

Expanded View Figures

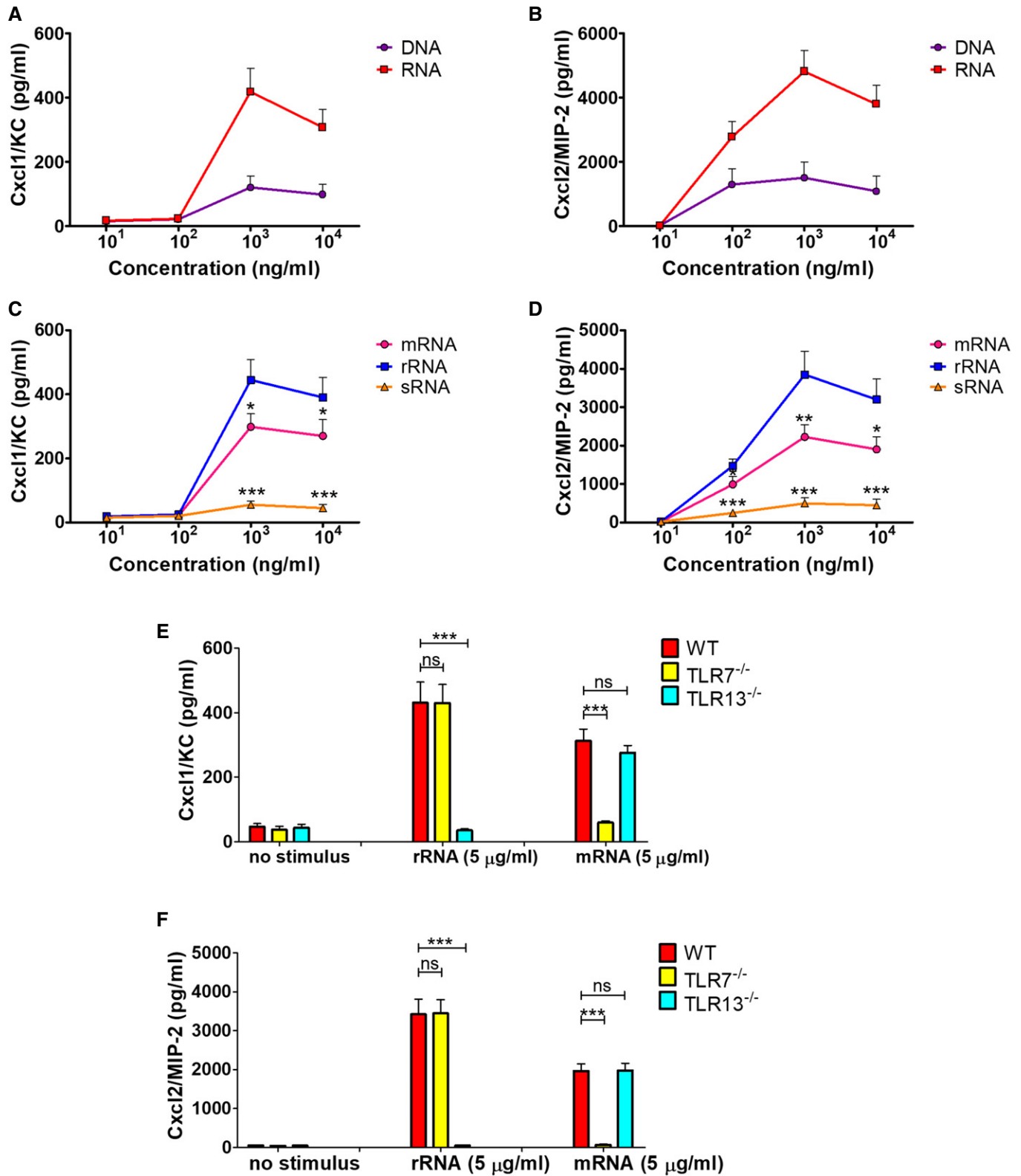


Figure EV1.

