Evaluation of *ex vivo* expanded human NK cells on anti-leukemia activity in a human CML/SCID-beige mouse model

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Natural-killer (NK) cells are lymphocytes committed to the innate immune defense. Their cytolytic activity targets mostly malignant or virus infected cells. The possibility of using NK cells in treatment of human hematological malignancies has increased in recent years. In part, this is a consequence of an increased understanding of how NK receptor-ligand matching influences lysis of target cells. Additionally, the introduction of methods for *ex vivo* generation of enriched populations of clinical grade NK cells has allowed obtainment of sufficient numbers of human NK cells for clinical trials. In the present study the safety and *in vivo* anti-tumor activity of human *ex vivo* expanded clinical grade NK cells were evaluated against human chronic myeloid leukemia (CML) cells in SCID-beige mice. CML was induced in irradiated mice by intravenous (i.v.) injection of $1x10^6$ K562 tumor cells. One day after tumor-cell injection mice were treated with $20x10^6$ *ex vivo* expanded human NK cells by i.v. injection. This procedure resembles treatment of human CML residual disease. Administration of NK cells was safe. NK cells were detected until day four after i.v. injection into the mice, and they were found in the same tissues infiltrated by CML tumor cells. Administration of NK cells prevented leukemia development, proving anti-leukemia activity *in vivo* of the present *ex vivo* expanded clinical grade NK-cell-enriched effector-cell population. In the untreated

group 80 % of mice developed leukemia within 80 days after tumor-cell injection, whereas 70 % of the mice treated with the NK-cell-enriched effector-cell population remained healthy within the period of observation (100 days). These results validate the present human clinical grade NK cells as a source for cellular immunotherapy, implying new possibilities for treatment of human hematological malignancies and possibly other forms of cancer.

Key words: NK cells, immunotherapy, chronic myeloid leukemia