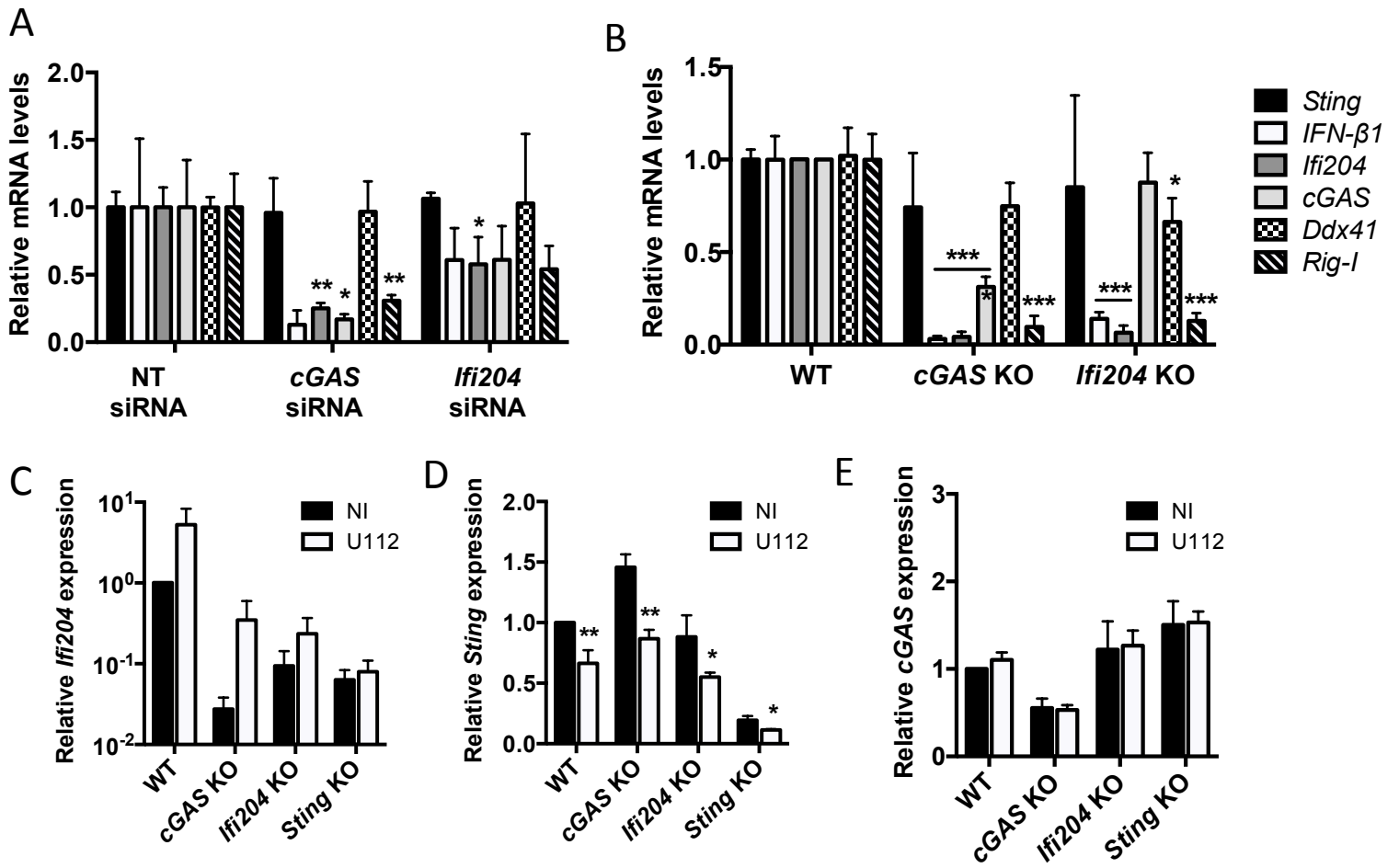


**Figure S1. Type I IFN response profiles in primary and immortalized macrophages.** (A) A standard curve for the L929 ISRE reporter assay using purified IFN-β is shown. (B) Type I IFN levels were measured from C57Bl/6 BMMs and RAW264.7 macrophages infected with indicated *F. novicida* strains at an MOI 10 and monitored overtime. Macrophages were infected with the indicated *F. novicida* strain at an MOI 10. Type I IFN activity was measured from C57Bl/6 BMM 12 h post-infection (C) or from RAW264.7 macrophages 24 h post-infection (D). Graphs show the mean ± standard deviation (SD) of triplicate wells and are representative of three independent experiments.



