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Robust GMP manufacturing process with IL-2 and a glycoprotein antibody cocktail generates highly active human NK cell batches for cancer immunotherapy

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Purpose/Objectives: NK cells are innate immune cells crucial for killing of infected and malignant cells. They are able to fight circulating tumor cells thereby preventing metastases formation which account for approximately 90% of all cancer-deaths. Moreover, infiltration of solid tumors with NK cells has been shown to correlate with a better prognosis for the patient. Thus, NK cells became interesting candidates for cancer immunotherapy and ex vivo manipulation and expansion of highly potent NK cells for adoptive transfer in patients is an aim of paramount importance. Using a novel approach for NK cell expansion, we aimed at providing an extensive characterization of the generated cells to evaluate their potential for cancer immunotherapy.

Materials/Methods: Human, CD3+ T cell-depleted PBMCs were expanded in a bioreactor with IL-2 and a glycoprotein antibody cocktail following GMP guidelines. Cells were analyzed for NK cell purity, expression of different chemokine receptors, cell adhesion molecules, activating receptors and death ligands as well as IFNγ production using flow cytometry. Further, cytotoxicity towards different tumor cell lines was assessed via LDH assays and flow cytometry-based degranulation assays.

Results: Upon expansion NK cell purity reached 85 to 96%. The cells showed expression of the chemokine receptors CXCR3, CXCR4 and CCR7 and the cell adhesion molecules L-selectin, LFA-1 and VLA-4. Further, they expressed the activating receptors NKp30, NKp44, NKp46, NKG2D, DNAM-1 and CD16, the death ligands Fas and TRAIL and produced IFN?. NK cells showed cytotoxicity towards the tumor cell lines K562 (leukemia), PaCa5061 (pancreatic cancer), and SKOV3 (ovarian cancer).

Conclusion: We describe a novel approach for ex vivo NK cell expansion generating a set of highly potent NK cells which represent promising candidates for cancer immunotherapy. The GMP manufacturing process allows the use of these cells in clinical trials, i.e. adoptive NK cell transfer in patients.