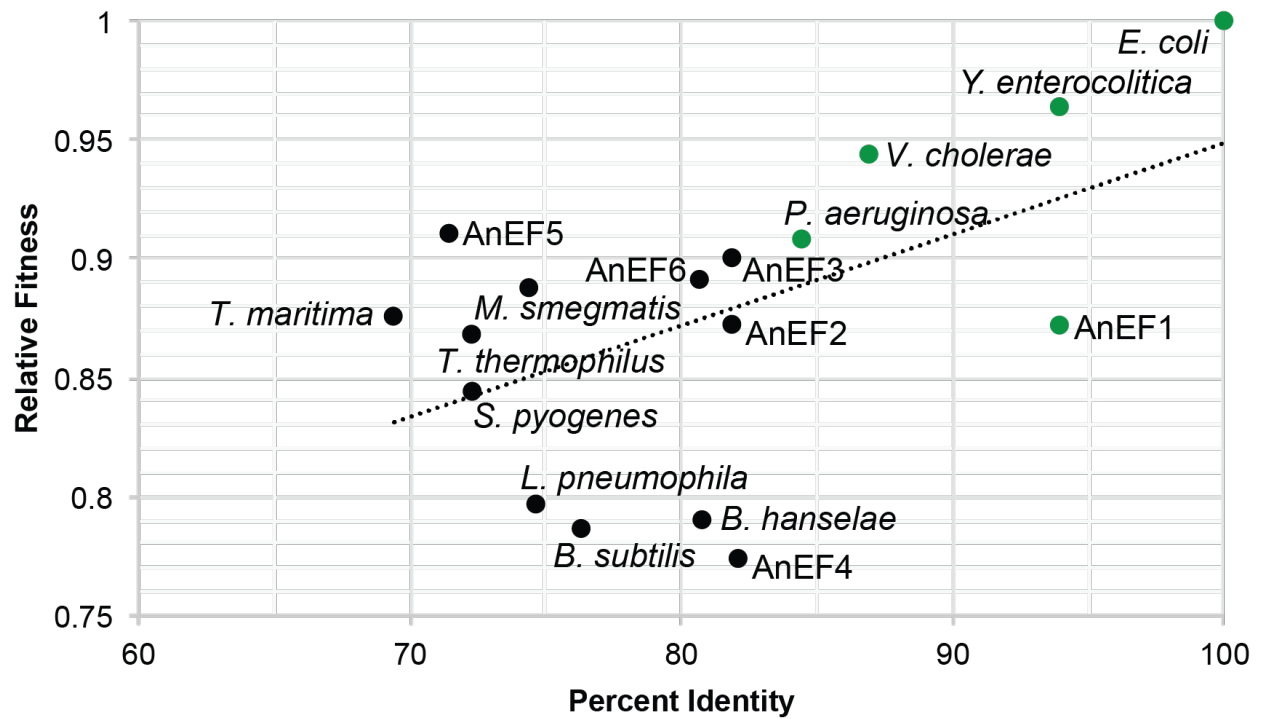
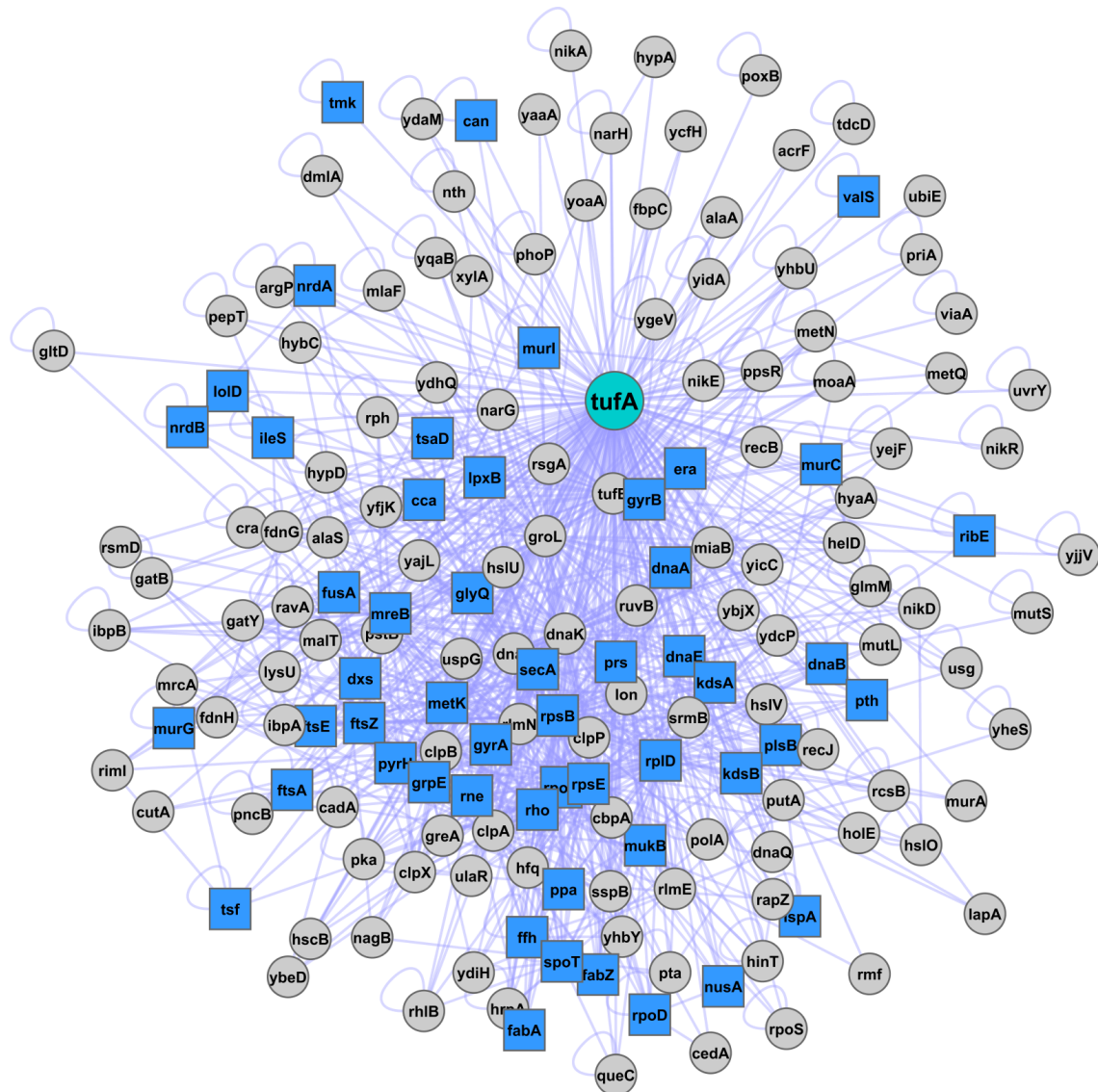


