Stroke is a grave neurological disorder resulting in the limitation of the blood flow in the brain, leading to an acute, global and focal, vascular brain disorders due to a death of brain tissue. Because of various clinical manifestations, stroke can be divided into hemorrhage stroke (HS) and ischemic stroke (IS), referring to 80-90% of all cases. According to data from WHO, strokes are the second, most common cause of death globally, and in the face of the progressive phenomenon of aging, the increasing number of patients in the near future can be expected. Furthermore, strokes are one of the main cause of disability. Neurological deficits lead to a disruption of the motor, intellectual and emotional processes, resulting in the significant reduction of life quality, which has not only medical dimension but social and economic either.

In the pathogenesis of IS, the major role is attributed to the progressing atherosclerotic process. Accumulation of fatty compounds in the arterial wall lead to an endothelium inflammation, and the secretion of inflammatory mediators responsible for recruitment of peripheral blood cells, lead to a deposition and rupture of the atherosclerotic plaque resulting in blood vessel blockage. An important role in this process is played by blood platelets, which due to interaction with endothelial cells mobilize leukocytes to migrate towards inflammatory endothelium affecting the enlargement and destabilization of atherosclerotic plaque.

The studies planned in this project are aimed to comparative analysis of both: platelet's protein profile and mRNA expression profile in the group of patients diagnosed with IS, in comparison to healthy volunteers without cardiovascular disorders. Results obtained from planned experiments are aimed to explain the molecular changes in blood platelets in two different pathways: proteomic and transcriptomic. Determining the differences overlapping from both analysis can contribute to identify the platelet molecular markers indicating human predisposition to the occurrence of IS. Searching for new, molecular markers is particularly important in the case of platelets, which despite the key role in IS have not been widely studied in this field. Proving the genetic basis of functional and structural changes in platelets, responsible for the occurrence of ischemic events, may expand the current state of knowledge on the molecular mechanisms underlying IS

The innovative character of this project lies in its twofold nature. Analysis of proteome and transcriptome of blood platelets will be conducted simultaneously, in order to determine the differences not only in protein levels, but also in the expression of correlated mRNA transcripts. This will eliminate the possible changes that could arise as a result of administrated antiplatelet pharmacotherapy, because these drugs will not affect the transcription process of unnucleated platelets. In the first stage of the research, a comparative analysis of the protein profile will be performed by 2-Dimensional Gel Electrophoresis in the patients with IS and in the control group. The identification of proteins showing different expression in both groups will be carried out by mass spectrometry. At the same time, a comparative analysis of the expression of the mRNA profile will be performed using the microarray technique. Validation of results for transcripts showing differences in both groups will be performed using the innovative technique – Droplet Digital PCR (ddPCR), which provides ultrasensitive quantification of nucleic acids, where each PCR reaction is divided into 20 000 single nano liter droplets. The obtained results will be compared to each other in order to determine the correlation between elevated protein levels and augmented expression of the mRNA transcripts. For molecules showing differences in both transcriptome and proteome analysis, quantitative study will be performed using the Bio-Plex immunoenzymatic method, and validation will be performed using Western Blot technique.