TUMOR REACTIVE, CYTOKINE INDUCED, ENGINEERED HUMAN NATURAL KILLER CELLS DEMONSTRATE SERIAL CYTOTOXICITY AGAINST LIQUID AND SOLID TUMOR CELL TARGETS

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Background Expression of PD-L1 on human natural killer (NK) cells identifies a subset that are selectively reactive to tumors. PD-L1(+) NK cells have increased CD107a expression, cytotoxicity, and interferon-gamma secretion vs. the PD-L1(-) fraction. PD-L1(+) NK cells are manufactured for patient administration using cytokine stimulation under GMP conditions and in vivo survival sustained by engineering these tumor-reactive, cytokine-induced killer (TRACK) NK cells to express soluble (s)IL-15. Cryopreserved, off-the-shelf allogeneic TRACK NK cells expressing sIL-15 are being assessed in a phase 1 trial in patients with relapsed/refractory non-small cell lung cancer (NSCLC; NCT05334329). Here, we assess phenotype and function of human TRACK NK cells and demonstrate serial natural cytotoxicity and antibody-dependent cellular cytotoxicity (ADCC) lymphoma and NSCLC targets.

Methods TRACK NK cells were manufactured from umbilical cord blood NK cells by activation and expansion of NK cells over a feeder cell line, engineering to secrete sIL-15, followed by cytokine activation before cryopreservation. Serial TRACK NK cell cytotoxicity was measured as killer frequency, defined as the average number of tumor target cells killed per single TRACK NK cell over 8–40 hours.1

Results Fresh TRACK NK cells expressed >90% CD16 immediately prior to cryopreservation; following thaw, resting NK cell CD16 expression was >60% at T0 but rose significantly to >75% following culture at 37°C for 6 h and >82% at 24 h (p<0.05), mimicking upregulations as would likely occur in vivo. Similar statistically significant upregulation of NKp44 and NKp46 (p<0.05) was observed on TRACK NK cells, whereas very high expression of NKp30, NKG2D, and DNAM-1 remained stable. CD57 and PD-1 expression remained low (~1–5%). TRACK NK cells performed robust ADCC against both CD20(+) at various E:T in the presence of anti-CD20 and anti-EGFR, respectively, significantly better than NK killing of either target. Substantial TRACK NK cell cytoReduction of NSCLC was also demonstrated in vivo. In addition, antibody independent serial cytotoxicity and antibody-dependent serial cytotoxicity by TRACK NK cells was demonstrated against both lymphoma and NSCLC lines.

Conclusions Under various conditions, each TRACK NK cell was able to kill a multitude of lymphoma cells via ADCC in the presence of anti-CD20 mAb or a multitude of NSCLC in the absence or presence of antibody. These data support the use of the TRACK-NK cells in the clinic using both their natural cytotoxicity and their ADCC mechanism of action against both liquid and solid tumors.

REFERENCE