

SUPPLEMENTAL MATERIAL

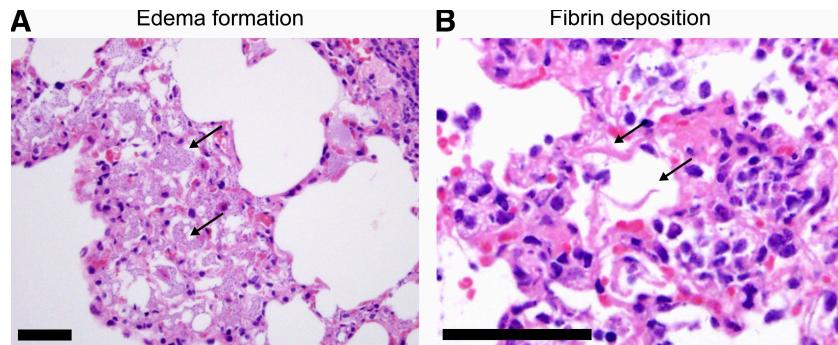
Nagamatsu et al., <http://www.jem.org/cgi/content/full/jem.20090494/DC1>

Figure S1. Edema formation and fibrin deposition were observed in the lungs of mice infected with *Bordetella* WT. (A and B) 2 d after infection with WT *Bordetella*, lung sections were stained with H&E. Arrows indicate edema formation (A) and fibrin deposition (B), respectively. Bar, 50 μ m.

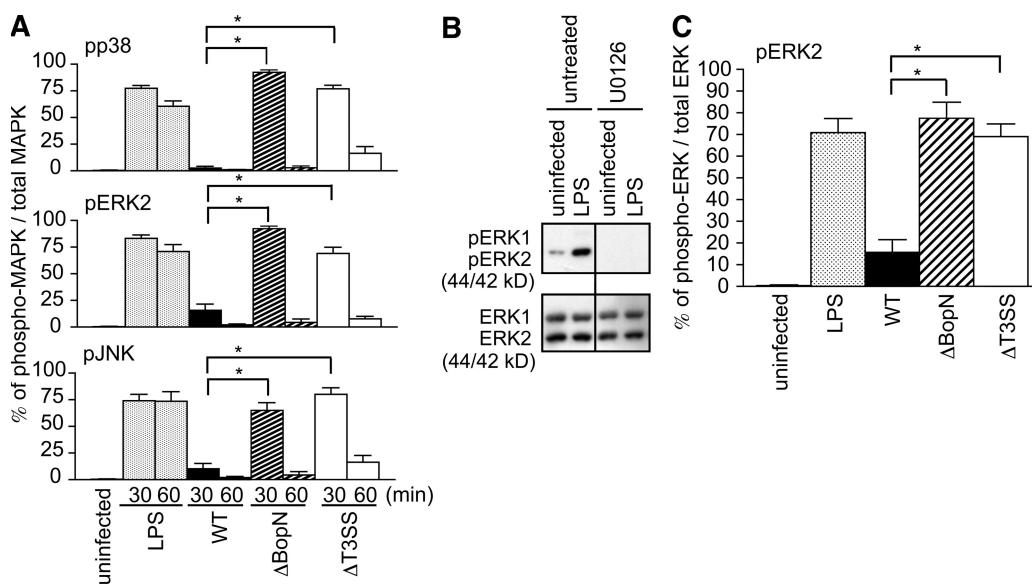


Figure S2. BopN regulates MAPK activities. (A) Band intensities of phospho-MAPK and total MAPK obtained from immunoblots (Fig. 7 A) were quantified using ImageJ, and percentages of phospho-MAPK/total MAPK were determined from three independent experiments. For JNK, intensities of the top bands were quantified. The values are means \pm SE. *, P < 0.05. (B) The MEK1/2 inhibitor U0126 was added to DC2.4 culture medium at 10 μ M for 1 h before LPS stimulation, and the antibody specificity against phospho-ERK was confirmed by immunoblot analysis. (C) The percentages of cells showing phospho-ERK/total ERK were determined from results of immunofluorescence analysis (Fig. 7 B). The percentages were based on a count of 50 cells. The values are means \pm SE from three independent experiments. *, P < 0.05.

