

**Table S1. Oligonucleotides used in this study**

Oligonucleotide	Sequence <sup>a</sup>	Reference
536-Nco-for	5' - AGA <u>CCA TGG</u> TTG AGA ATA ATG CGC TGA G	(Aoki <i>et al.</i> , 2010)
536-Xho-rev	5' - GAT <u>CTC GAG</u> TAC AAT TAT CTG ATT GAT TTT T	(Aoki <i>et al.</i> , 2010)
DUF-536-Nco-for	5' - TCT <u>CCA TGG</u> GCG TAG ATC CGT CGA AAC TGA C	This study
VENN-less-Nco-for	5' - AAT <u>GCC ATG GGT</u> CTG GTT GCC AGA GG	This study
tRNase-Nco-for	5' - ATA <u>CCA TGG</u> GTT CCG GGG CTG CCT C	This study
536-Cys(mut)	5' - CTT TAG TCC TGC TAG GTG CTG CGA CCG CAC TGC CTC TGG CAA C	This study
pET-Sph/Pst	5' - CAA GGA ATG GT <u>G CAT GCC TGC AGA</u> TGG CGC CC	(Garza-Sánchez <i>et al.</i> , 2006)
pET-Kpn/Nco	5' - AGT CCA TGG TAC CTC TCC TTC TTA AAG	This study
536-H178A-for	5' - GGA GGA TAT TGG GAT GCT ATG CAG GAA ATG C	(Diner <i>et al.</i> , 2012)
536-CT-Xho-rev	5' - TTA <u>CTC GAG</u> GTA ATC ATA TTC CAT A	This study
536-cdil-Eco-for	5' - AGG <u>GAA TTC</u> CAT ATG ATT ACC TTA CGT AAA	This study
536-cdil-Spe-rev	5' - GTG ACT AGT TAC AAT TAT CTG ATT GAT TT	This study
colE5-M429-Nco	5' - CTG CCA TGG AAA GCA GGA AGA AG	This study
imE5-Spe	5' - AAA ACT AGT CAT CTT TAA CGT GAT AAT GAA AGC	This study
immE5-Eco-for	5' - GGA <u>GAA TTC</u> TCT ATG AAG TTA TCA CC	This study
pET-Pst-rev	5' - CGG <u>CTG CAG</u> CAG CCA ACT CAG TGG	This study

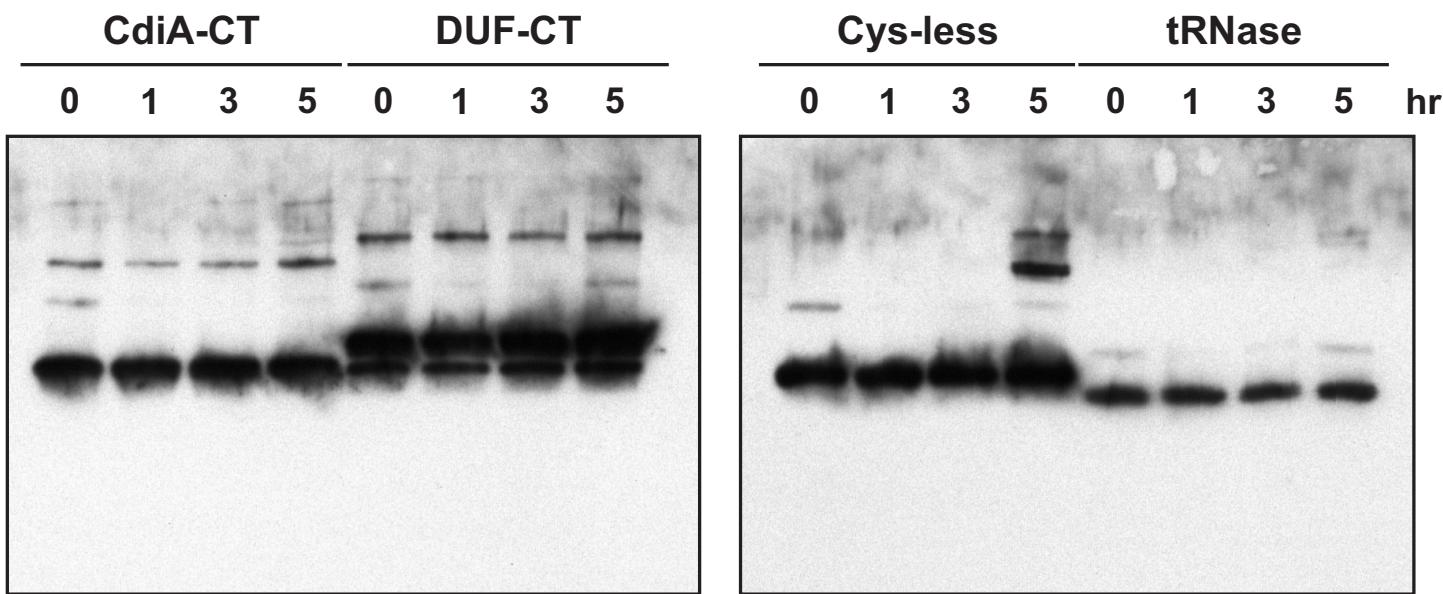
colE5-Nco-for	5' - GAG TTT <u>CCA TGG</u> GCG GTG GCG ATG G	This study
colE5-Bam-rev	5' - AGC <u>GGA TCC</u> TTT ATT ATC CTC TTT CTT CTT C	This study
colE5-Bam-for	5' - AAA <u>GGA TCC</u> AGA GAT GCT GAA GGC AAA C	This study
immE5-Xho-rev	5' - CAC <u>CTC GAG</u> CAT CTT TAA CGT GAT AAT G	This study
536-Bam-rev	5' - GCC <u>CCG GAT CCG</u> TAC TTA TC	This study
traA-Nco-for	5' – TAA <u>CCC ATG</u> GTT GCT GTT TTA AGT GTT CAG G	This study
traA-Spe-rev	5' – TTT ATA <u>CTA GTT</u> CAG AGG CCA ACG ACG GCC ATA CC	This study
traA-D74G	5' - TTC GGT AAG GGC TCC AGT GTT GTT AAA TGG G	This study
traA-G120C-Spe	5' - TAT <u>ACT AGT</u> TCA GAG GCA AAC GAC GGC C	This study
ECL-CT-Kpn-for	5' - GGG <u>GGT ACC</u> ATG GCT GAG AAT AAC TCG CTG GC	This study
ECL-cdil-Nhe-rev	5' - CTC <u>TGC TAG</u> CGT TGT TAA GAC TAT GAT AAA AAT C	This study
ECL-cdil-Eco-for	5' - GAT TAA <u>GGA ATT</u> CTG GTA TGT TTG G	This study
ECL-cdil-Sac-rev	5' - AGT <u>GAG CTC</u> TGT TTC AGT TGT TAA GAC	This study
argQ probe	5' - CCT CCG ACC GCT CGG TTC G	(Garza-Sánchez et al., 2006)
tyrU probe	5' - CTT CGA AGT CTG TGA CGG CAG	(Nikolakakis et al., 2012)
traA-cat-for	5' - ACA TTT AAT ACA CTC TAG TTT TAT TCA TTT ATC CGA AAT TGA GGT AAC TTA TGG AGA AAA AAA TCA CTG GAT ATA CC	This study
traA-cat-rev	5' - GGA AAC GAT ATT TCT TAA GTT TAT TCT CGT CTC CCG ACA TCG TTT TAT TTC CTG TTA CGC CCC GCC CTG CCA CTC	This study

