cancers

Supplementary Materials: HydrAd: A Helper-Dependent Adenovirus Targeting Multiple Immune Pathways for Cancer Immunotherapy

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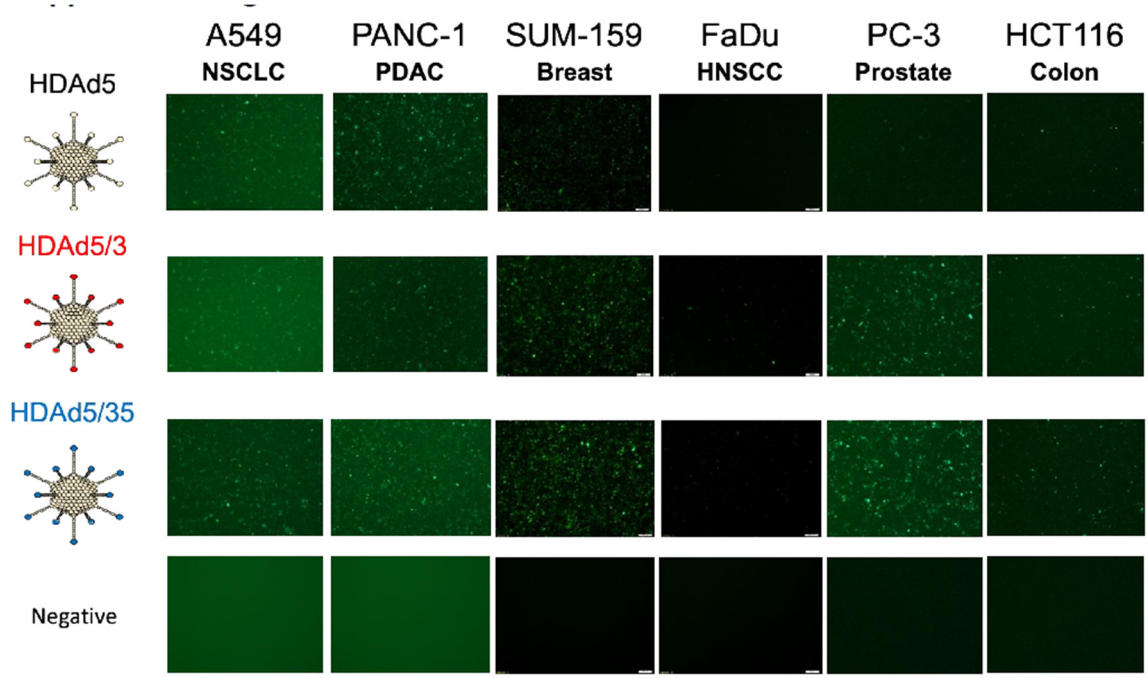

Figure S1. HDeGFP Transduction Microscopy. Representative fluorescent microscopy images of HD*eGFP* 100vp/cell infected solid tumor cell line panel.