**Figure S2. mRNA expression profiles of uninfected and *F. novicida*-infected RAW264.7 mutant cell lines.** mRNA expression levels for the indicated gene were measured by qRT-PCR from uninfected C57Bl/6 BMMs (A) transfected with siRNA for 36 h or (B) uninfected RAW264.7 (WT) macrophage cell lines. mRNA expression levels of (C) *Iff204*, (D) *Sting* (E), and *cGAS* were measured from uninfected and U112 infected macrophage cell lines at an MOI 10 for 8 h. mRNA levels were normalized to GAPDH and either the NT control in BMMs or WT RAW264.7 macrophages. \* $p < 0.05$ , \*\* $p < 0.01$ , \*\*\* $p < 0.001$ . Graphs show the mean  $\pm$  standard deviation (SD) of three independent wells.

**Table S1. Primers used for this study.**

qRT-PCR primers (5'-3')

Gapdh F	ACTTTGTGAAGCTCATTTCCTGGTA
Gapdh R	GTGGTTTGAGGGCTCTTACTCCTT
Ifi204 F	GGAAAGAGACAACCAAGAGC
Ifi294 R	TGGCTTGTAGTTGATGTAGG
Sting F	CCTAGCCTCGCACGAACTTG
Sting R	CGCACAGCCTTCCAGTAGC
IFN- $\beta$ 1 F	CTGGAGCAGCTGAATGGAAAG
IFN- $\beta$ 1 R	CTTGAAGTCCGCCCTGTAGGT
Ddx41 F	CCTACCATTCTGTCCGGTCCG
Ddx41 R	CTCTCGCTTGGAGAAAGGCA
Lrrfip1 F	CTGAGAACCCTCTGCCTGTG
Lrrfip1 R	GTTGCCGAAGTGAAGTGCAG
RIG-I F	TGCAAAGCGCTTTCCTGTA
RIG-I R	GTAAGCTCTCGCTCGGTCTC
cGAS F	AGTCGTAAGGGGACCTAGCC
cGAS R	ACTCCCGTTTCTGCATTCTG
TNF $\alpha$ F	GATCGGTCCCAAAGGGATG
TNF $\alpha$ R	TGGTTTGTGAGTGTGAGGGTC

Cloning primers (5'-3')

cGAS Kozak XhoI F	TTTCTCGAGACCATGGAAGATCCGCGTAGAAGG
cGAS FLAG Sall R	TTTGTCTGACTCACTTATCGTCGTCATCCTTGTAATCAAGCTTGTCAAAAATT
Ifi204 Kozak XhoI F	GGAAACCC
Ifi204 6xHIS Sall R	TTTCTCGAGACCATGGTGAATGAATACAAGAGAATTG
	TTTGTCTGACTCAGTGGTGTGATGATGCTTTCTAGCATTGATGACCTG

Sequencing primers (5'-3')

cGAS seq F	CTCTGCTGGTCCCCACTGGTG
cGAS seq R	CCGGATCCAGGATGGCCTGC
Ifi204 seq F	CACCTCTTCTGCTTTCACCTG
Ifi204 seq R	CTGTCTGTCTGTCCCACCTTC
Sting seq F	GTCAGTTCTAGATTGAGTGGG
Sting seq R	GGTAAGGTTTAATGAAGTGG

**Table S2. Construction of targeted mutations in RAW264.7 macrophages**

Gene Target	Target Sequence for CRISPR/Cas9 mutations	Clone Number	Nature of mutation(s)
cGAS	GTCGGGGCGCGCTTCGCGGA	1	c119del; c122_123del
		2	c303_310del; c304_313del
Ifi204	GACAATCAGACGAGCCGAAG	1	g14389_14396del
		2	g14387_14393del; g14393del
Sting	AGCAAAAACATCGACCGTGC	1	c940del
		2	c939_940del; c941_942del