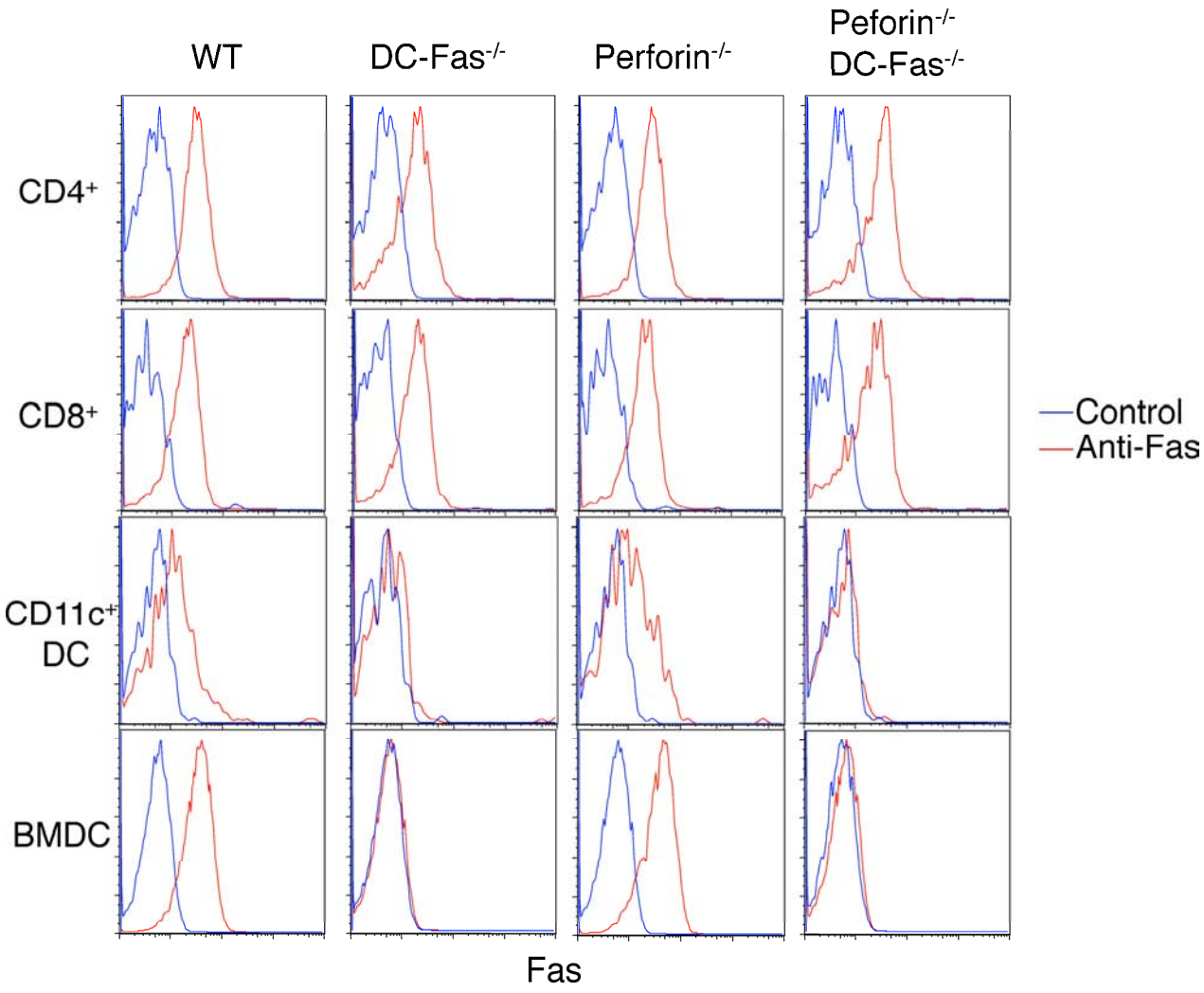

Figure S1. Expression of Fas on lymphocytes and DCs from perforin$^{-/-}$DC-Fas$^{-/-}$and control mice

Splenocytes from 4-week-old WT, perforin$^{-/-}$, DC-Fas$^{-/-}$, and perforin$^{-/-}$DC-Fas$^{-/-}$ mice were stained with APC-conjugated antibodies to CD4, CD8, or CD11c, followed by staining with PE–anti-Fas. Bone marrow derived DCs (BMDC) were also stained with PE–anti-Fas. The cells were analyzed by flow cytometry and Fas staining on different cell types was plotted.

