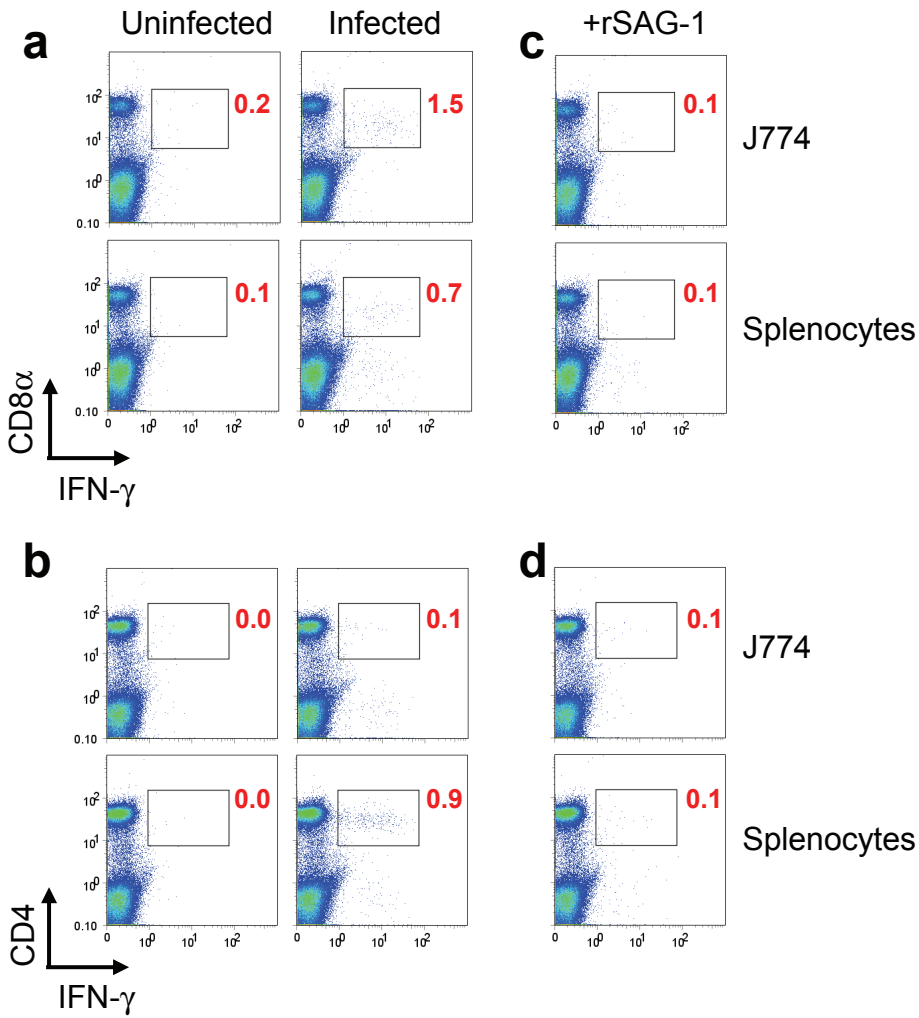
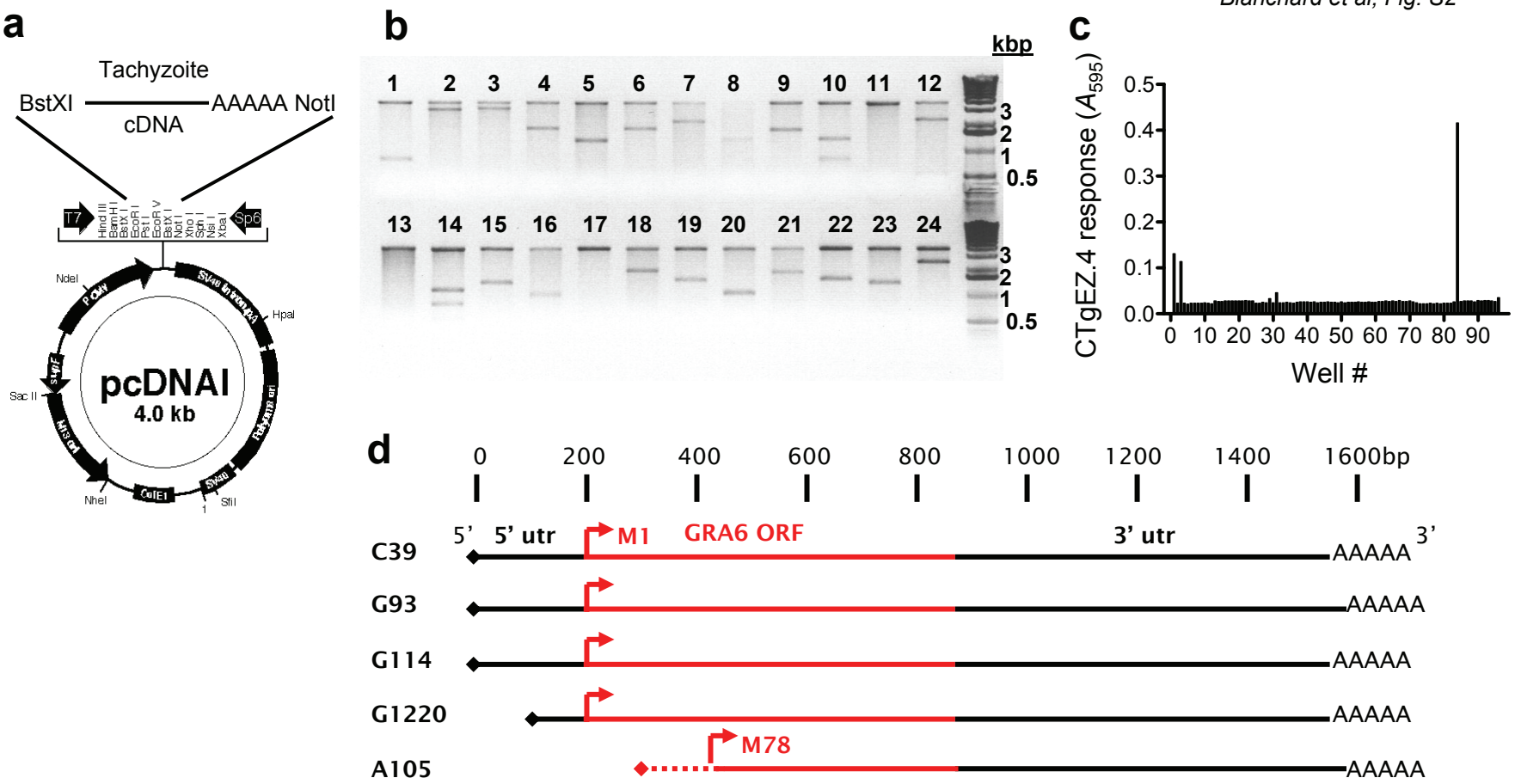
