Blood vessels are made up of three layers: inner, middle and outer. The inner layer is composed of a single layer of endothelial cells coated with a glycocalyx. The location of endothelial cells (ECs) makes them the natural border between blood and tissue, and thus they are the first to respond to all physical and chemical stimuli. In response to inflammation, ECs can change the amount of biologically active substances secreted, regulate the transport of substances through the vessel wall, regulate the behaviour of smooth muscle cells and the tension of blood vessel walls. The inflammatory process of ECs is observed during the development of life-style diseases, e.g. atherosclerosis, hypertension, and type 2 diabetes. Nowadays, a lot of the work is aimed at seeking the prevention and treatment of life-style diseases, however, without an appropriate research methodology leading to an effective biomedicine of ECs in the blood vessel, it seems impossible.

In this project**, propose the development of a unique research methodology for the multi-parameter assessment of endothelial phenotype in isolated blood vessels.** The proposed methodology based on the synergy and complementarity of imaging techniques: Raman, fluorescence, and atomic force microscopy (AFM) supported by additional techniques such as liquid chromatography with tandem mass spectrometry (LC-MS/MS), or functional assays will contribute to a comprehensive understanding of the chemical, biological, nanostructural, and functional alterations occurring in the activated/dysfunctional endothelium in an isolated murine blood vessel, and their dependence on vasoprotective drugs (sodium-glucose co-transports 2 inhibitors (SGLT2-I, and others).

Sodium-glucose co-transports 2 inhibitors (SGLT2-I) are a new generation of drugs for diabetes (including empagliflozin or dapagliflozin), however, their spectrum of action goes far beyond the systemic glucose reduction, and theirs protective effect on the ECs is postulated, both in hyperglycaemic conditions and in inflammatory state of the vasculature. Since the mechanism of action of SGLT2-I remains elusive so far, in this project I propose the development of a unique methodology aimed to uncover alterations of chemical, biological, nanostructural, and functional properties of activated/dysfunctional ECs in isolated blood vessels, which brings us closer to unravelling the mystery of how SGLT2-I actually work on the vascular wall. This first stage of the project includes the application of the developed research methodology to characterize the effect of SGLT2-I on inflammation- or hyperglycemia-activated endothelium, while the second stage includes the application of the developed research methodology to reveal the mechanisms of action of SGLT2-I and to verify the therapeutic efficiency of SGLT2-I to reverse endothelial dysfunction.