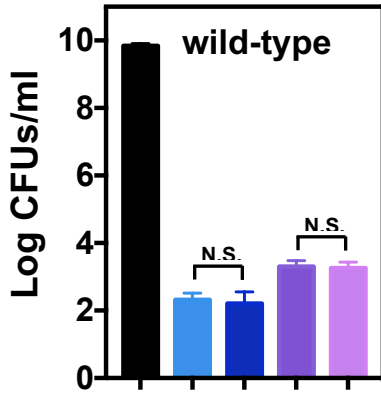
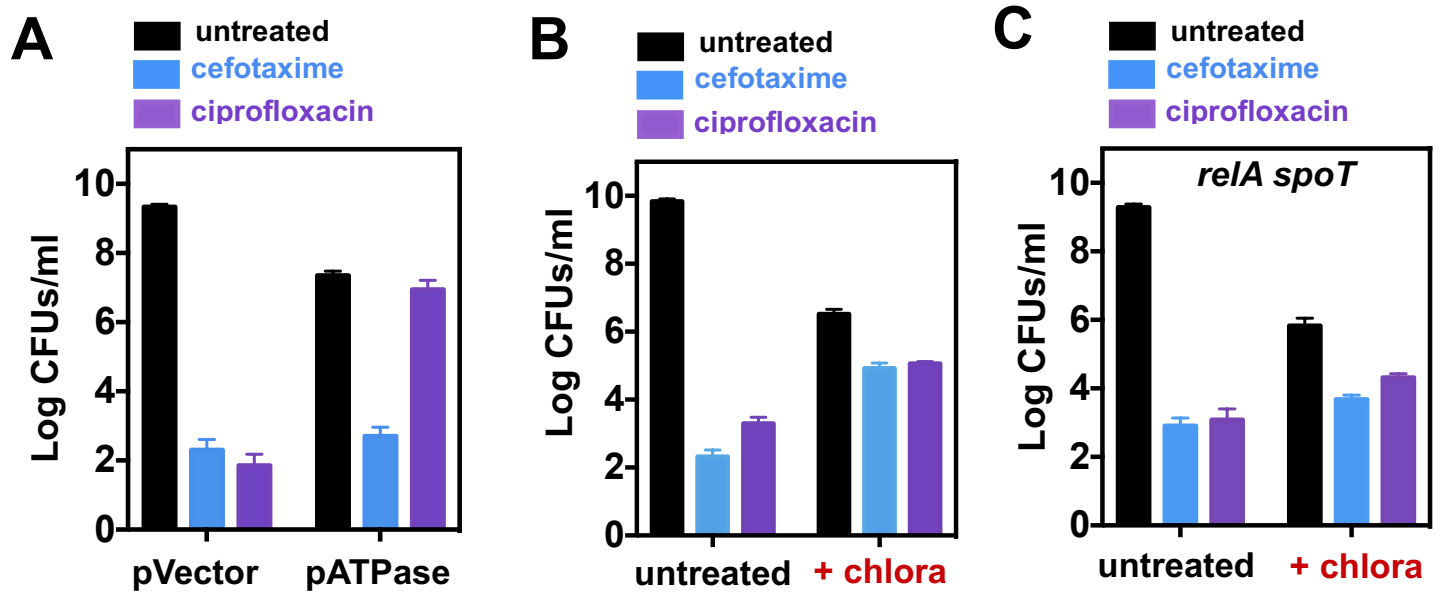


**Fig. S1. Determination growth rates and of the emergence of antibiotic resistant in cultures of *Salmonella* strains.** (A) Growth dynamics of wild-type (14028s),  $\Delta 12TA$  (MP1422) and *relA::Tn10 spoT* (MP342) *Salmonella* in LB and MOPS medium containing 2 mM K<sub>2</sub>HPO<sub>4</sub> and 10  $\mu$ M MgCl<sub>2</sub>. Error bars represent standard deviations. Growth curves are representative of typical experiments. Note log scale of *y* axis. (B) Number of surviving wild-type (14028s),  $\Delta 12TA$  (MP1422) and *relA::Tn10 spoT* (MP342) *Salmonella* following two rounds of 24 h exposure to cefotaxime (200  $\mu$ g/mL) or ciprofloxacin (2  $\mu$ g/mL) in LB medium. Error bars represent standard deviations (N = 6 biological replicates). Note log scale of *y* axis.