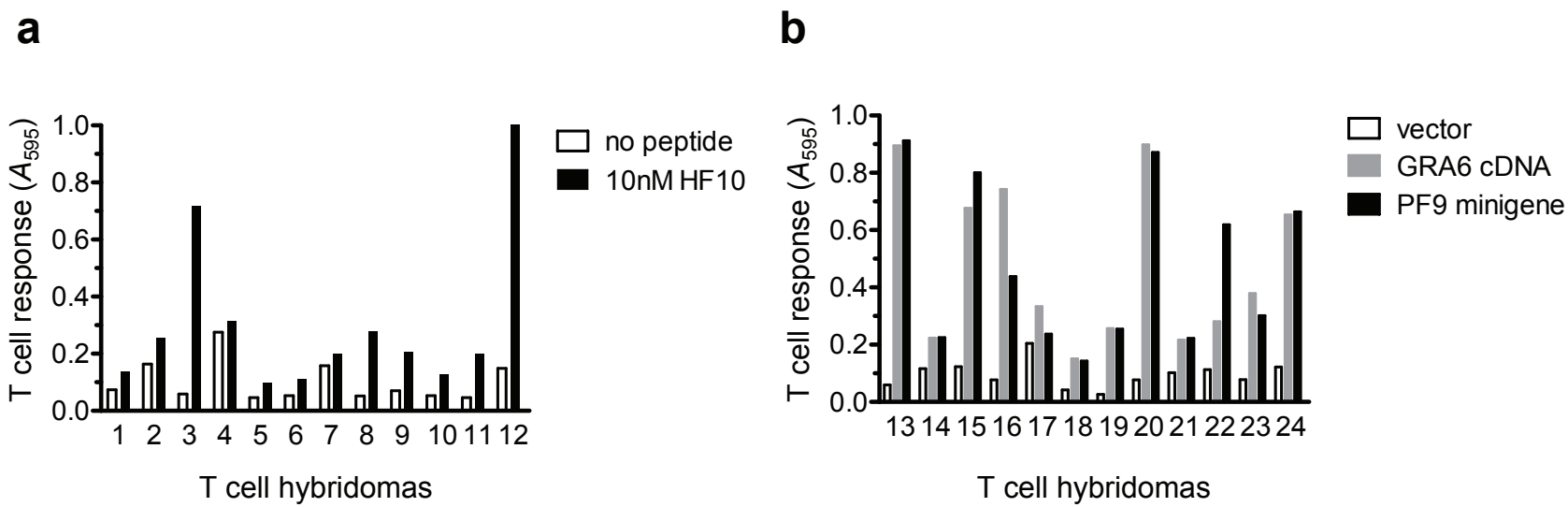


