

Expanded View Figures

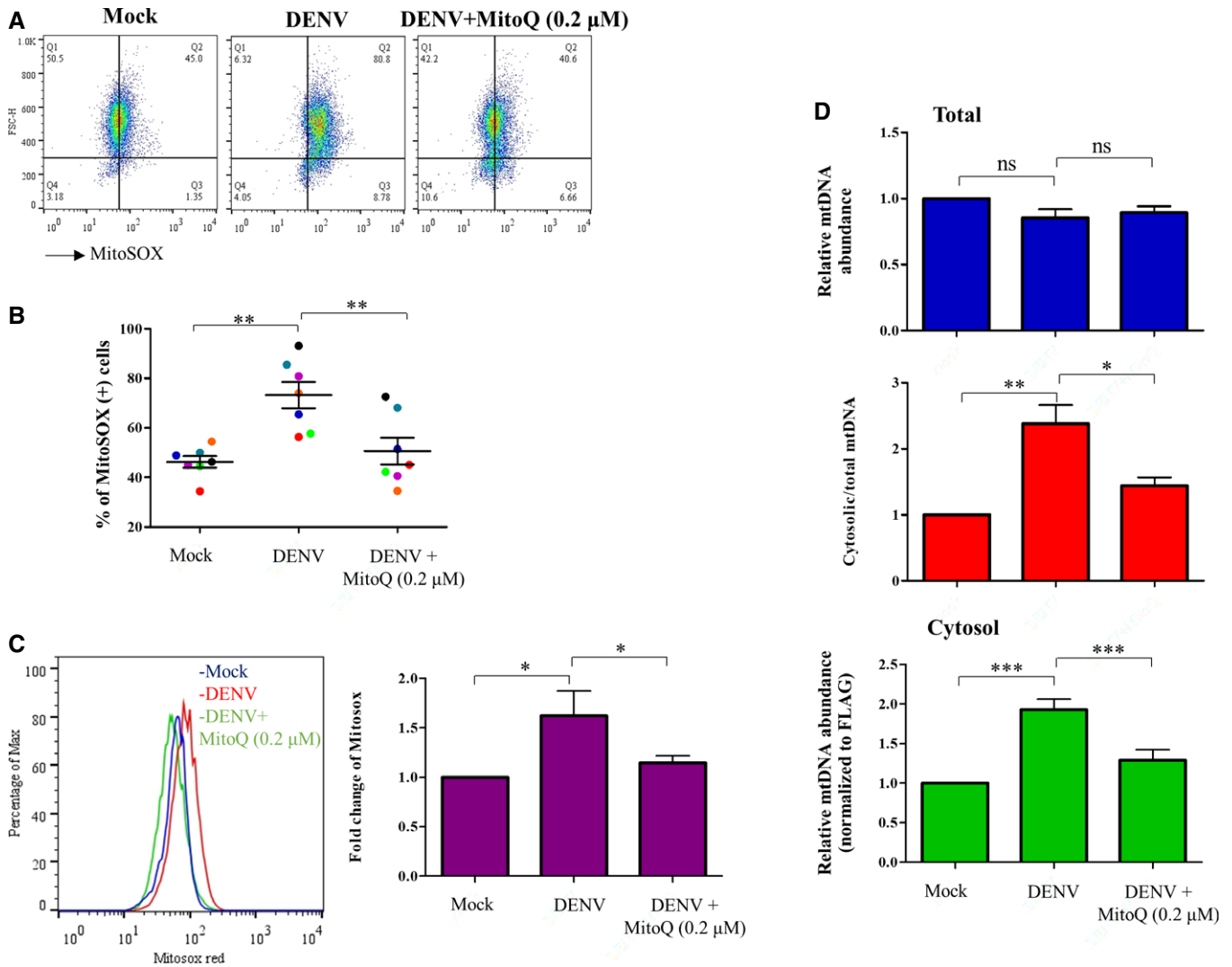


Figure EV1. Effects of a mitochondria-targeted antioxidant MitoQ in DENV-induced mtDNA release.

A–D Human DCs were infected with mock or DENV (MOI = 5) in the absence or presence of a mitochondria-targeted antioxidant MitoQ (0.2 μM). The intensity of MitoSOX fluorescence (A, $n = 7$), the percentages of MitoSOX-positive cells (B, $n = 7$), the percentages of Max (C, $n = 7$), and the relative abundance of mtDNA (D, $n = 7$) were determined. Values are means of individual measurements in each sample \pm SEM. * $P < 0.05$, ** $P < 0.01$, and *** $P < 0.001$ (paired, two-tailed Student's t -test).

