Vimentin is a protein that builds a network of intermittent filaments in cells, which interconnects the actin filaments with the microtubule (MT) network forming a cell cytoskeleton. This protein network has numerous functions and one of these functions is maintaining the shape of cells and their mechanical properties. The amount of vimentin in the cytoskeleton of normal and diseased cells varies. Overexpression of vimentin was found in fibroblasts (FBs) in fibrotic lung tissue. This pathology is a common symptom for idiopathic pulmonary fibrosis (IPF), and cystic fibrosis (CF). The cause of IPF is not clear, however, it commonly develops in the lungs of elderly people, and CF is a genetic disorder. In both cases, patients have trouble breathing due to stiffening of the lung tissue, which reduces lung compliance and decreases the amount of inhaled air (oxygen). There is no cure for IPF, and the treatments aim to slow disease progression, manage symptoms, and improve quality of life. In the case of CF, treatment has seen breakthroughs in recent years, especially with CFTR (cystic fibrosis transmembrane conductance regulator) modulator therapies, however, symptom-directed therapies are still applied.

In the project, we plan to test three selected vimentin intermittent filaments (VIF) targeting molecules. We will investigate how the modulation of the VIF network influences the rheological properties of cells and modeled lung tissue. The lung tissue model will be formed by embedding normal and diseased fibroblasts in three-dimensional collagen hydrogels. The lung model will be exposed to compression and tensile stress mimicking compression and decompression of the tissue during breathing. Next, FBs, exposed to VIF targeting agents, will be introduced to the tissue model. Rheological properties will be measured with a rheometer. Simultaneously, collagen matrix-FBs interactions (cell migration, elongation, or COL fibers pulling) will be observed via an optical/fluorescence microscope integrated with the rheometer. We have selected three VIF targeting agents: Withaferin A (VIF disassembly), trametinib (spatial spreading), and ALD-R491 (VIF stabilization). Microrheological characterization of untreated and treated FBs will be performed with atomic force microscopy (AFM, stress-relaxation, and frequency sweep measurements). Also, AFM will be used to image cell topography and compare the adhesion of FBs to ECM proteins after drug treatment.

This project aims to deepen our understanding of the role of vimentin intermediate filaments (VIF) in regulating the rheological properties of fibroblasts and their ability to remodel the extracellular matrix (ECM) in three-dimensional models of pulmonary fibrosis. The outcomes of this project have the potential to develop new therapeutic strategies for IPF and CF based on VIF targeting agents.