

Supplementary figures for

A co-formulated vaccine of irradiated cancer cells and cowpea mosaic virus improves ovarian cancer rejection

Authors:

Zhongchao Zhao^{1,2}, Oscar A. Ortega-Rivera¹, Young Hun Chung³, Andrea Simms¹, Nicole F. Steinmetz^{1,2,3,4,5,6,7}

¹Department of NanoEngineering, University of California, San Diego, 9500 Gilman Dr., La Jolla, CA, 92093 USA

²Center for Nano-ImmunoEngineering, University of California, San Diego, 9500 Gilman Dr., La Jolla, CA, 92093 USA

³Department of Bioengineering, University of California, San Diego, 9500 Gilman Dr., La Jolla, CA, 92093 USA

⁴Department of Radiology, University of California, San Diego, 9500 Gilman Dr., La Jolla, CA, 92093 USA

⁵Institute for Materials Discovery and Design, University of California, 9500 Gilman Dr., La Jolla, CA, 92093 USA

⁶Moores Cancer Center, University of California, San Diego, 9500 Gilman Dr., La Jolla, CA, 92093 USA

⁷Center for Engineering in Cancer, Institute for Engineering in Cancer, University of California, San Diego, 9500 Gilman Dr., La Jolla, CA, 92093 USA

Corresponding author: nsteinmetz@ucsd.edu

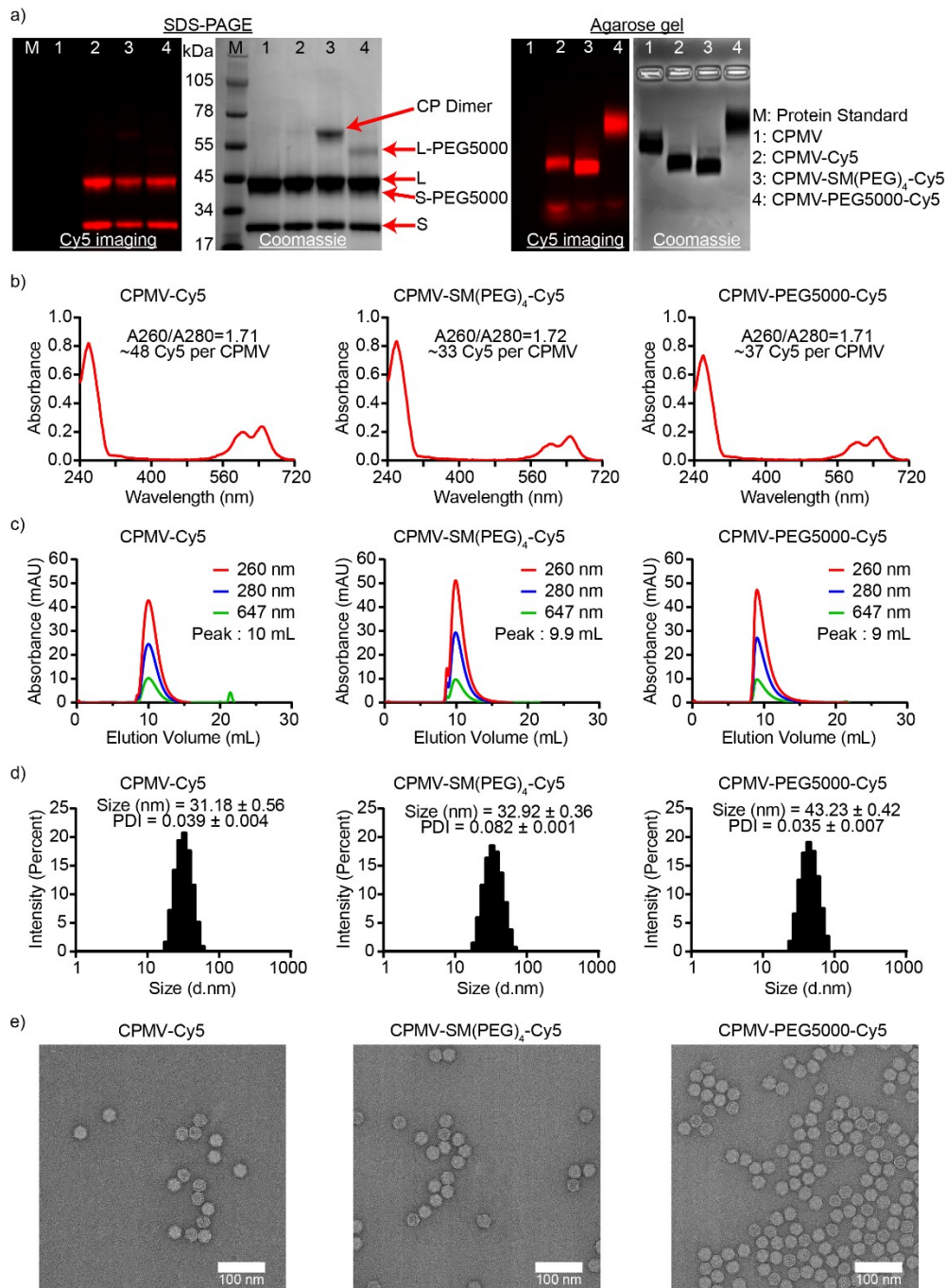
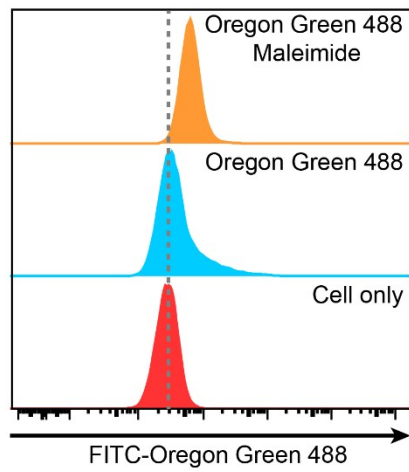


