The UHMK1 kinase regulates splicing of DNA damage response genes in melanoma cells

Lisa Latzko1, Isabela David Cardoso2, Vanessa C Arfelli1, Sebastian Vorsberg1, Philipp A Greif1, Leticia Fröhlich Archangelo2 1Department of Medicine III, University Hospital, LMU, German Cancer Consortium (DKTK), Munich, Germany, 2Ribeirao Preto Medical School - University of Sao Paulo (FMRP-USP), Ribeirao Preto, Brazil

Background: Aberrant alternative splicing contributes to transcriptional reprogramming during melanoma development. We previously demonstrated that UHMK1 kinase regulates the phosphorylation of multiple splicing factors and that altered UHMK1 expression affects the splicing of hundreds of transcripts. UHMK1 is highly expressed in primary melanoma tumors and metastases, suggesting that it may promote malignant phenotypes through dysregulated splicing of oncogenic pathways. Method: We used nanopore long-read sequencing to investigate the effects of UHMK1 overexpression (OE) and knockout (KO) on alternative splicing in two melanoma cell lines, A375 and WM793. In total, 79 samples were analyzed, including wild-type and kinase-dead UHMK1 OE compared with empty-vector controls, as well as four independent UHMK1 KOs per cell line compared with native controls. Differential splicing was defined as an adjusted p-value < 0.05 and |ΔPSI| > 0.1. Results: Across all conditions and cell lines, 700 unique differential splicing events were identified. In WM793 cells, splicing changes were limited, with four candidates detected following UHMK1 KO and three following UHMK1 OE. In A375 cells, five candidates were identified after UHMK1 KO, whereas 95 events were significant following UHMK1 OE. The top 10 OE-associated events were selected based on |ΔPSI|. The strongest alteration occurred in PARP2 (mean ΔPSI = 0.82), involving an alternative 5' splice site. Other top candidates included ERCC1 and CLK1, also associated with DNA damage response, while PCLAF, a regulator of DNA replication-associated damage response, was identified among the A375 KO candidates. Conclusion: Modulation of UHMK1 expression induces alternative splicing changes in melanoma affecting genes involved in DNA damage response and replication. Functional characterization of these isoforms will determine their contribution to melanoma progression and their potential as therapeutic targets