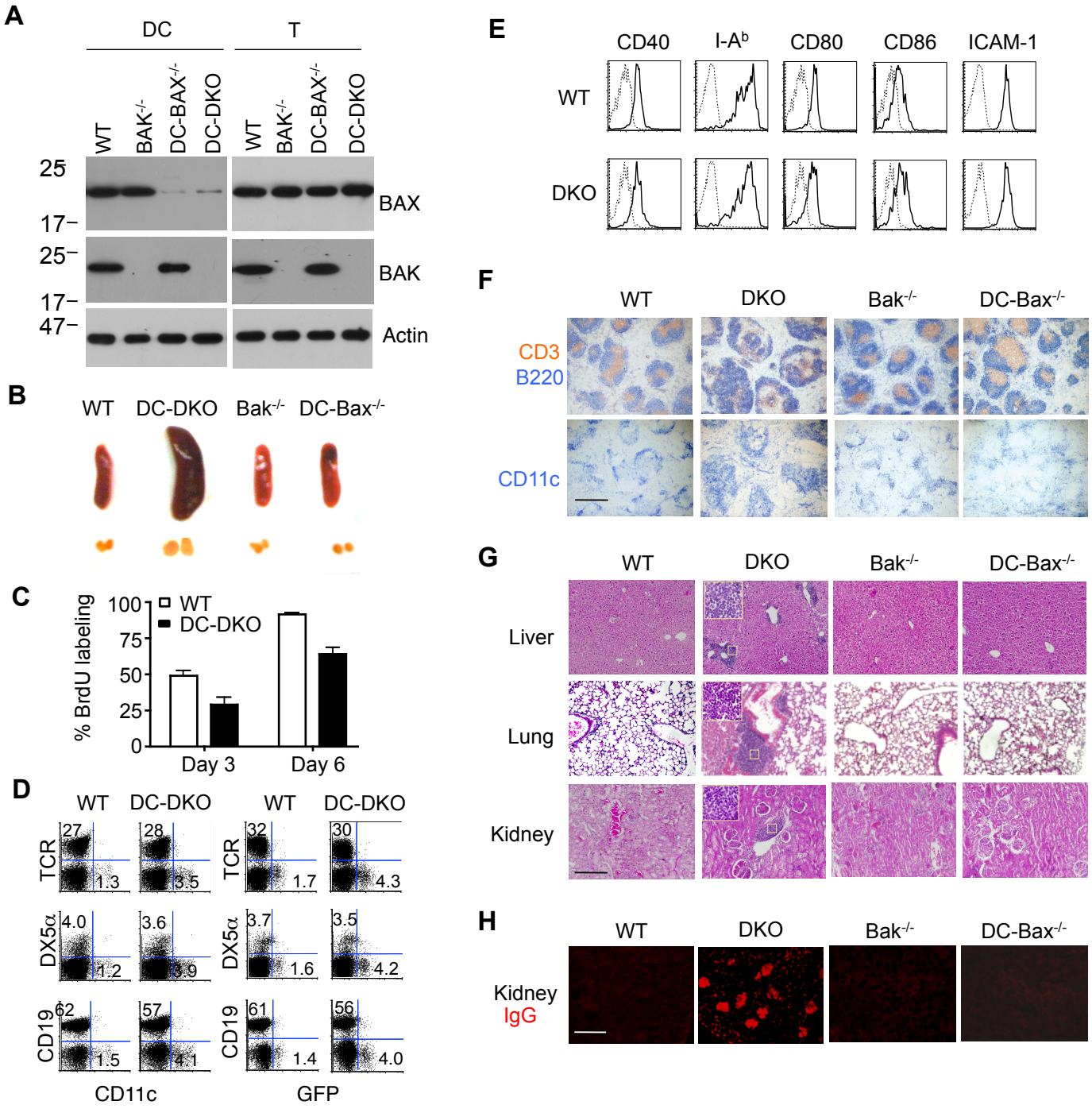
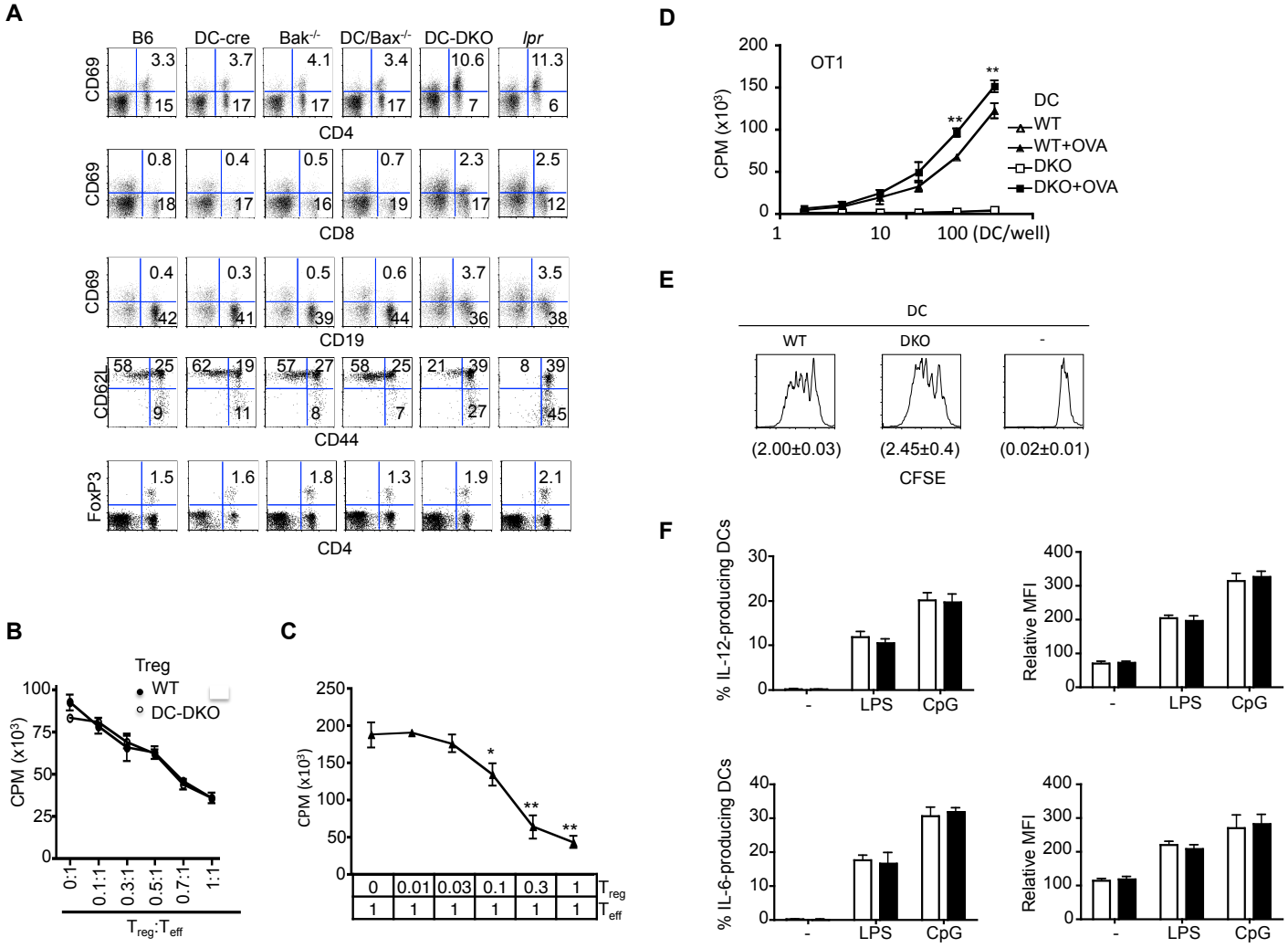
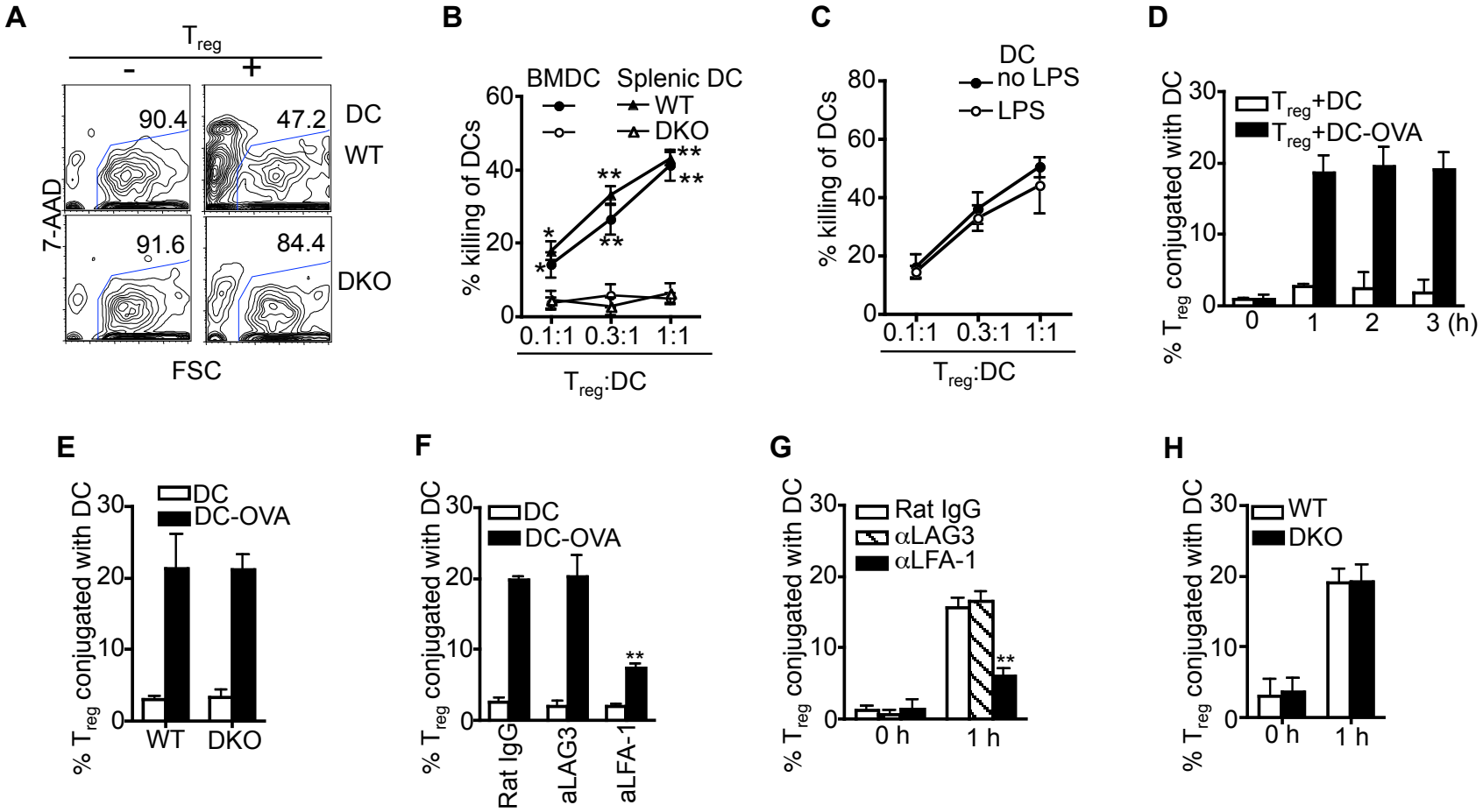


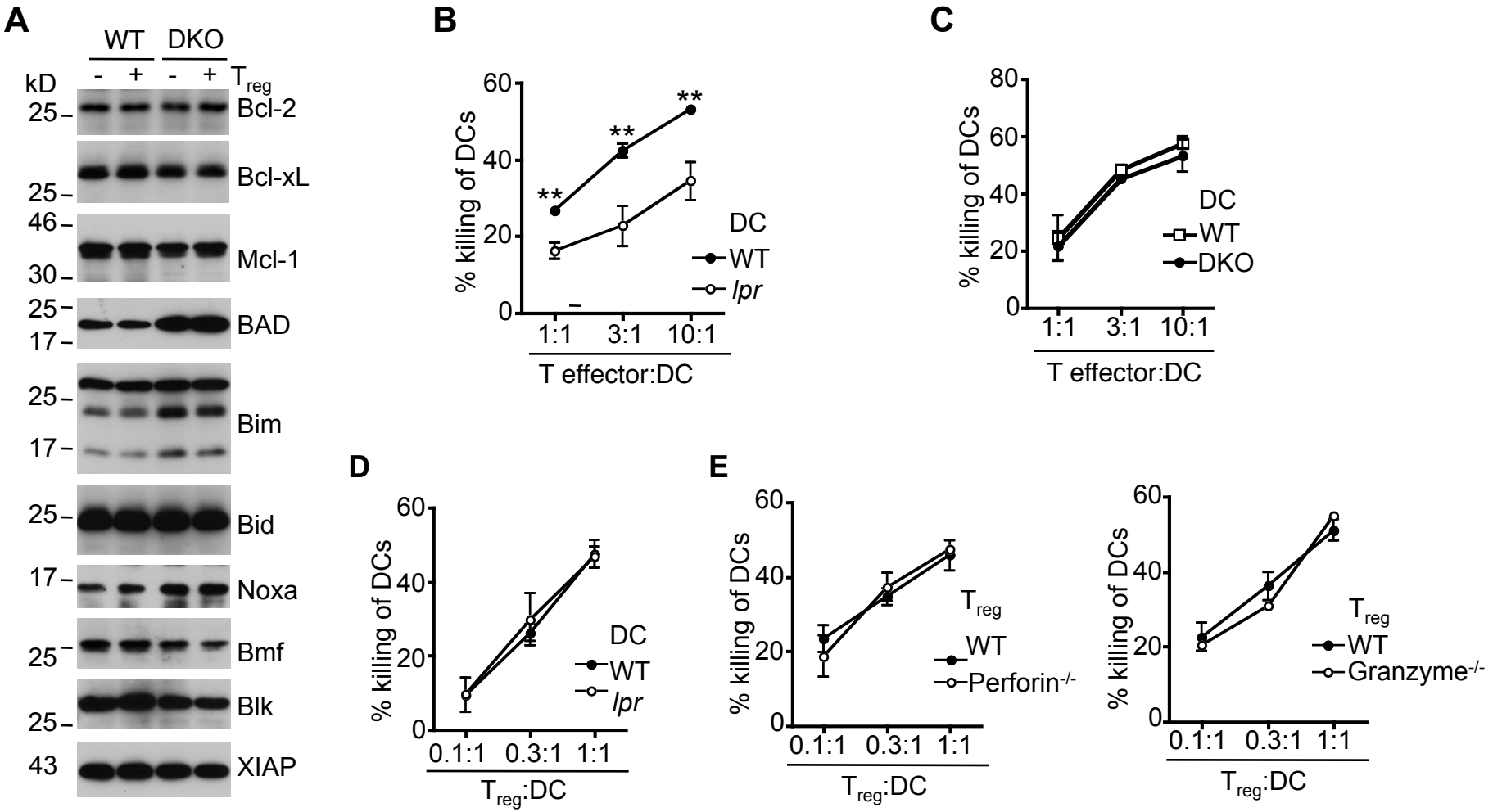
Supplemental Figure 1. **Analyses of DC-DKO mice.** (A) Western blot for Bax and Bak in DCs, T cells in DC-DKO mice or controls. Bax is deleted in DCs, but not in lymphocytes in DC-DKO mice. (B) Enlargement of the spleen and inguinal lymph nodes in six-month-old DC-DKO mice (N=7). (C) Splenocytes from mice labeled with BrdU for 3 or 6 days were stained with antibodies to CD11c and CD11b, followed by intracellular staining with FITC-anti-BrdU. Percentages of BrdU labeling of $CD11c^+CD11b^+$ DCs (mean ± SD) were determined by flow cytometry (6 mice/ group). (D) Splenocytes of WT and DC-DKO mice were stained