

Table S1. DNA primers used in this study.

<u>Name</u>	<u>Sequence</u>
<u><i>ΔydcI</i></u>	
P1ydcIkd3	GTC TGT TTG AAC GCG GGC GAC TGG GCG CGC AAT TAA CGG TGT GTA GGC TGG AGC TGC TTC
Kd3ydcIp2	GAC AGC CGC TAT CAC AGC GGC ATC GAT TTA CGG ATC GCC GCA TAT GAA TAT CCT CCT TAG TTC C
<u><i>lacZ</i> fusion</u>	
YdcIendP1	GCGGCTGTCGGAGAATAAACGCGCCCGCGGCTAAAATAGCTGTGTA GGCTGGAGCTGCTTC
YdcItrgP2	CATGATACTGGCGCGACGATGCGATCATCCTTCCTTTAGTCATATG AATATCCTCCTTAGTTCC
<u>Gel shift probes</u>	
For H3: 5Yhalf	CTTAACGGCAGTTGAATCATCAGTAAA
ydcIprom3	CGCAACCGAATGCGCTGACTAAACAGA
For H5: 5ydcIprom	ATCTCATCTGCCGTGATGGTGTTCGCC
Yhalf3	CGTGTTAAGCCGCGATCTGGTCGG
For 362-bp: 5ydcIprom	ATCTCATCTGCCGTGATGGTGTTCGCC
ydcIprom3	CGCAACCGAATGCGCTGACTAAACAGA
For <i>lacZ</i> : 150lacZ5	TGTGCGGCGAGTTGCGTGACTACCTAC
3lacZ150	TTCGACGTTTCAGACGTAGTGTGACGCG

RT-PCR

S. typhimurium

5ydcI CTGAACGAACTGGAACAACCTCACC

ydcI3a CATCGTATTGTTCATGGTCGCGACCTG

S. typhimurium NCR

5dcuSint CCGTGATGGGTAATTTATTTTCATCAGAATTAC

intdcuS3 GAAAACTAATCATAATAGATTAAATATTAGCGCTG

S. typhi

Ty5 ATGATGAATATCTCCCAGCCGGCGCTATCG

3Ty CAGGCTCAGAATCGCCTGTGATACGCAGTC

P. mirabilis

PM5 CCGCAAGGCTAAGCAATTGAATAATTTTCATC

3PM TCGGTAATACGGTGTTACCTGATAATTTGC

S. marcescens

SER5 GACTGAGCATGGTATTCAGCTTCTCGGTTATGCC

3SER CATCGTCGTGCTGCGCTTTACGCGCACGTC

K. pneumoniae

KP5 ATCGACATCGGCATTGGACGTATGTCCGAC

3KP GCGCGACAGGGAGGCGGAGAGGGTCTCGAT

C. koseri

CK5 AACCCTTAACCTGAGTCAACCTGCACTC

3CK CCTGGCCAGCGGTATTAAGCGCATCAAGCAC

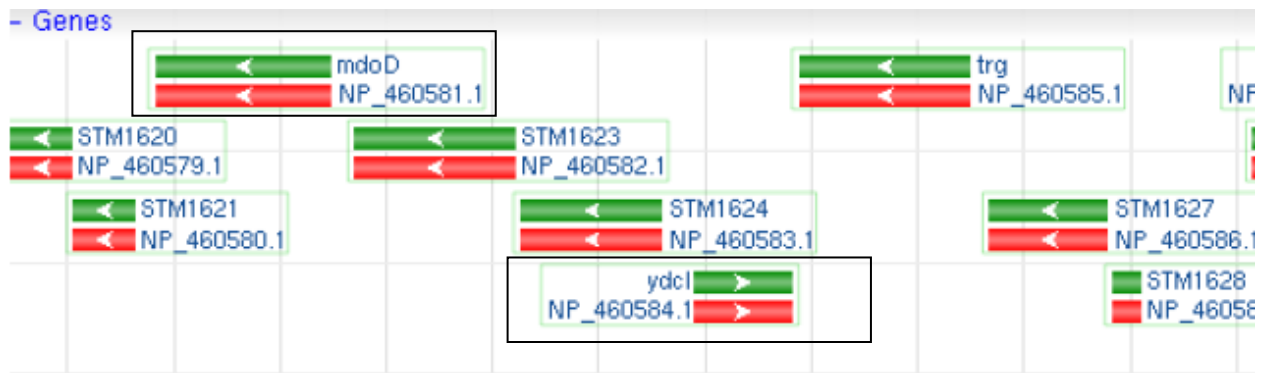
Figure S1.