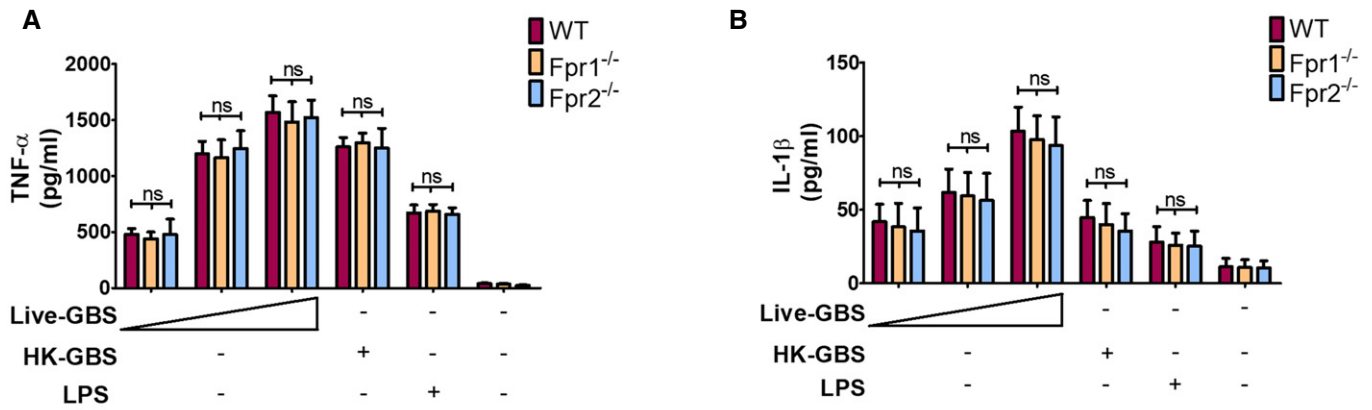
Figure EV1. Production of Cxcl1 and Cxcl2 in neutrophils stimulated with bacterial nucleic acids.

A–D Concentrations of Cxcl1 (A, and C) and Cxcl2 (B, and D) in supernatants of neutrophil cultures stimulated with increasing concentrations (10^1 , 10^2 , 10^3 , and 10^4 ng/ml) of the indicated nucleic acids. mRNA, messenger RNA; rRNA, ribosomal RNA; sRNA small RNA.

E, F Cxcl1 (E) and Cxcl2 (F) concentrations in supernatants of bone marrow-derived neutrophils from mice with genetic deficiency in TLR7 or TLR13 after stimulation with rRNA or mRNA. Chemokine levels in supernatants were measured at 24 h after stimulation.

Data information: Data are expressed as means + SD of three independent experiments conducted in duplicate. * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$ versus RNA (A and B), rRNA (C and D) or WT mice (E and F), determined by unpaired t-test.

Neutrophils



M-CSF-MΦ

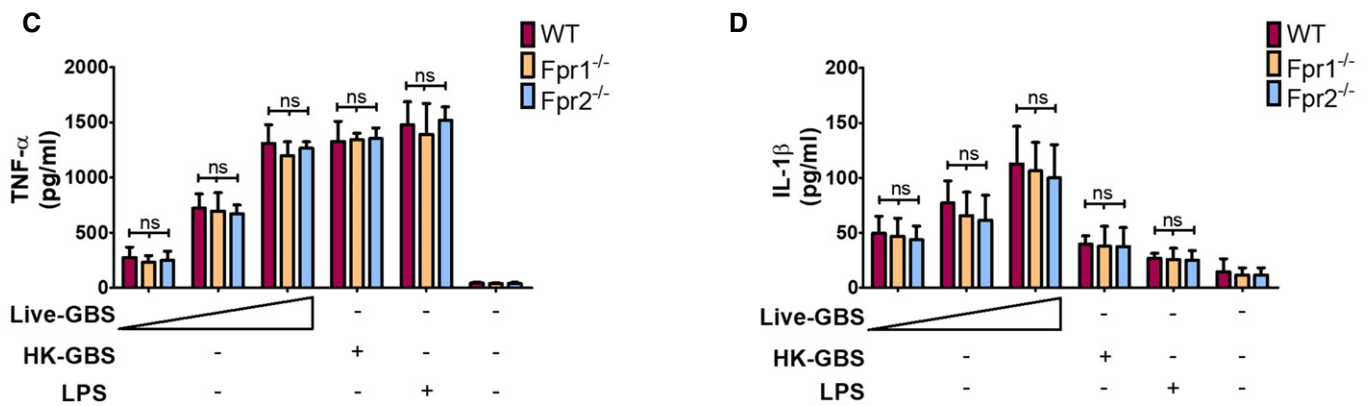


Figure EV2. Production of pro-inflammatory cytokines in Fpr-defective phagocytes.

A–D Concentrations of TNF- α (A and C) and IL-1 β (B and D) in 24-h culture supernatants of neutrophils (A and B) or M-CSF-polarized BMDMs (C and D) from WT or Fpr-deficient mice stimulated with live GBS (MOI of 2, 5, and 10) or HK-GBS (10 μ g/ml). LPS (100 ng/ml) was used as a positive control stimulus. Means + SD of data from three independent experiments conducted in duplicate. ns, non-significant, as determined by unpaired t-test.

