

1177

**MULTIFUNCTIONAL NATURAL KILLER CELL ENGAGER  
RELEASING CXCL10 AUGMENTS NATURAL KILLER CELL  
RECRUITMENT AND ANTI-TUMOR EFFICACY AGAINST  
GLIOBLASTOMA**Sandro Matosevic, Xue Yao\*. *Purdue University, West Lafayette, IN, USA*

**Background** The effectiveness of natural killer (NK) cell-based immunotherapy against solid tumors is limited by the inadequate infiltration of NK cells into the tumors and the heterogeneous immunosuppressive tumor microenvironment (TME), suggesting that a multi-specificity of targeting mechanisms is needed to achieve durable responses.

**Methods** Tumor-infiltrating NK cells and peripheral blood (PB) NK cells from glioblastoma multiforme (GBM) patients were isolated to measure the expression of chemokine receptor CXCR3. The effect of exogenous CXCL10 on NK cell migration and functions was also established. We further investigated the regulation of the CXCR3-CXCL10 axis on NK cells and tumor cells by establishing CXCR3-knockdown primary NK cells and CXCL10-overexpressing GBM cells. Based on these findings, we designed and synthesized a novel natural killer cell engager (NKCE) which not only targets NK cells and tumor cells, but also specifically releases CXCL10 locally and responsively in the tumor while sustaining NK cell activation and persistence via production of interleukin (IL)-15.

**Results** CXCR3 expression was upregulated on NK cells in PB from healthy donors compared to blood NK cells from GBM patients, while also being upregulated on tumor-infiltrating NK cells from GBM patients. One of the chemokine ligands for CXCR3, CXCL10, induced NK cell migration via CXCR3 but did not affect NK cell functions. In addition, CXCL10 overexpression showed no effect on tumor growth but resulted in enhanced NK cell migration into tumor sites and, in turn, improved the anti-tumor activity of NK cells. Furthermore, our novel NKCE activated NK cells and efficiently recognized and targeted GBM tumor cells, thus promoting the contact between NK cells and tumor cells. The engager's CXCL10-releasing domain, which is activated in the local TME, promotes the specific increase in CXCL10 concentration and, in turn, NK cell homing in the TME and efficient anti-GBM responses.

**Conclusions** The CXCR3-CXCL10 axis contributes to the recruitment of NK cells to GBM. Our novel NKCE which has the ability to responsively and locally release CXCL10 induced NK cell migration and boosted NK cell anti-tumor activity against solid tumors. Such a multi-specific approach not only activates NK cells locally, but promotes their recruitment and retention in the TME.

<http://dx.doi.org/10.1136/jitc-2023-SITC2023.1177>