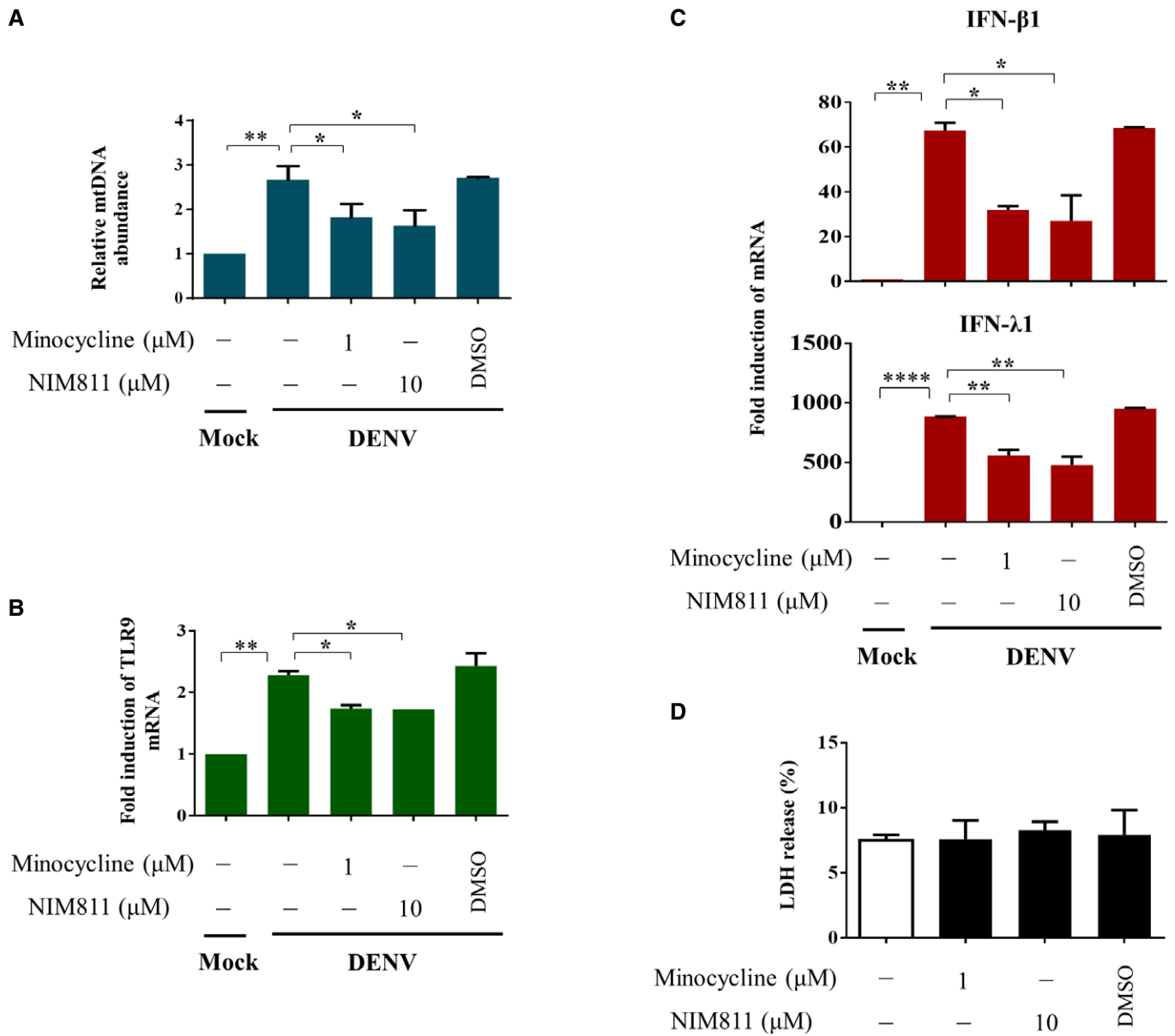


Figure EV2. Effects of minocycline and NIM811 on DENV-induced mtDNA release, TLR9 mRNA expression, and IFN mRNA expression.

A–D Human DCs (2×10^4) were infected with mock or DENV (MOI = 5) for 24 h in the absence or presence of minocycline (1 μM), NIM811 (10 μM) or DMSO. The relative abundance of mtDNA was measured using the exogenously added FLAG gene as a control for normalization (A, $n = 2$). The expression of TLR9 mRNA (B, $n = 2$), IFN-β1 and IFN-λ1 mRNA (C, $n = 2$), and LDH release (D, $n = 2$) were measured accordingly. Values are means of individual measurements in each sample ± SEM. * $P < 0.05$, ** $P < 0.01$, and **** $P < 0.0001$ (one-way ANOVA multiple comparisons with Bonferroni *post hoc* test).