Salmonella	MEK---NGLFSQRIRLRHLHTFVAVAQOGTLGRAAETLNLSQPALSKTINELEQLTGTRLFERGRLGAQITVPGEQFLTHA
Ralstonia	MENPAASGEALLSRIRFRHLSCFVAIAQERNLRRAAERLHLSQPAISKTLGELEALAGVRLVERGRQGARLTSAGEQFLRHA
Pseudomonas	-----MNLDTRIKYRHLFCFLEIARQGSILARAADSLISISQPAISKTLKELEDLLEARLFERSRQGVELTPAGTRFMRYA
Burkholderia	MHN-----RIADGRVKFRHLQCFLAVAQFGSVQKAAHLSITQPAVSKTIAELESILGVRLFERRGRQGARPTREGQLFMPHA
Rhizobium	-----MIDSRIKFRHLQTFVEVARQKSVMKAAELLHVSQPAVTKTIRELEQVLGVDFVERDGRGIKITRYGEVFLRHA
Agrobacterium	-----MVDQRIKFRHLQTFVEVARQKSVIRAAEILHVSQPAVTKTIRELEDVLGVSLLEREGRGIRISRYGEVFLRHA
Brucella	-----MIGNRIKFRHLHTFVEVARQKSVVKASEILNISQPAVTKTMRELEEILGVPVLERDGRGIRITRYGEVFLRHA
Ruegeria	-----MIDRRRIKFRHIQCFVEIAREGSLKTAALKFLTQPAWSKTLKELEEITGTTLMRRSRAGVELTKPGEVFLHFA
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Salmonella	VKVLDAINTAGQALNRKEDASADVVRVGALPTAALGILPAAIGRFHQQKSTSLQVATMNNMTMLLAGLKSGEIDLIGIRMSD
Ralstonia	VGVTQALEAATAALAGTGESSTPMVQVGALPTVASGLLPQAIARLHAERPHAGVRLRTGTNVELLAALKAGELDFVVGMRSE
Pseudomonas	GPSVQALRDGVSSLRG-EARAPSQVRIGVLSTVEGLLMPVLCRLHQRHEALVISVVTGVSQAQLLGQLHLGELELVVGRMTD
Burkholderia	SACVLALRQGVGLLAREGGGAAATLEIGMLPTVAASLAPAVLKTAAEWPRAVVRITTVANAELLERLKAGAIECAIGRLSE
Rhizobium	GAAALTALRQGLDSVSQEQAFAEAPPIRIGALPTVSSRIMPRAMELFLQEKTWRSRVKIVTGENAVILLEELRVGDLDDLVDVGRLAG
Agrobacterium	GATMTALRQAVDSVSQEAARAGPPVRVGALPTVSVRIMPKAMSGFLAECTGSPVKIVTGENAVILLEQLRVGDLDDLVDVGRILAA
Brucella	GTALTALRQGLDSVSQELDGSPPIRIGALPTVSTRIMPKAISLFLAECTGSKVKIVTGENAVILLEQPRVGDLDLVDVGRILAA
Ruegeria	RMSLASLQQGLDGIETEGRRQRETLSVGALPSVAAYLMPGAVSEFSRLAPHALLRIQDGPBGYLIERLRLGALDDLIVIGRMGP
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Salmonella	PELMGGLNYELLFLESCLKIVVRPGHPLLQET---ITLSRVMEWPVVVSPKGTVPQRNAEALLQSQGCCKMPAGCIETLSASLS
Ralstonia	EDMMQGLSFELLYAEPLALVVRPGHPLLSQLGAAASLQAVLDYPLVIATAGTVPRHHTALEFQTHGLRLPFGTTETLSVSVS