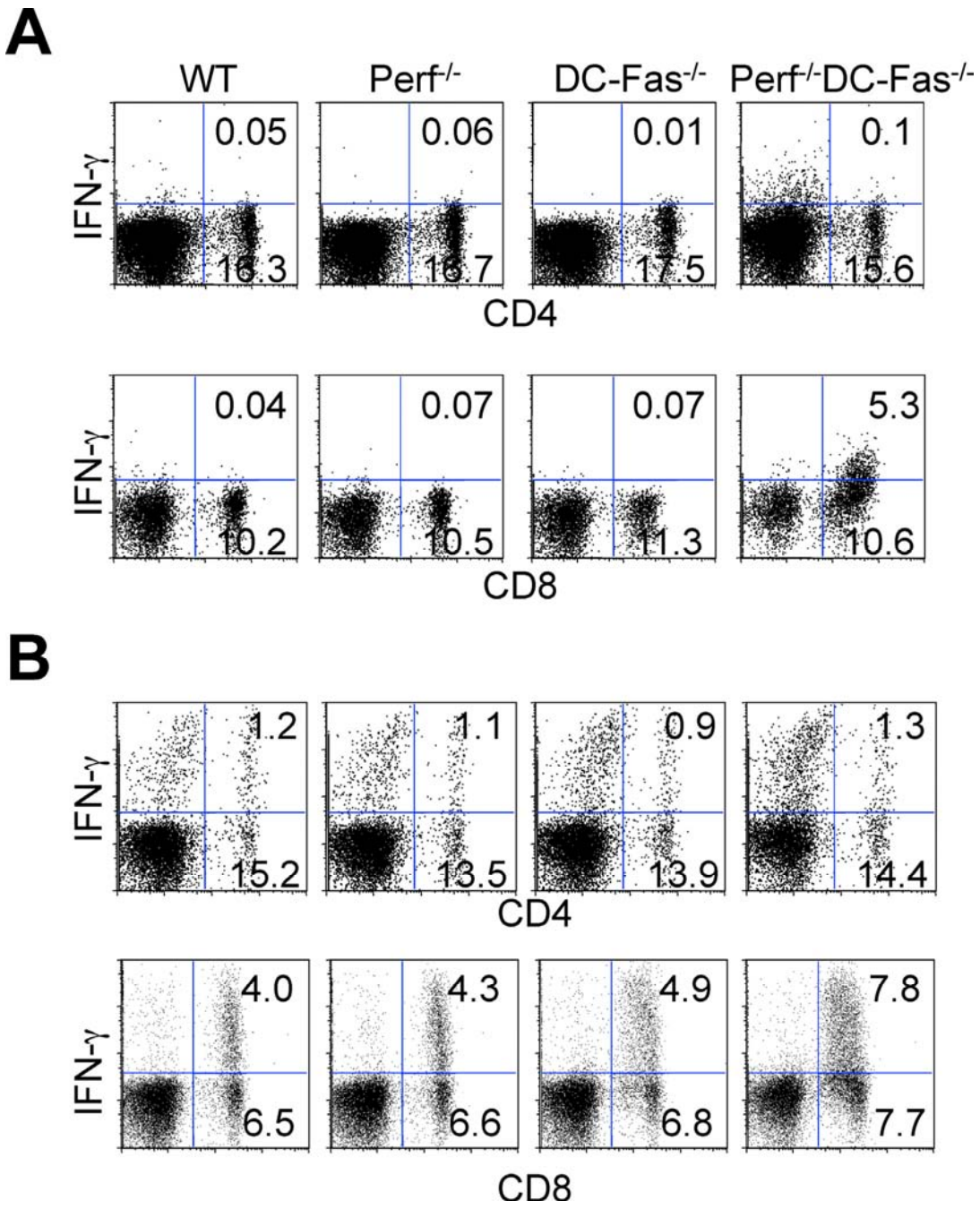

Figure S2. IFN-γ production by CD8$^+$ T cells in perf$^{-/-}$DC-Fas$^{-/-}$ mice
(A) Splenocytes from 4-week-old perf$^{-/-}$DC-Fas$^{-/-}$ mice were stained with APC–anti-CD4 and Cychrome–anti-CD8, followed by intracellular staining with PE–anti-IFN-γ. (B) Splenocytes were stimulated with 1 μg/ml anti-CD3 and anti-CD28 in the presence of brefeldin A for 5 h. The cells were stained as in (A).

