

SUPPORTING INFORMATION

for

Poly(propylacrylic acid)-Peptide Nanoplexes as a Platform for Enhancing the Immunogenicity of Neoantigen Cancer Vaccines

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Supporting Data:

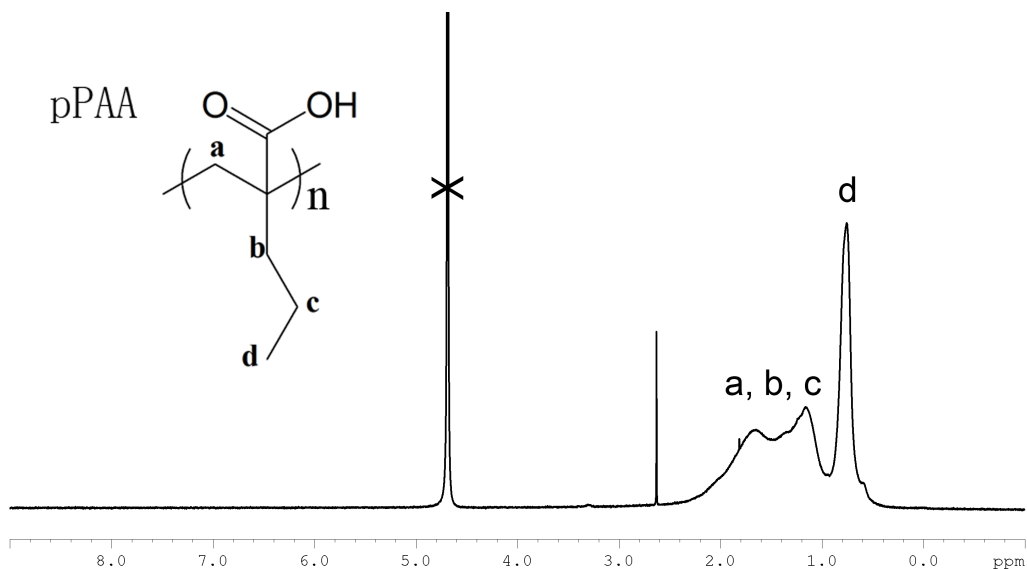


Figure S1: ¹H NMR spectrum of purified poly(propylacrylic acid) (pPAA) in DMSO-d₆.

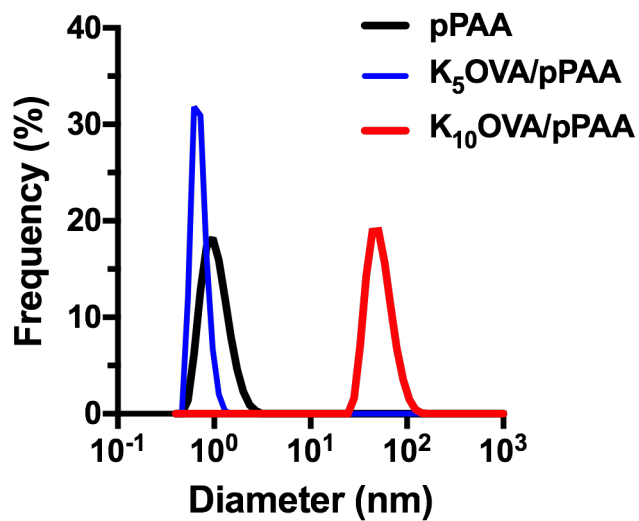


Figure S2: Representative size distribution (volume average) measured by DLS of soluble pPAA and complexes of pPAA with K₅OVA or K₁₀OVA at a 2:1 COOH:NH₂ ratio.