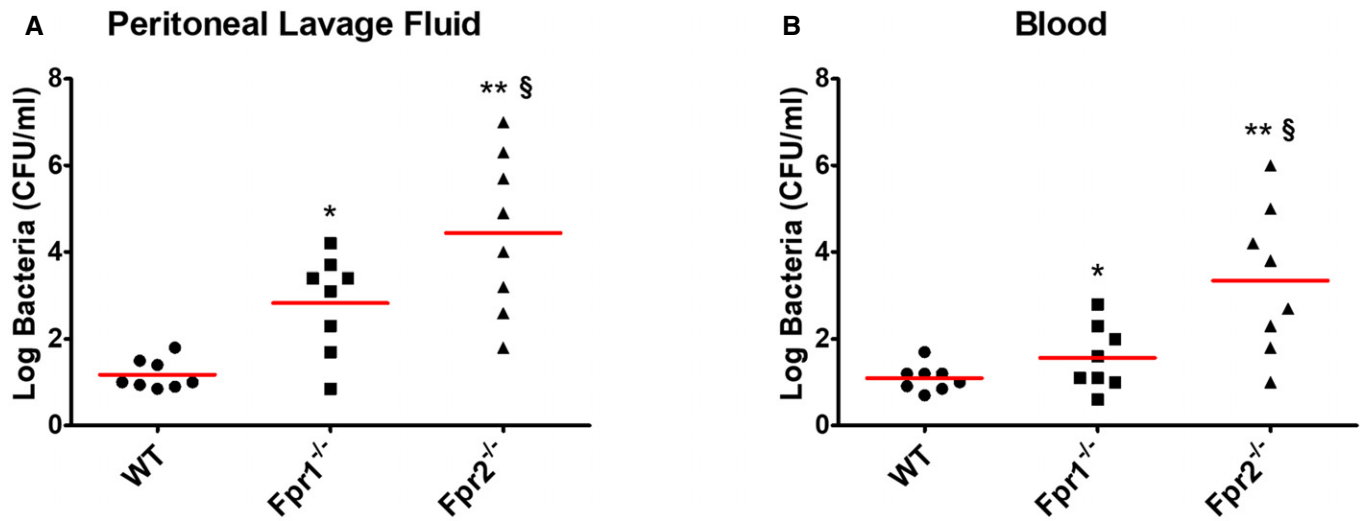


Figure EV3. Bacterial burden in Fpr-defective mice after challenge with GBS.

A, B CFU numbers in peritoneal lavage fluid (A) and blood (B) samples from wild type (WT), Fpr1, and Fpr2 KO mice at 24 h after i.p. infection with live GBS (2×10^5 CFU). Horizontal red bars indicate mean values. Each determination was conducted on a different animal in the course of two experiments, each involving 4 animals per group. * $P < 0.05$, ** $P < 0.01$ vs. WT mice, § $P < 0.05$ vs. Fpr1-deficient mice, as determined by the Wilcoxon test.