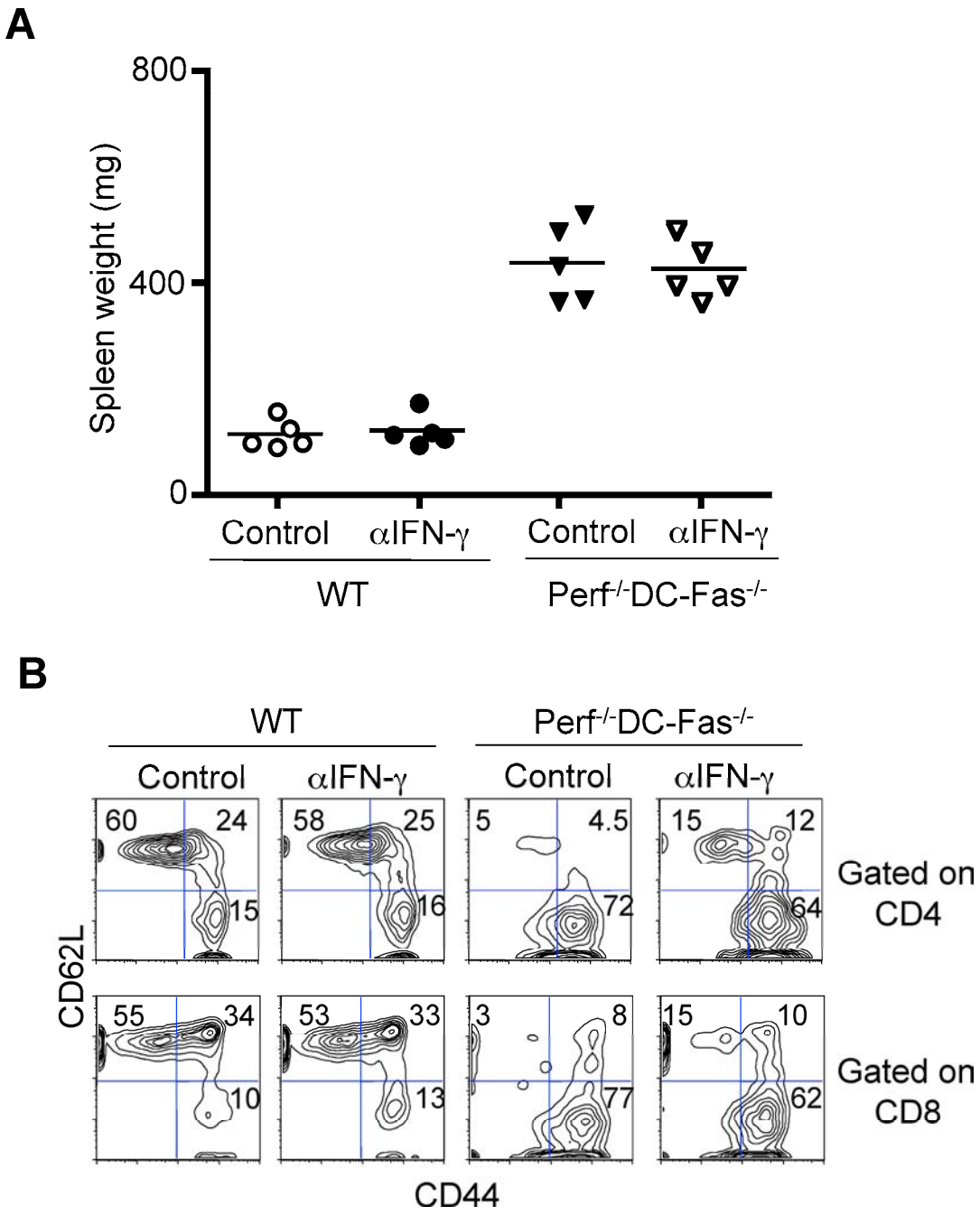

Figure S3. Analyses of Perf$^{-/-}$DC-Fas$^{-/-}$ mice with anti–IFN-γ treatment
(A) Spleen weight of 7-week-old wild type or Perf$^{-/-}$DC-Fas$^{-/-}$mice treated as in Fig. 7.
(B) Flow cytometry for CD44 versus CD62L on splenocytes of mice in (A).