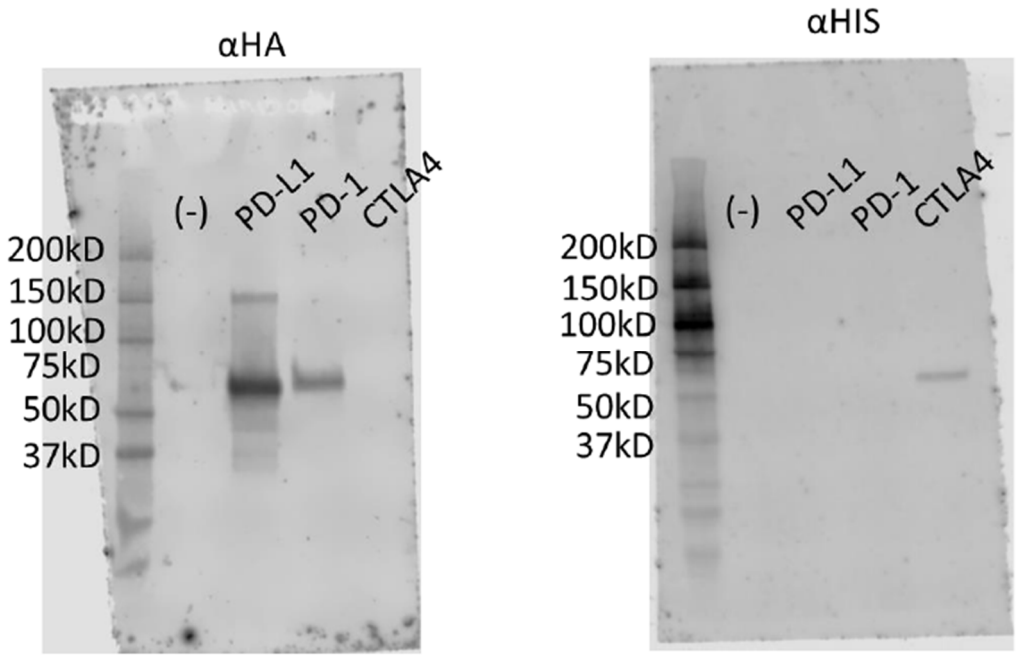

Figure S2. Complete, uncropped western blots for HD*PDL1*, HD*PD1*, HD*CTLA4* minibody expression.

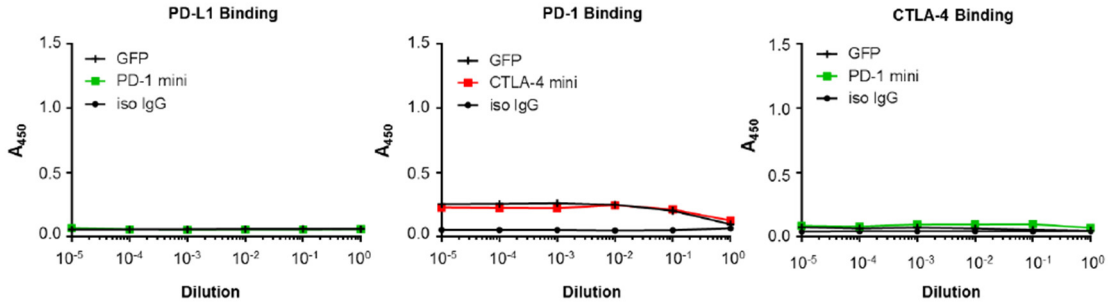

Figure S3. Minibody binding controls. A549 cells were transfected with GFP expression plasmid alongside minibody expression plasmids to serve as a control for minibody binding. Media containing GFP or an irrelevant minibody were assessed for binding to recombinant human protein (PD1minibody:PDL1, CTLA4minibody:PD1, PD1minibody:CTLA4). Isotype IgG were used as controls (10ug/mL; highest concentration).

