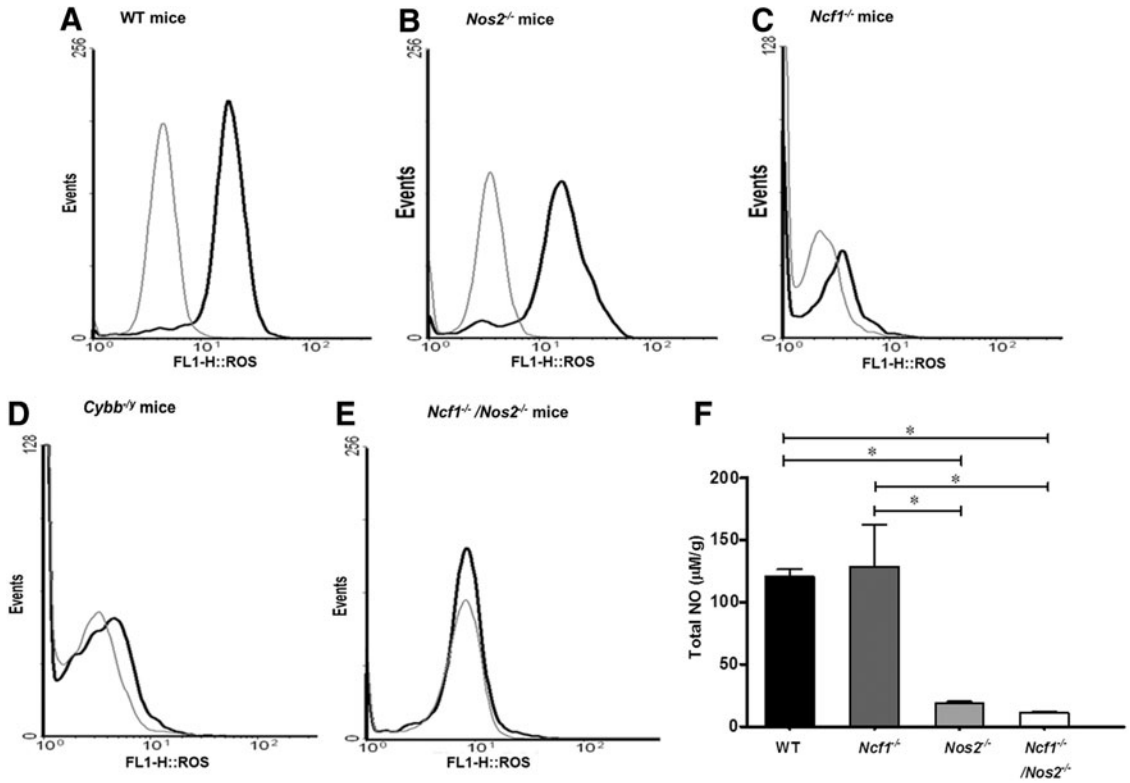
Supplementary Data



SUPPLEMENTARY FIG. S1. The functional defects of the leukocytes from their respective mouse strains were confirmed with direct measurement of ROS or NO from their leukocytes. The blood of mice was harvested from B6 wild-type, *Nos2*$^{-/-}$, *Ncf1*$^{-/-}$, *Cybb*$^{-/-}$, and *Ncf1*$^{-/-}$*/Nos2*$^{-/-}$ mice. **(A–E)** The red blood cells were lysed by RBC lysis buffer. The blood cells (1×10^6 cells/ml) were then incubated with H2DCFDA (5 μg/ml) in suspension buffer for 30 min at 37°C. PMA (400 ng/ml) was added 15 min before collecting cells. The fluorescent intensities were then analyzed by flow cytometry. Untreated cells with H2DCFDA were used as a negative control. The histogram overlays show the results of treated cells (*black lines*) compared with untreated cells (*gray lines*). **(F)** The sum of total NO (nitrate and nitrite) produced in mouse lungs of wild-type ($n=4$), *Ncf1*$^{-/-}$ ($n=4$), *Nos2*$^{-/-}$ ($n=3$), and *Ncf1*$^{-/-}$*/Nos2*$^{-/-}$ ($n=3$) mice. Total NO (nitrate and nitrite) levels were measured with the nitrate/nitrite colorimetric assay kit (No. 780001; Cayman Chemical, Ann Arbor, MI). Two steps were carried out before measurement with the Griess method. First, the lung homogenates were deproteinized using 10 KDa Amicon® Ultracentrifugal filters (Millipore, Bedford, MA). After deproteinization, an NADPH-dependent nitrate reductase was used to convert all nitrates to nitrites. The Griess reagents then converted the nitrite into deep purple azo compounds, and the absorbance of the azochromophore was measured spectrophotometrically at 540 nm. The statistically significant differences between groups by one-way ANOVA with the Bonferroni correction are indicated with * (*$p < 0.05$). The experiments were repeated twice with similar results. ANOVA, analysis of variance; ROS, reactive oxygen species.