**Fig. S2. Effect of Shx treatment on the frequency of persisters.** Concentration of surviving wild-type (14028s) *Salmonella* following 24 h treatment with either cefotaxime (200  $\mu\text{g}/\text{mL}$ ) or ciprofloxacin (2  $\mu\text{g}/\text{mL}$ ). Shx treatment (100  $\mu\text{g}/\text{mL}$ ) was carried out for 30 min prior to outgrowth as described. Error bars represent standard deviations (N = 6 biological replicates). Note log scale of y axis. Two-tailed t-test between populations at the edge of brackets: N.S. for no significance.



**Fig. S3. Effects of ATP depletion and inhibition of protein synthesis on antibiotic tolerance.** (A) Concentration of surviving wild-type *Salmonella* (14028s) following 24h exposure to cefotaxime (200  $\mu\text{g}/\text{mL}$ ) or ciprofloxacin (2  $\mu\text{g}/\text{mL}$ ). Bacteria expressed the soluble portion of the ATPase from plasmid pATPase or harbored the pVector control. Concentration of surviving (B) wild-type (14028s) or (C) *relA::Tn10 spoT* (MP342) *Salmonella* following cefotaxime (200  $\mu\text{g}/\text{mL}$ ) or ciprofloxacin (2  $\mu\text{g}/\text{mL}$ ) exposure in the presence of absence of chloramphenicol (50  $\mu\text{g}/\text{mL}$ ). Error bars represent standard deviations (N = 8 biological replicates). Note log scale of y axis.