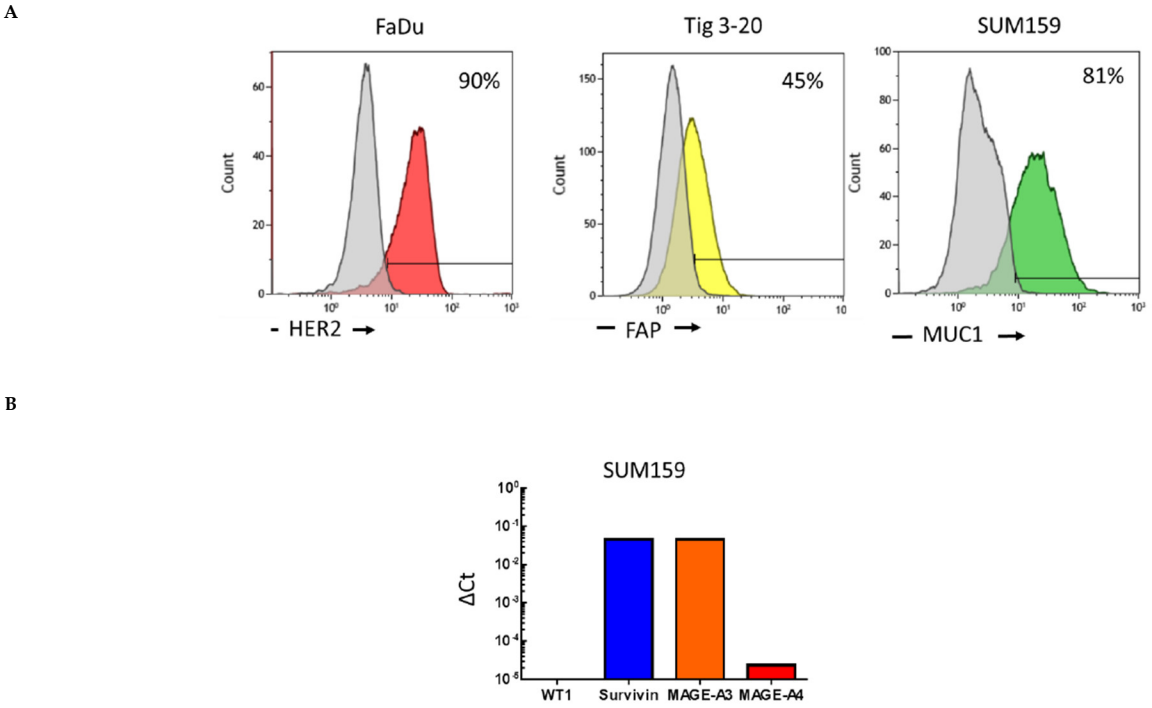

Figure S4. Tumor target antigen expression. (**A**) HER2, FAP, and MUC1 cell surface expression on FaDu, Tig-3-20, and SUM-159 cells respectively, were analyzed by flow cytometry. (**B**) RNA was extracted from SUM-159 cells and expression of the Cancer/Testis antigens WT1, Survivin, MAGE-A3, and MAGE-A4 was analyzed by qRT-PCR. Data were normalized with human B-actin.

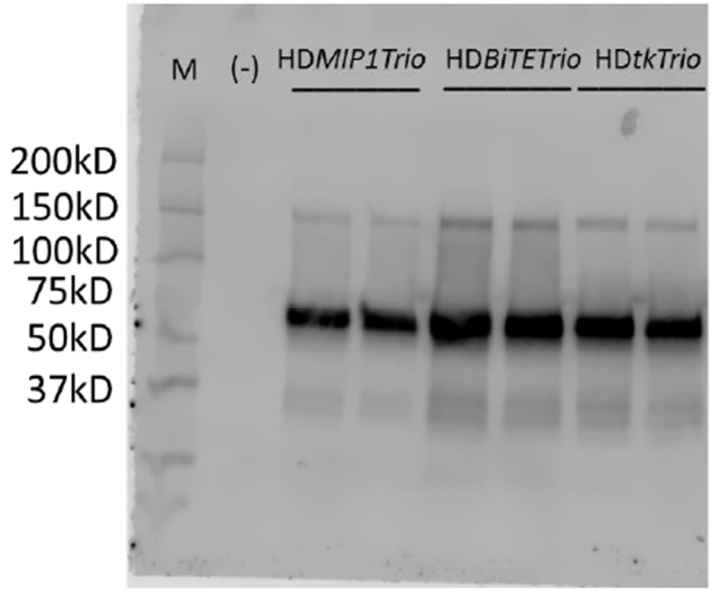

Figure S5. Complete, uncropped western blots for HD*Trios* minibody expression.

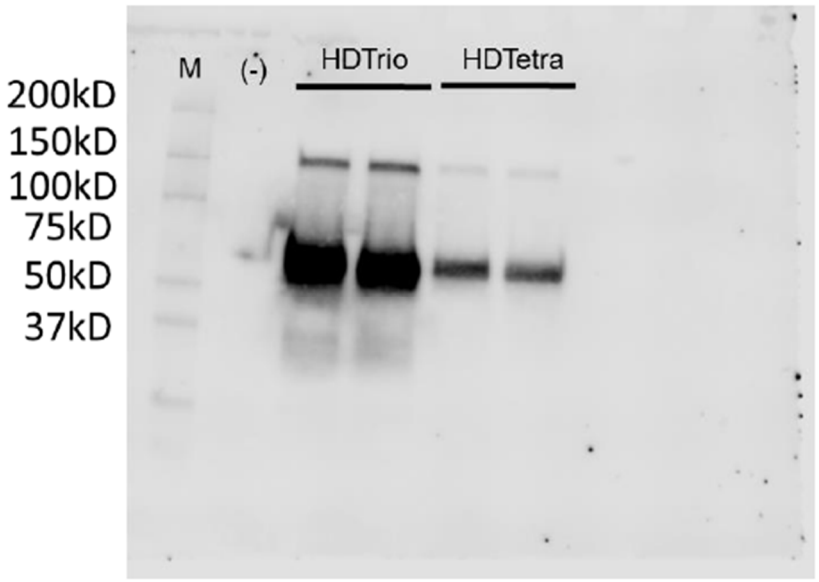

Figure S6. Complete, uncropped western blots for HD*Trio* and HD*Tetra* minibody expression.