

PROTEOMIC ANALYSIS OF NEUROSPHERES FORMED BY PATIENTS-DERIVED NEURAL STEM CELLS

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Introduction: Tridimensional models of human cells for in vitro research have been gaining relevance. These models offer significant advantages over bidimensional models, such as the simulation of cell-cell and cell-matrix interactions, microenvironments and the tridimensional nature itself of in vivo tissues. These factors influence cell behaviour and characteristics and are indispensable for a more realistic modeling of the in vivo environment. Patients-derived neurospheres are even capable of carrying the specific genetic features of an individual and are promising for the personalized medicine. **Objectives:** Characterize the proteome of patients-derived neurospheres and compare it with that of bidimensionally cultured neural stem cells. **Methods:** In this study, we utilized two sample groups: neural stem cells (NSCs) derived from patients' induced pluripotent stem cells and neurospheres formed by those same NSCs. The samples were lysed and the proteins were extracted via centrifugation. Proteins were reduced with dithiothreitol, alkylated with iodoacetamide and digested with trypsin. Resulting peptides were labeled with TMT 10plex, with each channel representing a unique sample (5 from NSCs and 5 from neurospheres). After labeling, samples were combined in a single tube, which was fractionated using HPLC. Fractions were analysed by mass spectrometry. Spectral data was processed with MaxQuant software. Statistical analyses were conducted with Perseus software, including Student's T test to identify the differentially expressed proteins. These proteins were further analysed by GO enrichment. **Partial results:** Analysis of the TMT validation revealed that neurospheres had 75% more neural-related enriched processes, in comparison with NSCs. **Perspectives:** We are currently analysing the remaining of the samples, since partial results are derived from the TMT validation. We aim to increase the number of identifications to provide more material for analysis, allowing us to draw conclusions regarding the differences between the two groups.

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