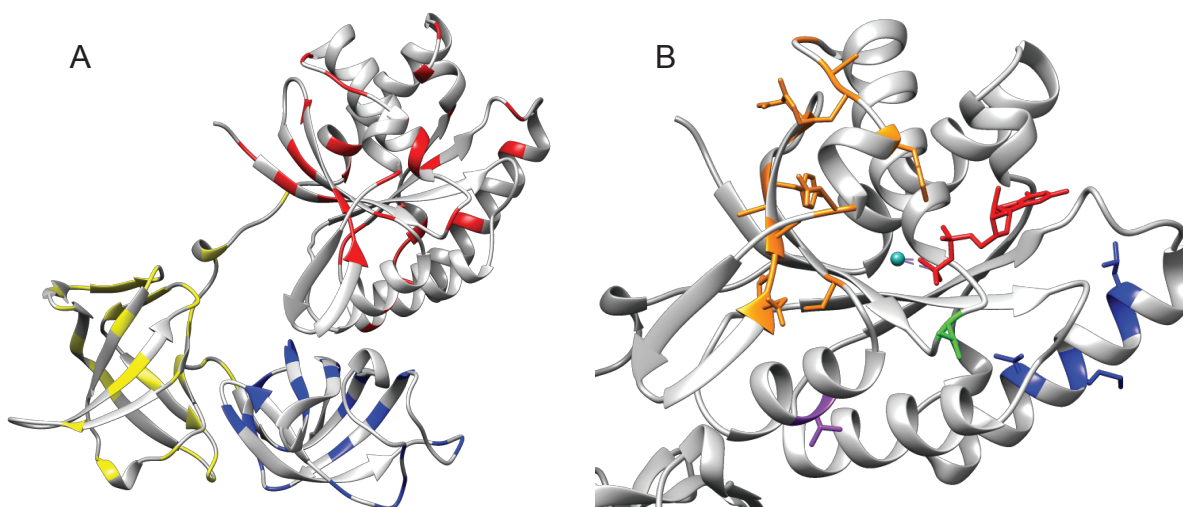
Supplementary Figure 1: Foreign *tuf* genes were integrated into the bacterial genome in a stepwise fashion as shown. A. Each of the foreign *tuf* genes was integrated into the *E. coli* chromosome (replacing *tufA*). B. In each strain carrying a foreign *tuf* gene at the *tufA* locus an attempt was made to remove the native *E. coli tufB* gene.



Supplementary Figure 2. Relative fitness of strains carrying two *tuf* genes (*tufA*-foreign and *tufB* *E. coli*) as a function of identity to *E. coli* EF-Tu (indicated as 100%). Strains shown in green carry a foreign *tuf* gene that can support viability when *E. coli* *tufB* gene has been deleted.



Supplementary Figure 3: The interaction partners of EF-Tu and all interactions among these first neighbors of EF-Tu. Nodes represent proteins (indicated by the relevant gene names) and each purple line represents an interaction. To easily distinguish *tufA* in the network, it has been colored cyan. Essential proteins are shaped rectangular and colored blue in this sub-network. Protein-protein interactions of *E. coli* were retrieved from the HitPredict database (Patil, et al. 2011) and the Cytoscape visualization tool was used to draw the network (Demchak, et al. 2014).



Supplementary Figure 4: Structural representation EF-Tu-GDP. EF-Tu is colored gray and residues that are conserved in all viable homologs but changed in at least one of the non-viable homologs are colored. A) Changes within the G-domain are colored in red, domain II in yellow and domain III in blue. B) A close-up view of the G-domain where the change in P-loop is shown in green, changes in the Effector region/Switch I are shown in orange, the change in Switch II in purple and changes in the D-helix in blue. GDP is shown in red and the Mg ion as a cyan sphere. Graphics were performed with the UCSF Chimera package (Pettersen, et al. 2004) using the PDB-ID: 1EFC (Song, et al. 1999).

Supplementary Table 1. List of organisms and plasmids serving as EF-Tu sources. *Synthetic gene sequences are the courtesy of EA Gaucher (Gaucher, et al. 2008).