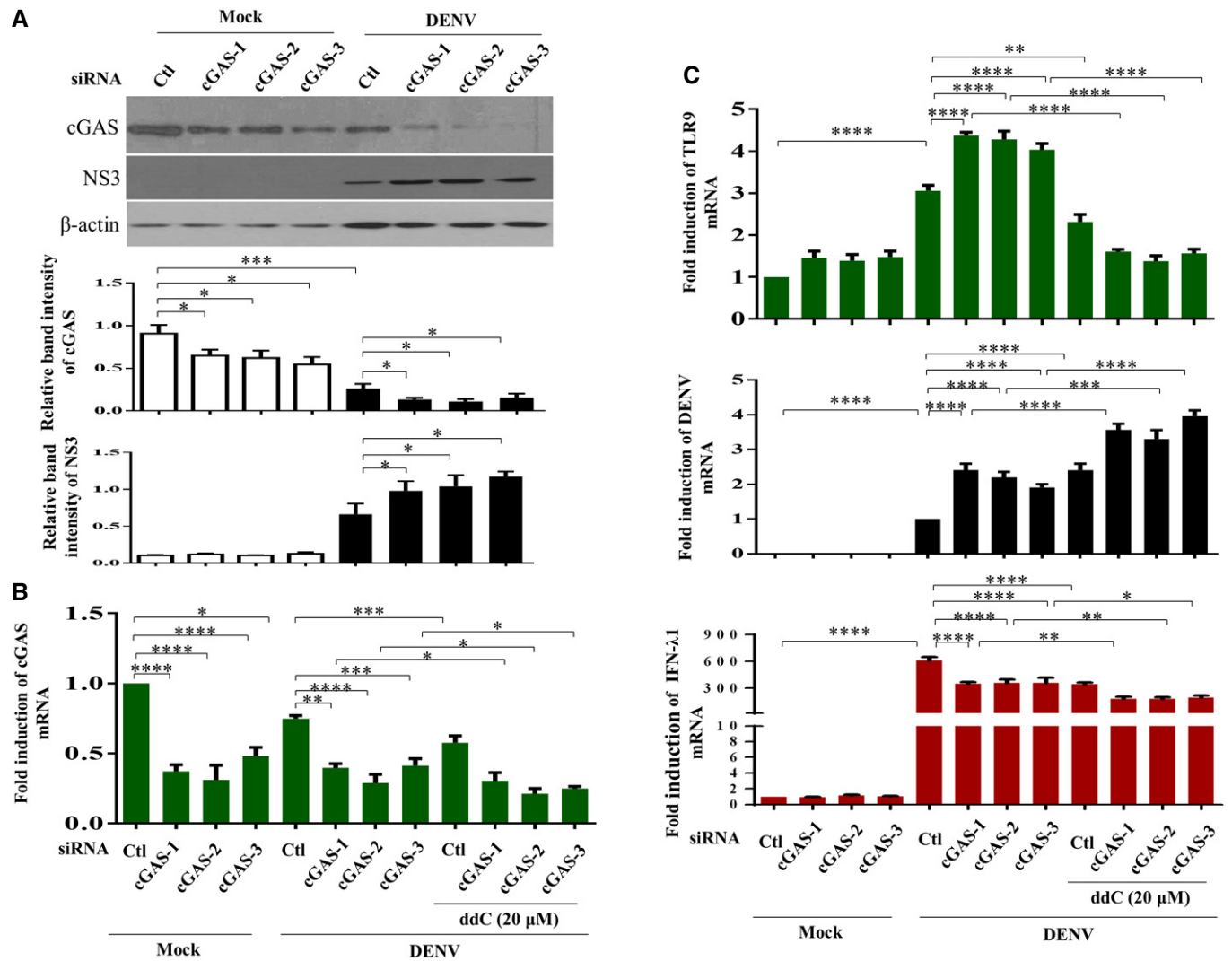


Figure EV3. Effects of cGAS knockdown with or without concomitant mtDNA depletion in DENV-infected DCs.

A–C Human DCs were transfected with control siRNA (Ctl) or three sets of cGAS siRNA duplexes for 24 h and then infected with mock or DENV (MOI = 5) for an additional 24 h. As indicated, ddC treatment to deplete mtDNA was introduced in some conditions. The cGAS, viral NS3 and β-actin protein levels (A, $n = 4$), cGAS mRNA (B, $n = 4$) and TLR9 mRNA, DENV mRNA, and IFN-λ1 mRNA (C, $n = 4$) were determined. Values are means of individual measurements in each sample \pm SEM. * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$, and **** $P < 0.0001$ (one-way ANOVA multiple comparisons with Bonferroni *post hoc* test).

