IDENTIFICATION OF AGE-RELATED TRACES OF AMYOTROPHIC LATERAL SCLEROSIS AS A BASIS FOR THE SEARCH FOR NOVEL DISEASE MODIFIERS

We live in the fast-growing aging society where already now nearly 10% of people is over 65 years old and this ratio is predicted to almost double by 2050. As life expectancy of the population increases, the prevalence of aging-associated disorders raises exponentially. This includes devastating neurodegenerative diseases for which aging is a major risk factor. Regrettably, despite having escalating affliction on society and representing a crucial and exponentially increasing challenge to health care systems throughout the world, neurodegenerative diseases remain largely uncurable.

Among neurodegenerative diseases, Amyotrophic Lateral Sclerosis (ALS) is the most prevalent motor neuron disease, characterized by the progressive degeneration of motor neurons in the brain and the spinal cord. Clinically, following focal weakness of voluntary skeletal muscles, the disease spreads fast, eventually leading to paralysis and death by respiratory failure. The condition is usually diagnosed between 51 and 66 years of age and mostly presents an aggressive nature, leading to death within 3–5 years after the disease onset. A clear genetic history exists in about 5 to 10 percent of cases, with *C9orf72*, *SOD1*, *TARDBP* and *FUS* being the most common genetic risk factors identified.

As for other neurodegenerative diseases, effective therapies against ALS are far from realization. In order to change that, the deep understanding of the underlying neuropathology is necessary, which will be possible only in valid disease models.

The goal of this project is the characterization of new human models of ALS, which will allow studies of aging-relevant signatures of the disease. Using this new platform, we will search for novel modifiers of cellular pathology related to ALS.

Identification of novel therapeutic strategies in neurodegeneration has been hampered by the insufficient translatability of available models. Animal models, although provided a crucial insight across the range of disease mechanism differ significantly in their genetics as compared to humans, which limits translatability. These limitations can now be overcome, at least to some extent, by using patient derived iPSC-based models that enable investigation of pathological mechanisms in the context of human genetics. However, induction of pluripotency resets the aging hallmarks of donor cells which cannot be reverted after differentiation, which allows monitoring of only earliest molecular pathogenic events in iPSC-based models. As it turned out, the direct cell reprogramming where somatic cells are converted into neurons with omitting the pluripotent state well retains multiple age-associated signatures stored in donor cells. Such models, developed using a novel state-of-the-art conversion method for Huntington's disease, tauopathies and Alzheimer's disease, were able to recapitulate unique aging-relevant disease signatures, difficult to be recovered in iPSC-based models. This strategy was not yet applied to model ALS.

Our aim is to implement the direct conversion method to produce age-retaining motor neurons from ALS patients' fibroblasts. Cells will be next extensively characterized to assess recapitulated disease phenotypes. Following novel models validation, the proteomic profiles of ALS-motor neurons will be determined using advanced mass spectrometry based method. Based on the results of the proteomic analysis, we will select candidates that will be next functionally validated as potential disease modifiers. Finally, we will assess the contribution of astrocytes to the neuronal pathology.

Described study plan proposes the validation of new ALS models retaining the human genetic and epigenetic background of the disease. Proteomic profiling of reprogrammed cells will reveal aging-specific signatures of the disease. Based on these analyses we envisage to discover factors with a potential to be further explored for therapeutic purposes in ALS.