

## Abstract

Neurodegenerative diseases are a wide group of disorders affecting the central (CNS) and peripheral nervous system. Despite enormous efforts of the scientific and medical communities, in many cases, the etiologies of neurodegenerative diseases remain unknown with no effective treatment available.

Recently, glial cells have entered the spotlight as important players in disease onset and progression, proving themselves as potentially interesting therapeutic targets. In healthy conditions, they maintain homeostasis in the CNS. However, upon its disturbance, they acquire reactive states, which affect the pathological processes in a positive or a negative manner. The reactive states are disease-specific, yet there are similarities linking reactive states of glia across pathologies, pointing to common mechanisms of coping with the disturbance of homeostasis in the CNS.

Single-cell RNA sequencing (scRNA-seq) belongs to modern high-throughput methods that enable studying transcriptional profiles of thousands of individual cells. Therefore, it can provide information on the representation of cell types across a variety of samples and reveal even small populations with unique, biologically interesting transcriptional signatures.

In this work, we applied scRNA-seq to investigate cell populations and transcriptional profiles in two neurodegenerative disorders with quite distinct etiologies – amyotrophic lateral sclerosis (ALS) and Alexander disease (AxD). ALS represents a neurodegenerative disease with a multifactorial cause. In this case, we used a well-established mouse model carrying a SOD1(G93A) mutation that causes only a minority of ALS cases. Addressing a controversial question of the extent to which the cortex is affected in the SOD1(G93A) mouse model, we found only limited signs of pathology in the SOD1 cortices. Thus, our in-depth transcriptomic analysis showed that this well-studied animal model does not recapitulate all aspects of human pathology, and another model should be used for the investigation of the cortex in ALS.

In contrast to ALS, Alexander disease belongs to rare diseases with well-defined and specific cause – mutations in the intermediate filament protein GFAP, which is primarily expressed by astrocytes and radial glia in the developing CNS. In this project, we focused on human induced pluripotent stem cell-derived (hiPSC) models in 2D co-culture as well as in brain organoids. We described these models using transcriptional profiling and found an interesting, early differentiation phenotype resulting in a reduction of mature astrocyte populations and overrepresentation of cells from other than neuroectodermal lineages. Thus, we revealed a previously unknown effects of a GFAP mutation on cell type differentiation, offering new insights into AxD pathogenesis that should be considered in the future research on this and related disorders.

Overall, the presented research projects demonstrate the usage of state-of-the-art transcriptomic methods to describe neurodegenerative diseases in high resolution, revealing information potentially important for future research on neurodegeneration with implications for the design of novel treatment strategies.