DNase I Intervention Enhances CAR-T Cell Therapy in Solid Tumors by Targeting Neutrophil Extracellular Traps in Metastatic Melanoma

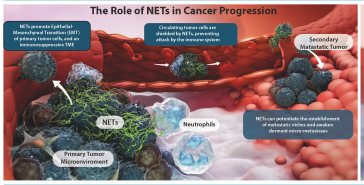
Alexey Stepanov[1] PhD, Wenjian Wang[1] PhD, Yingqin Hou[1] PhD, Ivan Chernikov[1] PhD, Reid Bissonnett[2] PhD, Curtis Lockshin[2] PhD, George Tetz[3] PhD, Grigory Borisenko[2] PhD and Dmitry Genkin[2] MD PhD

[1] Scripps Research, La Jolla, CA; [2] Xenetic Biosciences, Framingham, MA; [3] CLS Therapeutics, New York, NY

Abstract #123

Background

In recent years, chimeric antigen receptor (CAR) T cell therapy has emerged as a promising approach for treating various malignancies, most notably in hematologic cancers. However, its efficacy in solid tumors is often hindered by an immunosuppressive tumor microenvironment (TME), which, in particular, is frequently characterized by the presence of tumor-associated cell-free DNA (cfDNA), especially in the form of neutrophil extracellular traps (NETs). In adoptive immunotherapies NETs, composed primarily of neutrophil-derived DNA, histones and neutrophil granule enzymes (e.g., MPO, NE), act as physical barriers and induce the secretion of immunosuppressive factors that impair CAR T cell function.

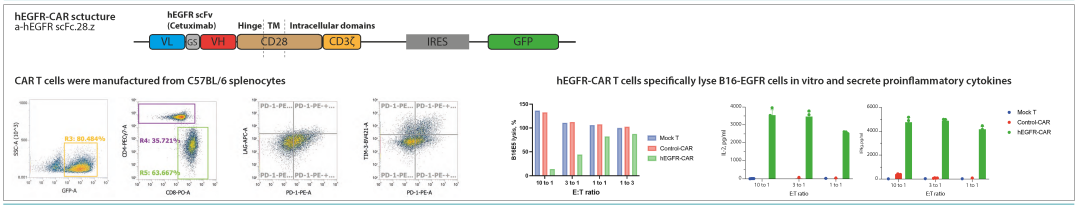


Materials&Methods

1. Female C57BL/6 mice 7-8 weeks old at start.
2. 0.8*10^6 B16-hEGFR tumor cells intravenously (i.v.).
3. Day before CAR T cells injection mice preconditioned by 5Gy irradiation or cyclophosphomide.
4. Day 7 Injection of 2*10^6 hEGFR-CAR T cells i.v.
5. Day 7 rhDNaseI treament 10 mg/kg started.
6. For TILs and Metastatic index analysis lungs colected at day 3 and 6 after CAR T cells infusion.

Summary

This research highlights the critical role of the NETs in modulating CAR T cell efficacy and provides a compelling rationale for incorporating DNase I as an adjunctive treatment to improve therapeutic responses in patients undergoing CAR T cell therapy. Further clinical studies are warranted to validate these findings and explore the translational potential of this combinatorial approach in enhancing cancer treatment.