Supplementary Figure 1. Immunization with irradiated tachyzoites activates *T. gondii*-specific CD4$^+$ and CD8$^+$ T cells which do not recognize recombinant SAG-1.

BALB/c (H-2^d) mice were immunized twice with 10^6 γ-irradiated Pru tachyzoites i.p. 21 days apart. One week after the second injection, IFN-γ produced by (**a**) CD8$^+$ and (**b**) CD4$^+$ cells upon stimulation with uninfected or infected APC (J774 or splenocytes) was analyzed by flow cytometry. (**c, d**) J774 and splenocytes were pulsed with 10 μg/ml recombinant SAG-1 (rSAG-1) protein and used to stimulate *T. gondii*-immunized spleen cells as above. IFN-γ was measured by flow cytometry.

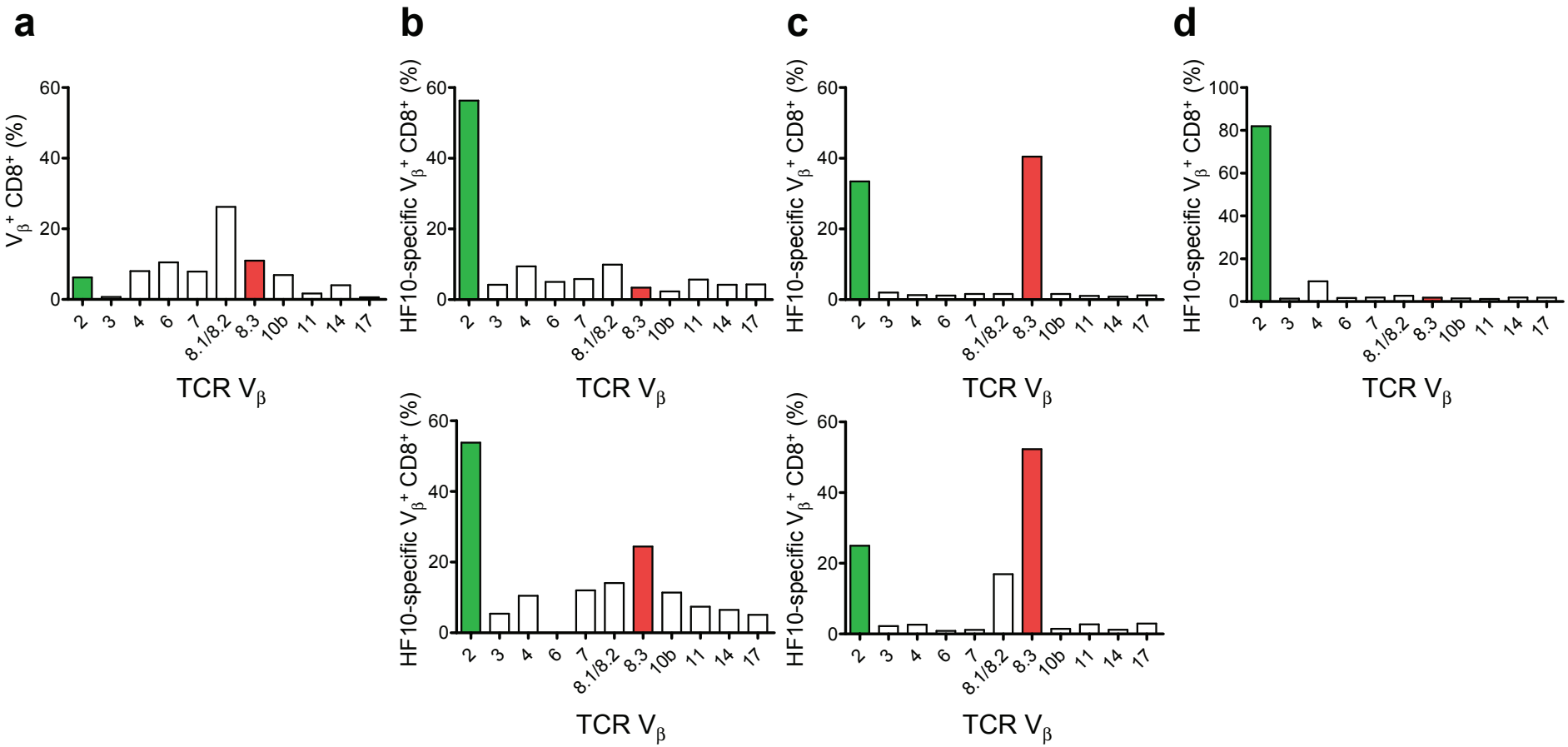


Supplementary Figure 2. Identification of GRA6 by screening a *T. gondii* tachyzoite cDNA library.
(a) A cDNA library was constructed using polyA+ mRNA isolated from Pru tachyzoites. The large cDNA fragments were cloned unidirectionally into the BstXI and NotI sites of the pcDNA I vector. **(b)** BstXI and NotI restriction digests of randomly picked plasmids showed cDNA inserts ranging from ~1 to ~3 kbp. (**c)** Approximately 16 colony forming units of the cDNA library were transfected into L^d-L cells in each well of a 96-well plate and tested for their ability to stimulate the CTgEZ.4 T cell hybrid. LacZ responses in a typical 96-well plate are shown. Each cDNA pool with antigenic activity was further subdivided, rescreened and sequenced. **(d)** Schematics of the nucleotide sequences and their coding potential for five positive cDNA clones isolated from the screens. One of these clones, A105, lacked the 5' UTR as well as the first 99 nucleotides of GRA6. Translation of the antigenic precursor was likely initiated at the internal AUG codon in a good Kozak context.