with APC-anti-CD11c and PE-conjugates anti-TCRβ, anti-DX5a or anti-CD19, followed by flow cytometry. The expression of CD11c or GFP under the control of the transgenic CD11c promoter (CD11c-cre-IRES-GFP) versus other cell surface markers was plotted. (E) Expression of surface molecules on DCs in DC-DKO vs WT mice (N=5). CD40, $I\text{-}A^b$, CD80 CD86 and ICAM-1 on $CD11c^+CD11b^+$ DCs (solid lines) were examined by flow cytometry. Dotted lines: isotype control. (F) Immunohistochemistry analyses for T cells ($CD3^+$), B cells ($B220^+$) and DCs ($CD11c^+$) in the spleens of 6-month-old DC-DKO and control mice (N=6). Scale bar: 50 μm. (G) Sections of lungs, livers and kidneys of 6-month-old DC-DKO or control mice (N=6) were stained with H&E. Scale bar: 50 μm. (H) Kidney sections of 6-month-old DC-DKO or control mice were stained with Alexa Fluor 594-conjugated anti-mouse IgG. Scale bar: 50 μm.



Supplemental Figure 2. **Activation of T cells and cytokine production by DCs.** (A) Splenocytes from 6-month-old DC-DKO and control mice were stained with antibodies to different markers and analyzed by flow cytometry. CD69 expression on $CD4^+$ T cells, $CD8^+$ T cells and $CD19^+$ B cells and CD44 and CD62L expression on $TCR\alpha\beta^+$ T cells were analyzed. Splenocytes were also stained with FITC-anti-CD4, followed by intracellular staining with PE-anti-FoxP3. Data are representative 5 mice per genotype. (B) $CD4^+CD25^{high}$ T_{reg} cells from DC-DKO and control mice were incubated with $CD4^+CD25^-$ T_{eff} cells at different