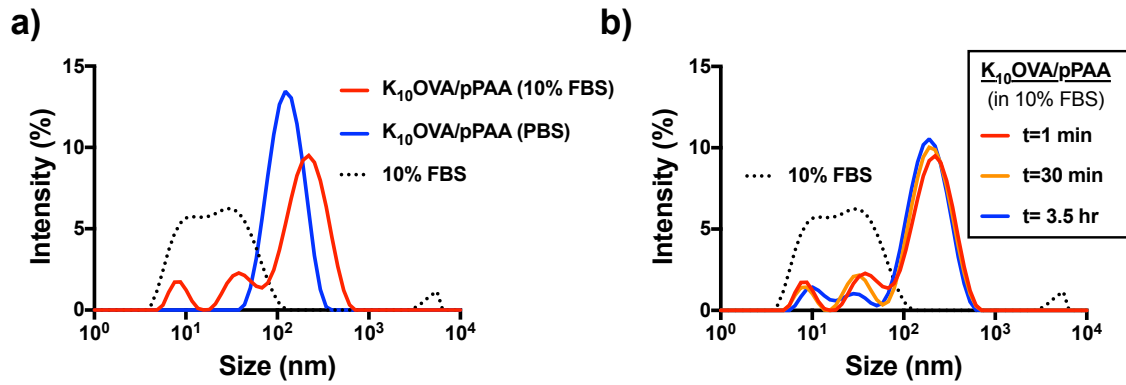


Figure S3: Dynamic light scattering (DLS) analysis of particle size distribution of nanoplexes in PBS containing 10% serum. **(a)** DLS analysis of $K_{10}OVA/pPAA$ nanoplexes in PBS or 10% serum (FBS). **(b)** DLS analysis of $K_{10}OVA/pPAA$ nanoplexes in 10% serum at 1 min, 30 min, or 3.5 hours.

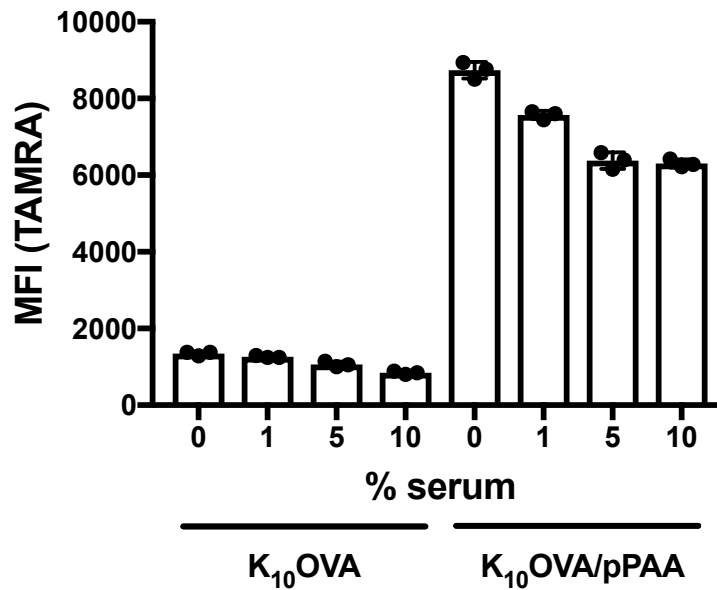


Figure S4: Effect of the percentage of serum (fetal bovine serum) in culture media on intracellular uptake of TAMRA-labeled $K_{10}OVA$ or $K_{10}OVA/pPAA$ nanoplexes by DC2.4 cells.

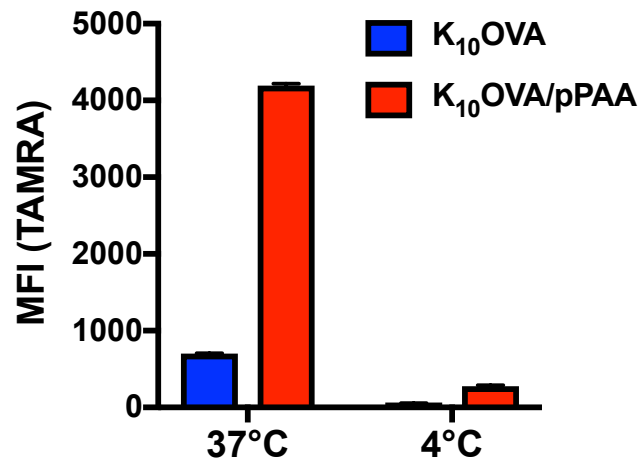


Figure S5: Median fluorescent intensity (MFI) values of DC2.4 cells incubated with TAMRA-labeled K₁₀OVA or K₁₀OVA/pPAA nanoplexes for 4h at 37°C or 4°C.

