Abstract (EN)

Since their initial discovery in *C. elegans* and human cells more than 30 years ago, proteins from the Bcl-2 family were intrinsically linked with the induction, regulation and suppression of the mitochondrial branch of apoptotic signaling in mammalian cells but they also appear to modulate non-apoptotic signaling pathways. In this study we aimed, through comprehensive analysis, to enhance current knowledge of non-apoptotic roles of the main pro-apoptotic Bcl-2 family proteins BAX and BAK, with a specific focus on their role in cellular metabolism. We employed the CRISPR/Cas9 technique to generate Bax/Bak-deficient cancer cells of diverse tissue origins and assed impact of combined Bax and Bak deficiency on mitochondrial respiration and cellular glycolysis. While the ablation of Bax and Bak expression had no discernible effect on glycolysis across all tested cell lines, it modulated mitochondrial respiration in cell-type-specific manner. Elimination of Bax and Bak expression in HCT-116 colorectal cancer cells had no impact on mitochondrial respiration, but it largely affected mitochondrial respiration in Bax/Bak-deficient glioblastoma (U87) and lymphoma (HBL-2, UPF1H, UPF1G) cells. Bax/Bak-deficient U87 cells notably upregulated mitochondrial respiration as well as accelerated their proliferation and tumor growth in NSG mice. In contrast, Bax/Bak-deficient HBL-2 cells showed attenuated respiration and slower growth both in vitro and in vivo. The alterations in respiration were in Bax/Bak-deficient U87 cells accompanied by changes in metabolic pattern, such as an increase in NAD+/NADH ratio, increased mitochondrial membrane potential (MMP), and higher production of ATP. Subsequent analyses revealed upregulation/downregulation of mitochondria-encoded subunits of the respiratory chain complexes and stabilization/destabilization of the mitochondrial transcription elongation factor TEFM in Bax/Bak-deficient U87 and HBL-2 cells, respectively.

Downregulation of TEFM expression using shRNAs resulted in attenuated mitochondrial respiration not only in Bax/Bak-deficient U87 cells but also in parental HBL-2 cells. Our findings revealed that (post)translational regulation of TEFM protein levels in Bax/Bak-deficient cells modulates the expression levels of mitochondrial respiratory complex subunits, thereby influencing cellular respiration, metabolism, and proliferation dynamics. In collaborative projects, we participated in the assessment of the role Bcl-2 family proteins and mitochondrial metabolism in hematopoietic malignancies uncovering their response and resistances to therapeutically relevant BH3 mimetics.