Supplementary Figure 3. All *T.gondii*-specific T cell hybridomas recognize a C-terminal peptide derived from the GRA6 antigen.

The lacZ response of a panel of T cell hybridoma clones cultured with **(a)** 10 nM HF10 synthetic peptide and J774 cells as APCs, or **(b)** H-2L^{d}-L cells transfected with full-length GRA6 cDNA or a PF9-encoding minigene cloned in a ubiquitin-based vector (see Methods). This method allows for cytosolic release of the PF9 nonamer in transfected APCs.



Supplementary Figure 4. H-2L^d-HF10-specific T cell receptors prefentially use the V_β2 and V_β8.3 variable chains. Analysis of TCRV_β usage on DimerX-HF10$^+$ CD8$^+$ spleen cells. Splenocytes from **(a)** naïve BALB/c mice, or mice orally infected for **(b)** 2 weeks, **(c)** 4 weeks or **(d)** 5 weeks were labeled with CD8, DimerX-HF10 and a panel of antibodies specific for the indicated V_β antibodies. Graphs show the proportion of HF10-specific CD8$^+$ cells (or total CD8$^+$ cells for the naïve) expressing the indicated V_β, as measured by flow cytometry. Each graph represents the data from one individual mouse. Note that expression of the V_β5.1/5.2, V_β9, V_β12 and V_β13 chains could not be assessed because the respective antibodies were of the same isotype as the DimerX immunoglobulin region and thus interfered with the secondary antibody.

