SYSTIMMUNE

Abstract Number: 5680

Tetra-specific antibody GNC-039: Guidance and Navigation Control (GNC) molecule development for treatment of EGFRvIII+ malignancies

Jahan Salar Khalili, Sa Xiao, Yi Zhu. SystImmune Inc., Redmond, WA, Sichuan Baili Pharmaceutical Co., Ltd., Chengdu, China

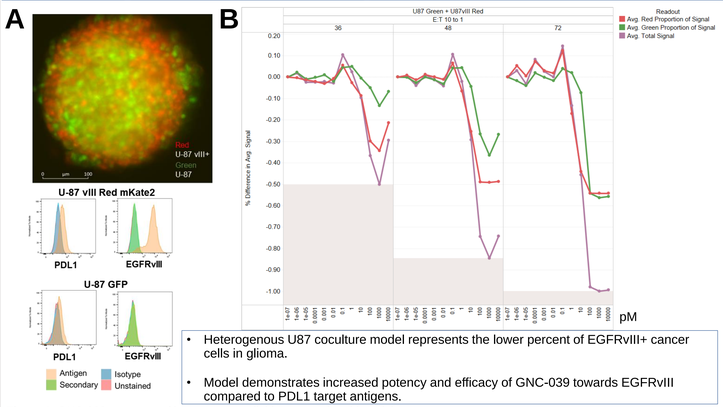
Abstract

One of the primary challenges in the targeting of the tumor specific antigen EGFRvIII is the expression of the antigen among tumor cells in cranial glioblastoma tumors. The development of GNC-039 is based on the capability of this protein to redirect T cell cytolysis toward the tumor specific antigen EGFRvIII and guide T cells in the tumor microenvironment. Here we demonstrate that the tetraspecific Guidance and Navigation Control (GNC) antibody, GNC-039, binds to EGFRvIII, CD3, PD-L1, and 4-1BB and mediates T cell cytolysis of the human glioblastoma cancer cell line U87 expressing EGFRvIII in the in vitro tumor spheroid model.

When delivered intravenously, the biodistribution of GNC-039 is an important factor in the development of this intracranial tumor targeting biologic. To better understand the biodistribution of GNC-039, Orthotopic Patient Derived Xenograft (PDX) models of Glioma were utilized and vivo-tag680XL-labelled GNC-039 was evaluable by total flux in the Brain area. Multiple IV infusions of GNC-039 were carried out over the study period (29 Days). Groups received either GNC-039 (n=5), GNC-039 with engrafted PBMC (n=5), Temozolomide(n=5), or Vehicle (n=5). The PDX model was sensitive to Temozolomide, 0/5 mice residual tumor, median overall survival (mOS) of 15 days. In the Vehicle treatment group, 5/5 mice had residual tumor, mOS was 26 days. Mice receiving GNC-039 without PBMC, could partially respond to the treatment, in this group 3/5 animal had residual brain tumors by end of study, but with only a mOS of 15 days. In these animals, the GNC-039 accumulated in the brain region to its maximal level by the third dose on Day 7 and stayed consistently at that level for the duration of the study period. However, in the mouse group with engraft PBMC, the level of GNC-039 in the brain region could exceed that of the drug when infused alone. Mice receiving GNC-039 with engrafted PBMC completely responded to treatment, 5/5 mice in the group had no residual brain tumor, and a mOS of 20 days. In the presence of the engrafted PBMC, the increased level of GNC-039 in the brain region was delayed compared with treatment in the absence of PBMC. As a point of comparison, the Day 7 levels from GNC-039 treatment alone were not reached until Day 15 in presence of PBMC. However, beyond this timepoint, the level of GNC-039 in the brain region was significantly increased due to the engrafted PBMC.

Collectively this data indicates the functionality of GNC-039 as a multi-specific T cell engager with the potential to target EGFRvIII+ cancer cell cytolysis in primary brain disease. The clinical phase I-b study of GNC-039 is under way and the available data exhibit strong signals of efficacy with acceptable tolerability.

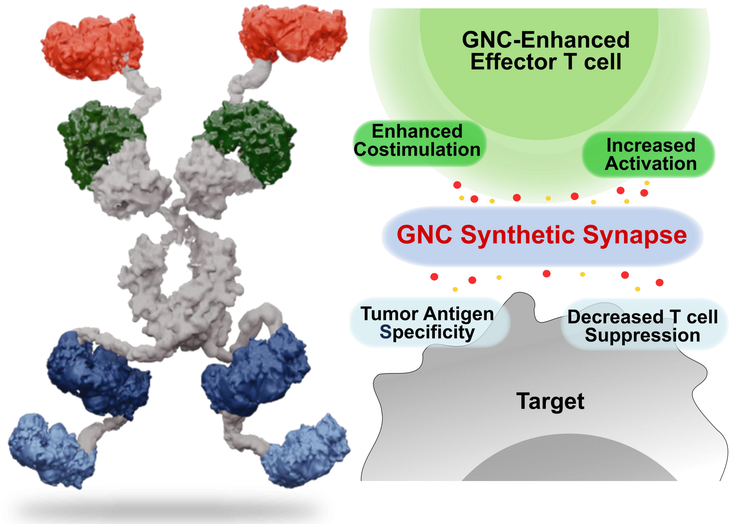
Heterogeneous EGFRvIII glioblastoma *in vitro* model



- Heterogenous U87 coculture model represents the lower percent of EGFRvIII+ cancer cells in glioma.
- Model demonstrates increased potency and efficacy of GNC-039 towards EGFRvIII compared to PDL1 target antigens.

