

Supplemental Figure 1. Total CD8 T cell population in the lung remains unchanged following VLP vaccination. BALB/c mice were treated i.n. with 2.5 µg VLPs in 50 µl PBS or 50 µl PBS as a control. On day 14 post vaccination, lungs were harvested and the total numbers of CD8α⁺/CD3ε⁺ T cells were determined by flow cytometry. The data are the mean values ± SEM from two independent experiments (n=9 mice/group). Data were analyzed using Student's t test.

Supplemental Figure 2. VLP vaccination does not alter the bulk or NP₁₄₇⁺ CD8 T cell population in the lungs following lethal IAV challenge. BALB/c mice were treated i.n. with 2.5 µg VLPs in 50 µl PBS or 50 µl PBS as a control. Thirty days later mice were infected with 5 LD₅₀ A/PR/8/34 and lungs were harvested on the indicated time points prior to (Day 0), and post, infection. Tetramer specific CD8α⁺/CD3ε⁺ cells were identified by flow cytometry. Representative flow plots from the lungs on day 8 post IAV challenge are shown in the left column. The data are the mean values ± SEM from three independent experiments (n=3-6 groups of 2-4 mice pooled). Data were analyzed using a two-way ANOVA followed by Bonferroni post test.

Supplemental Figure 3. Kinetics of the total CD8 T cell population following anti-CD8α depletion. BALB/c mice were infected with .1 LD₅₀ A/PR/8/34 and treated 30 days later with anti-CD8a depleting antibody i.p. (250 µg) and i.n. (100 µg). Prior to, and at multiple times following, depletion (48 hs, 4 wks, 6 wks, 11 wks, and 14 wks) mice were sacrificed and lungs, dLN, spleen and blood were collected. Representative flow plots show CD8α⁺/CD3ε⁺ cells identified by flow cytometry. (n=2-3 mice/group)

Supplemental Figure 4. Kinetics of HA₅₃₃-specific CD8 T cell populations in the draining lymph nodes and lungs following anti-CD8α depletion. BALB/c mice were infected with .1 LD₅₀ A/PR/8/34 and treated 30 days later with anti-CD8a depleting

antibody i.p. (250 μ g) and i.n. (100 μ g). Prior to, and at multiple times following, depletion (48 hs, 4 wks, 6 wks, 11 wks, and 14 wks) mice were sacrificed and dLN and lungs were collected. Tetramer-specific CD8 α^+ /CD3 ϵ^+ cells were identified by flow cytometry (n=2-3 mice/group). The dashed line indicates the mean number of HA₅₃₃-specific CD8 T cells present in a naïve dLN or lung (numbers from PBS control dLN and lungs on day 14 post treatment).

