A robust cellular platform for studying of Epithelial-Mesenchymal Transition created by gene editing

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Epithelial-mesenchymal transition (EMT) and, its inverse process, mesenchymal-epithelial transition (MET), are physiological processes that also play important roles in disease. Mounting evidence correlates EMT with the poor outcome of multiple neoplasias, as well as metastasis and drug resistance. Thus, the molecular mechanisms that regulate EMT are an important area of cancer research and drug targeting. In order to study EMT, different exogenous reporter systems that can signal phenotypical changes are commonly used. Nonetheless, the use of exogenous reporters that are unable to describe molecular events, including the interference of alternative promoters, regulatory elements or epigenetic mechanisms that modulate promoter activation, making some data unreliable. Trying to overcome those limitations, we developed and validated a reporter cell line by modifying the endogenous VIMENTIN gene (a gene associated with EMT) using gene editing CRISPR/Cas9 technology. Specifically, we successfully knocked-in a DNA sequence, containing a self-cleaving peptide followed by a fluorescent protein in-frame with VIMENTIN into the genome of H2170 lung cancer cells. Our data uniquely illustrate that the Vimentin reporter cells are a reliable model for studying EMT and MET. The Vimentin reporter cells allow spatio-temporal observation of cellular plasticity with respect to their mesenchymal/epithelial in vitro and in the future in vivo. This is an excellent model to study the molecular mechanisms of EMT/MET as well as a robust platform to screen for new anti-cancer drugs.

Biography

Michał Kiełbus completed his Ph.D. thesis in 2016, was focused on finding the prognostic factors in brain cancer. As a post-doc he partially remained in the cancer field and his current project investigates the molecular basis of Epithelial-Mesenchymal Transition (EMT). He acquired experience in designing and conducting all the steps of well-planned functional experiments, starting from virtual cloning, genome editing and ending with different assays including e.g. cell imaging or gene expression assessment. His particular interests are, exploring the molecular mechanisms that change cell phenotype and result in the induction of a disease process.

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