Organism/node	Strain	Accession number	Annotation of gene
<i>Vibrio cholerae</i>	N16961 El Tor	NC_016445	<i>tufB</i>
<i>Mycobacterium smegmatis</i>	MC2 155	NC_018289	<i>tuf</i>
<i>Bacillus subtilis</i>	186	NC_000964	<i>tuf</i>
<i>Thermus thermophilus</i>	HB8	ATCC 27634	<i>tuf</i>
<i>Thermotoga maritima</i>	DSM3109 [MSB8]	ATCC 43589	<i>tuf</i>
<i>Streptococcus pyogenes</i>	MGAS5005	NC_007297	<i>tuf</i>
<i>Legionella pneumophila</i>	Philadelphia 1	AE017354	<i>tufB</i>
<i>Bartonella hansenellae</i>	Houston 1	BX897699	<i>tuf2</i>
<i>Yersinia enterocolitica</i>	8081	NC_008800	<i>tufA</i>
<i>Pseudomonas aeruginosa</i>	PAO1	NC_002516	<i>tufA</i>
Node 317 (700mya)	Synthetic gene*	N/A	AnEF1
Node 253 (~1.2bya)			AnEF2
Node (~1.8bya)			AnEF3
Node 168 (~2.5bya)			AnEF4
Node 184 (~3.5bya)			AnEF5
Node 262 (~2.5bya)			AnEF6

Supplementary Table 2: The elongation factor EF-Tu is among the top ten most connected proteins in the overall *E. coli* interactome. The proteins listed below are ranked according to their connectivity using the Uniprot database. The mean average degree of interaction in the whole interactome is 12.

Rank	Uniprot Code	Gene Name	Product Description	Degree of Connectivity
1	P0A6F5	<i>groL</i>	GroEL, chaperone Hsp60, peptide-dependent ATPase	702
2	P18843	<i>nadE</i>	NH(3)-dependent NAD(+) synthetase	483
3	P60422	<i>rplB</i>	50S ribosomal protein L2	386
4	P0A6Y8	<i>dnaK</i>	Chaperone protein DnaK	363
5	P60723	<i>rplD</i>	50S ribosomal protein L4	244
6	P0A7V8	<i>rpsD</i>	30S ribosomal protein S4	242
7	P0A7V3	<i>rpsC</i>	30S ribosomal protein S3	225
8	P0A7V0	<i>rpsB</i>	30S ribosomal protein S2	206
9	P0A7W1	<i>rpsE</i>	30S ribosomal protein S5	192
10	P0CE47	<i>tufA</i>	Elongation factor TuA	172

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Supplementary Table 3: Strains and genetic markers used in this work and their origin.

Strain	Species, Genotype or Characteristics
CH1464	<i>Escherichia coli</i> K12 MG1655
CH3139	<i>Pseudomonas aeruginosa</i> PAO1
CH3998	MG1655 <i>yfaH</i> ::[TP22-amilCP_opt-kan-sacB-T0]
CH1940	MG1655 / pSIM5-tet
CH4765	MG1655 <i>tufA</i> ::[TP22-amilCP_opt-kan-sacB-T0]
CH4766	MG1655 <i>tufB</i> ::[TP22-amilCP_opt-kan-sacB-T0]
CH4849	MG1655 <i>tufA</i> ::[TP22-amilCP_opt-kan-sacB-T0] / pSIM5-tet
CH4931	MG1655 <i>tufA</i> :: <i>tufA</i> from <i>Salmonella enterica</i> serovar Typhimurium LT2
CH4932	<i>Bacillus subtilis</i>
CH4935	<i>Streptococcus pyogenes</i>
CH4961	MG1655 <i>tufA</i> :: <i>tuf</i> from <i>Mycobacterium smegmatis</i>
CH4962	MG1655 <i>tufA</i> :: <i>tuf</i> from <i>Streptococcus pyogenes</i>
CH4963	MG1655 <i>tufA</i> :: <i>tuf</i> from <i>Thermus thermophilus</i>
CH4964	MG1655 <i>tufA</i> :: <i>tufB</i> from <i>Vibrio cholerae</i>
CH4965	MG1655 <i>tufA</i> :: <i>tuf</i> from <i>Bacillus subtilis</i>
CH4966	MG1655 <i>tufA</i> :: <i>tuf</i> from <i>Thermotoga maritima</i>
CH4967	MG1655 <i>tufA</i> ::AnEF4
CH4968	MG1655 <i>tufA</i> ::AnEF5

CH4969 MG1655 *tufA*::AnEF6

CH4970 MG1655 *tufA*::AnEF1

CH4971 MG1655 *tufA*::AnEF2

CH4972 MG1655 *tufA*::AnEF3

CH5016 MG1655 *tufB*::[TP22-amilCP_opt-kan-sacB-T0], *tufA*::*tufA* from *Salmonella enterica* serovar Typhimurium LT2

CH5018 MG1655 *tufB*::[TP22-amilCP_opt-kan-sacB-T0], *tufA*::*tuf* from *Mycobacterium smegmatis*

CH5019 MG1655 *tufB*::[TP22-amilCP_opt-kan-sacB-T0], *tufA*::*tuf* from *Streptococcus pyogenes*

CH5020 MG1655 *tufB*::[TP22-amilCP_opt-kan-sacB-T0], *tufA*::*tuf* from *Thermus thermophilus*

CH5021 MG1655 *tufB*::[TP22-amilCP_opt-kan-sacB-T0], *tufA*::*tufB* from *Vibrio cholerae*

CH5022 MG1655 *tufB*::[TP22-amilCP_opt-kan-sacB-T0], *tufA*::*tuf* from *Bacillus subtilis*

CH5023 MG1655 *tufB*::[TP22-amilCP_opt-kan-sacB-T0], *tufA*::*tuf* from *Termotoga maritima*

