

Supplementary Fig.1: Screening of B-cell supernatants against UL16 and UL141 revealed five mAbs that bound UL16 and nine mAbs that bound UL141. (A) HFFF-hCARs were transduced with RAds expressing native or ER retention signal-truncated UL16 or UL141. An identical vector lacking a transgene was used as a control. Transduced cells were stained with a mAb specific for the HA-tag engineered into the N-terminus of each protein. (B, C) HFFF-hCARs were transduced with RAds expressing ER retention signal-truncated UL16 (B) or UL141 (C). An identical vector lacking a transgene was used as a control. Transduced cells were stained with an anti-human IgG-AF647 secondary mAb. Positive clone supernatants and a representative negative clone supernatant are shown for each protein (total n = 60). ctrl, control. Data are shown as flow histograms and are representative of at least two experiments (A–C).





Supplementary Figure 2: The sequences of the heavy (A) or light (B) chain of the B-cell receptor for each antibody were aligned, then a neighbour joining tree constructed using CLC Main.

А



Supplementary Figure 3: Viral Fc receptors do not have a major impact on ADCC activity at 48 hours post infection. HF-TERTs were infected with HCMV strains Merlin or Merlin Δ Fc (A-C) or Merlin Δ UL141 (D, E). HF-CAR were infected with RAd expressing UL141, or control RAd lacking a transgene (C). (A) Infected cells were stained with fluorochrome-labeled Cytotect (100 µg/ml). Data are shown as flow histograms. (B) Percent degranulation of CD56+ CD57+ NK cells among PBMCs in the presence of infected cells and either Cytotect or seronegative IgGs (each at 40 µg/ml). Infected cells alone were included as a control. Data are shown as mean ± SD of triplicate samples. (C) Plasma membrane proteins (PMP) were oxidised and aminoxy-biotinylated, before being immunoprecipitated with streptavidin beads, and lysed in SDS-PAGE buffer. Whole-cell lysates (WCL) prior to IP were lysed directly in SDS-PAGE buffer. Proteins were separated by SDS-PAGE, western blotted, and stained using anti-UL141 monoclonal antibodies. (D, E) Infected cells were stained with the native or Fc-modified forms of the UL141-specific mAbs B2 (C) or G11 (D), each at a concentration of 10 µg/ml, and analyzed for binding to viral FcRs. Data are shown as flow histograms. All experiments were performed 48 h after infection (A–D). *p < 0.05 (two-way ANOVA).