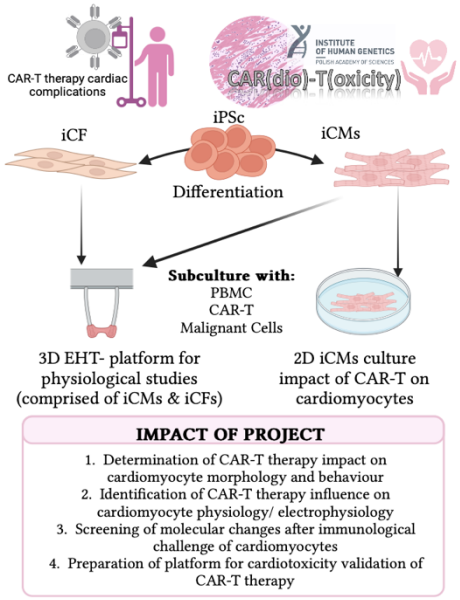
CAR(dio)-T(oxicity)
Engineered Heart Tissue model for CAR-T cardiotoxicity evaluation

Cardiac studies has always been considered as an Achilles' heel of science- mainly, because adult cardiomyocytes do not proliferate and it is technically difficult to continually culture harvested cardiomyocytes *in vitro*. Modelling of disease conditions using stem cells is one of the greatest achievements of the last decade. Thanks to the appropriate conditions created in the laboratory, scientist are able to obtain heart cells *in vitro*- from the patients stem cells. This creates enormous opportunities for cardiovascular research to track the consequences of applied therapies.

The aim of the project is to obtain cardiomyocytes and 3D engineered heart tissue constructs reflecting the electrophysiological properties of the heart. Preparation of such platform will allow to investigate the impact of CAR-T immunotherapy on the biology and physiology of cardiomyocytes- due to the fact that the use of CAR-T has main side effect - cardiotoxicity caused by cytokine storm (CRS).



IMPACT OF PROJECT

1. Determination of CAR-T therapy impact on cardiomyocyte morphology and behaviour
2. Identification of CAR-T therapy influence on cardiomyocyte physiology/ electrophysiology
3. Screening of molecular changes after immunological challenge of cardiomyocytes
4. Preparation of platform for cardiotoxicity validation of CAR-T therapy

At the beginning we plan to obtain induced pluripotent stem cells form PBMC population isolated from whole blood. The pluripotency allows us to differentiate the iPSc toward cardiomyocytes. Cardiomyocytes cultured *in vitro* in monolayer will be diligently examined in the aspects of CAR-T cardiotoxic influence on their morphology, basic biological functions and physiology including: size, organization of contractile apparatus, beating characteristics. Sadly, according to the literature, 2D cultures of cardiomyocytes are not mature enough to be reliable source of information for physiological studies. To get a better insight in the physiology of cardiomyocytes we will prepare Engineered Heart Tissue (EHT) system. EHT is a macro-sized model, with direct interaction and mechanical training that is applied to a mixture of cell types (including cardiomyocytes and cardiac fibroblasts), which enables precise pharmacological-grade, force-driven measurements of drug/therapy responses. In technical aspects - the EHT models allow to conduct precise translational studies in terms of force development, tissue stiffens/ elasticity, contraction and relaxation measurements to elucidate the impact of CAR-T therapy on cardiac muscle.

To be able to recreate all the components that are participating in the immunological challenge of cardiomyocytes we are planning to use not only a CAR-T cells but also a population of PBMC that contribute to CRS. This will ensure the complexity of proposed model.

To achieve the highest standard of the studies we will use cutting-edge methods like: confocal microscopy, atomic force microscopy and molecular analysis including high-throughput next-generation sequencing.

We believe that, this type of translational research may have a great socio-economic impact by progressing the science in cardiac field and by increasing the safety and effectiveness of CAR-T therapy for patients.