traT-cat-for	5' - AAA ACA AGA AGT TAT CAA GAG TAA AAT AAA AGA TAT TAG AGA GTA AAT ATA TGG AGA AAA AAA TCA CTG GAT ATA CC	This study
traT-cat-rev	5' - CAA AGC GAG GCG TCA GTC AGG AGG CCG GTC AGA CCA GCC TCC GGA AGA TAA TTA CGC CCC GCC CTG CCA CTC	This study
trbl-cat-for1	5' - GGA AGA ACA TGA GAA ATA CAG GAG TGT GGC ATG AGT GAG AAA AAA ATC ACT GGA TAT ACC	This study
trbl-cat-for2	5' - AAG AAG TCA GGG CCG GAA ATG GCT TCG CTG GAA GCC TGG CTG GAA GAA CAT GAG AAA TAC	This study
trbl-cat-rev1	5' - GCC CCC ATA TCA GCA GGG CAA TCA GCC CCC GGC ATC TCA TGC CCC TCC CTG CCA CTC ATC	This study
trbl-cat-rev2	5' - CAC AGA TCG CCC CAG GTA CCA AGA TCG GCG GCG GCC ACA CTC TGC CCC CAT ATC AGC AGG	This study
trbl-Kpn-for	5' - TTT <u>GGT ACC</u> ATG AGT TCA ACG CAG AAC	This study
trbl-Xho-rev	5' - TTT <u>CTC GAG</u> TCT CAT GGT TCC GCC CTC	This study