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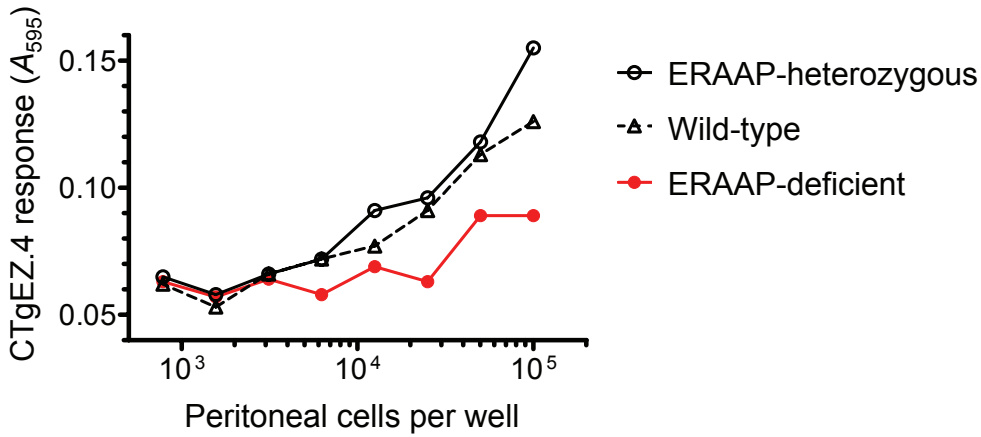
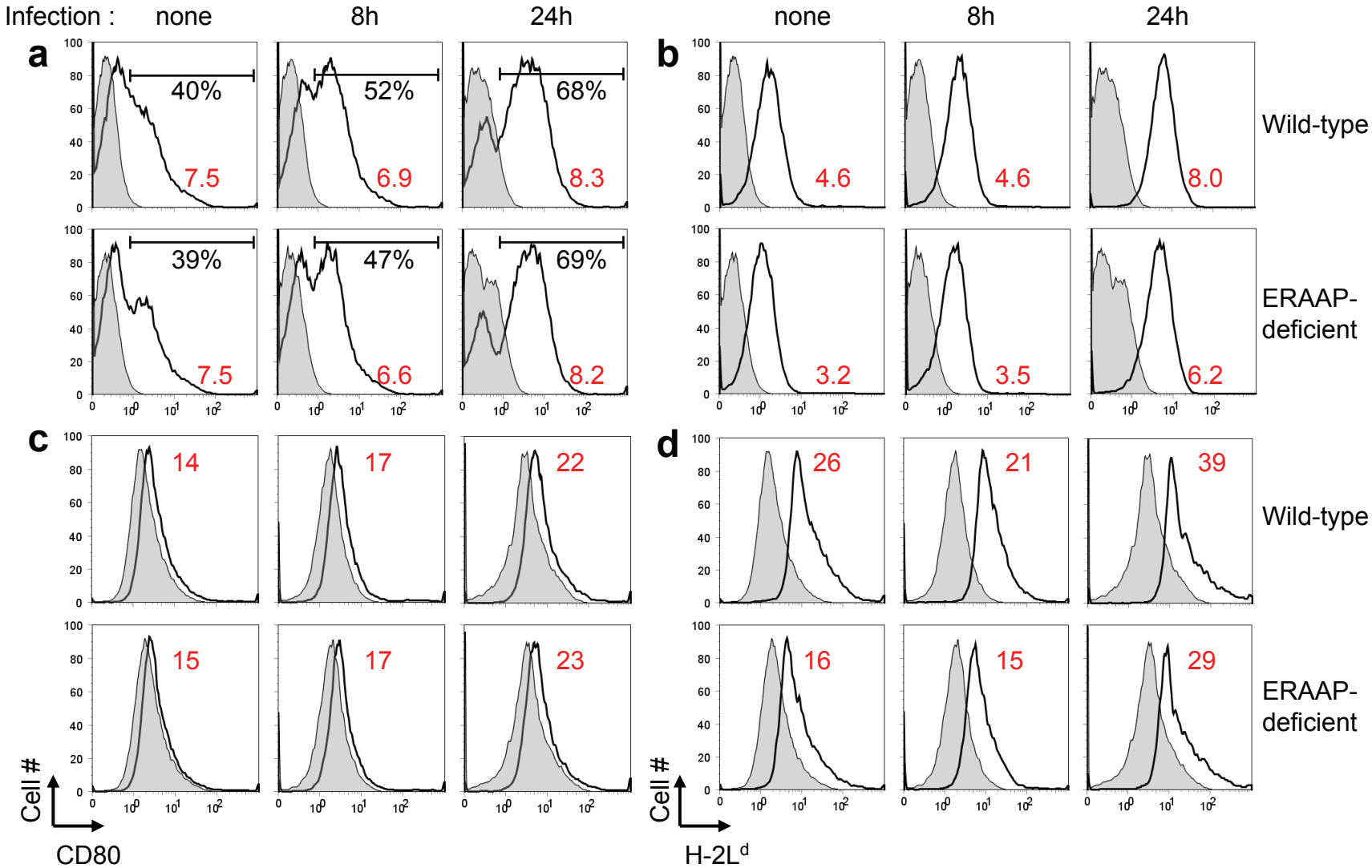


Supplementary Figure 5. ER proteolysis is required to generate H-2L^d-HF10 complexes in peritoneal macrophages.

Peritoneal cells were harvested from ERAAP-deficient, ERAAP-heterozygous and wild-type animals and infected for 24h at a 6:1 MOI with γ-irradiated Pru tachyzoites. Presentation of H-2L^d-HF10 complexes were assessed by measuring β-galactosidase induction in the CTgEZ.4 hybridoma.



Supplementary Figure 6. ERAAP deficiency affects the surface expression of H-2L^d molecules but not CD80. Surface expression of H-2L^d and CD80 on (**a,b**) BMDCs and (**c,d**) BMDMs was analyzed by flow cytometry after infection with γ-irradiated Pru-GFP tachyzoites for 8h or 24h at a 8:1 MOI. Tinted histograms, isotype control. Numbers in red represent the mean fluorescence intensity of the H-2L^d or CD80 stainings. Numbers in black (**a**) represent the percentage of CD80^{high} cells. Data are representative of at least 2 experiments.