LYMPH NODE TARGETED BOOSTING WITH COGNATE AMPHIPHILE-PEPTIDE VACCINES ENHANCES TCR-T CELL THERAPY TO ERADICATE SOLID TUMORS

Dylan Drakes*, Abdurraouf Abbas, Jacqueline Shields, Peter DeMuth. *Elicio Therapeutics, Boston, MA, USA*

Background Clinical results from TCR-T Cell therapies demonstrate anti-tumor efficacy, although therapeutic benefits remain transient due to suboptimal T Cell functional persistence and tumor infiltration alongside antigen escape mechanisms.^{1 2 3 4}

⁵ Amphiphile (AMP) vaccines improve lymph node targeting of cancer immunogens, stimulating an enhanced endogenous anti-tumor response.^{6 7} We describe an approach to generate robust and durable anti-tumor responses by combining AMP lymphatic targeting with TCR-T Cell therapy. AMP cognate peptides traffic to lymph nodes and improve TCR-T Cell activation, persistence, and function compared to soluble (SOL) peptide vaccination or TCR-T Cells alone, inducing a superior anti-tumor effect.

Methods C57BL/6J mice were subcutaneously implanted with B16F10 10 days prior to transduced pmel-1 T cell transfer or 75 days after T cell treatment for rechallenge experiments. Tumor-bearing mice received 5 doses, 2x/week of AMP-GP100/AMP-CpG, SOL-GP100/SOL-CpG, or PBS by tail-base vaccination. Caliper measurements determined tumor progression and overall survival. TCR-T Cell persistence was assessed bi-weekly through retro-orbital bleeds. Tumors and lymph nodes from treated mice were excised and analyzed by Nanostring for differential gene expression and flow cytometry for TCR-T Cell functional persistence and T cell epitope spread. Human T Cells (HTCs) and Dendritic Cells (DCs) were isolated from autologous PBMCs, transduced with KRAS-specific TCRs, and cultured with AMP-KRAS-peptide pulsed DCs before assaying T Cell boosting.

Results We demonstrate that AMP vaccination expands tumor specific TCR-T Cells in vivo up to 46-fold while enhancing the activation, cytokine secretion, and pro-inflammatory gene expression of tumor-infiltrating TCR-T Cells. Endogenous tumor-infiltrating T cells from AMP vaccinated mice produced up to 17-fold greater cytokine secretion following re-stimulation with non-targeted tumor epitopes. These results correspond to the eradication of established B16F10 tumors and a resistance to secondary tumor challenge in cured mice. Providing clinical relevance, HTCs transduced with KRAS-specific TCRs and boosted with AMP-KRAS-peptide pulsed DCs exhibited enhanced T cell activation, Th1 cytokine secretion, and cytolytic capacity compared to HTCs exposed to unlabeled DCs.

Conclusions AMP vaccination delivers cognate peptides to lymph nodes providing in vivo activation of tumor-specific TCR-T Cells which amplifies anti-tumor potency of such adoptively transferred cells. AMP vaccination significantly enhanced TCR-T Cell anti-tumor response and led to durable cures of solid tumors in an established, syngeneic tumor model. Additionally, AMP-peptide pulsed autologous DCs enhanced the function of clinically relevant KRAS-specific TCR-T cells in vitro. Taken together, these studies provide direct rationale and evidence for the combination of AMP vaccination with TCR-T Cell therapies to augment clinical responses.

REFERENCES

1. Robbins PF, Morgan RA, Feldman SA, Yang JC, Sherry RM, Dudley ME, Wunderlich JR, Nahvi AV, Helman LJ, Mackall CL, et al. Tumor regression in patients

with metastatic synovial cell sarcoma and melanoma using genetically engineered lymphocytes reactive with NY-ESO-1. *J Clin Oncol* 2011;**29**:917–924. doi: 10.1200/JCO.2010.32.2537.

2. Rapoport AP, Stadtmauer EA, Binder-Scholl GK, Golubeva O, Vogl DT, Lacey SF, Badros AZ, Garfall A, Weiss B, Finklestein J, et al. NY-ESO-1-specific TCR-engineered T cells mediate sustained antigen-specific antitumor effects in myeloma. *Nat Med* 2015;**21**:914–921. doi: 10.1038/nm.3910.
3. Doran SL, Stevanovic S, Adhikary S, Gartner JJ, Jia L, Kwong MLM, Faquin WC, Hewitt SM, Sherry RM, Yang JC, et al. T-Cell Receptor Gene Therapy for Human Papillomavirus-Associated Epithelial Cancers: A First-in-Human, Phase I/II Study. *J Clin Oncol* 2019;**37**:2759–2768. doi: 10.1200/JCO.18.02424.
4. Chandran SS, Klebanoff CA. T cell receptor-based cancer immunotherapy: Emerging efficacy and pathways of resistance. *Immunol Rev* 2019;**290**:127–147. doi: 10.1111/imr.12772.
5. D'Angelo SP, Melchiori L, Merchant MS, Bernstein D, Glod J, Kaplan R, Grupp S, Tap WD, Chagin K, Binder GK, et al. Antitumor Activity Associated with Prolonged Persistence of Adoptively Transferred NY-ESO-1 (c259)T Cells in Synovial Sarcoma. *Cancer Discov* 2018;**8**:944–957. doi: 10.1158/2159-8290.CD-17-1417.
6. H Liu, KD Moynihan, Y Zheng, GL Szeto, AV Li, B Huang, DS Van Egeren, C Park, DJ Irvine. Structure-based programming of lymph-node targeting in molecular vaccines. *Nature* 2014;**507**: 519–522. doi: 10.1038/nature12978.
7. KD Moynihan, CF Opel, GL Szeto, A Tzeng, EF Zhu, JM Engreitz, RT Williams, K Rakhra, MH Zhang, AM Rothschilds, S Kumari, RL Kelly, BH Kwan, W Abraham, K Hu, NK Mehta, MJ Kauke, H Suh, JR Cochran, DA Lauffenburger, KD Wittrup, DJ Irvine. Eradication of large established tumors in mice by combination immunotherapy that engages innate and adaptive immune responses. *Nat Med* 2016;**22**: 1402–1410. doi: 10.1038/nm.4200.

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