ratios, and cultured in the presence of 0.25 μg/ml soluble anti-CD3 and irradiated T-cell-depleted spleen cells. Proliferation was quantified by ^{3}H-thymidine incorporation 4 days later. (C) $CD4^+FoxP3^+$ T_{reg} cells expanded *in vitro* were mixed with freshly sorted $CD4^+FoxP3^-$ T_{eff} cells from GFP-FoxP3 mice at different ratios in the presence of 0.25 μg/ml soluble anti-CD3 and irradiated spleen cells depleted of T cells and DCs ($5x10^4$/well). Cell proliferation were quantitated 4 days later by ^{3}H-thymidine incorporation. Statistic comparison to control with no T_{reg} cells: **P*<0.05, ***P*<0.01. (D) WT or DKO BMDCs were unpulsed or pulsed with $OVA_{SIINFEKL}$ peptide. Various numbers of DCs were then incubated with MACS beads purified OVA-specific $CD8^+$ transgenic OT1 T cells ($5x10^4$/well). Cell proliferation was measured by ^{3}H-thymidine incorporation 3 days later. WT+OVA versus DKO+OVA: ***P*<0.01. (E) CFSE-labeled $CD8^+$ OT1 T cells ($5x10^4$/well) were labeled with CFSE and incubated with DCs pulsed with $OVA_{SIINFEKL}$ peptide (300 DCs/well). Cell proliferation was measured by CFSE dilution 4 days later. Average numbers of cell cycle ± SD are shown. WT versus DKO: *P*=0.03. (F) WT or DKO BMDCs were stimulated with LPS or CpG for 24 h, followed by intracellular staining of IL-12 or IL-6. The percentage of IL-12- or IL-6-producing cells and the mean fluorescence staining (MFI) (means ± SD) for IL-12 or IL-6 staining were plotted. The statistic difference between WT and DKO: not significant.