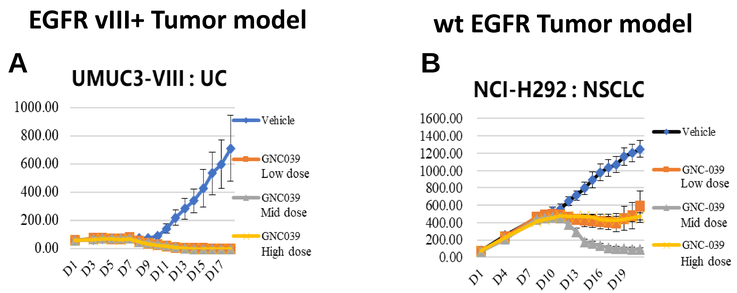
Figure 1. GNC-039 treatment of heterogenous glioblastoma spheroid model
Glioblastoma spheroid coculture models were established using EGFR+, EGFRvIII- U87 (green) and EGFR+, EGFRvIII+ U87 (red) cell lines (**A**). Redirected T cell cytotoxicity assay performed to evaluate GNC-mediated cytolysis of EGFRvIII-, EGFRvIII+ and total U87 cancer cell signal. Data are shown for 36, 48 and 72-hour timepoints (**B**).

GNC-039: Tetra-specific T cell engager



2nd tetra-specific antibody therapy in human trials

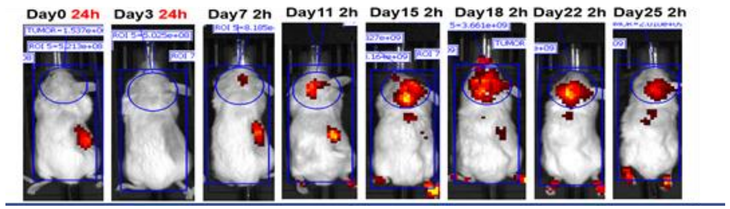
Subcutaneous EGFRvIII+ Bladder Cancer and wild type EGFR Lung Cancer xenograft models



- GNC-039 induces antigen-specific tumor size reduction in EGFRvIII+ and EGFR- subcutaneous cancer cell xenografts at all drug doses tested.
- Improved activity is observed in EGFRvIII+ xenograft, while GNC retains PDL1-specific killing in EGFRvIII- xenograft, indicating *in vivo* activity against both EGFRvIII positive and negative tumors.

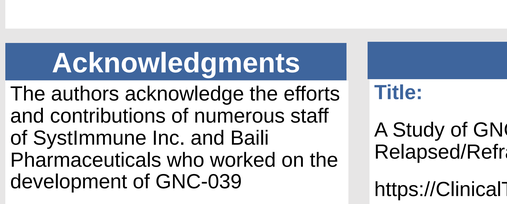
Figure 2. GNC-039 treatment in mouse xenograft model
NCG mice were implanted with **A**) UMUC-vIII bladder cancer and **B**) NCI-H292 Non-small cell lung cancer tumor xenografts to measure reduction in tumor volume during treatment with multiple doses of GNC-035 . Error bars represent SEM.

T cell mediated GNC Homing to cranial PDX Glioma



BN2287: Orthotopic Glioma PDX

Group	Residual tumor in brain (No.)	mOS (Day)
Vehicle	5/5	26 [10-29]
GNC-039	0/5	20 [14-29]
TMZ	0/5	15 [10-20]
GNC-039, No PBMC	3/5	15 [8-26]



- **Partial responses** observed in mice receiving GNC-039 in **absence of PBMC**. Maximal GNC accumulation at Day 7.
- Median overall survival of **15 days**. **Complete response** observed in mice receiving GNC-039 and **engrafted PBMC.** Maximal GNC accumulation beyond Day 15 in presence of PBMC. Complete clearance of tumor and median overall survival of **20 days**.

Figure 3 GNC-039 treatment in mouse PDX model
Orthotopic Patient Derived Xenograft (PDX) models of Glioma were utilized and vivo-tag680XL-labelled GNC-039 was evaluable by total flux in the Brain area. Multiple IV infusions of GNC-039 were carried out over a 29-day study period. Groups received either GNC-039 (n=5), GNC-039 with engrafted PBMC (n=5), Temozolomide(n=5), or Vehicle (n=5). Error bars represent SEM.

Summary

- GNC-039 exhibits increased potency and efficacy of GNC-039 towards EGFRvIII compared to PDL1 target antigens in in vitro coculture model of glioblastoma.
- GNC-039 exhibits improved activity in EGFRvIII+ and EGFRvIII- xenograft *in vivo.*
- In a PDX model of glioblastoma, mice receiving GNC-039 with engrafted PBMC completely responded to treatment, 5/5 mice in the group had no residual brain tumor, and a mOS of 20 days.
- Levels of GNC-039 in the brain region were significantly increased in the presence of engrafted PBMC
- Collectively this data indicates the functionality of GNC-039 as a multi-specific T cell engager with the potential to target EGFRvIII+ cancer cell cytolysis in primary brain disease.

Acknowledgments

The authors acknowledge the efforts and contributions of numerous staff of SystImmune Inc. and Baili Pharmaceuticals who worked on the development of GNC-039

References

Title:

A Study of GNC-039, a Tetra-specific Antibody, in Participants With Relapsed/Refractory or Metastatic Solid Tumors

https://ClinicalTrials.gov/show/NCT04794972