SUPPLAMENTARY FIGURES

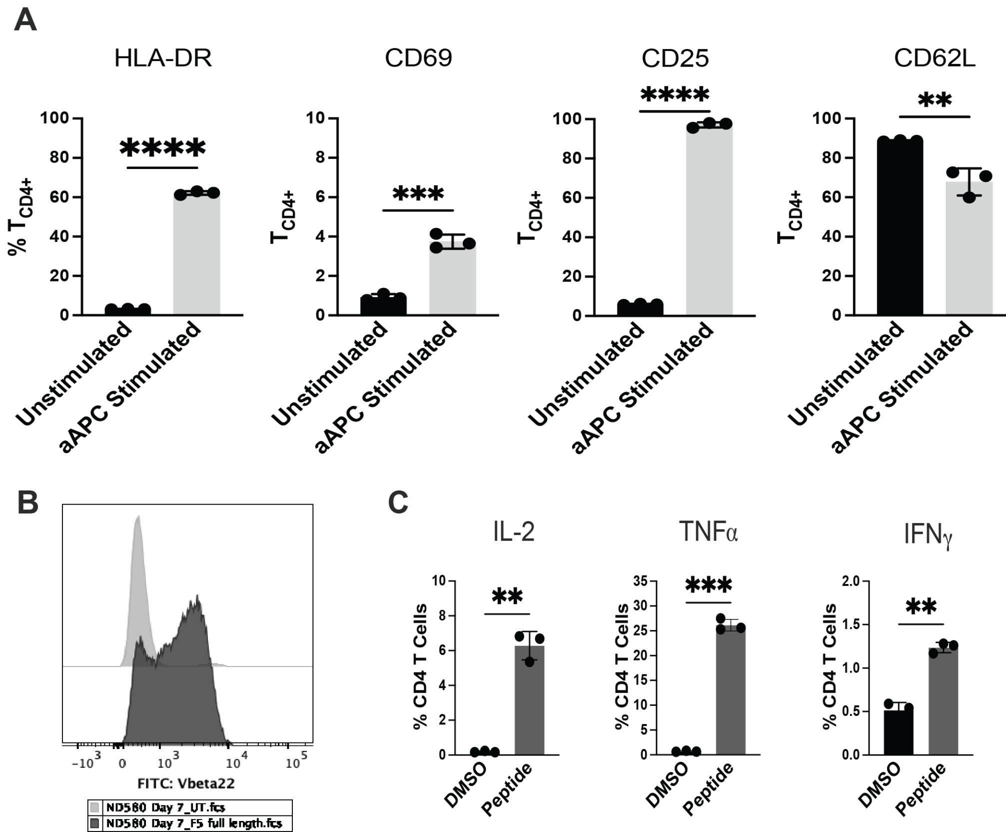

Figure S1. T_{CD4+} activated with aAPC express typical T_{CD4+} activation markers and are transducible with HIV-1-specific TCR. (**A**) T_{CD4+} were isolated from the same healthy donor and stimulated with K562.OKT3.86.64 cells (artificial APCs (aAPCs)) for 7 days in the presence of 100 U IL2/mL. Cells were then analyzed for expression of activation markers by flow cytometry. (**B**-**C**) T_{CD4+} were activated and were transduced with a lentivirus encoding the F5 HIV-1-specific TCR as described in Materials and Methods. Cells were assessed for TCR expression and functionality 6 days post transduction. (**B**) Transduced TCR expression at the T_{CD4+} surface 7 days after transduction. (**B**) HIV-1-specific TCR-transduced T_{CD4+} secrete IFNγ in response to peptide-loaded DCs Each dot represents a technical replicate. Bars represent mean ± SD. One way ANOVA, *p<0.05, **p<0.01, ***p<0.005, ****p<0.001.