Supplemental Figure 3. T_{reg} cells in the killing of and conjugate formation with DCs. (A) BMDCs were incubated in the absence or presence of Treg cells. An example of forward scattering (FSC) versus 7-AAD staining of DCs was plotted. (B) T_{reg} cells in the killing of BMDCs or splenic DCs from wild type or DC-DKO mice. WT versus DKO: **P*<0.05, ***P*<0.01. (C) T_{reg} cells in the killing of BMDCs with or without LPS stimulation. (D) OT2 FoxP3$^+$ Treg cells and DCs labeled with CFSE and CMTMR, respectively, were mixed (1:1) and centrifuged at 500 g for 5 min, followed by incubation at 37 ^{o}C for different time. The cells were collected and analyzed by flow cytometry. Percentage of T_{reg} cells that formed conjugates with DCs was plotted. Groups with OVA versus no OVA at 1, 2 and 3 h, *P*<0.01. (E) Percentages of OT2 FoxP3$^+$ Treg cells forming conjugates with OVA-pulsed WT or DKO DCs after 1 h of incubation. (F) OT2 FoxP3$^+$ T_{reg} cells and OVA-pulsed DCs were labeled with CFSE and CMTMR, respectively, and cultured as in (D) in the presence of 10 μg/ml of different antibodies for 1 h. Anti-LFA-1 versus IgG: ***P*<0.01. Each group with DC-OVA versus no OVA: *P*<0.01. (G) CD4$^+$FoxP3$^+$ T_{reg} cells expanded *in vitro* were mixed with DCs in the presence of rat IgG, anti-LAG3 or anti-LFA-1 for 1 h, followed by quantification of cell conjugates. Anti-LFA-1 versus IgG: ***P*<0.01. (H) Percentages of CD4$^+$FoxP3$^+$ T_{reg} cells as in (G) forming conjugates with WT or DKO DCs.



Supplemental Figure 4. T_{reg}-mediated cell death in DCs. (A) Wild type or DKO BMDCs were incubated with or without T_{reg} cells (T_{reg}:DC at 0.5:1) for 6 h. $CD11^+$ DCs were isolated using MACS beads and lysed for Western blot. Data are representative of 2 independent experiments. (B, C) $CD4^+FoxP3^-$ T effector cells were activated with anti-CD3/anti-CD28 Dynabeads and incubated with wild type or *lpr* (B) or $Bax^{-/-}Bak^{-/-}$ (C) BMDCs in the presence of 1 μg/ml anti-CD3 for 4 h. Killing of DCs was measured by flow cytometry. WT versus *lpr*: $^{**}P<0.01$. (D) Killing of wild type or *lpr* BMDCs by activated $CD4^+FoxP3^+$ T_{reg} cells. (E) T_{reg}-mediated killing of DCs. WT and $PFP^{-/-}$ (left panel) or granzyme $A^{-/-}$/granzyme $B^{-/-}$ (right panel) T_{reg} cells were incubated with CFSE-labeled DCs at a ratio of 0.1:1, 0.3:1 or 1:1 for 6 h, followed by measurement of killing of DCs.