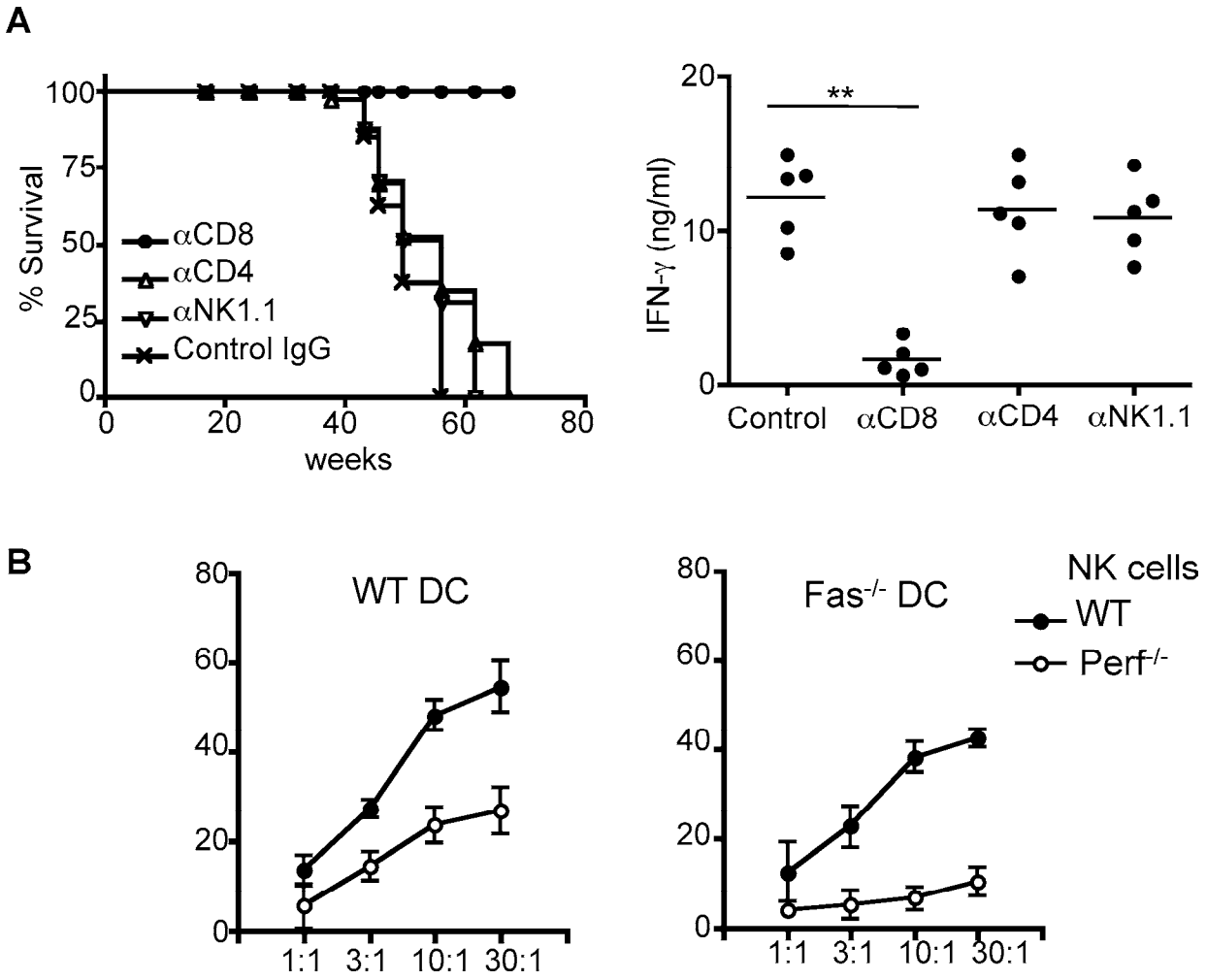

Figure S4. Effect of depletion of T cells or NK cells in Perf$^{-/-}$DC-Fas$^{-/-}$ mice and the killing of DCs by NK cells

(A) Perf$^{-/-}$DC-Fas$^{-/-}$ mice were injected with antibodies to CD8 (53-6.7), CD4 (GK1.5), NK1.1 (PK136), or control IgG from 3 weeks to 10 weeks of age or until mouse died (100 μg/mouse, I.P. every 3 days). The percentages of survival of the mice were plotted. IFN-γ in the sera of these mice at 6 weeks of age was measured. **P<0.01. (B) Wild type or perforin$^{-/-}$ NK cells were cultured in 1000 U/ml IL-2 for 6 days. Killing of wild type or Fas$^{-/-}$ DCs by NK cells was determined.