Pseudomonas	SPQIQGLSFEHLYSEMSLVVRPGHPLLASTP--VERGQVGRYPLVLPPAGTTIRQHADSLSFVQCGIQMPAQRLETLSLALS
Burkholderia	PERMVGLSFEHLYNEPLVAVVRAGHPLAASAS---PAAQLARYPVVLPYPYGTMIHQAAEQLLSACGAPPLESFVEVLVSVVA
Rhizobium	AEKMAGFSFEHLYSEQVVFVVRAGHPLLDGRQ--SLFSAFRDYTVLMPTRGSIIRPVVENFLIANGVSSLENIQIETVSDAFG
Agrobacterium	PQKMAGFSFEHLYSEKVRVVRAGHPLLSPGL--SVFDHLHEYFVLMPTTRQSVIGPVVEQFLIANGVPALPIRIETVSDAFG
Brucella	PEKMTGFSFEHLYSERVFLVVRAGHPLLDDD---TIFDHLANYFVLMPTTRNSIIRPFVERLLITNGVGSILPTQIETVSDAFG
Ruegeria	PETMQGLSFTQLYQEQQVVFVVRPGHPLLAAP---DLARVADWPVIYPPPAAIRPLVEQMLIANGIGEFENRLETVSGAFG
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Salmonella	RQLTVDYDYVWFVPSGAVKEDLRQATLVSLFVPTQSAGEPIGILTRVDIPLSTGAQMLIAAIRKSMPL-----
Ralstonia	RLACRSDAVWITPERTARDDLEHGWLARLDIPTSGTKEPVGLLLRSAAESTALARAFMETLAEELARMP-----
Pseudomonas	RRYLLGSDGLWVAPRDAVLLDLRRGELVELDLGVREPGGSVIGICRNAALPLSLPGQWVGEVLREVAGEYREGRYP-
Burkholderia	RALALENDVAVFVPRYAAEFDLAAGTLTRLALPVDGADEPVGLILRTDAQSPVARALIDAVRAIARRRLAGAGT-
Rhizobium	RAFLRASDAIWIISNGVVAGDVADGRLALLPVETGETRGPVGLTMRADAVPSAPQSILMQTIREAARELS-----
Agrobacterium	RAFLRTSDAIWIISEGVVAADVADGILAILPVETGDTSGPVGLTVRADTQPSLPLSLLMQAIREAAGELFDGRTEG
Brucella	RAFVRDSDAVWIISEGVAAHDVAEGKLVALPIDTSETTGVPVGLTMRMDMVQTAPLQILVQTIREAADH-----
Ruegeria	RVHARRSDAVWIIISRGVVAEIEAGRLVLEFGTELTRGPVGLMTRDPESAVAQLFALALRNIVIGAGAGA----
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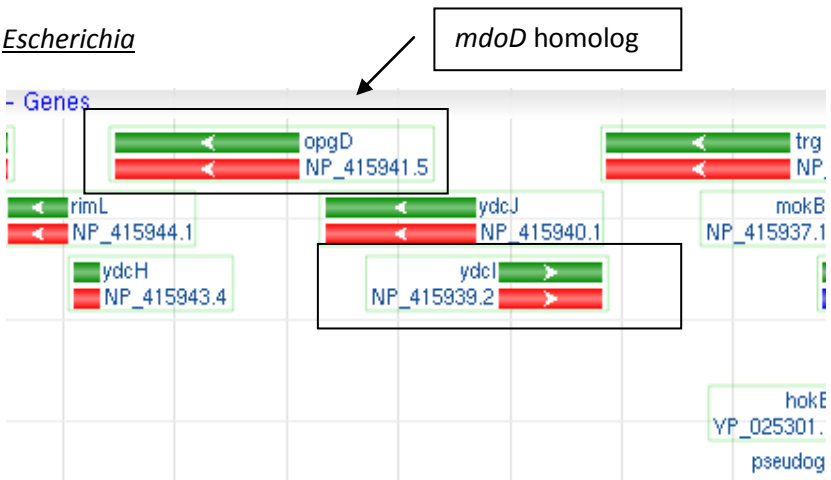
Figure S1. Alignment of the *S. typhimurium* YdcI protein with homologs present in a range of other Gram negative genera more distantly related to *Salmonella* spp. Yellow highlight indicates amino acid identity (in at least five of the proteins). The e-values for the aligned proteins are between 10^{-71} and 10^{-49} .