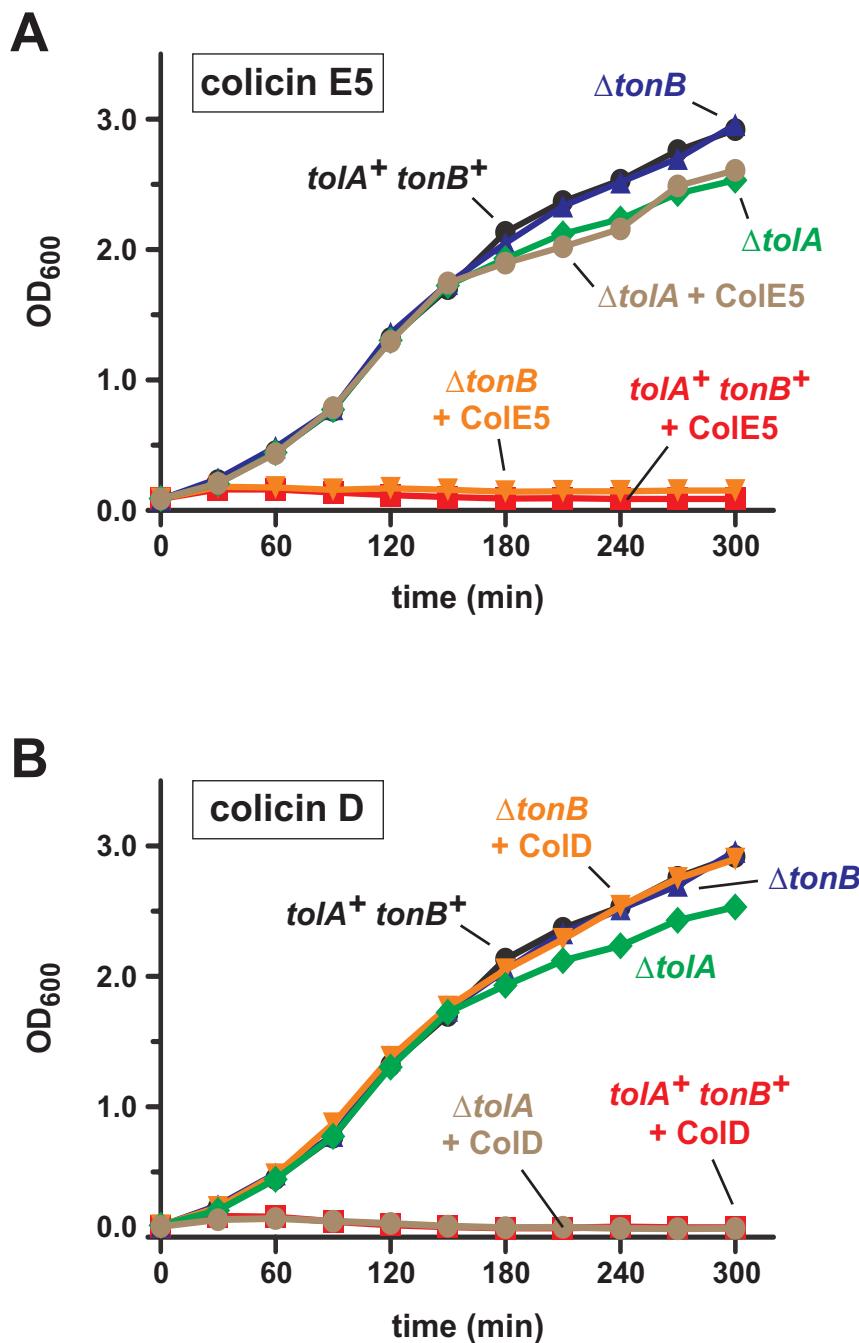
<sup>a</sup>Underlined residues correspond to restriction endonuclease sites.

## References

- Aoki, S. K., E. J. Diner, C. T. de Roodenbeke, B. R. Burgess, S. J. Poole, B. A. Braaten, A. M. Jones, J. S. Webb, C. S. Hayes, P. A. Cotter & D. A. Low, (2010) A widespread family of polymorphic contact-dependent toxin delivery systems in bacteria. *Nature* 468: 439-442.
- Diner, E. J., C. M. Beck, J. S. Webb, D. A. Low & C. S. Hayes, (2012) Identification of a target cell permissive factor required for contact-dependent growth inhibition (CDI). *Genes Dev* 26: 515-525.
- Garza-Sánchez, F., B. D. Janssen & C. S. Hayes, (2006) Prolyl-tRNA(Pro) in the A-site of SecM-arrested ribosomes inhibits the recruitment of transfer-messenger RNA. *J Biol Chem* 281: 34258-34268.
- Nikolakakis, K., S. Amber, J. S. Wilbur, E. J. Diner, S. K. Aoki, S. J. Poole, A. Tuanyok, P. S. Keim, S. Peacock, C. S. Hayes & D. A. Low, (2012) The toxin/immunity network of *Burkholderia pseudomallei* contact-dependent growth inhibition (CDI) systems. *Mol Microbiol* 84: 516-529.



**Figure S1. CdiA-CT<sup>536</sup> toxins are stable in *E. coli* cultures.** Purified CdiA-CT<sup>536</sup> toxins were added at 100 nM to *E. coli* X90 cultures and incubated with shaking at 37 °C. Samples were removed at the indicated times and the culture supernatant analyzed by immunoblot using polyclonal antibodies to CdiA-CT<sup>536</sup>.



**Figure S2.** *E. coli*  $\Delta\text{tolA}$  and  $\Delta\text{tonB}$  mutants are resistant to colicin E5 and D, respectively.

**A)** *E. coli* X90 was treated with colicin E5 (ColE5, group A) at 0 min and cell growth monitored by optical density at 600 nm ( $\text{OD}_{600}$ ). **B)** *E. coli* X90 was treated at 0 min with colicin D (ColD, group B) and cell growth monitored by  $\text{OD}_{600}$ . Strains carried  $\Delta\text{tolA}$  or  $\Delta\text{tonB}$  deletions where indicated.