CH5024 MG1655 *tufB*::[TP22-amilCP_opt-kan-sacB-T0], *tufA*::AnEF4

CH5025 MG1655 *tufB*::[TP22-amilCP_opt-kan-sacB-T0], *tufA*::AnEF5

CH5026 MG1655 *tufB*::[TP22-amilCP_opt-kan-sacB-T0], *tufA*::AnEF6

CH5027 MG1655 *tufB*::[TP22-amilCP_opt-kan-sacB-T0], *tufA*::AnEF1

CH5028 MG1655 *tufB*::[TP22-amilCP_opt-kan-sacB-T0], *tufA*::AnEF2

CH5029 MG1655 *tufB*::[TP22-amilCP_opt-kan-sacB-T0], *tufA*::AnEF3

CH5088 MG1655 *tufB*::[TP22-amilCP_opt-kan-sacB-T0], *tufA*::*tufA* from *Salmonella enterica* serovar Typhimurium / pSIM5-tet

CH5089 MG1655 *tufB*::[TP22-amiCP_opt-kan-sacB-T0], *tufA*::*tufA* from *Vibrio cholerae* / pSIM5-tet

CH5091 MG1655 Δ *tufA*

CH5130 MG1655 *tufB*::[TP22-amiCP_opt-kan-sacB-T0], *tufA*::AnEF1 / pSIM5-tet

CH5132 MG1655 Δ *tufB*, *tufA*::*tufA* from *Salmonella enterica* serovar Typhimurium LT2

CH5133 MG1655 Δ *tufB*, *tufA*::*tufA* from *Vibrio cholerae*

CH5226 MG1655 *tufB*::[TP22-amiCP_opt-kan-sacB-T0] / pSIM5-tet

CH5227 MG1655 *tufA*::*tufB* from *Legionella pneumophila* Philadelphia 1

CH5228 MG1655 *tufA*::*tuf2* from *Bartonella henselae* Houston 1

CH5229 MG1655 *tufB*::*tuf* from *Bacillus subtilis*

CH5234 MG1655 Δ *tufB*

CH5235 MG1655 Δ *tufB*, *tufA*::AnEF1

CH5239 *Yersinia enterocolitica*

CH5240 MG1655 *tufB*::[TP22-amiCP_opt-kan-sacB-T0], *tufA*::*tufB* from *Legionella pneumophila* Philadelphia 1

CH5241 MG1655 *tufB*::[TP22-amiCP_opt-kan-sacB-T0], *tufA*::*tuf2* from *Bartonella henselae* Houston 1

CH5244 MG1655 *tufA*::*tufA* from *Yersinia enterocolitica*

CH5249 MG1655 *tufA*::*tufA* from *Pseudomonas aeruginosa*

CH5256 MG1655 *tufB*::[TP22-amiCP_opt-kan-sacB-T0], *tufA*::*tufA* from *Yersinia enterocolitica*

CH5258 MG1655 *tufB*::[TP22-amiCP_opt-kan-sacB-T0], *tufA*::*tufA* from *Pseudomonas*

aeruginosa

CH5269 MG1655 *tufB*::[TP22-amiI_{CP}_opt-kan-sacB-T0], *tufA*::*tufA* from *Yersinia enterocolitica* / pSIM5-tet

CH5271 MG1655 *tufB*::[TP22-amiI_{CP}_opt-kan-sacB-T0], *tufA*::*tufA* from *Pseudomonas aeruginosa* / pSIM5-tet