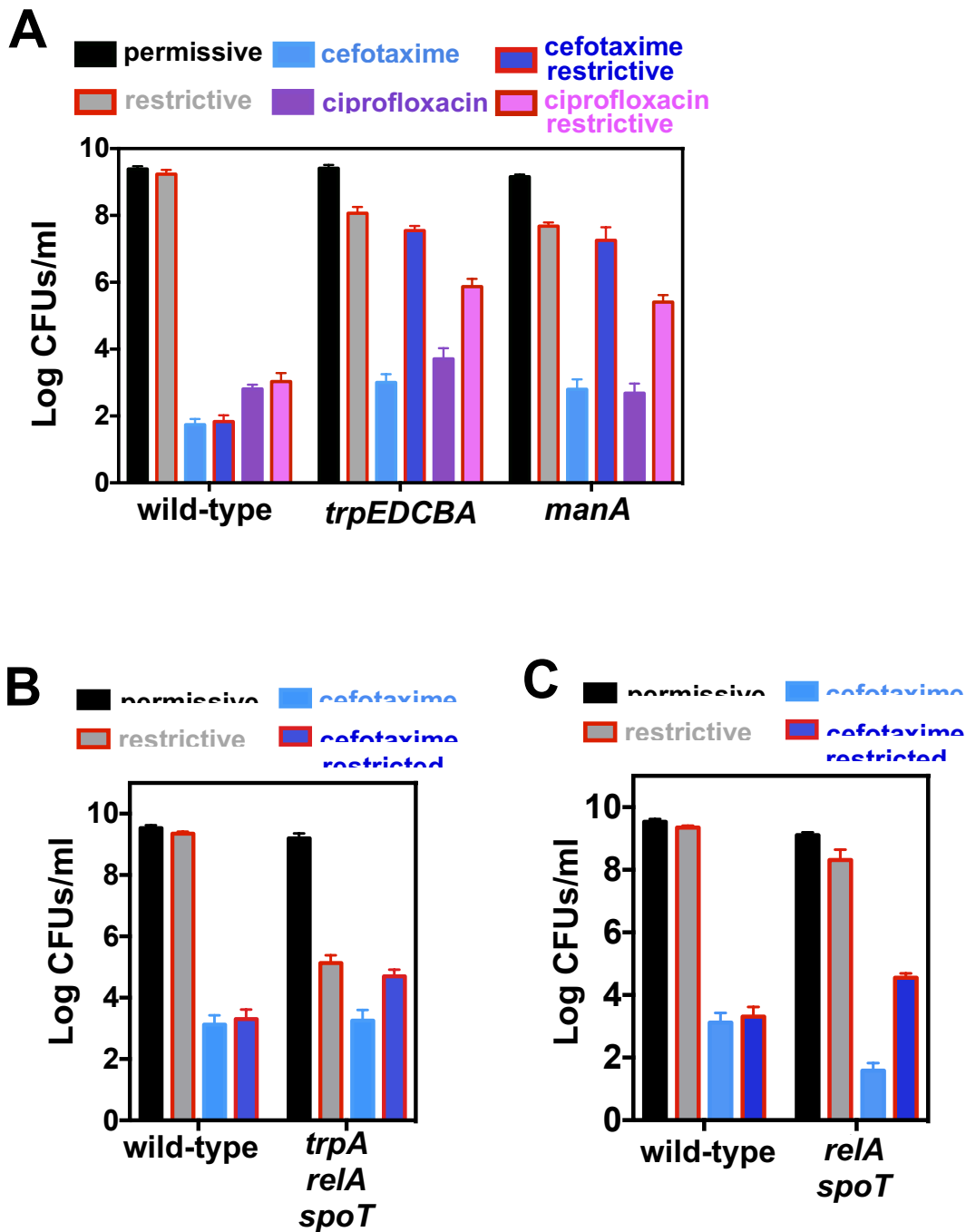


Fig. S4. Effect of growth inhibition on antibiotic tolerance in *Salmonella*. (A) Concentration of surviving wild-type (14028s), *manA* (MP50) and *trpEDCBA::Tn10* (AS301) *Salmonella* following 24 h treatment with either cefotaxime (200  $\mu\text{g}/\text{mL}$ ) or ciprofloxacin (2  $\mu\text{g}/\text{mL}$ ) under growth-permissive and growth-restrictive conditions.

For the *manA* (MP50) strain, growth restriction was achieved in MOPS minimal medium containing mannose as the sole carbon source (see Materials and Methods). For *trpEDCBA::Tn10* (AS301) strain, growth restriction was achieved in MOPS minimal medium lacking tryptophan (see Materials and Methods). **(B)** Concentration of surviving wild-type (14028s) and *relA::Tn10 spoT trpA::kan* (MP1494) *Salmonella* following 24 h treatment with either cefotaxime (200  $\mu\text{g}/\text{mL}$ ) or ciprofloxacin (2  $\mu\text{g}/\text{mL}$ ) under growth-permissive and growth-restrictive conditions. For the *relA::Tn10 spoT trpA::kan* (MP1494) strain, growth restriction was achieved in MOPS minimal medium lacking tryptophan (see Materials and Methods). **(C)** Concentration of surviving wild-type (14028s) and *relA::Tn10 spoT* (MP342) *Salmonella* following 24 h treatment with cefotaxime (200  $\mu\text{g}/\text{mL}$ ) under growth-permissive and growth-restrictive conditions (see Materials and Methods). For the *relA::Tn10 spoT* (MP342) strain, growth restriction was achieved in N-minimal medium lacking various amino acids and containing glycerol as the sole carbon source (see Materials and Methods). Error bars represent standard deviations (N = 8 biological replicates). Note log scale of *y* axis.