

Supplemental Material to:

Soon-Keat Ooi, Tian-Yeh Lim, Song-Hua Lee and Sheila Nathan

Burkholderia pseudomallei kills Caenorhabditis elegans through virulence mechanisms distinct from intestinal lumen colonization

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http://www.landesbioscience.com/journals/virulence/article/21808/

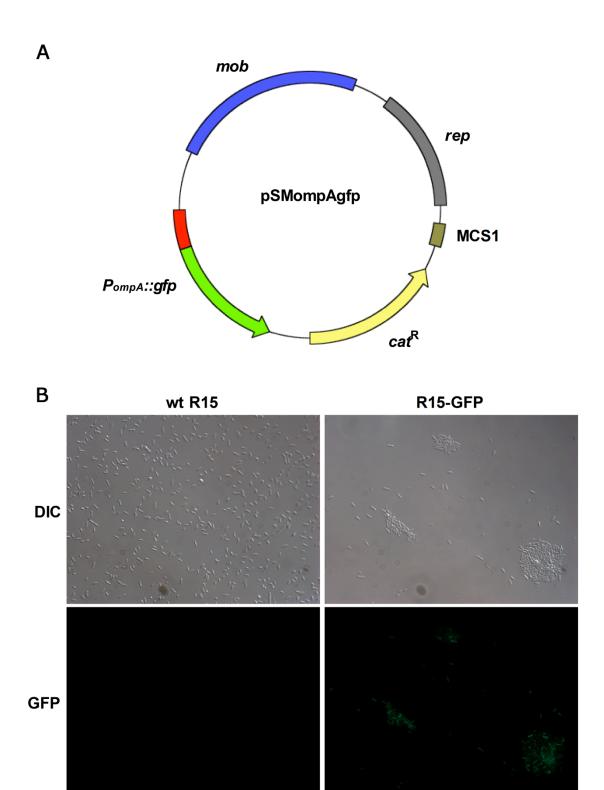


Figure S1. *B. pseudomallei* R15 electroporated with pSMompAgfp constitutively expresses GFP. (A) Shown is the map of pSMompAgfp, refer to **Table 1** for additional information.

Abbreviations: *mob*, mobilization gene; *rep*, origin of replication; MCS1, multiple cloning site 1; cat^R , chloramphenicol acetyltransferase gene (confers chloramphenicol resistance phenotype).

(B) Representative photomicrographs of wild-type *Bp* R15 and R15-GFP captured under DIC optics and GFP2 filter cube at 1000x magnification, showing that R15-GFP specifically expresses GFP *in vitro*.

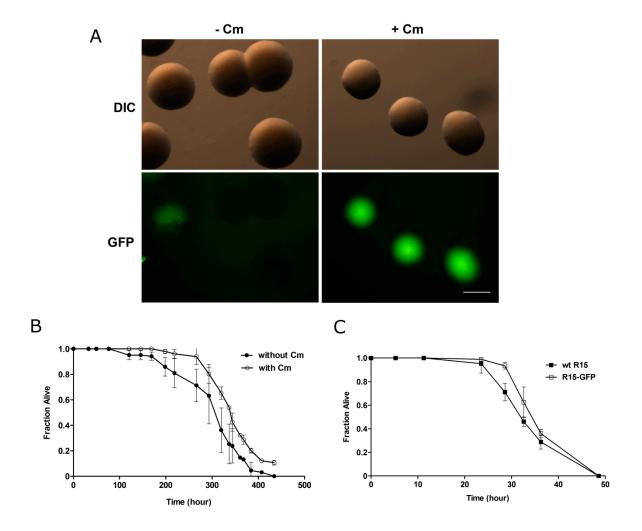


Figure S2. Assessment of the suitability of R15-GFP as a surrogate strain for *Bp* R15. (A) Loss of fluorescence observed in R15-GFP colonies after a passage of cells without chloramphenicol (Cm) selection showed that pSMompAgfp was unstable in *Bp* R15. Nevertheless, the bright green fluorescence was retained with the supplementation of Cm. Shown are photomicrographs taken at 100x magnification with the scale bar represents 200 μm. (B) The addition of 100 μg/ml Cm in OP50/NGM plates did not compromise the lifespan of worms. As live *E. coli* is pathogenic to aged nematodes, the extension of the curve with open circles is likely due to bacteriostatic effect of Cm. (C) R15-GFP displays *C. elegans* N2 killing kinetics similar to *Bp* R15 on NG/Cm agar, validating the use of pSMompAgfp in reporting colonizing bacteria within

C. elegans. Graphs in (B) and (C) show the mean \pm S.D. of alive worm fraction from a sample size of 120 worms.

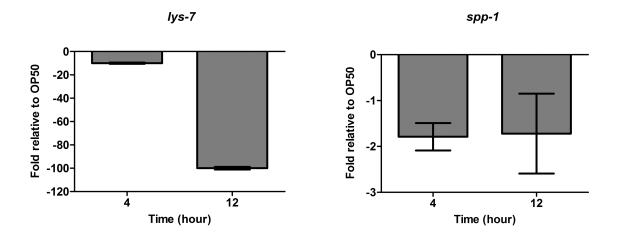


Figure S3. Bp R15 did not significantly colonize the C. elegans intestinal lumen despite its ability to suppress the expression of host antimicrobial peptide genes. Bars correspond to the mean \pm S.D. of fold change levels of host lys-7 (left) and spp-1 (right) genes of infected worms relative to E. coli OP50-fed worms at 4 and 12 hours post-infection.