CH5277 MG1655 Δ *tufB*, *tufA*::*tufA* from *Yersinia enterocolitica*

CH5279 MG1655 Δ *tufB*, *tufA*::*tufA* from *Pseudomonas aeruginosa*

Supplementary Table 4. Sequences for ancestral *tuf* genes

Ancestral gene	Sequence
AnEF1	<p>gtgTCCAAAGAGAAATTTGAACGCACGAAACCACATGTAAACGTAGGTA CCATTGGCCACGTTGATCATGGCAAACCTACCCTGACTGCGGCAATTA CCACGGTACTGGCGAAAACCTTACGGCGGTGCAGCTCGTGCTTTTCGACC AGATCGATAACGCTCCGGAGGAGAAAGCTCGTGGCATCACCATCAATA CGTCCCACGTTGAATACGATACGCCTACCCGTCATTACGCTCATGTAGA TTGTCCGGGCCACGCGGACTATGTTAAGAACATGATCACGGGCGCAGC TCAGATGGACGGTGCAATCCTGGTTGTGGCTGCTACCGATGGCCCGAT GCCGCAGACGCGTGAACACATTCTGCTGGGCCGTCAGGTTGGCGTACC GTACATTATCGTGTCTTCTGAACAAATGCGACATGGTCGACGATGAAGA ACTGCTGGAGCTGGTCGAAATGGAGGTGCGCGAACTGCTGTCCAGTA CGACTTCCCGGGCGATGATACCCCGATCATCCGTGGTTCTGCTCTGAAG GCTCTGGAAGGCGAAGCAGAATGGGAGGCGAAAATTATCGAGCTGGC CGAAGCGCTGGACAGCTACATCCAGAACCGGAACGTGCAATCGATC AGCCGTTTCTGCTGCCGATTGAAGATGTTTTTCAGCATCTCCGGTCGTGG TACGGTAGTTACCGGCCGTGTGGAACGTGGCATCGTAAAAGTTGGTGA AGAAGTCGAAATTGTTGGTATCAAAGATACTACCAAACCTACCTGCAC CGGCGTGAAATGTTCCGCAAACCTGCTGGACGAAGGCCGTGCGGGCG AAAACGTAGGTGTCTGCTGCGCGGCACCAAACGCGACGAAATCGAG CGTGGTCAGGTTCTGGCGAAACCGGGCTCCATCACCCCTCACACGACT TTTGAATCCGAGGTGTACGTGCTGTCTAAAGACGAAGGTGGCCGTAC ACTCCATTCTTCAAGGGCTATCGTCCGCAGTTCTACTTTTCGCACCACGG ACGTGACCGGTACCATTTGAACTGCCTGAAGGTGTTGAGATGGTTATGC CGGGCGACAACATTAATAATGACCGTGACTCTGATCCACCCTATTGCGA TGGACGAAGGTCTGCGTTTTGCCATCCGTGAAGGTGGTCGTACCGTTG GCGCCGGTGTGGTTGCGAAAATCATCGCT_{taa}</p>
AnEF2	<p>gtgGCCAAAGAGAAATTTGAACGCACGAAACCACATGTAAACATCGGTA CCATTGGCCACGTTGATCATGGCAAACCTACCCTGACTGCGGCAATTA CCAAGGTAAGGCTCCGGAGGAGAAAGGCAAGCTGAATTTAAGGCTTACGACC AGATCGATAAAGGCTCCGGAGGAGAAAGAACGTGGCATCACCATCAGC ACGGCCCACGTTGAATACGAGACTGAAAACCGTCATTACGCTCATGTA GATTGTCCGGGCCACGCGGACTATGTTAAGAACATGATCACGGGCGCA GCTCAGATGGACGGTGCAATCCTGGTTGTGTCTGCTGCCGATGGCCCG ATGCCGCAGACGCGTGAACACATTCTGCTGGCCCGTCAGGTTGGCGTA CCGTACATTGTCTGTTTCTGAACAAAGTCGACATGGTCGACGATGAA GAACTGCTGGAGCTGGTCGAAATGGAGGTGCGCGAACTGCTGTCTCTCT TACGACTTCCCGGGCGATGATATCCCGATCATCAAAGGTTCTGCTCTGA AGGCTCTGGAAGGCGACGAAGAAGGCGAGGAGGCAATTATGAAGCTG ATGGACGCGGTGGACAGCTACATCCAGAACCGGAACGTGCAATCGAT AAGCCGTTTCTGATGCCGATTGAAGATGTTTTTCAGCATCTCCGGTCGTG GTACGGTAGTTACCGGCCGTGTGGAACGTGGCATCGTAAAAGTTGGTG AAGAAGTCGAAATTGTTGGTATCCGTGATACTCAGAAAACCTACCTGCA CCGGCGTGAAATGTTCCGCAAACCTGCTGGACGAAGGCCAAGCGGGC GACAACGTAGGTGTCTGCTGCGCGGCACGAAACGCGAAGATGTGCA GCGTGGTCAGGTTCTGGCCAAACCGGGCTCCATCAAACCTCACACGAA ATTTAAAGCCGAGGTGTACATCCTGACTAAAGAAGAAGGTGGCCGTCA CACTCCATTCTTCAACGGCTATCGTCCGCAGTTCTACTTTTCGCACCACG</p>

	GACGTGACCGGTGTGATTACCCTGCCTGAAGGTGTTGAGATGGTTATG CCGGGCGACAACGTTACTATCACCGTGGAACTGATCGCCCCTATTGCG ATGGAAGAAGGTCTGCGTTTTGCCATTTCGTGAAGGTGGTCGTACCGTT GGCGCCGGTGTGGTTTTCCGAAATCATCGAAtaa
AnEF3	gtgGCCAAAGAGAAATTTGAACGCACGAAACCACATGTAAACATCGGTA CCATTGGCCACGTTGATCATGGCAAACCTACCCTGACTGCGGCAATTA CCAAGTTCTGGCGGAGAAAGGCAAAGCTGAATTTTCGTGCTTACGACC AGATCGATAAGGCTCCGGAGGAGAAAGAGCGTGGCATCACCATCAAC ACGGCCCACGTTGAATACGAGACTGAAAACCGTCATTACGCTCATGTA GATTGTCCGGGCCACGCGGACTATGTTAAGAACATGATCACCGGCGCA GCTCAGATGGACGGTGCAATCCTGGTTGTGTCTGCTGCCGATGGCCCG ATGCCGCAGACGCGTGAACACATTCTGCTGGCCCGTCAGGTTGGCGTA CCGTACATTGTTCGTGTTTCTGAACAAATGCGACATGGTCGACGATGAA GAACTGCTGGAGCTGGTTCGAAATGGAGGTGCGCGAACTGCTGTCCGAG TACGACTTCCCGGGCGATGATATCCCGATCATCCGCGGTTCTGCTCTGA AGGCTCTGGAAGGCGATGAAGAATGGGTGGAAAAGATTATGGAGCTG ATGGACGCGGTGGACAGCTACATCCCAGAACCAGGAACTGACATCGAT AAGCCGTTTCTGATGCCGATTGAAGATGTTTTCAGCATCACCGGTCGTG GTACGGTAGTTACCGGCCGTGTGGAACGTGGCATCGTAAAAGTTGGTG ACGAAGTCGAAATTGTTGGTCTGCGTGATACTCGTAAAACCTGTCGTCA CCGGCGTGGAAATGTTCCGCAAACCTGCTGGACGAAGGCCAGGCGGGC GACAACGTAGGTGTCCTGCTGCGCGGCATCAAACGCGAAGATGTGAG CGTGGTCAGGTTCTGGCCAAACCGGGCTCCATCAAACCTCACACGAAA TTAAAGCCGAGGTGTACGTGCTGACTAAAGAAGAAGGTGGCCGTCAC ACTCCATTCTTCAACGGCTATCGTCCGAGTTCTACTTTCGACCACGG ACGTGACCGGTGTGATTACCCTGCCTGAAGGTGTTGAGATGGTTATGC CGGGCGACAACGTTACTATGACCGTGGAACTGATCCACCCTATTGCGA TGGAAGAAGGTCTGCGTTTTGCCATCCGTGAAGGTGGTCGTACCGTTG GCGCCGGTGTGGTTTTCCGAAATCATCGAAtaa
AnEF4	gtgGCCAAAGAGAAATTTGTACGCACGAAACCACATGTAAACGTCGGTA CCATTGGCCACGTTGATCATGGCAAATCTACCCTGACTGCGGCAATTA CCAAGTATCTGTCTCTGAAAGGCCTGGCTCAATATGTTTCCTTACGACCA GATCGATAAGGCTCCGGAGGAGAAAGCTCGTGGCATCACCATCAACAT TACCCACGTTGAATACGAGACTGAGAAACGTCATTACGCTCATATCGA TTGTCCGGGCCACGCGGACTATATTAAGAACATGATCACCGGCGCAGC TCAGATGGACGGTGCAATCCTGGTTGTGGCTGCTACCGATGGCCCGAT GCCGCAGACGCGTGAACACGTTCTGCTGGCCCGTCAGGTTGGCGTACC GTACATGATCGTGTATCAACAAAACCGACATGGTCGACGATCCAGA ACTGATCGAGCTGGTCGAAATGGAGGTGCGCGATCTGCTGTCCCAGTA CGAATATCCGGGCGATGAAGTCCCGGTCATCAAGGTTCTGCTCTGAA GGCTCTGGAAGCCAACCATGAAGCGTATAAACCAATTCAGGAGCTGCT GGACGCGATGGACAACCTACATCCCAGACCCGCAACGTGACGTCGATA AGCCGTTTCTGATGCCGATTGAAGATGTTTTTCAGCATCACCGGTCGTGG TACGGTAGTTACCGGCCGTATCGAACGTGGCCGCATCCGTCCTGGTGA CGAAGTCGAAATTATTGGTCTGTCTGAGATTCGTAAAACCTGTCGTCACC AGCGTGGAATGTTCCGCAAAGAGCTGGACGAAGGCATTGCGGGCGA CAACGTAGGTTGCCTGCTGCGCGGCATCGACAAAGATGAAGTCGAGCG TGGTCAGGTTCTGGCCGCACCGGGCTCCATCAAACCTCACAAGCGTTTT AAAGCCGAGGTGTACGTGCTGAAGAAAGAAGAAGGTGGCCGTCACAC TCCATTCTTCAAAGGCTATAAACCGCAGTTCTACATTCGACCACGGAC

