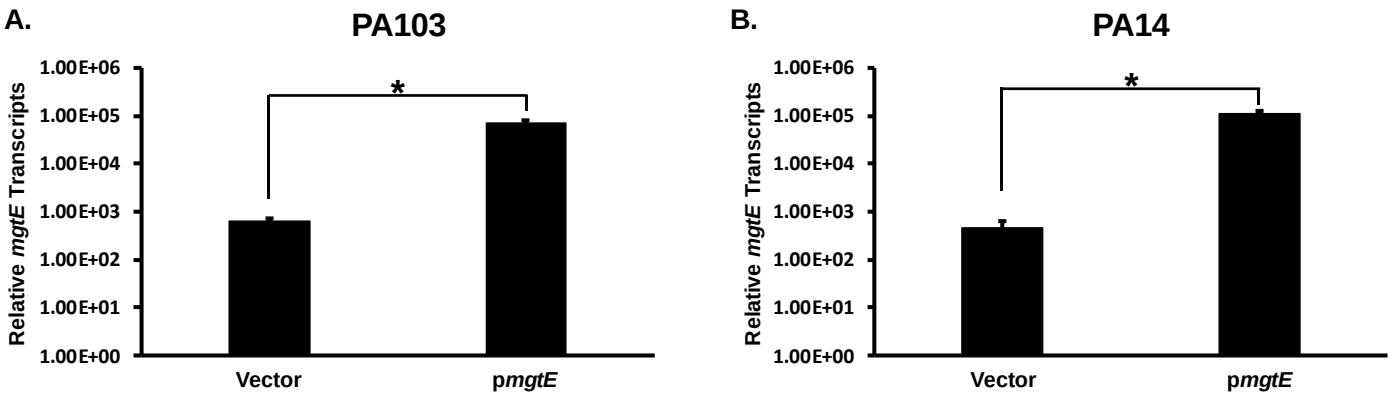
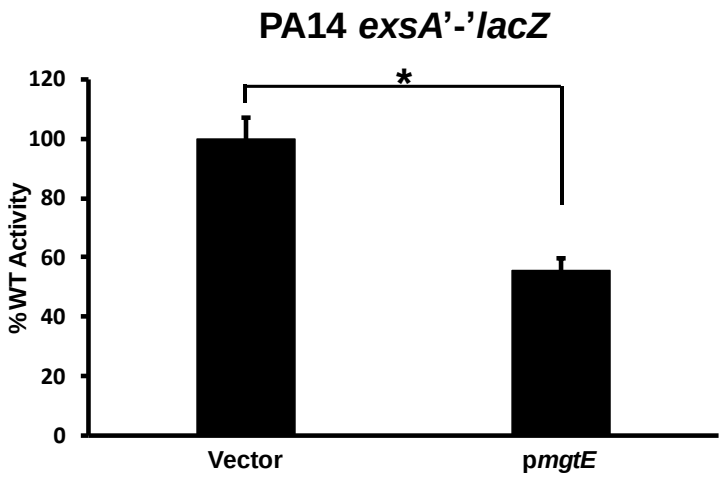
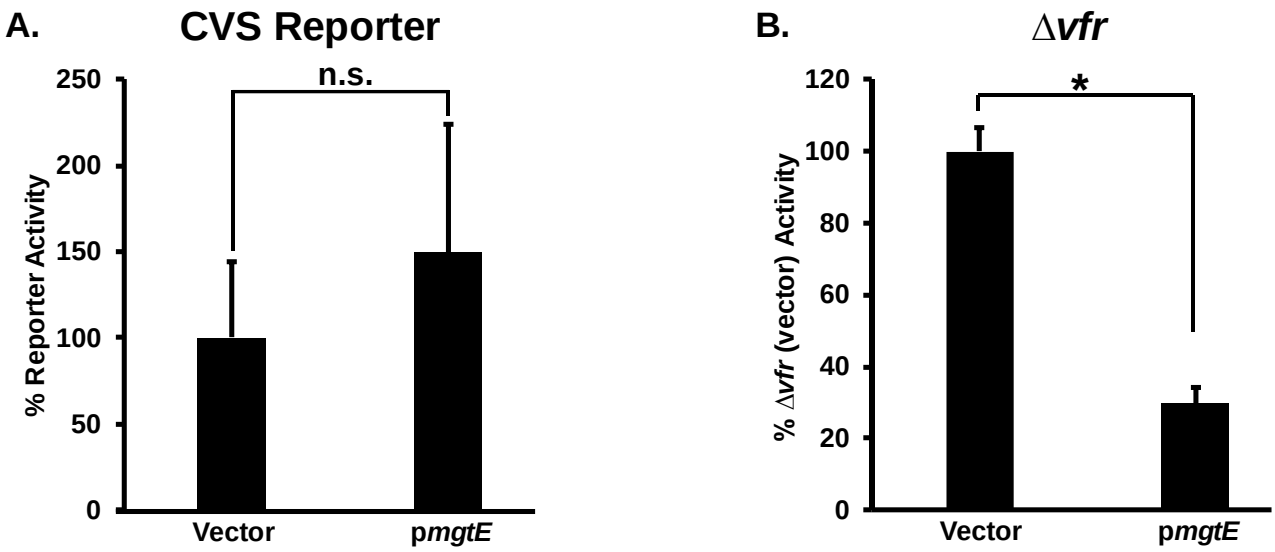