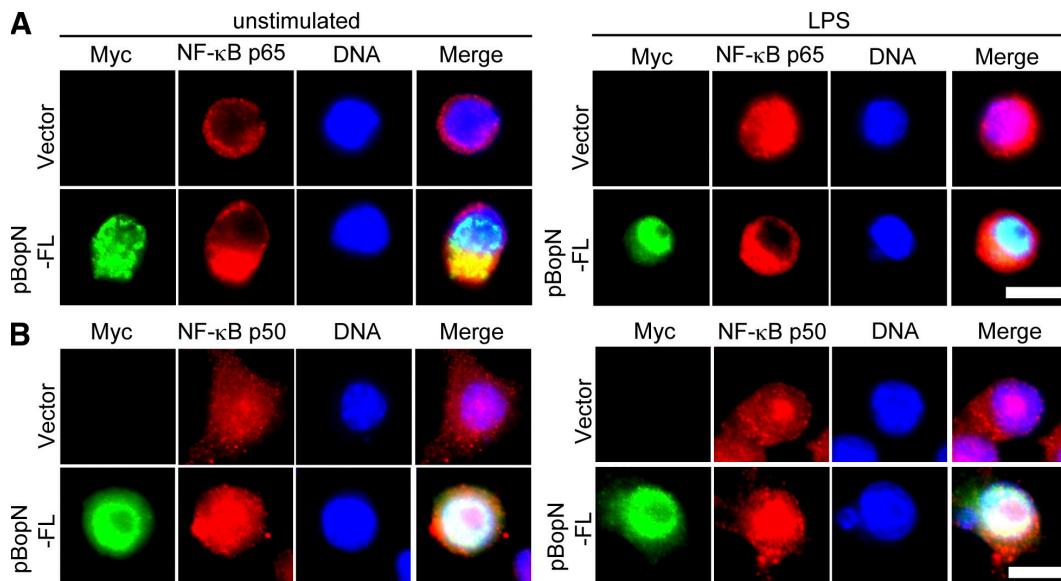


Figure S3. The nuclear localization of NF- κ B subunits in a macrophage cell line was modulated by introduction of a *bopN* clone. A mammalian expression vector, pcDNA3.1/His(-)A, encoding full-length BopN-FL (aa 1–365) with a Myc-tag fused at its C terminus was introduced into RAW264.7 cells. 24 h after transfection, cells were stimulated with 10 μ g/ml LPS for 10 min, and then fixed and stained with anti-Myc antibodies to detect BopN (green), anti-NF- κ Bp65 (A, red), or anti-NF- κ Bp50 (B, red), and DAPI for nuclei (blue). Bars, 10 μ m.

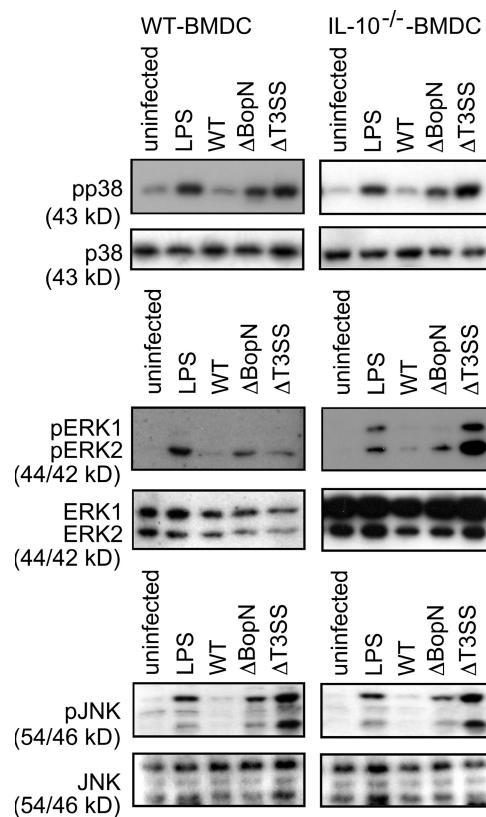


Figure S4. IL-10 production is not involved in the down-regulation of MAPKs in BMDCs during *Bordetella* infection. BMDCs from either WT or IL-10^{-/-} mice were stimulated with 10 μ g/ml LPS or infected with the indicated strains for 30 min. Immunoblot analyses of MAPK activities in the lysates of BMDCs were performed. Three independent experiments were performed and a representative immunoblot is shown.

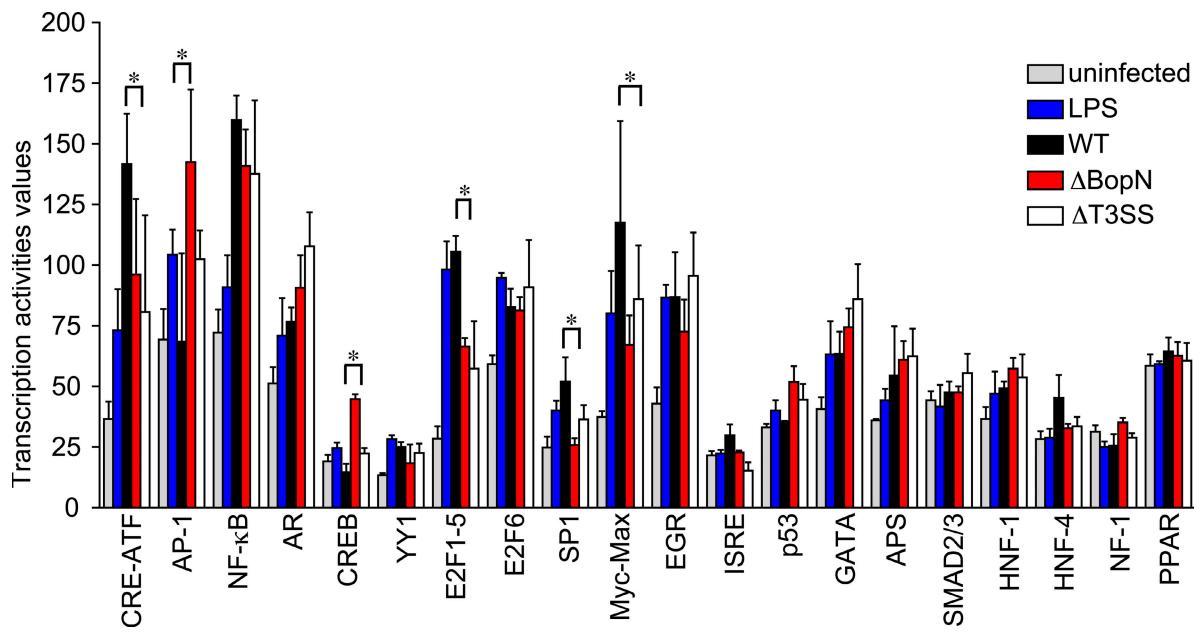


Figure S5. Activities of transcription factors are altered by *Bordetella* infection. DC2.4 cells were stimulated with 10 µg/ml LPS or infected with *B. bronchiseptica* WT, ΔBopN, and ΔT3SS for 30 min; resulting DCs were separated into nuclear fractions, and resulting fractions were analyzed by Multiplex transcription factor profiling kit. Differences in activation of transcription factors were determined by Bio-Plex. The values are means ± SE from three independent experiments. *, P < 0.05.