	GTGACCGGTGAGATTGTCCTGCCTGAAGGTGTTGAGATGGTTATGCCG GGCGACCACGTTGAAATGGAAATCGAACTGATCTACCCTGTTGCGATT GAAAAGGGTCAGCGTTTTTGCCATCCGTGAAGGTGGTTCGTACCGTTGGC GCCGGTGTGGTTACCGAAGTCATCGAAtaa
AnEF5	gtgGCCAAAGAGAAATTTGAACGCACGAAACCACATGTAAACATCGGTA CCATTGGCCACGTTGATCATGGCAAACCTACCCTGACTGCGGCAATTA CCAAGACTCTGGCGGCGAAAGGCAAAGCTGAAGCTCGTGCTTACGACC AGATCGATAAGGCTCCGGAGGAGAAAGCTCGTGGCATCACCATCAAC ACGGCCCACGTTGAATACGAGACTGAAAACCGTCATTACGCTCATGTA GATTGTCCGGGCCACGCGGACTATGTTAAGAACATGATCACCGGCGCA GCTCAGATGGACGGTGCAATCCTGGTTGTGTCTGCTGCCGATGGCCCG ATGCCGCAGACGCGTGAACACATTCTGCTGGCCCGTCAGGTTGGCGTA CCGTACATTGTCGTGTTTCTGAACAAATGCGACATGGTCGACGATGAA GAACTGCTGGAGCTGGTTCGAAATGGAGGTGCGCGAACTGCTGTCCGAG TACGAATTTCCGGGCGATGATATCCCGATCATCCGCGGTTCTGCTCTGA AGGCTCTGGAAGGCGAAAATGAATGGGTGGACAAAATTTGGGAGCTG ATGGACGCGGTGGACAGCTACATCCCAACCCCGGAACGTGACGTCGAT AAGCCGTTTCTGATGCCGTTGAAGATGTTTTACCATCACCGGTTCGTG GTACGGTAGCTACCGGCCGTGTGGAACGTGGCACCGTAAAAGTTGGTG ACGAAGTCGAAATTGTTGGTCTGCGTGATACTCGTAAAACCTGTCTGTA CCGGCGTGGAAATGTTCCGCAAACCTGCTGGACGAAGGCATGGCGGGC GACAACGTAGGTGTCCTGCTGCGCGGCATCCAACGCGAAGATGTTCGAG CGTGGTCAGGTTCTGGCCAAACCGGGCTCCATCAAACCTCACACGAAA TTTGAAGCCGAGGTGTACGTGCTGACTAAAGAAGAAGGTGGCCGTCAC ACTCCATTCTCAACGGCTATCGTCCGAGTTCTACTTTCGACCACGG ACGTGACCGGTGTGATTACCCTGCCTGAAGGTGTTGAGATGGTTATGC CGGGCGACAACGTTACTATGACCGTGGAACCTGATCCACCCTATTGCGA TGGAAGAAGGTCTGCGTTTTTGCCATCCGTGAAGGTGGTTCGTACCGTTG GCGCCGGTGTGGTTTCCAAAATCATCGAAtaa
AnEF6	gtgGCCAAAGAGAAATTTGAACGCACGAAACCACATGTAAACATCGGTA CCATTGGCCACGTTGATCATGGCAAACCTACCCTGACTGCGGCAATTA CCAAGACTCTGGCGGCGAAAGGCAAAGCTGAAGCTCGTGCTTACGACC AGATCGATAAGGCTCCGGAGGAGAAAGCTCGTGGCATCACCATCAAC ACGGCCCACGTTGAATACGAGACTGAAAACCGTCATTACGCTCATGTA GATTGTCCGGGCCACGCGGACTATGTTAAGAACATGATCACCGGCGCA GCTCAGATGGACGGTGCAATCCTGGTTGTGTCTGCTGCCGATGGCCCG ATGCCGCAGACGCGTGAACACATTCTGCTGGCCCGTCAGGTTGGCGTA CCGTACATTGTCGTGTTTCTGAACAAATGCGACATGGTCGACGATGAA GAACTGCTGGAGCTGGTTCGAAATGGAGGTGCGCGAACTGCTGTCCGAG TACGAATTTCCGGGCGATGATATCCCGATCATCCGCGGTTCTGCTCTGA AGGCTCTGGAAGGCGAAAATGAATGGGTGGACAAAATTTGGGAGCTG ATGGACGCGGTGGACAGCTACATCCCAACCCCGGAACGTGACGTCGAT AAGCCGTTTCTGATGCCGTTGAAGATGTTTTACCATCACCGGTTCGTG GTACGGTAGCTACCGGCCGTGTGGAACGTGGCACCGTAAAAGTTGGTG ACGAAGTCGAAATTGTTGGTCTGCGTGATACTCGTAAAACCTGTCTGTA CCGGCGTGGAAATGTTCCGCAAACCTGCTGGACGAAGGCATGGCGGGC GACAACGTAGGTGTCCTGCTGCGCGGCATCCAACGCGAAGATGTTCGAG CGTGGTCAGGTTCTGGCCAAACCGGGCTCCATCAAACCTCACACGAAA TTTGAAGCCGAGGTGTACGTGCTGACTAAAGAAGAAGGTGGCCGTCAC ACTCCATTCTCAACGGCTATCGTCCGAGTTCTACTTTCGACCACGG