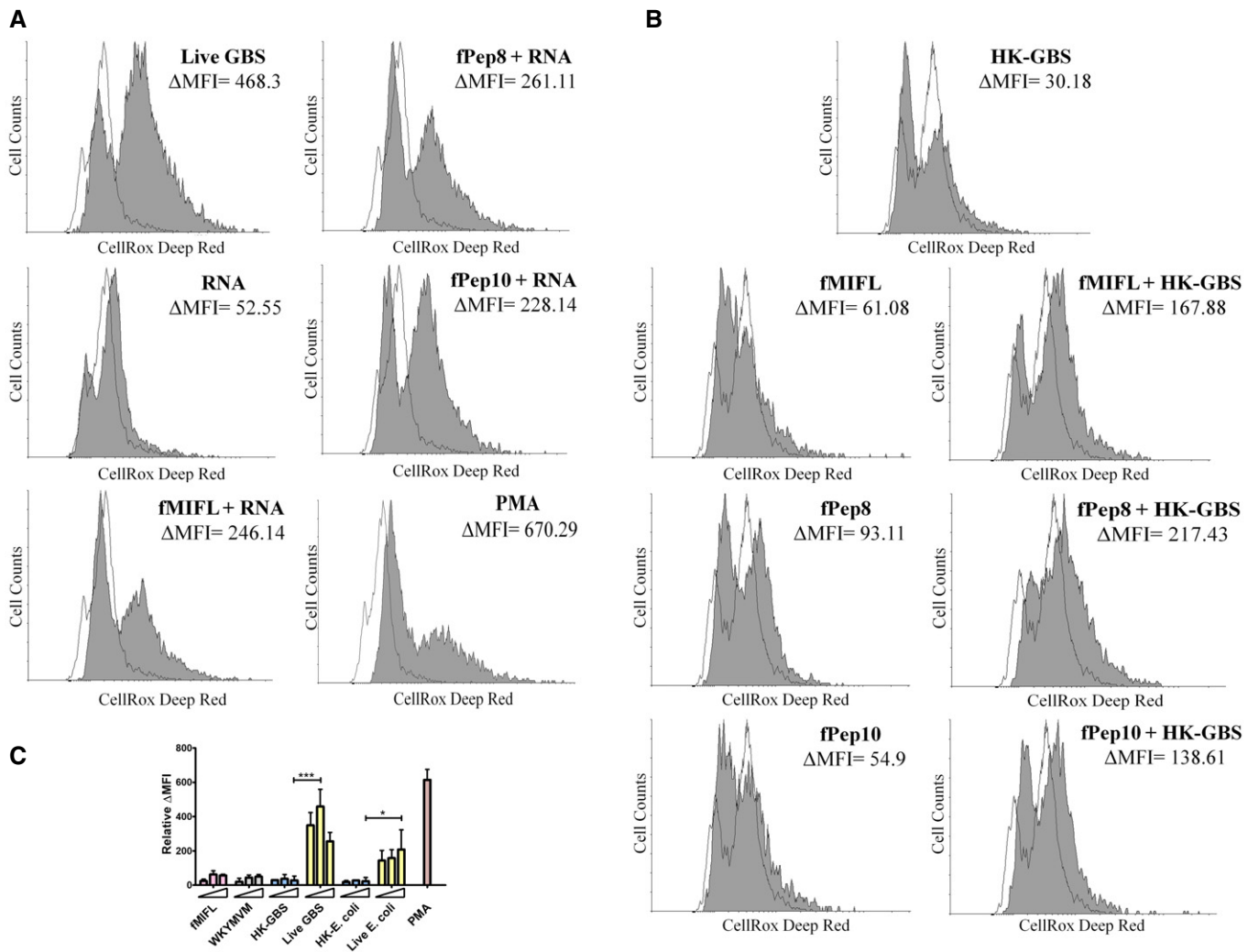


Figure EV4. ROS production in neutrophils stimulated with Fpr and TLR agonists.

A, B ROS production in neutrophils stimulated with live GBS (MOI 200) or combinations of Fpr agonists (fMIFL, WKYMVM, or GBS formylated peptides, 50 μM) and TLR agonists (HK-GBS, 10 $\mu\text{g}/\text{ml}$ or bacterial RNA, 1 $\mu\text{g}/\text{ml}$). Phorbol-12-myristate-13-acetate (PMA, 25 ng/ml) was used as a positive control stimulus. Data from one representative experiment of three producing similar results. Cells were stained with the CellROX fluorescent reagent. The shadowed area indicates stimulated cells.

C ROS production in neutrophils stimulated with increasing concentrations of fMIFL or WKYMVM (10, 20, and 50 μM), HK-bacteria (GBS or *E. coli*, 10, 20, and 40 $\mu\text{g}/\text{ml}$) or increasing numbers of live bacteria (MOIs of 100, 200, and 400). PMA (25 ng/ml) was used as a positive control stimulus. Data are expressed as means + SD from three experiments conducted in duplicate. * $P < 0.05$, *** $P < 0.001$, as determined by unpaired *t*-test.

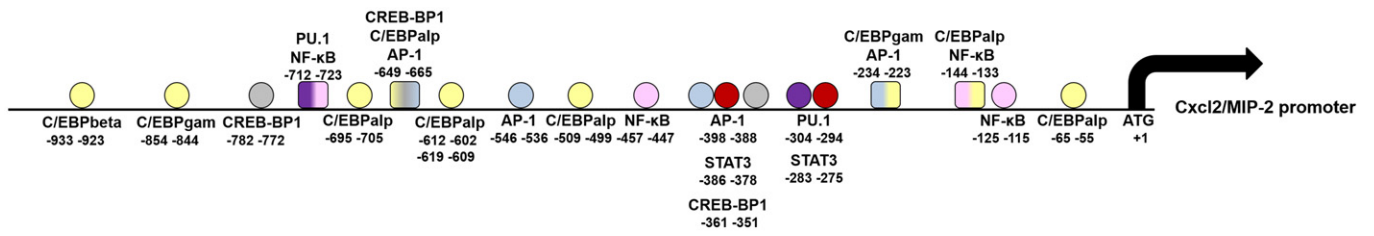


Figure EV5. Schematic representation of the Cxcl2 promoter.

The sequence 1,000 nt upstream of the start site was analyzed for the presence of transcription factor-binding sites using the Alibaba2.1 software (Grabe, 2002).