Figure S2.

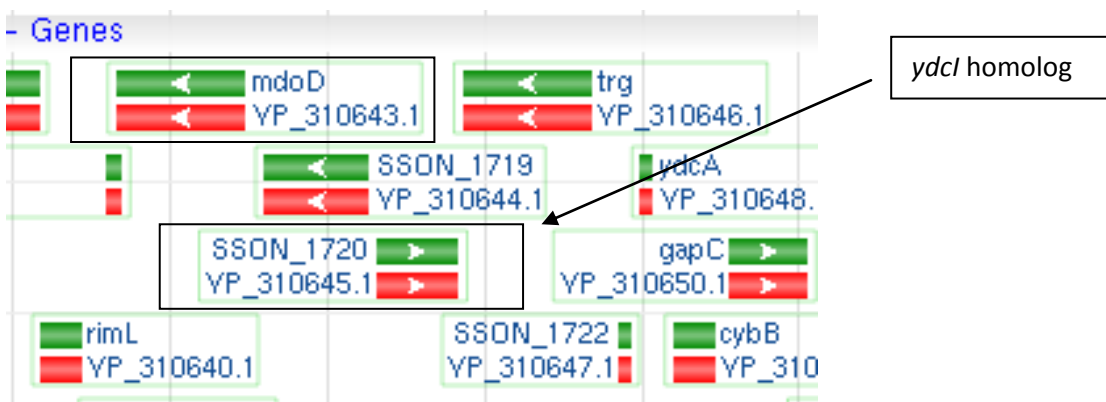
Salmonella



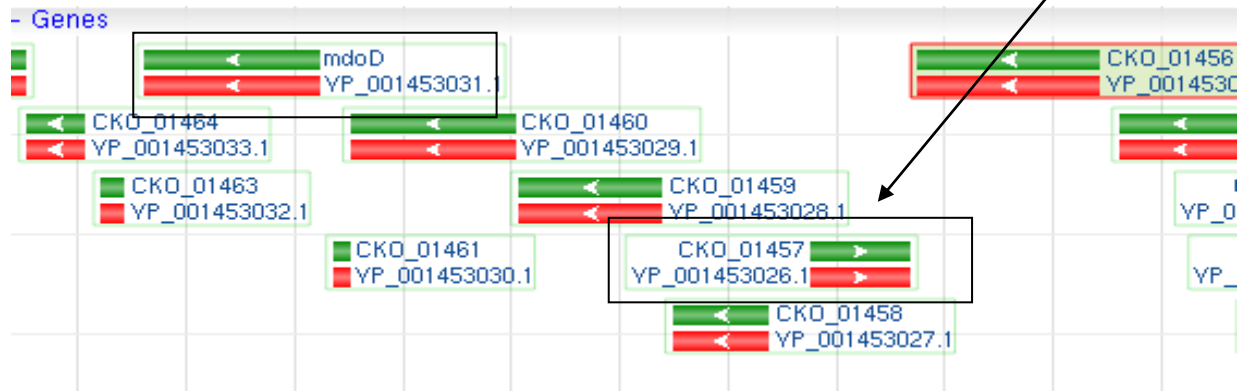
Escherichia



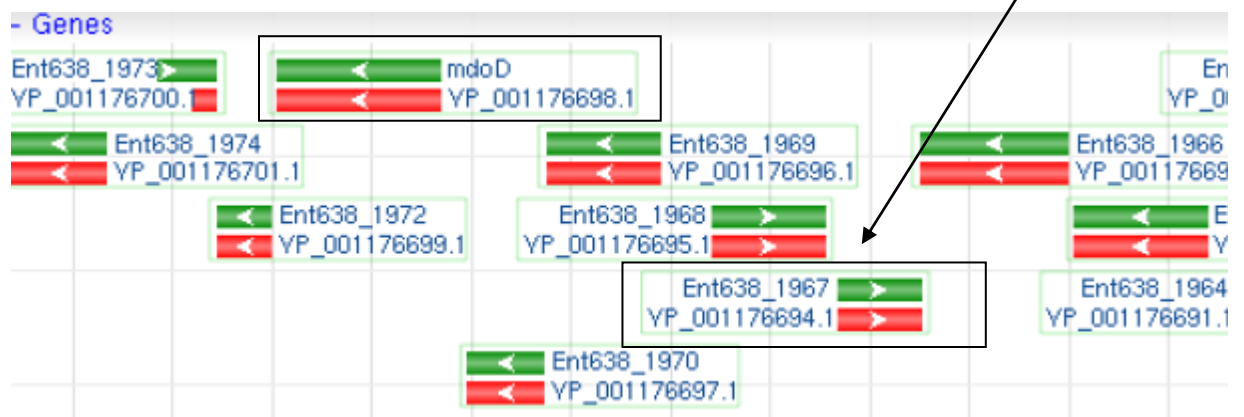
Shigella



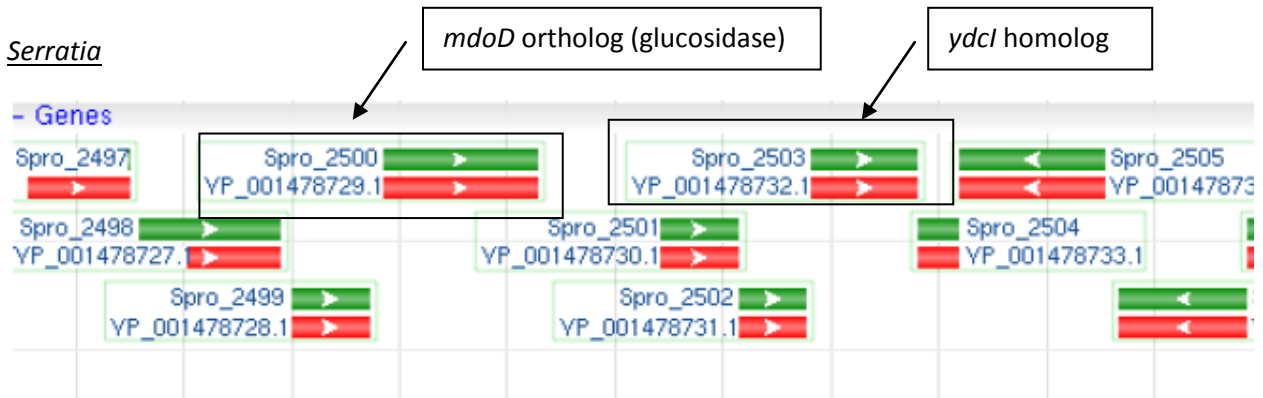
Citrobacter



Enterobacter



Serratia



Klebsiella

- Genes

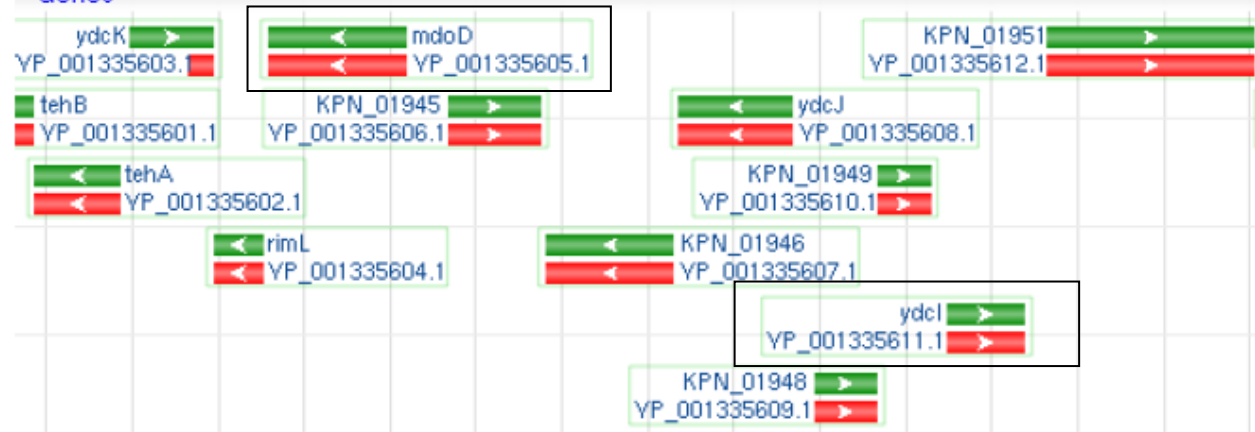


Figure S2. Genomic organization of the *ydcI* gene locus in a range of Gram negative genera. Maps of the genes associated with the *ydcI* gene homolog in the genomes of the indicated genera are provided. The *ydcI* genes and the *mdoD* genes are boxed for convenient visual reference.

Figure S3.