Figure S1. Characterization of CPMV-Cy5, CPMV-SM(PEG)₄-Cy5, and CPMV-PEG5000-Cy5. a) SDS-PAGE (left) and agarose gel electrophoresis (right) of CPMV nanoparticles. For both SDS-PAGE and agarose gel, conjugated Sulfo-Cy5 on CPMV was first imaged using the MultiColor Red filter with the imager, confirming successful Cy5 conjugation. Same as in **Figure 1**, SM(PEG)₄ and PEG5000 were successfully conjugated to CPMV after Coomassie stain. b) UV-vis spectra of three CPMV nanoparticles. The insets showed the absorbance ratio of 260 nm/280 nm and number of conjugated Cy5 per CPMV. c) SEC elution profiles showed co-elution of RNA (260 nm), CPMV protein (280 nm), and Cy5 (647 nm). The

inset showed the elution peak d) DLS of three CPMV nanoparticles. The insets showed the average sizes and PDI of three measurements. e) TEM images of the three CPMV nanoparticles.

a)



b)

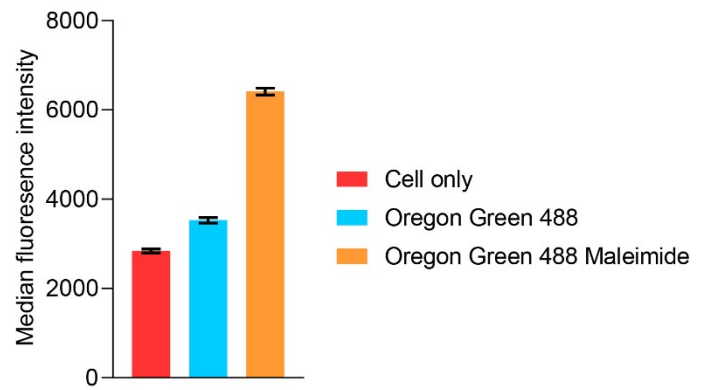


Figure S2. Flow cytometry analysis of free thiols on ID8-Defb29/Vegf-a-Luc cell surface. Free thiols were detected by incubating ID8-Defb29/Vegf-a-Luc cells with maleimide functionalized Oregon Green 488.

