

Potentiating cancer immunotherapies with modular albumin-hitchhiking nanobody–STING agonist conjugates

Authors: Blaise R. Kimmel¹⁻³, Karan Arora³, Neil C. Chada⁴, Jeffrey C. Rathmell^{7,9-11}, Ann Richmond^{6,11}, W. Kimryn Rathmell^{2,5,9-11}, Justin M. Balko^{5,7,9-11}, and John T. Wilson^{3,4,8-12}

Affiliations:

1, Department of Chemical and Biomolecular Engineering, The Ohio State University, Columbus, OH, 43210
2, The Ohio State University Comprehensive Cancer Center, The Ohio State University, Columbus, OH, 43210

3, Department of Chemical and Biomolecular Engineering, Vanderbilt University, Nashville, TN 37232, United States

4, Department of Biomedical Engineering, Vanderbilt University, Nashville, TN 37232, United States

5, Department of Medicine, Vanderbilt University Medical Center, Nashville, TN 37232, United States

6, Department of Pharmacology, Vanderbilt University Medical Center, Nashville, TN, 37232, United States

7, Department of Pathology, Microbiology, and Immunology, Vanderbilt University Medical Center, Nashville, TN 37232, United States

8, Vanderbilt Center for Innovative Technology, Vanderbilt University, Nashville, TN 37235, United States

9, Vanderbilt Center for Immunobiology, Vanderbilt University Medical Center, Nashville, TN 37232

10, Vanderbilt Institute for Infection, Immunology and Inflammation, Vanderbilt University Medical Center, Nashville, TN 37232

11, Vanderbilt Ingram Cancer Center, Vanderbilt University Medical Center, Nashville, TN 37232, United States

12, Vanderbilt Institute of Chemical Biology, Vanderbilt University, Nashville, TN 37232, United States

Abstract: The enhancement of antitumor immunity via agonists of the stimulator of interferon genes (STING) pathway is limited by pharmacological barriers. Here, we show that the covalent conjugation of a STING agonist to anti-albumin nanobodies via site-selective bioconjugation chemistries prolongs the circulation of the agonist in the blood and increases its accumulation in endocrine tumor tissue, stimulating innate immune programs that increase the infiltration of activated natural killer cells and T cells, which potently inhibited the growth of mouse tumors. The technology is modular, as demonstrated by the recombinant integration of a second nanobody domain targeting programmed death-ligand 1 (PD-L1), which further increased the accumulation of the agonist in tumors while blocking immunosuppressive PD-1/PD-L1 interactions. The bivalent nanobody–STING agonist conjugate stimulated robust antigen-specific T-cell responses and long-lasting immunological memory and conferred enhanced therapeutic efficacy. It was also effective as a neoadjuvant treatment to adoptive T-cell therapy. As a modular approach, hitchhiking STING agonists on serum albumin may serve as a broadly applicable strategy for augmenting the potency of systemically administered cancer immunotherapies in metastatic endocrine cancers.

Funding: This research was supported by grants from the Susan G. Komen (CCR19609205 to J.T.W.), the National Institutes of Health (R01 CA245134, R01 CA266767 and R01 CA274675 to J.T.W.; R01 CA11601 to A.R.; R01 CA217987 to J.C.R.; NCI SPORE 2P50CA098131–17 to J.M.B.; the National Science Foundation (CBET-1554623 to J.T.W.), a VICC Ambassador Discovery Grant (J.T.W.), VICC Support Grant P30 CA68485, a Department of Defense Era of Hope Scholar Award (BC170037 to J.M.B.), the Department of Veterans Affairs (101BX002301 and IK6B005225 to A.R.) and funds provided by the Vanderbilt University School of Engineering (J.T.W.). B.R.K. acknowledges postdoctoral funding support from the PhRMA Foundation Postdoctoral Fellowship in Drug Delivery.

COI: J.T.W., K.A., and B.R.K. are inventors on United States Patent Application PCT/US2023/079884 'NANOBODY-DRUG CONJUGATES AND METHODS OF PREPARING THEREOF' which describes nanobody conjugation and delivery technologies.