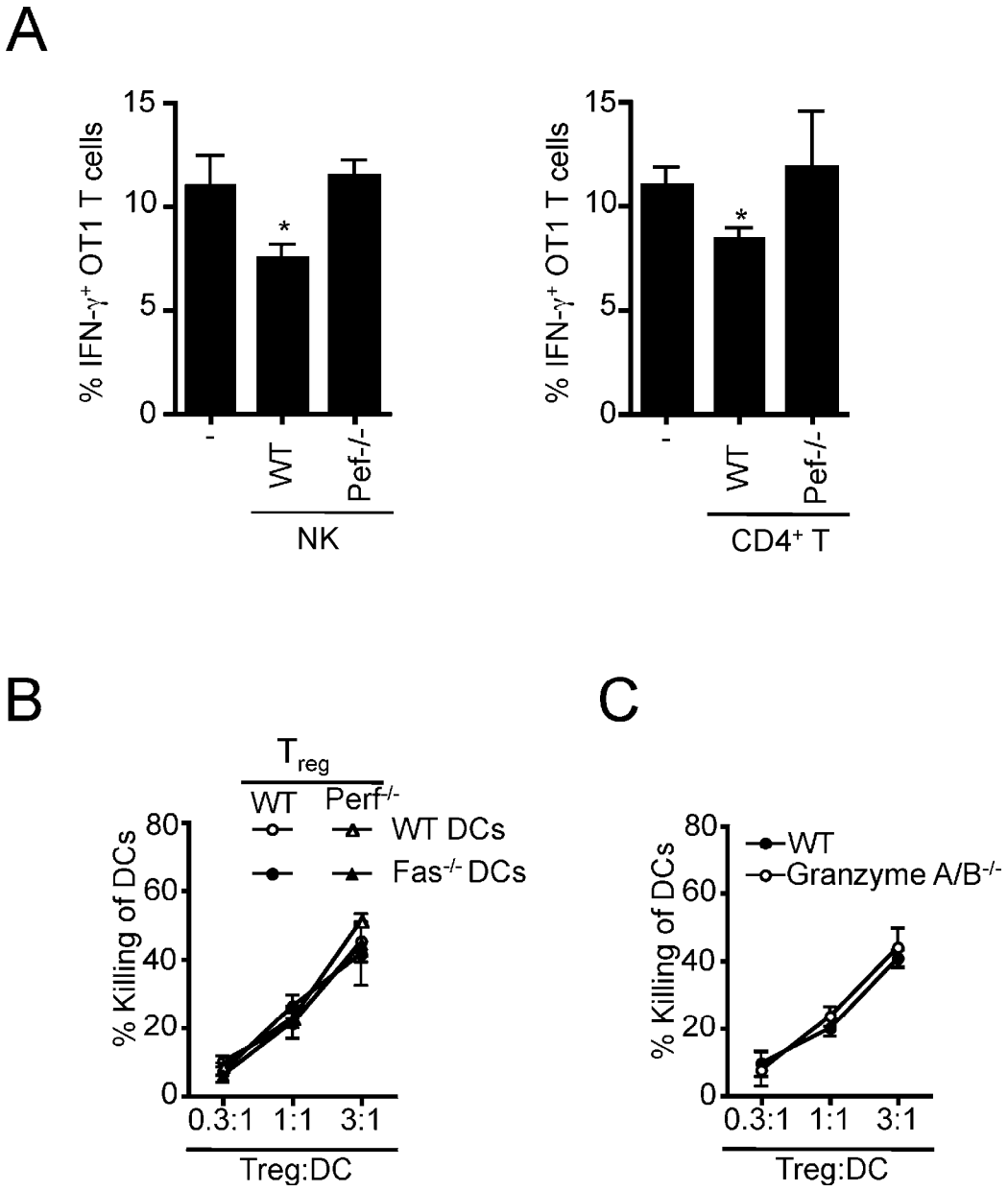

Figure S5. Effects of perforin deficiency in NK cells and T-cell subsets
(A) Wild type or perforin$^{-/-}$ NK cells were cultured with 1,000 U/ml IL-2 for 5 days. Perforin$^{-/-}$ OT1 T cells and antigen-pulsed Fas$^{-/-}$ DCs were transferred into recipient mice, together with wild type or perforin$^{-/-}$ NK cells (left panel). Fas$^{-/-}$ DCs were also pulsed with $OVA_{SIINFEKL}$ and $OVA_{323–339}$ peptides. Perforin$^{-/-}$ OT1 T cells were transferred into recipient with antigen-pulsed Fas$^{-/-}$ DCs, together with wild type or perforin$^{-/-}$ OT2 T cells (right panel). IFN-γ staining in transferred OT1 T cells were analyzed three days later. *P<0.05. (B) Killing of wild type or Fas$^{-/-}$ DCs by wild type or perforin$^{-/-}$ T_{reg} cells. (C) Killing of DCs by wild type or granzyme A/B$^{-/-}$ T_{reg} cells.