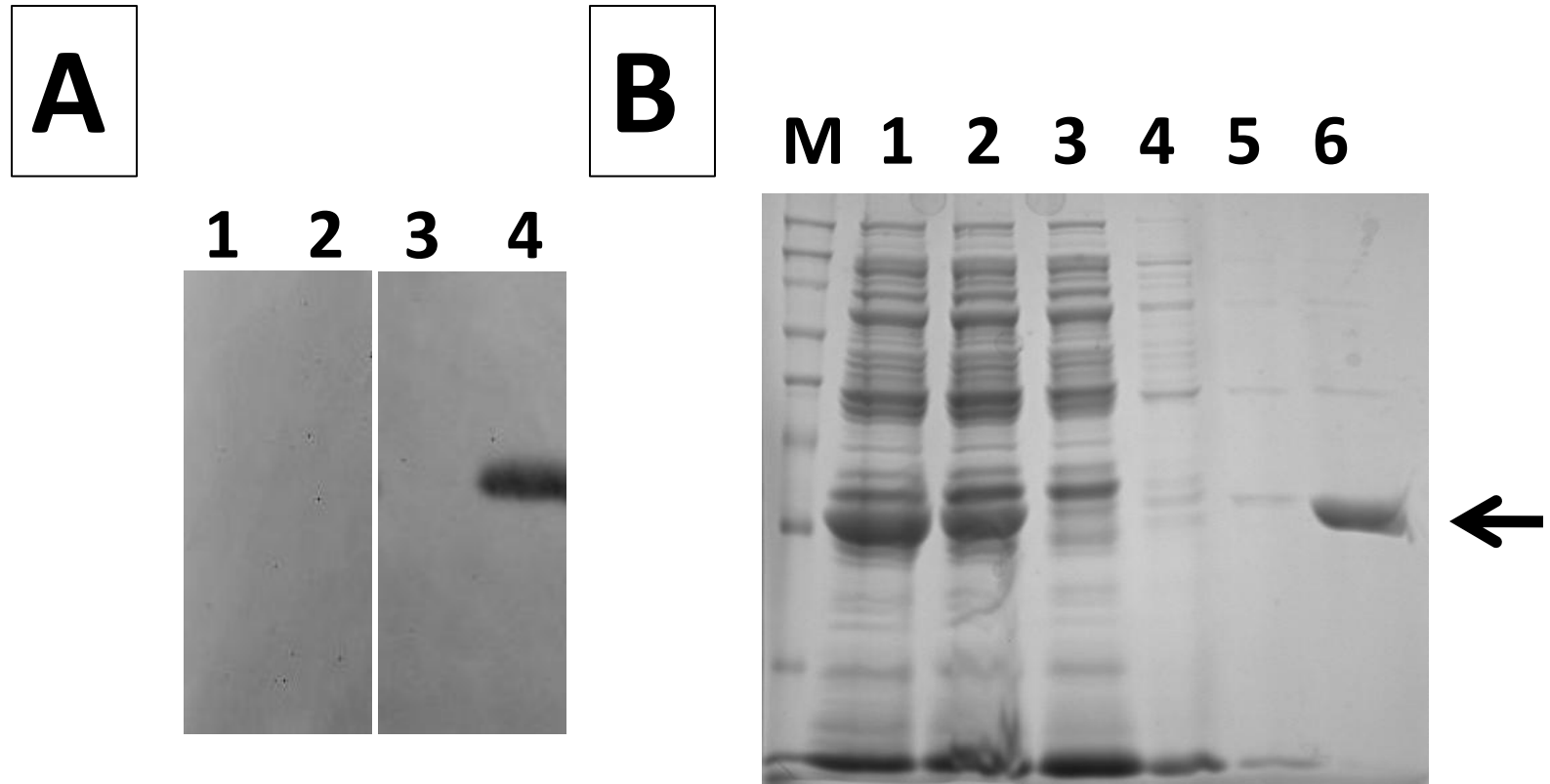


Figure S3. Expression and purification of YdcI protein. Panel A. Broth cultures of *E. coli* TOP10 strains containing pBAD18 or pBAD18 + *ycdI* grown in the presence and absence of arabinose were harvested for total cell protein. A Western blot of these samples was probed with anti-6x-histidine antibodies. Lanes correspond to: 1. pBAD18, - ara; 2. pBAD18, + ara; 3. pBAD18+*ycdI*, - ara; 4. pBAD18+*ycdI*, + ara. Panel B. Coomassie-stained SDS-PAGE gel of samples from purification of 6x-histidine-tagged YdcI from strain TOP10 (pBAD18 + *ycdI*). Lanes: 1. Total cell protein; 2. Soluble protein fraction; 3. Column flow-through; 4. First wash; 5. Second wash; 6. Elution.

Figure S4.

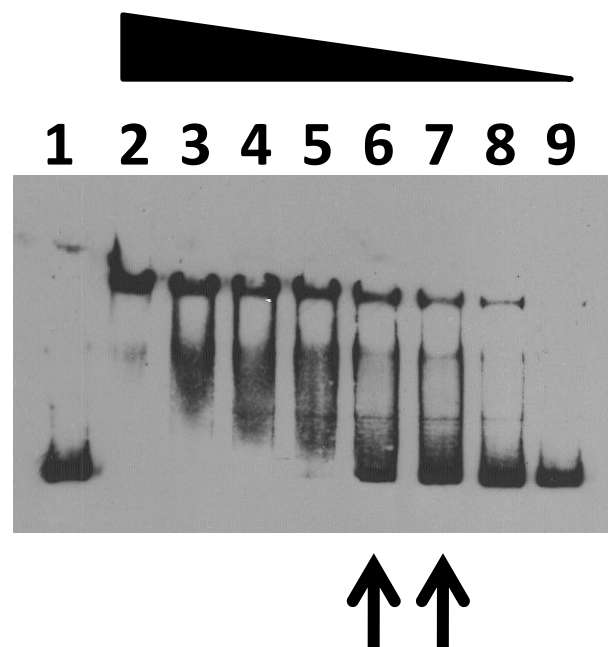


Figure S4. Determination of apparent dissociation constant (K_d) of YdcI protein binding to *ydcI* promoter probe. Lanes: 1. Probe alone; 2-9. Reactions with constant amount of probe (1.6 ng) and 74, 37, 18, 9.2, 4.6, 2.3, 1.2, and 0.6 nM YdcI protein, respectively. Arrows indicate the range at which the likely apparent K_d occurs (between 2.3 and 4.6 nM). The probe is a 362-bp fragment containing the *S. typhimurium ydcI* promoter (genomic coordinates 1714935 – 1714573). The Alphaview software package (Cell Biosciences, Santa Clara, CA) was used for quantification to confirm the apparent K_d range.

Figure S5.