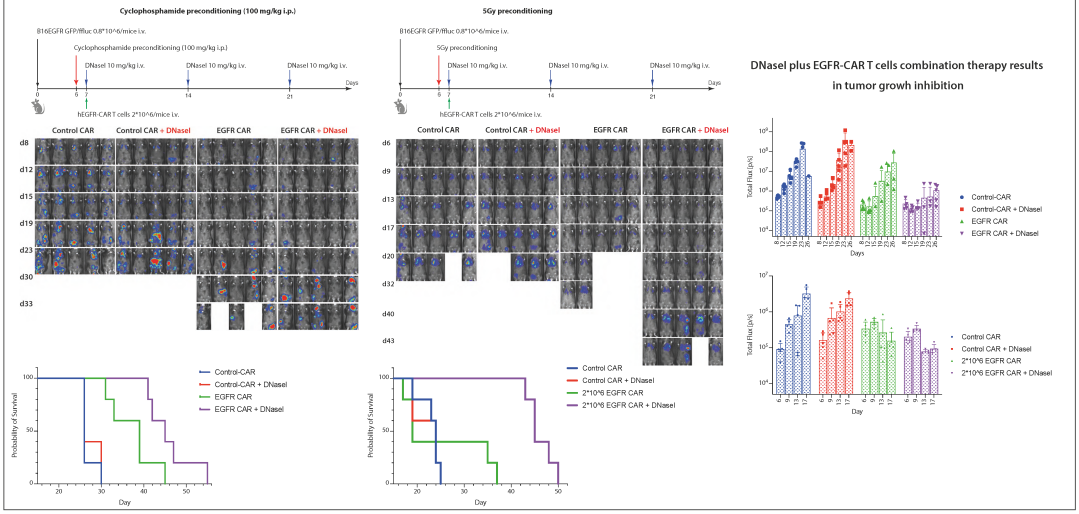


Results

This study investigates the synergistic effects of co-administering deoxyribonuclease I (DNase I) with CAR T cells in a syngeneic B16 melanoma model of lung metastasis. Using bioluminescent imaging, we observed that a single injection of DNase I (10 mg/kg) effectively suppressed B16-EGFR lung metastasis at early stages compared to vehicle controls. However, while DNase I demonstrated efficacy in reducing tumor growth, it did not significantly improve survival when administered alone.

In contrast, the combination treatment of DNase I with murine EGFR-CAR T cells led to a marked suppression of tumor burden, a decrease in the number of metastatic foci, and substantial prolongation of survival compared to CAR T cell monotherapy. This therapeutic enhancement was associated with an increase in tumor-infiltrating T and CAR T cells, indicating improved immune engagement. Notably, analysis of the CD8 T cell population from DNase I-treated groups revealed a notable decrease in PD-1 and TIM-3 expression, markers of T cell exhaustion, suggesting that DNase I effectively mitigates the immunosuppressive effects of the TME.

DNaseI plus EGFR-CAR T cells combination therapy results in tumor growh inhibition and enhanced survival in mice bearing B16-EGFR lung metastatic melanoma tumors.



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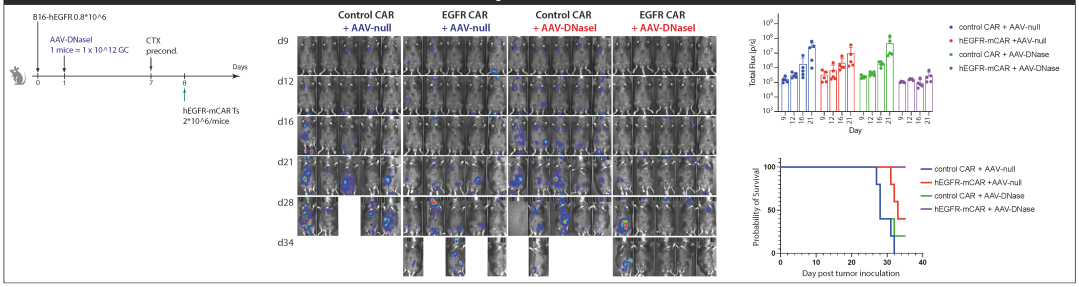
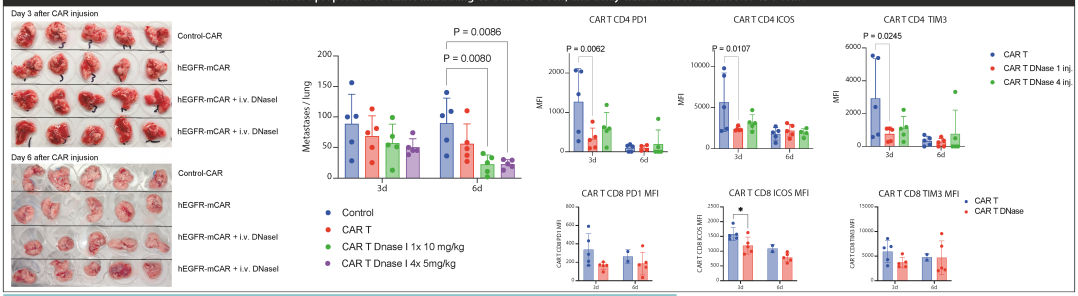


hEGFR-CAR T cells combination with i.v. DNase I supress the lung metastases in melanoma murine model. Increase proportion of tumor infiltrating CD4 and CD8 cels, and delay exhaustion of the effector CD8 cells.



Conclusions

Our findings illuminate the crucial role of NETs in limiting CAR T cell function and underscore the potential of DNase I as an adjunctive treatment to enhance immunotherapeutic responses. By degrading NETs, DNase I not only facilitates increased T cell infiltration but also restores T cell functionality, paving the way for more effective cancer treatment strategies. Further clinical studies are warranted to validate these findings, explore the translational potential of this combination approach, and assess its efficacy in various solid tumor contexts.

Xenetic BIOSCIENCES

Scripps Research