Additional File 1: Target cells used for the in vivo CTL assay express NKG2D ligands. Spleens were harvested from euthanized mice and prepared for use in the in vivo CTL assay. Briefly, spleens were subjected to ammonium chloride-based osmotic shock for red blood cell lysis, peptide loading, CFSE labelling, and adoptive transfer into a host mouse. Twelve hours after transfer, spleens were recovered from host mice. Expression of NKG2D ligands was confirmed by flow cytometry using NKG2D-Fc chimeras in CFSE + cells. Unstained and secondary antibody alone are shown.