Source data are available online for this figure.

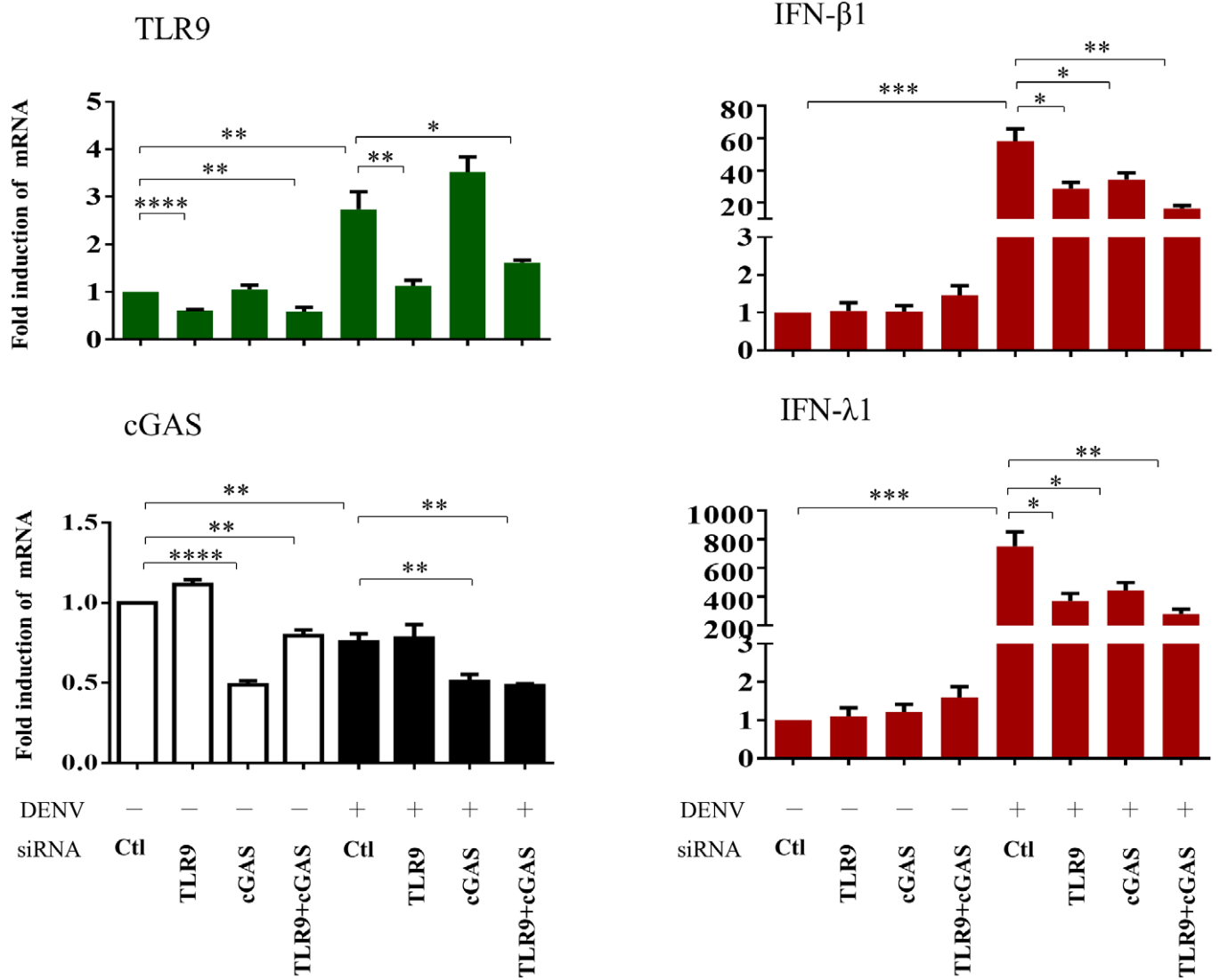


Figure EV4. Side-by-side comparisons of the effects of TLR9 and cGAS knockdown in DENV-induced IFN mRNA expression.

Human DCs were transfected with control siRNA (Ctl), TLR9 siRNA, cGAS siRNA, or both TLR9 and cGAS siRNA duplexes for 24 h and then infected with mock or DENV (MOI = 5) for an additional 24 h. The mRNA levels of TLR9, cGAS, IFN-β1, and IFN-λ1 were determined (*n* = 4). Values are means of individual measurements in each sample ± SEM. **P* < 0.05, ***P* < 0.01, ****P* < 0.001, and *****P* < 0.0001 (unpaired, two-tailed Student's *t*-test).