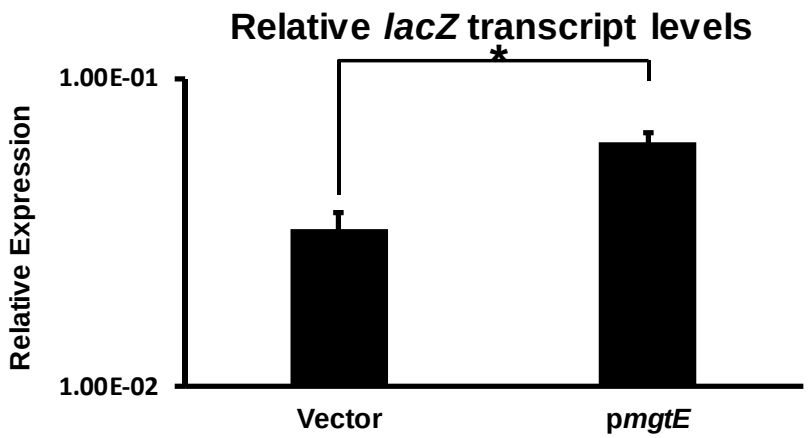
Supplemental Figure 1. Measurement of *mgtE* transcript levels in PA103 and PA14. *mgtE* levels were quantified by qRT-PCR in PA103 and PA14 containing either vector control or p*mgtE. fbp* was used as a normalization control transcript. **p*<0.05. These fold increases are on the same order of magnitude as seen after antibiotic treatment or under magnesium limitation conditions (29, 30).



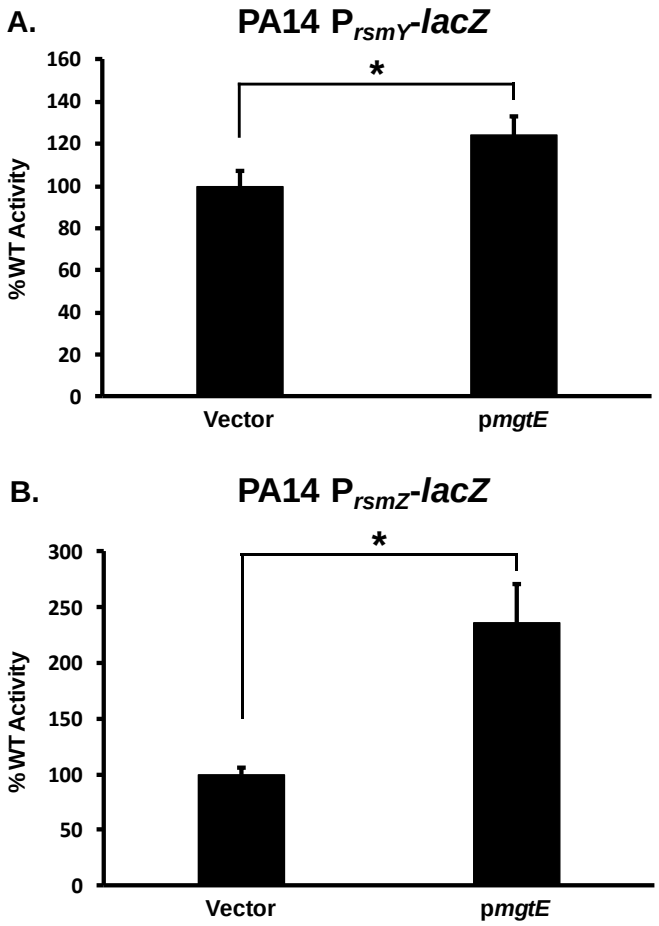
Supplemental Figure 2. *mgtE* expression represses *exsA* translation in *P. aeruginosa* PA14. *exsA* translational reporter strain PA14 *exsCEBA*'-'*lacZ*, with either vector control or p*mgtE* were assayed under T3SS inducing (+EGTA) conditions for beta-galactosidase activity. Percent activity was calculated considering the *lacZ* activity from the respective strain with the blank vector as 100%. The reporter constructs were transcribed from a constitutive P_{lacUV5} promoter. *p<0.05.



Supplemental Figure 3. The CVS cascade is not required for *mgtE*-mediated inhibition. (A) *mgtE* expression does not repress CVS reporter activity. PA103 CVS reporter strains with either vector control (Vector) or p*mgtE* were assayed for beta-galactosidase activity from the CVS reporter under T3SS inducing conditions (+EGTA). n.s.=not significant. (B) *mgtE* inhibits T3SS gene transcription in a Δ*vfr* background. Δ*vfr* strains with either vector control (Vector) or p*mgtE* were cultured under T3SS inducing (+EGTA) conditions and assayed for beta-galactosidase activity from the P_{exsD}-*lacZ* reporter construct. Percent activity was calculated considering the P_{exsD}-*lacZ* activity in the Δ*vfr* strain with blank vector as 100%. **p*<0.005.



Supplemental Figure 4. *mgtE* expression does not repress *exsA* transcript stability. mRNA was isolated from the *exsA*'-'*lacZ* translational reporter strain with either the blank vector or with p*mgtE* and *exsA*'-'*lacZ* transcript stability was analyzed by qRT-PCR. *lacZ* transcript abundance was normalized to that of the control transcript *fbp*. **p*<0.05.



Supplemental Figure 5. *mgtE* expression affects *rsm* transcription in *P. aeruginosa* PA14. *rsmY* and *rsmZ* transcriptional reporter strains (A) PA14 P$_{rsmY}$-*lacZ* and (B) PA14 P$_{rsmZ}$-*lacZ*, with either vector control or p*mgtE* were assayed under T3SS inducing (+EGTA) conditions for beta-galactosidase activity. Percent activity was calculated considering the *lacZ* activity from the respective reporter strains with the blank vector as 100%. **p*<0.05.