Abstract

ADP-ribosylation is a crucial post-translational modification that regulates various cellular processes, including DNA repair. It is catalysed by poly-ADP-ribose polymerases (PARPs) and involves the transfer of ADP-ribose moieties from the redox cofactor NAD^+ to proteins, including histones. To maintain cellular homeostasis, ADP-ribose chains need to be rapidly degraded by ADP-ribosyl glycohydrolases. While poly-ADP-ribose glycohydrolase (PARG) is highly efficient, it cannot cleave the terminal ADP-ribose moiety. For the removal of the terminal mono-ADP-ribose, two glycohydrolases, TARG1 and ARH3, are involved. This removal process is necessary because it enables DNA repair factors to access the site of DNA damage. The primary goal of this thesis is to characterise cells derived from patients with homozygous ARH3 mutations and to develop appropriate tools to improve our understanding of the molecular mechanism by which ARH3 mutations affect ADP-ribosylation and how it contributes to the onset of the associated neurological disease. To achieve this, I measured the levels of ARH3 protein and detected increased mono-ADP-ribosylation in *ARH3*-mutated patient-derived fibroblasts. Furthermore, I assessed the sensitivity of these cells to different PARP inhibitors, which hold potential for the therapeutic treatment. To gain a deeper insight into how exactly dysregulated ADP-ribosylation affects brain architecture and development, it is crucial to establish a more relevant study model. Therefore, I generated induced pluripotent stem cells (iPSCs) from patient-derived fibroblasts, confirmed their pluripotency and measured ADP-ribosylation. Moreover, I used generated iPSCs for the cultivation of cerebral organoids. The generation of patient-derived iPSCs and cerebral organoids will provide a valuable model system for studying the effects of dysregulated ADP-ribosylation on brain architecture and development and offer new insights into the pathogenesis of neurological disorders.

Key words: ARH3, SSB, SSBR, neurodegeneration, ADP-ribosylation, DNA damage, iPSCs, organoids.