	ACGTGACCGGTGTGATTACCCTGCCTGAAGGTGTTGAGATGGTTATGC CGGGCGACAACGTTACTATGACCGTGGAAGTATCCACCCTATTGCGA TGGAAGAAGGTCTGCGTTTTGCCATCCGTGAAGGTGGTCGTACCGTTG GCGCCGGTGTGGTTCCAAAATCATCGAAtaa
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Supplementary Table 5. Primers used in this study

Oligo name	Oligo sequence (5' to 3')
tufB::kansacolor_fw	GTGATATCACCGATTTATCCGTGTCTTAGAGGGACAATCGATCA AAGGGAAAACGTGCCA
tufB::kansacolor_rv	ATGCCCTTTTAGTGCGCATTGCGTCAAATGTTATCGGCCAAAAA TGAGACGTTGATCGGCACG
tufB_coli_fw	TTCTTTTCTCCTCCCTGT
tufB_coli_rv	GGCAAACCAAATCGAAAC
tufA::topo1.317_fw	CCGAAGCGCCCTTTTCAATTCAAACAACTAATTAACGTGTAAttaAGCGATGATTTT CGCAACCA
tufA::topo1.317_rv	CTCTCCTGAAGGGGAGAGCACTATAGTAAGGAATATAGCCgtgTCCAAAGAGA AATTTGAACG
tufA::topo2.262_fw	CCGAAGCGCCCTTTTCAATTCAAACAACTAATTAACGTGTAAttaTTCGATGATTTTG GAAACCA
tufA::topo2.262_rv	CTCTCCTGAAGGGGAGAGCACTATAGTAAGGAATATAGCCgtgGCCAAAGAGA AATTTGAACG
tufA::topo2.253_fw	CCGAAGCGCCCTTTTCAATTCAAACAACTAATTAACGTGTAAttaTTCGATGATTTT GGAAACCA
tufA::topo2.253_rv	CTCTCCTGAAGGGGAGAGCACTATAGTAAGGAATATAGCCgtgGCCAAAGAGA AATTTGAACG
tufA::topo1.170_fw	CCGAAGCGCCCTTTTCAATTCAAACAACTAATTAACGTGTAAttaTTCGATGATTTT GGAAACCA
tufA::topo1.170_rv	CTCTCCTGAAGGGGAGAGCACTATAGTAAGGAATATAGCCgtgGCCAAAGAGA AATTTGAACG
tufA::topo1.168_fw	CCGAAGCGCCCTTTTCAATTCAAACAACTAATTAACGTGTAAttaTTCGATGACTTC GGTAACCA
tufA::topo1.168_rv	CTCTCCTGAAGGGGAGAGCACTATAGTAAGGAATATAGCCgtgGCCAAAGAGA AATTTGTACG
tufA::topo2.184_fw	CCGAAGCGCCCTTTTCAATTCAAACAACTAATTAACGTGTAAttaTTCGATGATTTTG GAAACCA
tufA::topo2.184_rv	CTCTCCTGAAGGGGAGAGCACTATAGTAAGGAATATAGCCgtgGCCAAAGAGA AATTTGAACG
tufA::tufA.sty_fw	CCGAAGCGCCCTTTTCAATTCAAACAACTAATTAACGTGTAATTAGCCCAGAACTTT AGCAA
tufA::tufA.sty_rv	CTCTCCTGAAGGGGAGAGCACTATAGTAAGGAATATAGCCGTGTCTAAAGAAA AATTTGA
tufA::tufB.vcholerae_fw	CCGAAGCGCCCTTTTCAATTCAAACAACTAATTAACGTGTAATTAAGCGATGATCT TAGCTACAA
tufA::tufB.vcholerae_rv	CTCTCCTGAAGGGGAGAGCACTATAGTAAGGAATATAGCCGTGTCTAAAGAGA AATTTGAACG
tufA::tuf.msmeg_fw	CCGAAGCGCCCTTTTCAATTCAAACAACTAATTAACGTGTAATTACTTGATGATCTT GGTGACGC
tufA::tuf.msmeg_rv	CTCTCCTGAAGGGGAGAGCACTATAGTAAGGAATATAGCCGTGGCGAAGGCG AAGTTCCGACG