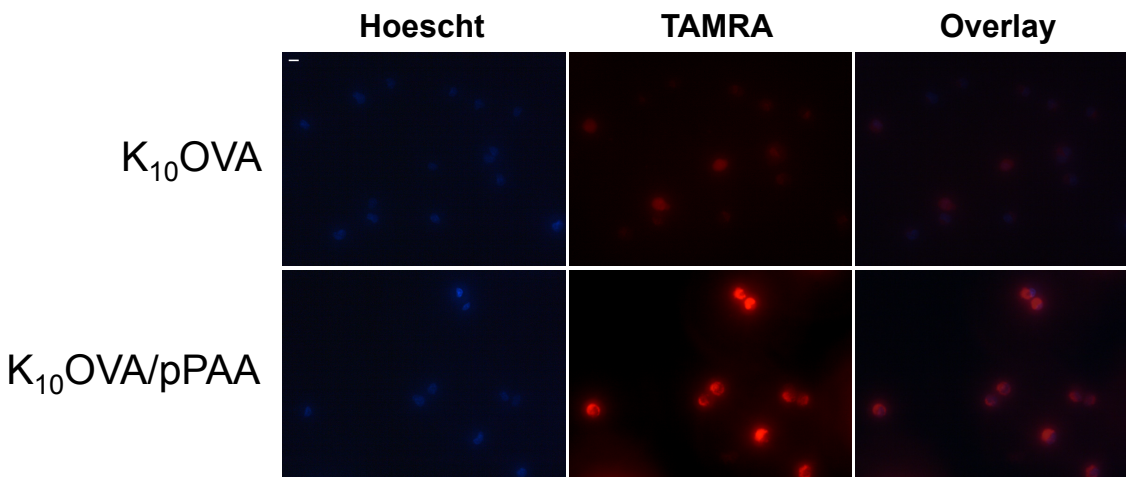


Figure S6: Representative fluorescent micrographs of DC2.4 cells incubated with TAMRA-labeled K₁₀OVA or K₁₀OVA/pPAA nanoplexes for 4h prior to washing and imaging. Scale bar (top left) = 10 μm.

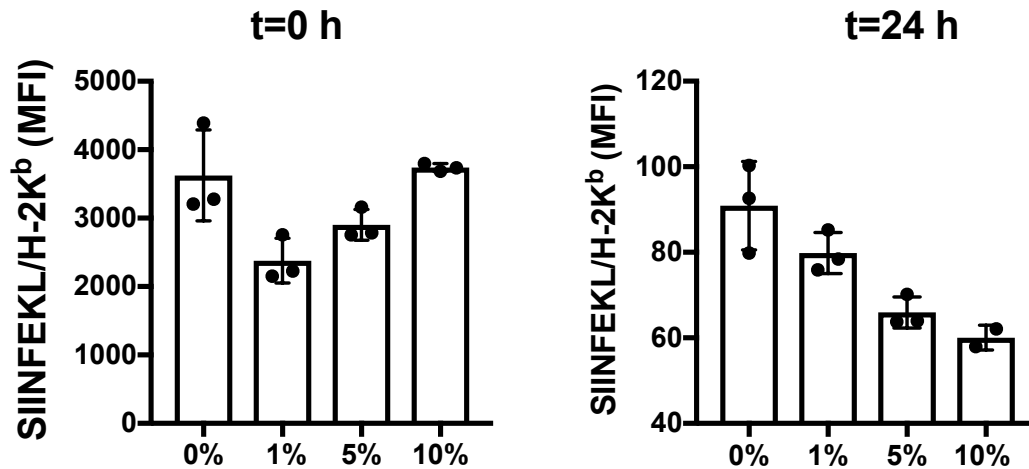


Figure S7: Effect of the percentage of serum (fetal bovine serum) in culture media on MHC-I (H-2K^b) SIINFEKL presentation after 4 h incubation with K₁₀OVA/pPAA polyplexes at 10 μM peptide, followed by washing, culture for 0 h or 24 h, and staining with an antibody (25-D1.16) specific to the SIINFEKL/H-2K^b complex. Note that measurements for t=0 and t= 24 h were collected using two different flow cytometers and therefore MFI values between time points cannot be directly compared.