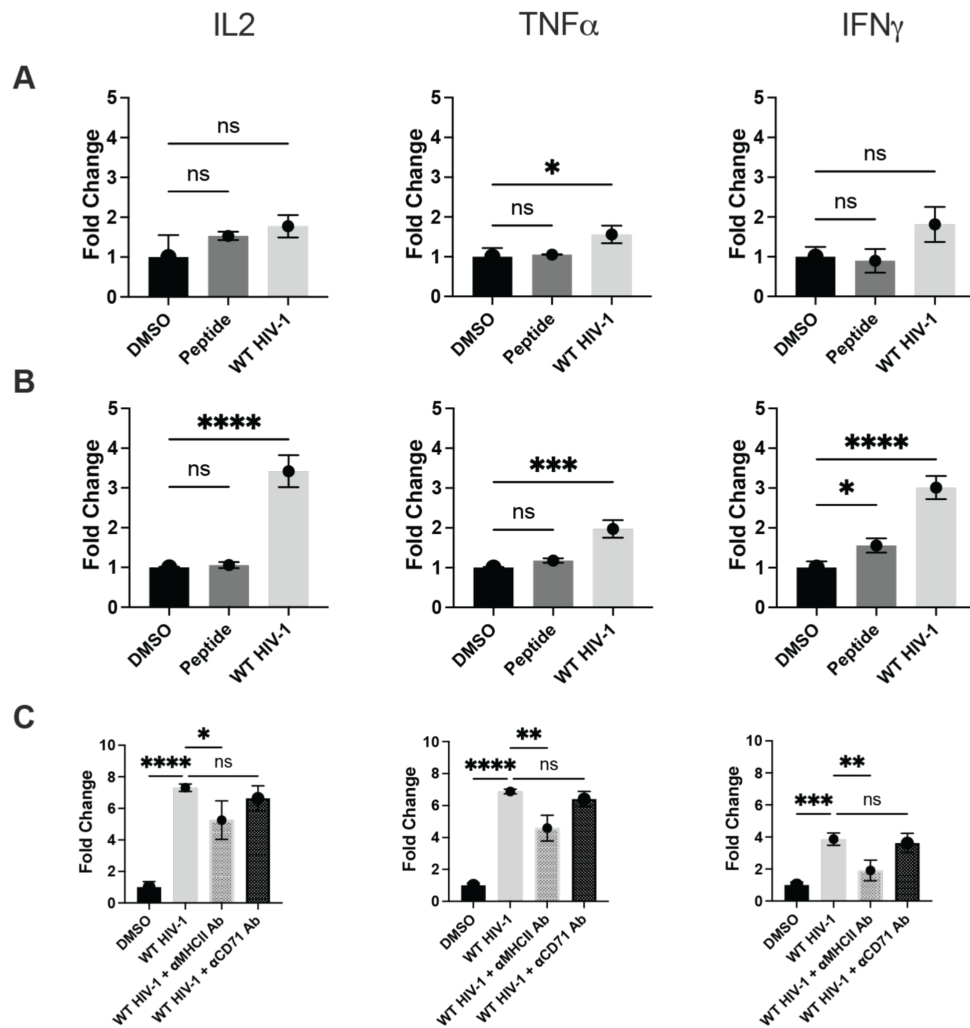

Figure S2. Presentation of synthetic peptide and WT HIV-1 by activated T_{CD4+} to responding T_{CD4+} is largely epitope-specific and MHCII-dependent. (**A**-**B**) Activated T_{CD4+} were cultured and infected as described in Materials and Methods and were then assessed for their ability to present synthetic peptide and WT HIV-1 to (**A**) ZnT8-specific T_{CD4+} and (**B**) HIV-1-specific T_{CD4+}. Activated T_{CD4+} were cocultured with either ZnT8 TCR-transduced T_{CD4+} or HIV-1-specific T_{CD4+} for 8 hours. IL-2, TNFα, and IFNγ expression was evaluated by flow cytometry. (**C**) Activated T_{CD4+} were cultured and infected as described in Materials and Methods and were then incubated with 50 μg/mL MHCII or CD71 blocking antibody for 2 hours prior to the beginning of the assay. Activated T_{CD4+} were assessed for their ability to present WT HIV-1 in the presence of MHCII or CD71 blocking antibody. IL-2, TNFα, and IFNγ expression was assessed by flow cytometry. Fold induction of each cytokine is shown, using DMSO as a baseline. Bars represent mean ± SD. One way ANOVA, *p<0.05, **p<0.01, ***p<0.005, ****p<0.001.