Kacar, *et.al.* Supporting Information

tufA::tuf.bsub_fw	CGAAGCGCCCTTTTCAATTCAAACAACTAATTAACGTGTAATTACTCAGTGATTGTA GAAACAAC
tufA::tuf.bsub_rv	CTCTCCTGAAGGGGAGAGCACTATAGTAAGGAATATAGCCGTGGCTAAAGAA AAATTGACCG
tufA::tuf.thermophilus_fw	CCGAAGCGCCCTTTTCAATTCAAACAACTAATTAACGTGTAATTATCCAGGATTTT GGTAACCA
tufA::tuf.thermophilus_rv	CTCTCCTGAAGGGGAGAGCACTATAGTAAGGAATATAGCCGTGGCCAAAGGT GAATTTATCCG
tufA::tuf.maritima_fw	CCGAAGCGCCCTTTTCAATTCAAACAACTAATTAACGTGTAATTATTCGATGACTTC GGTAACCA
tufA::tuf.maritima_rv	CTCTCCTGAAGGGGAGAGCACTATAGTAAGGAATATAGCCGTGGCCAAAGAG AAATTTGTACG
tufA::tuf.spyogenes_fw	CCGAAGCGCCCTTTTCAATTCAAACAACTAATTAACGTGTAATTAAGCTTCGATTTTC TGAAACGA
tufA::tuf.spyogenes_rv	CTCTCCTGAAGGGGAGAGCACTATAGTAAGGAATATAGCCGTGGCAAAGAA AAATACGATCG
del_kansacblue_tufB_coli	CTTTTAGTGCGCATTGCGTCAAATGTTATCGGCAACGATTGTCCCTCTAAGACA CGGATAAATCGGTGAT
del_kansacblue_tufA_coli	GCGCCCTTTTCAATTCAAACAACTAATTAACGTGTAAGGCTATATTCTTACTATAG TGCTCTCCCCTTCAG
tufs_check_fw	CAAAACAAAAGTCCTACGCA
tufs_check_rv	AGTATGATGAAGCGCCGA
check_deltufA_coli_fw	GTCAACAAATGCAAAAAGGG
check_deltufA_coli_rv	AAACTCCGGAAGAGAACAC
check_deltufB_coli_fw	TCCCCACCACCAATTTTC
check_deltufB_coli_rv	CCCACTTCATCGCTTCCA
tufA::tuf.legionella_fw	CCGAAGCGCCCTTTTCAATTCAAACAACTAATTAACGTGTAATTACTCGATTATTTT AGCGA
tufA::tuf.legionella_rv	TCTCTCCTGAAGGGGAGAGCACTATAGTAAGGAATATAGCCgTGGCGAAGGAA AAATTTGAACG
tufA::tuf.bartonella_fw	CCGAAGCGCCCTTTTCAATTCAAACAACTAATTAACGTGTAATTACTCAATGATCTT AGAAA
tufA::tuf.bartonella_rv	CTCTCCTGAAGGGGAGAGCACTATAGTAAGGAATATAGCCgTGGCGAAGAGC AAATTTGAACG
tufA::tuf.yersinia_fw	CCGAAGCGCCCTTTTCAATTCAAACAACTAATTAACGTGTAATTAAGCGATAACTTT AGCAA
tufA::tuf.yersinia_rv	CTCTCCTGAAGGGGAGAGCACTATAGTAAGGAATATAGCCGTGTCTAAAGAAA AATTTGAACG
tufA_yersinia_fw	TTGAATGGTATGTGGGTG
tufA_yersinia_rv	TGGAGTTCCTGAAGTATG
tufA::tuf.pseudomonas_fw	CCGAAGCGCCCTTTTCAATTCAAACAACTAATTAACGTGTAATTACTCGATGATCTT GGCAA
tufA::tuf.pseudomonas_rv	ATGCCCTTTTAGTGCGCATTGCGTCAAATGTTATCGGCAATTACTCGATGATCTT GGCAA
tufA_pseudomonas_fw	CCGGATTTGTTGGTTTTG

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tufA_pseudomonas_rv	TGGGTGAGATGTTTCGGTT
tufA_coli_qPCR_fw	ACTTTGATGATACCGCGTT
tufA_coli_qPCR_rv	ATTGACAAGCCGTTCTG
tufA_sty_qPCR_fw	ACATTTCAACGCCAGTACAG
tufA_sty_qPCR_rv	CGTTGTTACCGGTCGTGT
tufA_yersinia_qPCR_fw	GATTGGCAGCAGGAATGG
tufA_yersinia_qPCR_rv	AGGCGTTAAAGAGTGGGA
tufB_vibrio_qPCR_fw	CGATTGGCATCAGGAATG
tufB_vibrio_qPCR_rv	GTGGGAAGCGAAGATTGT
tufA_pseudo_qPCR_fw	CCAACGTTCTCACCAGCA
tufA_pseudo_qPCR_rv	AAGGTCCAGGAAGAAGTG
tuf_317_qPCR_fw	CGGTGCAGGTAGTTTTGGT
tuf_317_qPCR_rv	TTTCAGCATCTCCGGTCGT

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