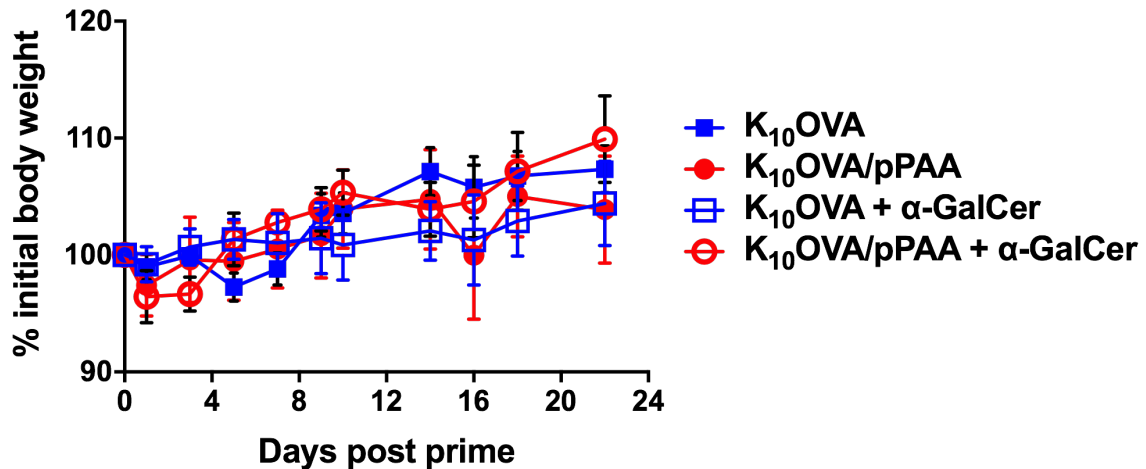


Figure S8: Change in relative body weight of mice immunized intranasally with the indicated formulation on d0 and d14.

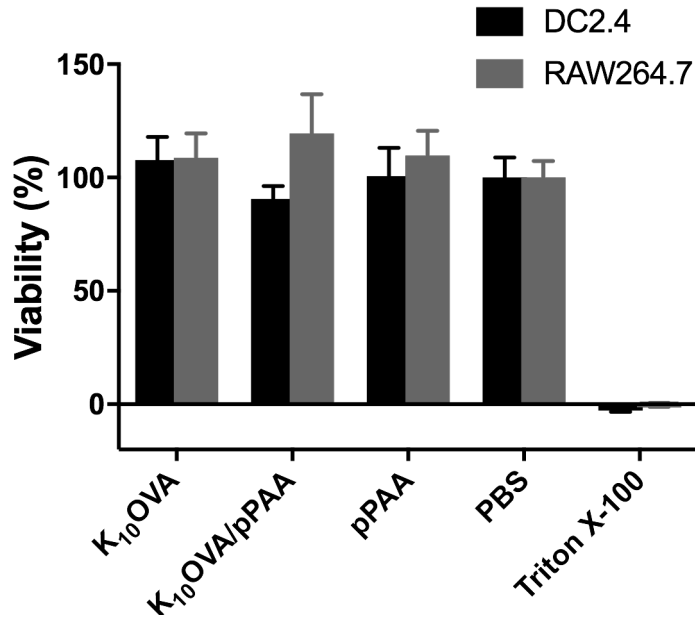


Figure S9: Viability of DC2.4 dendritic cells and RAW264.7 macrophages after 24 h incubation with indicated treatment group.

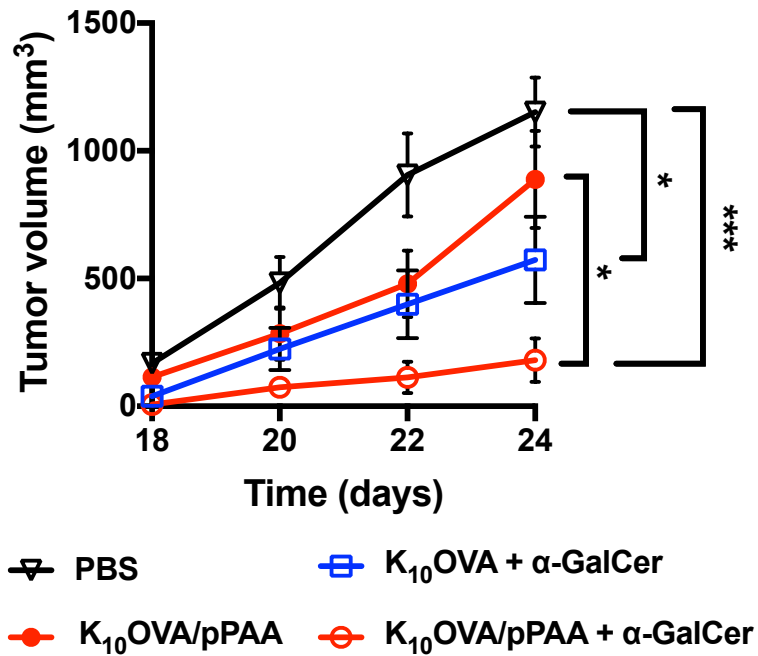


Figure S10: Mice were inoculated with B16-OVA cells subcutaneously (SC) 3 days prior to IN administration of indicated formulations. Mean (\pm SEM) tumor volume ($n=4-8$ mice/per group) is shown. * $p < 0.05$, *** $p < 0.005$ by one-way ANOVA with Tukey post-hoc test on day 24.