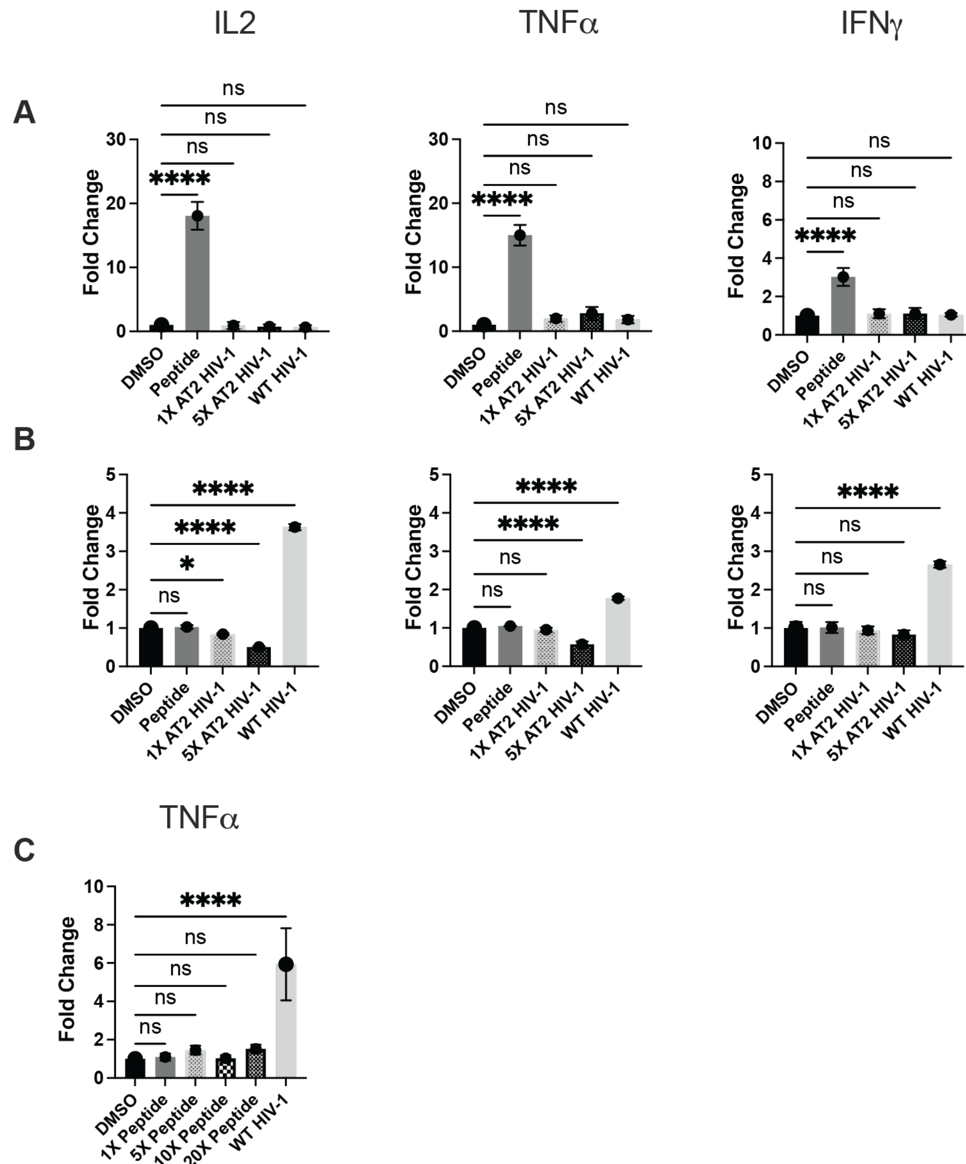

Figure S3. DCs and activated T_{CD4+} do not present a higher dose of AT2-inactivated HIV-1 and T_{CD4+} do not present high doses of synthetic peptide. DCs and activated T_{CD4+} were cultured and infected with WT HIV-1 as described in Materials and Methods. AT2-treated HIV-1 was added to DCs and activated T_{CD4+} 12 hours prior to the beginning of the assay (**A**) DCs and (**B**) T_{CD4+} were assessed for their ability to present low and high doses of AT2-treated HIV-1 after 8 hours of coculture with HIV-1-specific T_{CD4+}. (**C**) Activated T_{CD4+} do not present synthetic peptide, even at extremely high doses. IL-2, TNFα, and IFNγ expression was assessed by flow cytometry. Fold induction of each cytokine is shown, using DMSO as a baseline. Bars represent mean ± SD. One way ANOVA, *$p<0.05$, **$p<0.01$, ***$p<0.005$, ****$p<0.001$.