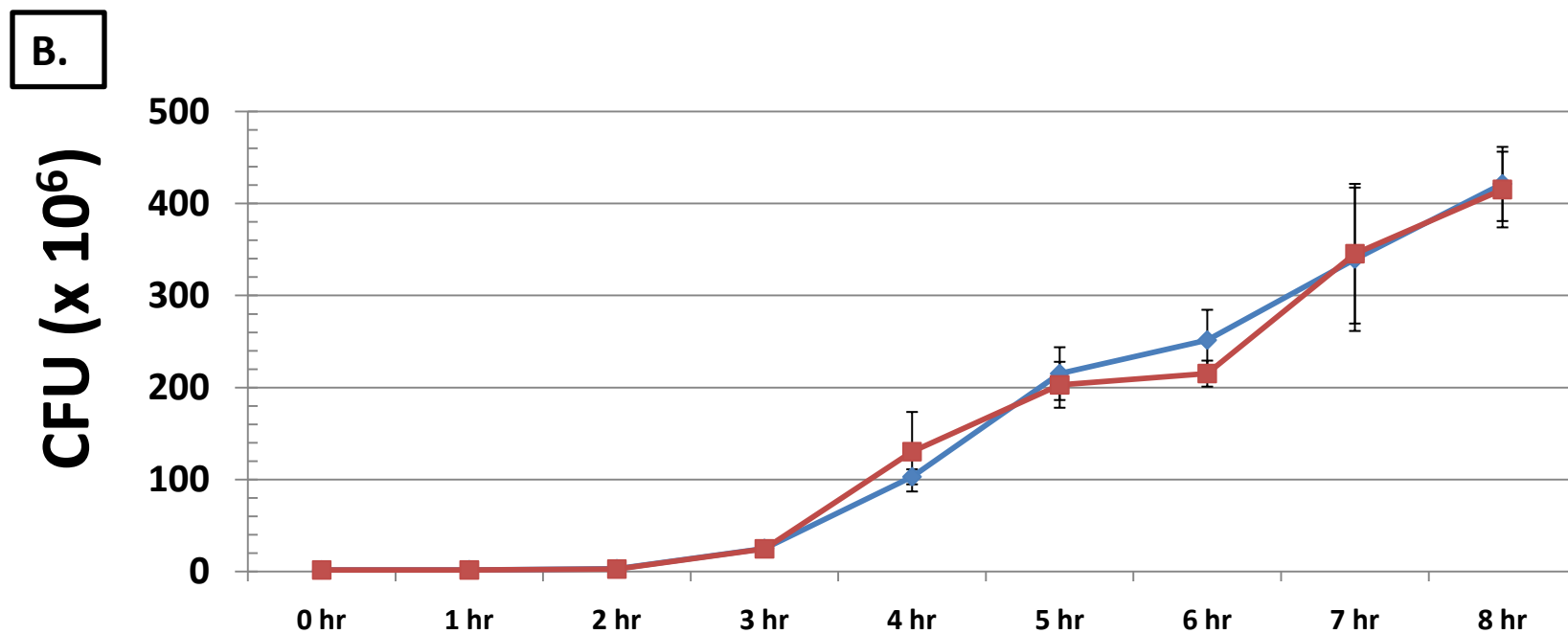
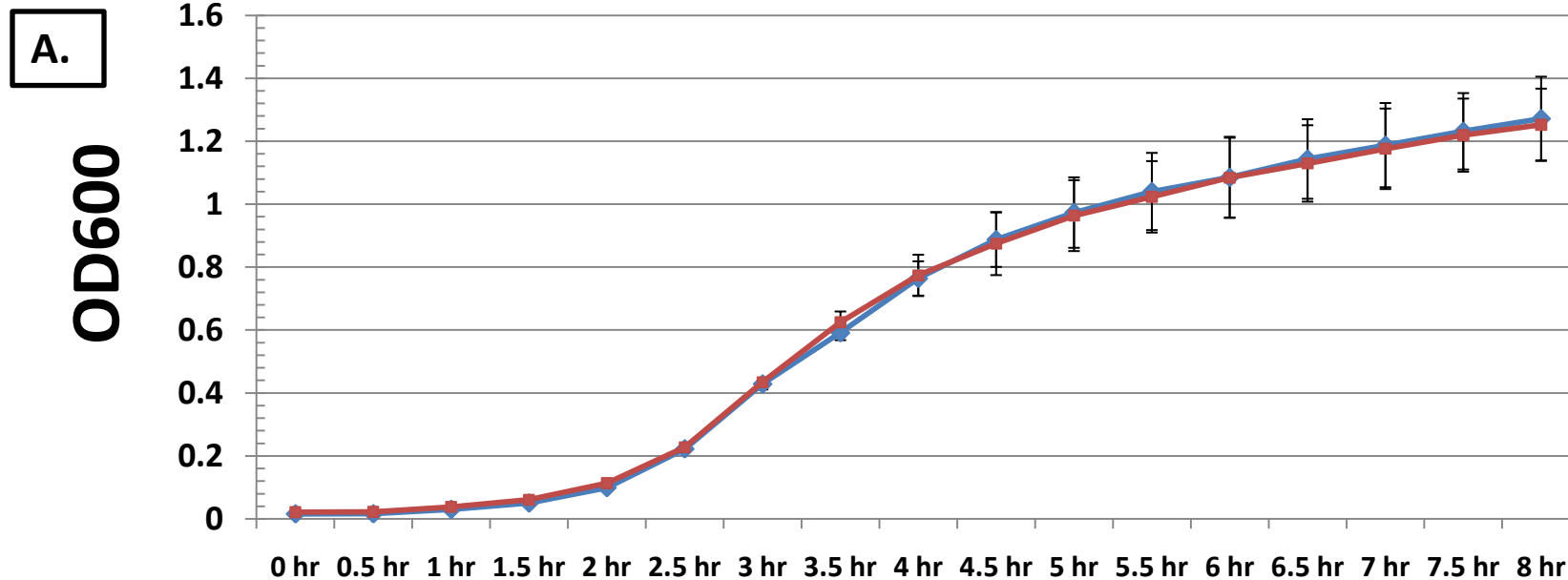


Figure S5. Growth curves of isogenic *S. typhimurium* WT and $\Delta ydcI$ strains. Cultures of WT and $\Delta ydcI$ strains were diluted into fresh LB media, grown shaking at 37 degrees C, and sampled for growth at the indicated time points for OD600 (Panel A) and CFU (Panel B) measurements. The data represent the mean and standard deviation from three independent cultures. The red and blue lines indicate the WT and $\Delta ydcI$ strains, respectively.

Figure S6.

