Myosin VI as a new regulator of nuclear biomechanics and chromatin organization in skeletal muscle

Motor proteins (walking proteins), thanks to chemical energy conversion, can generate movement within a cell, as well as the movement of whole cells or their parts and, as a result, tissues, and organs. Key bodily functions such as signal transmission at synapses, fertilization, or cell division would not be possible without motor proteins. Contraction and relaxation of skeletal muscles allowing for movement and respiration in humans and animals take place due to the presence of contractile apparatus proteins - actin and belonging to motor proteins, actin-dependent myosins. The myosin superfamily comprises more than 30 classes of proteins, almost all of which move toward the growing end (plus end) of actin filaments. The exception is the unconventional myosin VI (MVI) that steps toward the shortening end (minus end) of actin filaments. Myosins are involved in a huge number of processes necessary for normal cell function, including adhesion, migration, differentiation, intracellular transport, cytoskeletal organization, transcription, chromatin remodeling, and cancer cell metastasis. New functions of myosins are still being discovered, and research of the nuclear functions of these proteins is expanding intensively. Our research group has shown that MVI regulates the adhesion, fusion, and differentiation of myoblasts and is found in the nuclei of muscle cells. In recent years, myosin VI has also been shown to exhibit various nuclear functions in other cell types. Preliminary results from our laboratory suggest that lack of MVI causes changes in the shape of skeletal muscle cell nuclei and may affect key regulators of nuclear function and DNA organization. Given the essential contribution of myosins to muscle physiology, this project focuses on the novel role of MVI in the organization and structural changes of skeletal muscle nuclei. The proper shape of nuclei and their resistance to muscle contraction and stretch is often disrupted in diseases caused by nuclear protein mutations. In this project, we will use a variety of research models to investigate the effects of the lack of MVI on the organization and function of myonuclei. We will use state-of-the-art super-resolution imaging methods and automated image analysis tools, electron microscopy and new generation sequencing for cross-sectional characterization of structural and molecular changes, occurring in the nuclei of muscle cells lacking MVI. We will investigate the effect of MVI on the myonuclei mechanosensing using atomic force microscopy. We expect that MVI will affect the structure of the nuclear envelope and the organization of genetic material in the nucleus, as they are tightly linked. In addition, we will verify how the absence of MVI in muscle cells affects gene expression and DNA organization and analyze the localization and the role of the identified proteins and their interactions with other protein partners. Based on the preliminary studies conducted by me and other researchers, I expect that the identified MVI-dependent genes will be involved, among others, in processes such as the transport and maturation of messenger RNA (mRNA), transcription, translation, and DNA damage repair. We will also conduct experiments to restore the function of MVI in muscle and study its effects on myonuclei. Using inhibitors, we will also verify whether MVI's interaction with actin and its motor functions are necessary for changes in the organization of myonuclei. This will allow us to determine whether the observed changes directly depend on MVI, which domains of MVI are crucial, and what molecular mechanism controlled by this protein is responsible for regulating the function of nuclei in muscle. The results of our innovative study will shed new light on the role of MVI in muscle physiology and may help identify the causes of nuclear dysfunction in muscle pathology and muscle diseases.