Aggressive blood cancers (acute myeloid leukemias, AML) are characterized by ability to reprogram the ways how to produce energy, becoming dependent on intensive oxygen utilization called oxidative phosphorylation (OxPhos). OxPhos is therefore considered leukemia's core property and plays an important role in its progression and poor therapy response. Poor therapy response is partially due influence leukemia cells (blast) have on immune cells function, causing their impairment and weakening their properties to fight against blasts. Although blocking OxPhos with new drugs, called OxPhos inhibitors, in fight against leukemia, gave initially promising results, was very soon considered as toxic, because of leukemia cells ability to escape this blockade by switching to oxygen-independent way of energy production- fermentation. Leukemic blasts forced to fermentation by OxPhos blockade excrete an extensive amount of lactic acid (lactate) that causes sour/acidic microenvironment for all cells surrounding them and by releasing lactate into blood stream, causing in patients a severe side effect called lactate acidosis. Lactate accumulation around the leukemic cells impacts negatively performance of macrophages, cells responsible for removing all sick or damaged cells. Here however upon lactate effect, macrophages change their features from good guys (known as type M1) to bad guys (type M2). Instead of fighting against blasts, like M1, bad guys M2 start to protect them. **To understand processes of blast-macrophages communication might have therapeutic significance and therefore needs to be intensively investigated.** Our preliminary analysis of genes important in leukemia development on single cell resolution level indicated that leukemic monocytes, that give origin to macrophages, have an elevated expression of genes involved in metabolism of amino acids (building blocks of proteins), lactate transport (MCT4 and MCT1) and amino acids (ASCT2 and LAT1). Our observation suggests a potential role of ASCT2, LAT1 and MCT4 in blast-macrophages communication, in switching back macrophages from M2 to M1 state. We showed already, that the blockade of OxPhos and MCT1 prolonged significantly life of mice transplanted with T-lymphoblastic leukemia (T-ALL), due to lactate trapping inside the cells together with irreversible severe damages in genetic material leading ultimately to cell death. These already generated data show, that dual blockade of OxPhos/MCT1 could lift up the negative effects of acidification on macrophages, increase the pool of macrophages M1 and enhance their fight activity against blast. We propose new investigations, which could extend patients survival, minimize observed side effects of treatment and improve treatment tolerability in group of older patients, unfit for aggressive chemotherapy. We propose the hypothesis that **blocking MCT1/MCT4 on leukemic cells and/or MCT4 on macrophages together with blockade of LAT1 and/or ASCT2 could constitute a backbone of novel safe therapy.**

To test this hypothesis, we will conduct tests in cell lines and cells obtained from patient samples and later will verify our results in mice models, executing five specific aims: investigation of 1) the MCT1/MCT4 blockade on OxPhos activity in leukemic cells; 2) the effect of dual blockade MCT1/MCT4 with OxPhos blockade on blasts; 3) the MCT1/MCT4/OxPhos blockade on macrophages activity and role of ASCT2 and LAT1; 4) the blockade of MCT1/MCT4 and OxPhos on gene expression and metabolism in both blast and macrophages; 5) the efficacy of MCT1/MCT4 and OxPhos blockade to eradicate leukemia in mice models and benefits resulted from consolidation treatment based on healthy macrophage transplantation post completion of treatment with inhibitors. Our study is planned for 4 years. To execute proposed aims, we will utilize modern methods of molecular biology and complex techniques to provide confidently proof of principles. The results generated from basic experimental studies, will be evaluated in mice, which will receive human leukemic cells and will be treated with our experimental OxPhos/MCT1/MCT4 inhibitors. We will undertake also an attempt to block ASCT2 and/or LAT1on macrophages with antibodies and ASCT2 and LAT inhibitors, aiming to increase the pool of M1 macrophages and to restore function of immune system to fight with leukemia.

Finally, we will investigate possibilities to deepen therapeutic effect of MCT1/MCT4/Oxphos blockade, providing in examined mice models a transfusion of macrophages obtained from healthy blood donors , which will help to eliminate from blood circulation and bone marrow remaining residual leukemic cells. The evaluation of efficacy of this innovative therapy to eradicate the minimal residual disease in mice models will constitute a basis for initiation of clinical trials.

Given the fact that energetic needs of leukemic cells are universal, the proposed therapeutic strategy could be universally applied in all patients, including those who are older or ineligible for aggressive chemotherapy.