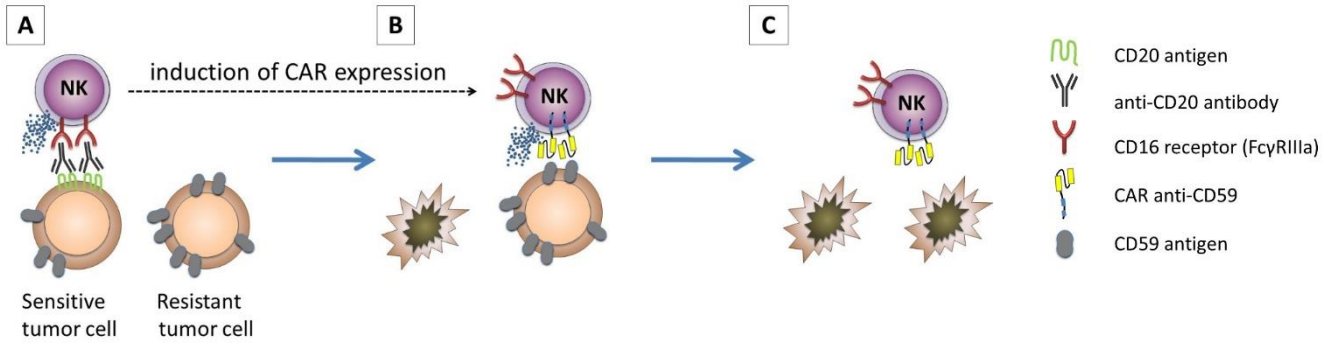
The malignant tumors are second cause of death in Poland, after cardiovascular diseases. While the mortality from cardiovascular diseases has dropped significantly in recent years, the incidence of cancer and the death rate has significantly increased, including the high incidence of malignancies originating form hematopoetic cells. Currently available treatment for tumors originating from B-lymphocytes, such as leukemia or lymphoma, includes application of chemotherapy combined with immunotherapy, which is based on antibodies, directed against molecules on the surface of malignant cells, such as CD20 antigen. High response rates have been reported with rituximab, an anti-CD20 antibody. The binding of the antibody to CD20 molecule results in direct death of tumor cell, either by the action of complement cascade or by engaging mechanisms of innate immune response such as cytotoxic action of natural killer (NK) cells.

Some patients tolerate well the therapy, however others develop a drug resistance and need to be treated with multi-drug combinations, which provide also some side effects. Therefore the hematologic community is looking for new safe combination schemes with minor side effects, such as chemotherapy-free approaches. The introduction of any new drug to the clinic needs however to be preceded by basic research studies at the molecular level. Active research is also needed in order to understand the changes in the malignant B-cell microenvironment, in order to overcome the mechanisms of resistance to therapies.

We propose a basic research study that would improve our understanding of the intercellular signaling in NK cells, induced by contact with malignant B-cells. We will use the knowledge gained during the initial steps of the project in order to design a tool with potential impact on the future immuno-therapeutical schemes. We plan to engineer NK cells with expression of Chimeric Antigen Receptors (CARs) inducible by anti-CD20 antibodies bound to malignant cells. Importantly, the CAR proteins will be able to specifically recognize and target malignant B-cells resistant to anti-CD20 monoclonal antibodies, such as rituximab. The resistance of lymphoma cells to immunotherapy with anti-CD20 mAbs is largely dependent on the expression level of complement-inhibitory proteins. We therefore propose to use this approach for a specific targeting of tumor cells that express high surface levels of complement-inhibitory proteins.



Simplified schema of the proposed experimental approach. (A) NK cell becomes activated by anti-CD20 antibody-coated malignant B-cell and causes death of sensitive tumor cell; the resistant cell lacking CD20 expression remains alive; (B) activated NK cell express the CAR protein; CAR recognizes and targets the complement-inhibitory protein on the surface of resistant tumor B-cell; (C) both the sensitive and the resistant malignant B cell are effectively eliminated.

At the completion of the research studies, we expect to acquire a comprehensive understanding of the molecular mechanisms effecting NK cells response to contact with malignant B cells during the antibody-mediated therapy. This initial phase of research will pave the road for building an armed NK cells with inducible CAR receptors. Their "built-in" capability will allow the NK cells to sense the presence of malignant B-cells and engage them in a kiss that brings them death despite their resistance to therapies. We hope that our system with inducible expression of the CAR receptors will help in efficient elimination of tumor cells resistant to traditional therapies with monoclonal antibodies. We also believe that such a tool will participate to the development of new anti-tumor approaches.