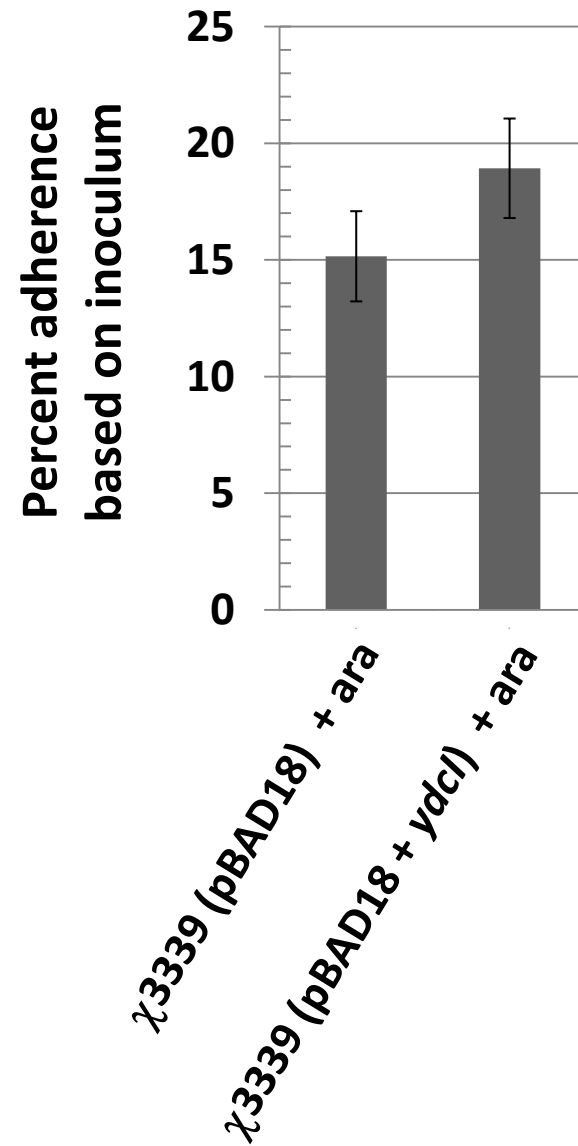


Figure S6. Attachment of *S. typhimurium* χ 3339 (pBAD18) and χ 3339 (pBAD18 + *ycdI*) strains to Int407 cells. Samples of the indicated strains were added to culture wells containing Int407 cells (at MOI of approximately 30) and allow to adhere for 30 minutes. Following washes, cells were harvested using deoxycholate, serial diluted in PBS, and plated for CFU counts. The percent bacteria adhering to the tissue culture cells based on inoculum is presented. Data represent the mean and standard deviation from four independent trials each performed in triplicate wells.

Figure S7.

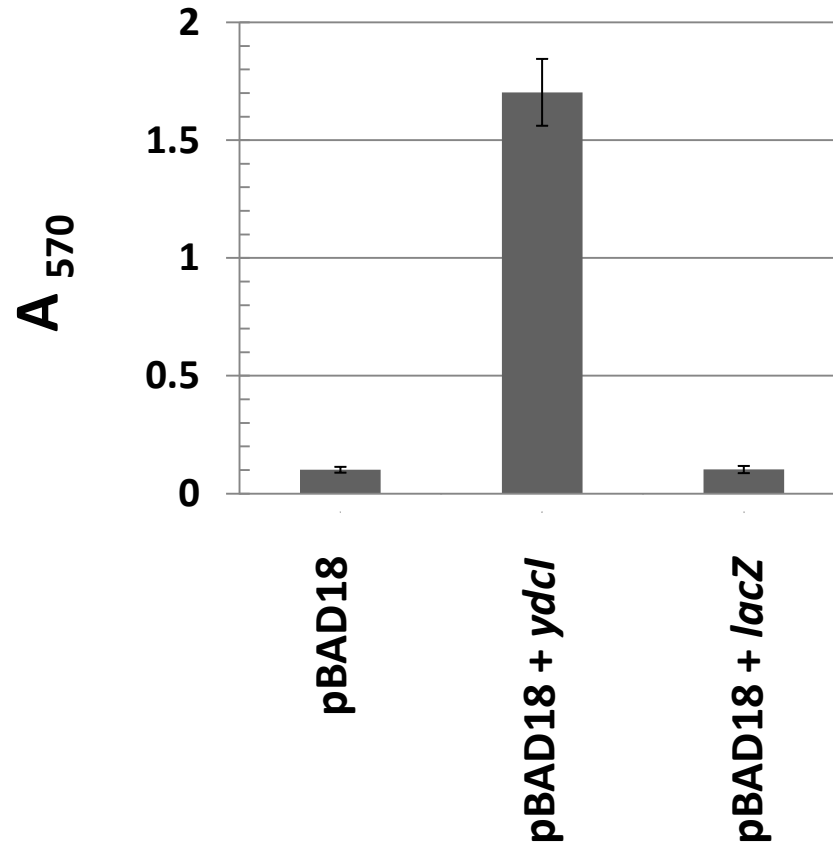


Figure S7. Quantitation of biofilm formation. Separate cultures of *S. typhimurium* strain χ 3339 containing the indicated plasmids were grown for biofilm as indicated in Figure 9 of the main text. After washing with PBS, the culture flasks were stained with crystal violet, and then the sides of the flasks were washed with 33% acetic acid as described previously in the text. Absorbance of the sample was determined at 570 nm wavelength and plotted. The results are obtained from three or four independent experimental cultures for each strain.