

ABSTRACT

Charles University, Faculty of Pharmacy in Hradec Králové

Department of Biological and Medical Sciences

Student: Bc. Eliška Hrachovcová

Supervisor: PharmDr. Jana Rathouská, Ph.D.

Consultant: doc. RNDr. Pavlína Kušnierová, Ph.D.

Title of diploma thesis: Laboratory diagnostics of microRNAs in different diagnostic groups

Background: Identification of potential miRNA targets for differential diagnosis of patients with atherosclerosis and dementia. Comparison of miRNA expression and uptake in serum samples with cerebrospinal fluid samples. Selection of appropriate miRNAs for differential diagnosis in patients with atherosclerosis and dementia. Furthermore, the selection of miRNA targets with the most significant fold-change and also the lowest p-value for subsequent validation studies.

Methods: To identify potential miRNAs, the received samples were centrifuged, aliquoted and stored at - 80°C. miRNAs were isolated using the iCatcher Circulating cfRNA 1000 kit and the CatchGene isolator. Reverse transcription and amplification with detection were then performed. Data were processed in Excel and then statistically processed in GenEx using Mann-Whitney test.

Results: The results were evaluated for 2 groups of patients: group A (samples of patients with Parkinson's disease and dementia) and group B (with atherosclerosis and dementia). Purity and concentration in serum and liquor samples were assessed. The most appropriate endogenous controls for both serum and liquor were evaluated. A total of 6 common miRNA targets were found for both groups. In patients, some miRNAs were statistically significantly upregulated or downregulated, specifically hsa-miR-23a-3p-TT-PRI, hsa-miR-30b-5p-TT-PRI, hsa-miR-142a-5p-TT-PRI, hsa-miR-146a-5p-TT-PRI, hsa-miR-29c-3p-TT-PRI, hsa-miR-151a-3p-TT-PRI.

Conclusions: The results obtained in this work point to miRNAs that could be a part of differential diagnosis in the future. We can already say with confidence that these miRNAs represent a major step towards early diagnosis of these diseases. The results of this thesis could serve as a starting point for future research and development in the diagnosis of atherosclerosis and dementia.