Breast cancer metastasis-induced endothelial-mesenchymal transition alongside ageing; implications for therapy

Breast cancer is one of the most common malignancies in Poland and its mortality rates increase with age. The reason of death is usually associated with formation of distant metastases in vital organs. Surprisingly, the elderly breast cancer patients also often die from cardiovascular diseases inevitably linked to impairment of endothelial function. Indeed, in the course of life a progressive decline in endothelial function is developed that is usually manifested with impairment of NO-dependent vascular relaxation and lower endothelial barrier integrity due to lower expression of endothelium-specific proteins: endothelial isoform of nitric oxide synthase (eNOS) and vascular endothelial cadherin (VE-CAD), respectively. Decreased expression of endothelium-specific proteins is, most probably, associated with progressive mesenchymal transformation of endothelial cells (EndMT). However, the progression of EndMT alongside ageing have not been characterised so far. The age-related decline in endothelial function might be associated with age-related alterations in platelet phenotype. Indeed, although circulating platelets are known mainly from their role in thrombus formation, the plethora of growth factors released from platelets of healthy individuals also continuously support endothelium well-being. This function of platelets was found to be especially important in the lungs that became leaky after experimental platelet depletion. Both ageing and cancer alter platelet phenotype and the age-/cancer-altered platelets may rather drive than prevent EndMT and disruption of endothelial barrier.

The hypothesis of the project has been formed on the basis of the presumption that EndMT is a cornerstone of age-related endothelial dysfunction and its advancement determines endothelium status of the individual as well as the outcome of the metastatic breast cancer. The aim of the project is to investigate EndMT progression in the systemic (aorta) and metastatic organ (lungs) of mice alongside ageing in the model of experimental breast cancer metastasis in association with age- and/or cancer-driven changes in platelet EndMT promoting/endothelium barrier disrupting function that will be verified in *in vitro* assays. Furthermore, proteomic analysis of age- and/or cancer-altered platelet releasates will allow to select the candidate protein(s) secreated from platelets that might drive EndMT/disrupt endothelial barrier integrity in ageing mice experiencing breast cancer metastasis. The role of the candidate protein(s) in EndMT progression/endothelial barrier disruption will be verified *in vivo* by knocking down their expression with specific morpholino-based oligonucleotides.

Implementation of the project will allow to verify the project hypothesis, in particular whether platelet-derived proteins promoting EndMT and disruption of endothelial barrier in the lungs could become therapeutic target(s) to prevent galloping progression of EndMT in ageing individuals experiencing breast cancer metastasis.