The aim of this project is to provide a new insight into the role of ROS generated during a single bout of maximal exercise by endothelial NOX2 and NOX4 enzymes (*Nicotinamide Adenine Dinucleotide Phosphate* (NADPH) *Oxidase*) in post-exercise oxidant stress and adaptation/maladaptation of endothelium, and in regulation of exercise capacity and post-exercise haemostatic/thrombotic parameters using unique animal models of endothelial-specific NOX knock-out (KO) mice.

Reactive oxygen species (ROS), as a by-products of metabolism resulting from of oxygen reduction are a large group of molecules, e.g.: superoxide anion ($^{\bullet}O_2^-$), peroxynitrite ($ONOO^-$) and hydrogen peroxide (H_2O_2). Overproduction of ROS substantially exceeding the ability of natural endogenous anti-oxidant mechanisms to eliminate them or their formation in non-physiological locations in the body is defined as oxidative stress. That may lead to the damage of cellular structures, proteins, lipids and DNA and has been involved in a wide range of pathologies. On the other hand, ROS play an important role as signaling molecules in regulation of physiological processes. There are several sources of ROS generation: NADPH oxidases (NOX), xanthine oxidase (XO), uncoupled nitric oxide synthase (NOS) or mitochondrial respiratory chain.

Physical exercise results in excessive generation of ROS. That ROS mediate health-promoting effects of regular physical activity by triggering many adaptive responses. However, overproduction of ROS may lead to impaired skeletal muscle contractility and muscular fatigue. Furthermore, $^{\bullet}O_2^-$ generated during exercise can directly inactivate nitric oxide (NO) produced by endothelial cells. Significantly reduced bioavailability of NO due to exercise-induced production of $^{\bullet}O_2^-$ may lead to transient endothelial dysfunction. Therefore, it is tempting to speculate that, in case of intense exercise, ROS are overproduced and may reduce exercise performance. However, human and animal studies do not fully confirm this idea because supplementation with antioxidants does not improve performance during a single bout of intense exercise. It is now known that the effects of ROS cannot be defined as beneficial or detrimental by merely measuring their overall formation but that their positive or negative role can also depend on the sites of their formation in the cells or tissues as well as on the type of ROS being formed. It seems that one of the major sources of ROS and oxidative stress during exercise is increased activity of NADPH oxidases in endothelial cells. Where NOX1, NOX2, NOX4 and NOX5 are functionally active. All of them generate $^{\bullet}O_2^-$ except NOX4 that, unlike other isoforms existing in endothelium, produces H_2O_2 rather than $^{\bullet}O_2^-$. Superoxide generated by NOX1, NOX2 and NOX5 in endothelial cells inactivates directly eNOS-derived NO and what results in endothelium dysfunction. In turn, NOX4 is incapable to inactivate NO and appears to have a protective role in cardiovascular system.

To achieve the aim of the proposed project wild-type control and endothelial-specific NOX2-KO and NOX4-KO mice will be subjected to a single bout of treadmill maximal exercise. To assess post-exercise oxidant stress, endothelial function and post-exercise haemostatic/thrombotic parameters blood, plasma and isolated aorta will be analyzed. Exercise capacity of mice will be assessed by comparison of distance, duration, maximal velocity and maximal oxygen consumption (VO'_{2max}).

The identification of beneficial or detrimental role of ROS generated in response to exercise depending on their enzymatic sources in endothelium may contribute to development of innovative pharmacological endothelium-targeted strategy which could improve exercise tolerance and protect against dangerous consequences of acute intense exercise in patients with endothelial dysfunction.