KNOCKOUT OF THE INHIBITORY RECEPTOR TIGIT ENHANCES ANTI-TUMOR RESPONSE OF EX VIVO EXPANDED NK CELLS AND PREVENTS FRATRICIDE WITH THERAPEUTIC FC-COMPETENT TIGIT ANTIBODIES

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Background Antibody blockade of the Natural Killer cells (NK cells) and T cell inhibitory receptor TIGIT has been shown to enhance tumor control and survival in preclinical mouse models and early clinical trials; however, there is a lack of understanding of the effect of TIGIT engagement on anti-tumor functions of activated primary human NK cells. Additionally, the majority of TIGIT antibodies in clinical development have a humanized IgG, which induces antibody-dependent cellular cytotoxicity (ADCC) and likely contributes to their efficacy. However, the potential consequences of these Fc-competent antibodies when binding NK cells, such as fratricide, have not been well characterized. Fratricide could deplete NK cells upon treatment, a detrimental effect to the overall efficacy since NK cells play a critical role in the success of checkpoint blockade immunotherapies. Recent efforts have focused on developing adoptive NK cell therapy and combinatorial immune-oncology therapies in order to enhance response. Adoptive transfer of TIGIT KO NK cells could provide a treatment strategy to mitigate potential negative effects of TIGIT blockade on NK cells. In this study, TIGIT knockout in ex vivo PM21-particle expanded human NK cells was performed and the effect on anti-tumor activity alone or in combination with Fc-competent TIGIT antibody blockade was evaluated and compared to WT NK cells.

Methods CRISPR was used to make a targeted TIGIT knockout (KO) in ex vivo PM21-particle expanded NK cells (PM21-NK cells). TIGIT KO NK cells were compared to wild type (WT) NK cells to determine changes cytotoxicity, ADCC, and IFNγ, TNFα, and the degranulation marker CD107a expression. Glycolytic rate and mitochondrial stress were measured. TIGIT KO or WT PM21-NK cells were combined with Fc-competent or non-Fc-competent anti-TIGIT and fratricide and cytotoxicity were measured.

Results TIGIT KO PM21-NK cells showed improved killing compared to WT against 3D spheroids from multiple cancer cell lines. ADCC increased proportionally in TIGIT KO NK cells. TIGIT KO PM21-NK cells had increased CD107a surface expression after cancer spheroid exposure and increased basal glycolytic rate after stimulation. TIGIT Knockout prevented Fc-competent anti-TIGIT driven NK cell fratricide and prevented decrease in NK cell cytotoxicity when combined with Fc-competent anti-TIGIT.

Conclusions Knockout of TIGIT in ex vivo expanded PM21-NK cells resulted in NK cells with improved anti-tumor activity and metabolic fitness. TIGIT KO prevented ADCC driven NK cell fratricide and inhibition of cytotoxicity when combined with Fc-competent anti-TIGIT. TIGIT KO PM21-NK cells are a potential cellular product for therapeutic use in combination with TIGIT blockade.

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