

Supplemental method:

1. Generation of the p114-mCherry-ClyLuc plasmid

Plasmid pClyA-GFP expressing Cytolysin A-Green fluorescent protein fusion was a generous gift from Dr. David Putnam (Cornell University, New York). This plasmid was linearized through a PCR reaction to amplify a 5.7 Kb fragment spanning the region from the *clyA* gene to the pBR322 origin. The medium strength constitutive promoter J23114 (http://parts.igem.org/Part:BBa_J23114), together with *mCherry* gene was then PCR-amplified and inserted upstream of the *ClyA-GFP* fusion gene to generate the p114-mCherry-ClyGFP plasmid. Subsequently, to generate the p114-mCherry-ClyLuc plasmid, the *gfp* gene was replaced with the nanoluciferase (*nluc*, Promega) gene, and a linker peptide “GGGSGGGG” sequence was inserted in-between the *clyA* gene and the *nluc*. The map and sequence of p114-mCherry-ClyLuc are shown in **Supplemental Figure S1**. The PCR-amplified inserts were cloned into the linearized vectors using ligation-independent cloning (In-Fusion cloning). The ligated plasmids were transformed into chemically competent commercial *E. coli*, verified through Sanger sequencing, and then extracted and subsequently transformed into *E. coli* MP1 through electroporation.