

## Supplemental Text

**Table S1.** Primers used in this study. “FAM” and “TAM” refer to fluorescent modifications added to probes used in qPCR. Y refers to randomly inserted C or T nucleotide. R refers to randomly inserted A or G nucleotide.

Primer Name	Sequence (5'-3')
NG1427 5	CCGTTCCGCCCGTTCCTTTC
NG1427 6	GTCGTTTAAACCGAGAGACGGACTGTATGTG
NG1427 7	GTACGTTTAAACGCAGCCAATCGATACTACAC
NG1427 8	CGAAATGGCCAAACAAGAG
NG1427 Ab2.2	CCGCCGGCTAGCATGTTTACTTTAGATGATTGA
NG1427 nostop2	GGCGGCAAGCTTAACCGTCTGCCGAACCACTC
NG1427 comp5' end	CCGCCGTTAATTAATTTACTTTAGATGATTGA
NG1427 comp3' end	GGCGGCGGCCGGCCTTCAAACCGTCTGCCGAACCACTC
pacI-NG1427-CF	GGCCGCTTAATTAATAATTTTGTTTAACTTT
fseI-NG1427-CR	GGCCGGGGCCGGCCCAACTCAGCTTCCTTTCGGGCT
NG1428 1 primer	GGAATTGTCCCAATAGATAAG
NG1428 2H primer	GCCGGCGGTYRACCGCAGTATAGATAAGATATTTGC
NG1428 3H primer	GCCGGCGGTYRACCTTGAACGAACAGGAAATGCTC
NG1428 4 primer	CAATCAAGCGTTGGTCGAAGTCTTC
NG1427 qPCR Forward Primer	GTTCCCTCATGACCGACAAACTG
NG1427 qPCR Reverse Primer	ATCCCTTGCTTACGATACCTTGG
NG1427 qPCR Probe Primer	FAM-CCGTATCCCGCAGCCGCCCG-TAM
NG1428 qPCR Forward Primer	TTTACCTCATTCTGCTTTGCCTTC
NG1428 qPCR Reverse Primer	CTTTGGAAAGGGGAGACTTGTTCTG
NG1428 qPCR Probe	FAM-ACCGTTCTGAAGCCAAACAGCCCG-TAM
RecN qPCR Forward Primer	ATGCTTCTAACACTTTCTTTGCGTG
RecN qPCR Reverse Primer	AAATAAACTAATTTTCGACGCAAG
RecN qPCR Probe	FAM-ATTGAACCCGAAGAGTGGCCTGCCAA-TAM
RecA qPCR Forward Primer	ACAGGCTTTGGAAATCTGCGACAC
RecA qPCR Reverse Primer	TTTGATGTGTCCGGTCAGTTTGCG
RecA qPCR Probe	FAM-AAAGCCGAAATCGAAGGCGATATGGG-FAM

Omp3 qPCR Forward Primer	TATTCGTTGCATTGCTCGCTTCCG
Omp3 qPCR Reverse Primer	TCTACGCGACCTTGGCTTGCTTTA
Omp3 qPCR Probe	FAM-ACCGTAAGCGGCCAATCGAACGAAAT-FAM
NG1427::cat 5' end qPCR Forward Primer	GCAGCCAATCGATACTACACCC
NG1427::cat 5' end qPCR Reverse Primer	GGGAGCGGAAGGATATTTATGGAAA
NG1427::cat 5' end qPCR Probe Primer	FAM-TGCTGAAGCCC GCAATCGTCATTTC A-TAM
G113D point mutation Forward Primer	CGGGCGGCTGCGGACTACGGGCAGTTT
G113D point mutation Reverse Primer	AAACTGCCCGTAGTCCGCAGCCGCCCG
C64S point mutation Forward Primer	TTAAGCAGTTGAAGGGGAGTAGTATCGAATGGCTG
C64S point mutation Reverse Primer	CAGCCAATCGATACTACTCCCCTTCAACTGCTTAA
IGR 1427 Primer	GGCCGCTTAATTAATTTTCTATCCTTTTTCTG
Comp3 HA	GGCGGCGCCGGCCTCATCAGGCGTAGTCCGGGACGTC GTAG-GGGTAAACCGTCCTGCCGAACCACTC
IGR1427/1428 Primer F	TCAATCATCTAAAGTAAACA
IGR1427/1428 Primer R	TTTCTATCCTTTTTCTGTCAA
IGR-RecN Primer F	TGAAATTCCAAATGCCG
IGR-RecN Primer R	TGTGAGGATTCCTTAGC

## Supplemental Methods:

### *Gc survival assays*

The oxidative killing assays were performed as described in previous publications from our lab (Stohl et al., 2005). Briefly, FA1090 1-81-S2 Gc in amended GCBL were grown to mid-log phase as described above, well-vortexed, diluted 1:10 in GCBL, and treated with a range of H<sub>2</sub>O<sub>2</sub>. Colonies were counted after overnight growth and survival for each concentration of H<sub>2</sub>O<sub>2</sub> was calculated relative to the untreated dose.

The protocol for assaying Gc survival following treatment with diamide (Sigma) was similar to the procedure described above, using FA1090 1-81-S2<sub>nv</sub> Gc. Following 1:10 dilution of mid-log phase Gc in GCBL, 1 ml aliquots of diluted Gc was added to 15 ml Falcon tubes. The dosage of diamide used in these experiments was 0 mM, 10 mM, and 20 mM. Following a 30 minute incubation in a drum rotator at 37°C, cultures were serially diluted in GCBL and plated. Survival was measured relative to the untreated samples.

Growth conditions were followed as described above for the MMS assay using FA1090 1-81-S2<sub>nv</sub> Gc. Following 1:10 dilution of mid-log phase GC in GCBL, 2ml aliquots of diluted Gc were added to 15ml Falcon tubes. Methyl methanesulfonate (MMS; Sigma) was added to a final concentration of 0, 0.001%, 0.005%, and 0.01%. Following a 60 minute incubation in a drum rotator at 37°C, cultures were serially diluted in GCBL and plated. Survival was measured relative to the untreated samples. The UV and PMN-mediated killing assay were performed identical to those described in (Skaar et al., 2002, Stohl et al., 2005) using FA1090 1-81-S2<sub>nv</sub> Gc.

**Figure S1. Examination of *ng1427* contribution to Gc survival.** Exponential phase *ng1427::cat*, *ng1428::tetM*, and parental Gc grown in liquid culture were exposed to: A: H<sub>2</sub>O<sub>2</sub>, B: Diamide, C: UV light, and D: PMNs with survival quantified relative to each respective untreated sample. IPTG-inducible complement strains with wild-type (E) or the G113D complement (F) were treated with H<sub>2</sub>O<sub>2</sub> with survival quantified relative to each respective untreated sample. Statistics were done using two-tailed Student's *t*-test with significance determined at *p*<0.05. Asterisk denotes *p*<0.05 between parent strain and *ng1427::cat*, and while statistical significance between parent and *ng1427::cat* occurred, in each case it was considered biologically irrelevant due to lack of major phenotype or failure to obtain significance upon further repetition. Error bars are +/- s.e.m.