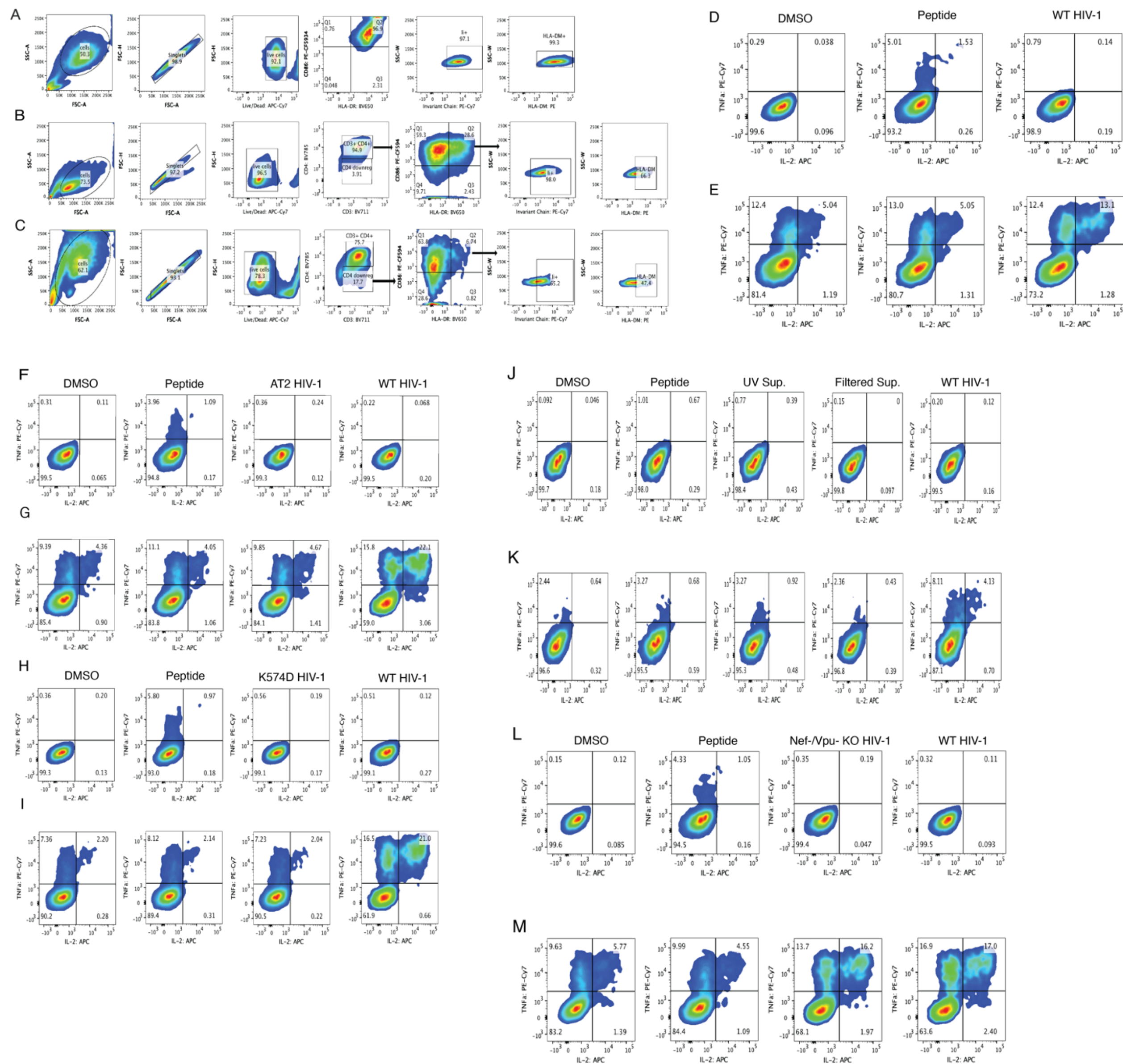

Fig. S4. Representative flow cytometry gating strategy for Figures 1a-d, 2, 3, 4, and 5. (A) gating strategy used to obtain DC data, **(B)** gating strategy used to obtain aAPC-activated T_{CD4+} data, and **(C)** gating strategy used to obtain HIV-1 infected T_{CD4+} data. (**D**-**E**) DCs and activated T_{CD4+} were cultured and infected as described in Materials and Methods. (**D**) DCs and (**E**) T_{CD4+} were assessed for their ability to present peptide and WT HIV-1 after 8 hours of coculture with HIV-1-specific T_{CD4+}. IL-2 and TNFα secretion was evaluated by flow cytometry. (**F**-**I**) DCs and activated T_{CD4+} were cultured and infected as described in Materials and Methods. AT2-treated HIV-1 and K574D fusion-deficient HIV-1 were added to DCs and activated T_{CD4+} 12 hours prior to the beginning of the assay. (**F**) DCs and (**G**) T_{CD4+} were assessed for their ability to present AT2-treated HIV-1 after 8 hours of coculture with HIV-1-specific T_{CD4+}. IL-2, TNFα, and IFNγ expression was assessed by flow cytometry. (**H**) DCs and (**I**) activated T_{CD4+} were assessed for their ability to present fusion-deficient HIV-1 in a similar manner. (**J**-**K**) DCs and activated T_{CD4+} were cultured and infected with WT HIV-1 as described in Materials and Methods. Supernatant from WT HIV-1-infected T_{CD4+} was collected and treated with UV light or filtered through a 100 kDa filter and then added to DCs and activated T_{CD4+} 12 hours prior to the beginning of the assay. (**J**) DCs and (**K**) T_{CD4+} were cocultured with HIV-1-specific T_{CD4+} for 8 hours. IL-2 and TNFα secretion was assessed by flow cytometry. (**L**-**M**) DCs and activated T_{CD4+} were cultured and infected with Nef-/Vpu- HIV-1 as described in Materials and Methods. (**L**) DCs and (**M**) T_{CD4+} were cocultured with HIV-1-specific T_{CD4+} for 8 hours. IL-2 and TNFα secretion was assessed by flow cytometry.