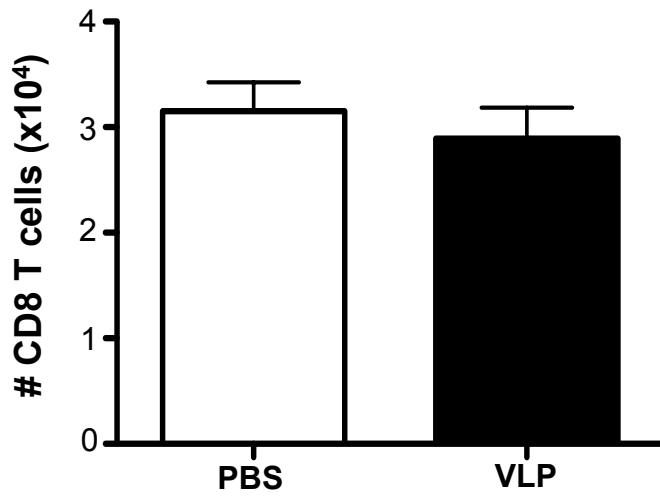


Fig S1

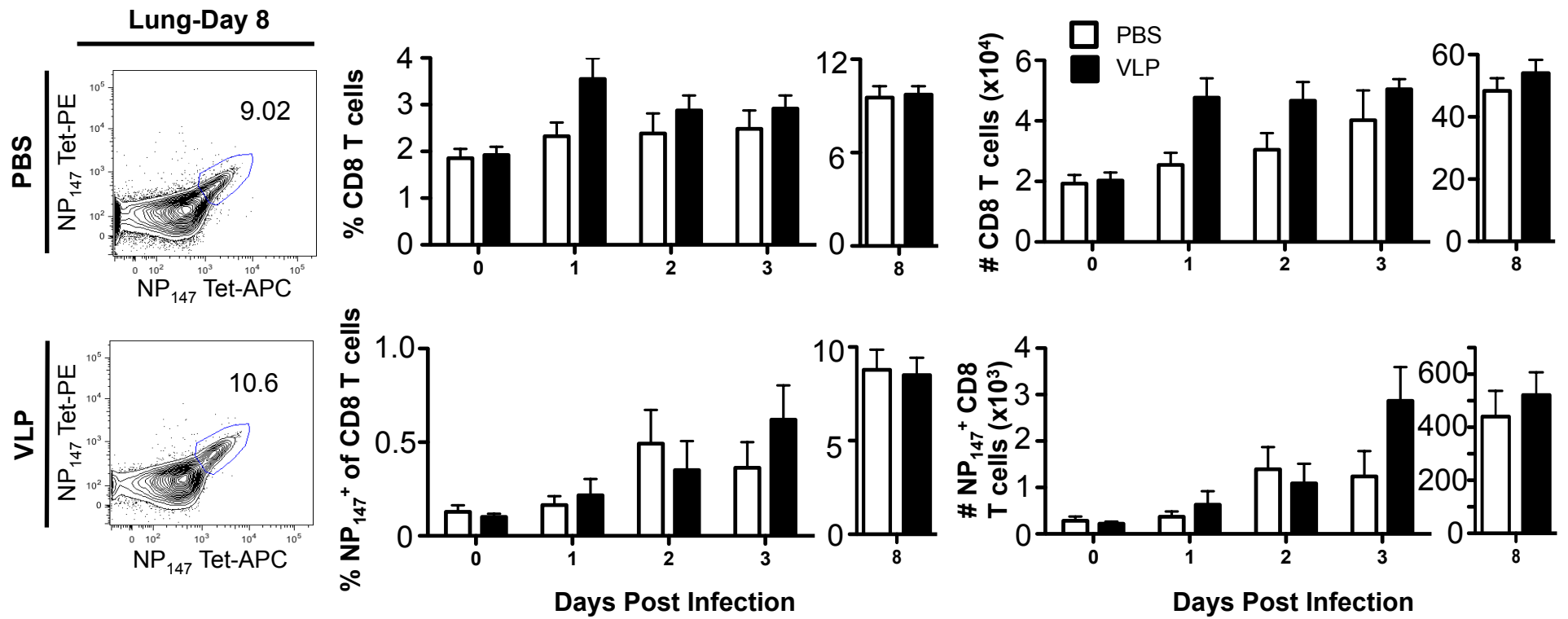


Fig S2

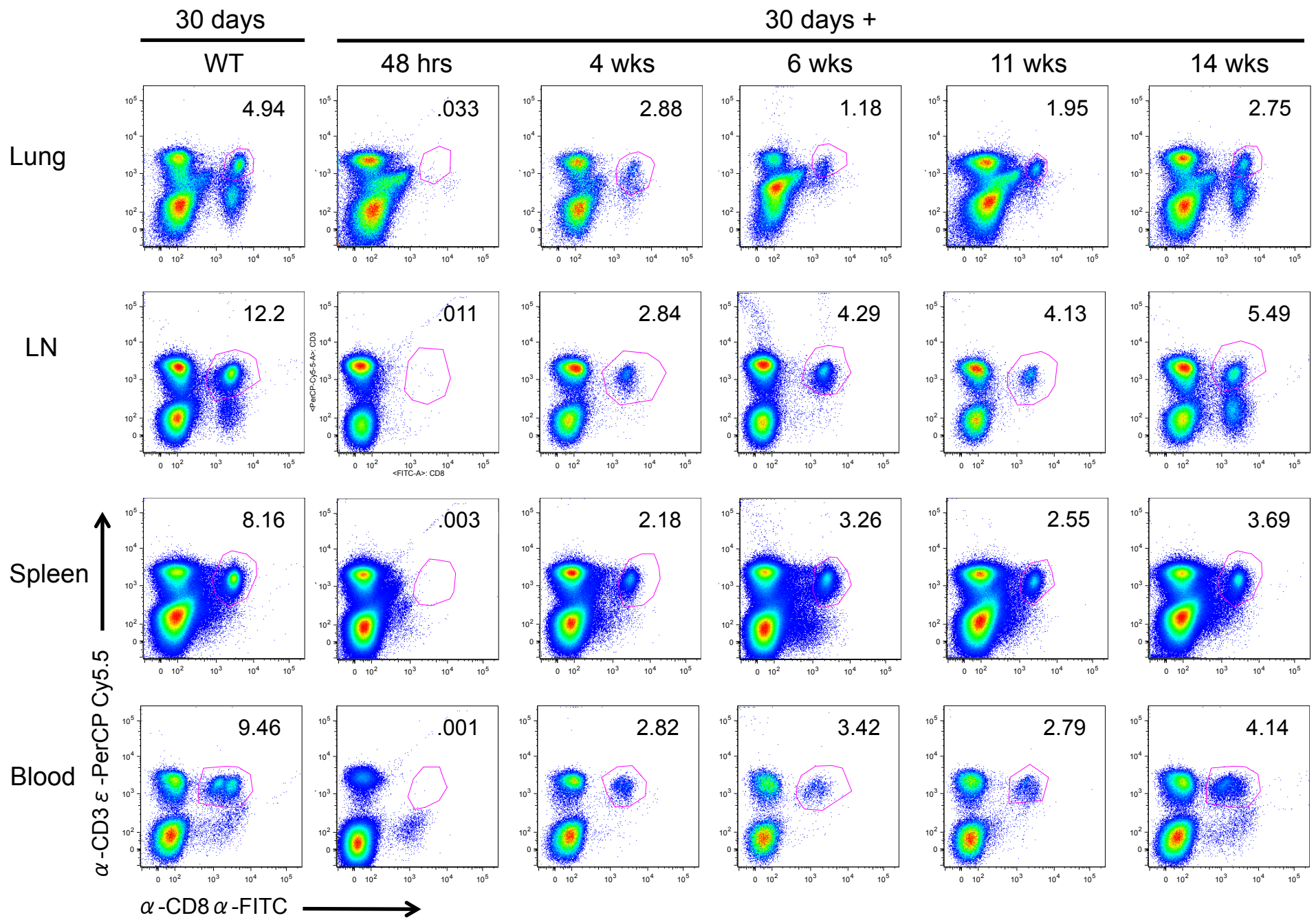


Fig S3

Fig S4