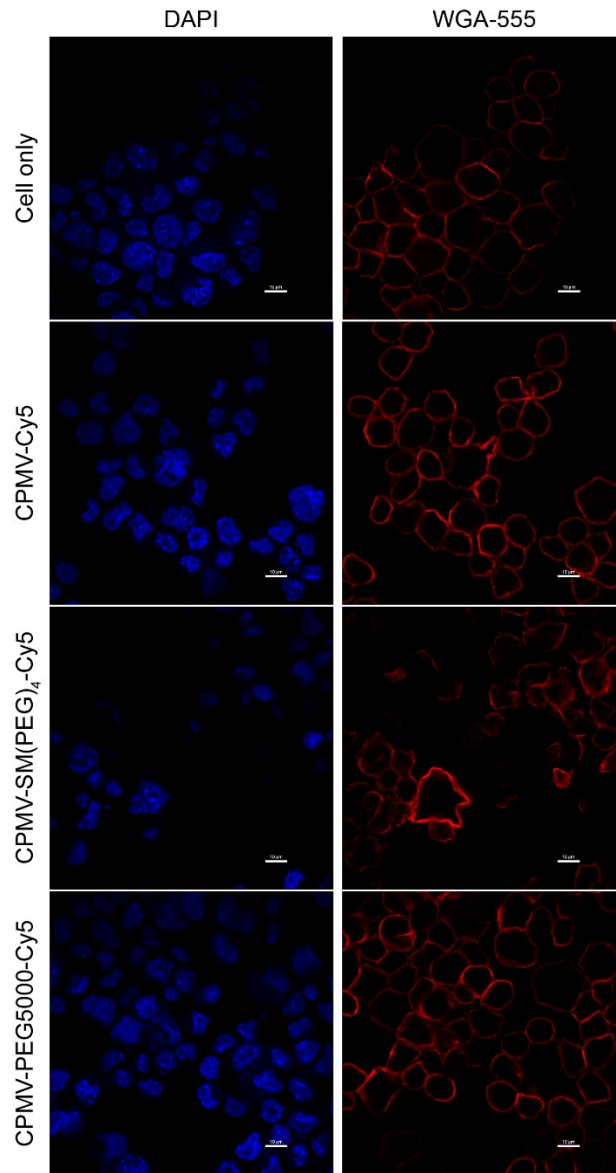


Figure S3. Confocal microscopy images of the nucleus stain and membrane stain for ID8-Defb29/Vegf-a-Luc cells in **Figure 4**.

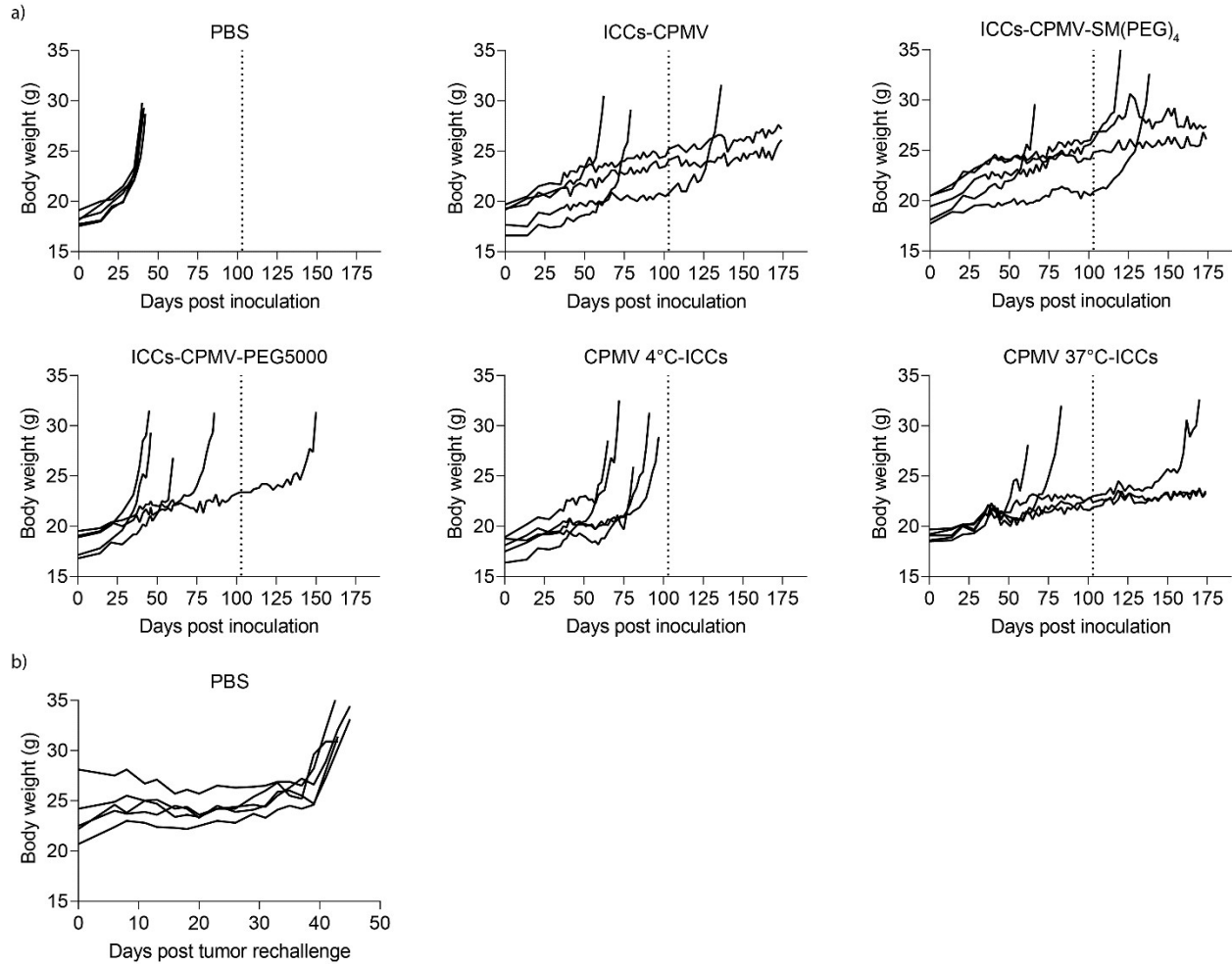


Figure S4. a) Body weights of each individual mice from the 6 groups were monitored as a sign of tumor progression. b) Body weights of individual mice with the same age of other vaccinated mice for the rechallenge experiment. Figures were generated using GraphPad Prism 8.