Abstract:

Mitochondrial DNA (mtDNA) changes occur more frequently in the cell than alterations in nuclear DNA (nDNA) due to factors such as proximity to reactive oxygen species (ROS) or fewer DNA repair pathways. While nDNA damage is known to play a role in colorectal cancer (CRC), there is a lack of studies concerning mtDNA damage. Changes in mtDNA copy number, a possible indirect marker of mtDNA damage known as mtDNA content, have been reported in CRC with conflicting results. Various changes in mtDNA have been observed in multiple cancer types and proposed as potential biomarkers, including CRC; however, its exact role in disease progression, patients' prognosis, or prediction of treatment is yet to be determined.

The primary hypothesis of this study was that mitochondrial dysfunctions resulting from mtDNA changes play a role in colorectal carcinogenesis and could serve as potential CRC biomarkers. To test this hypothesis, we measured mtDNA damage, mtDNA content, and the expression of selected DNA repair genes in both tumor and adjacent non-malignant mucosa. Initially, we conducted a Pilot study involving 7 patients, utilizing RNA sequencing and qPCR. Subsequently, a Validation study was performed on a larger cohort of 50 patients using qPCR.

Our findings revealed that adjacent mucosa exhibited higher levels of mtDNA damage compared to tumor tissue in both the Pilot set (10.267 ± 0.40 and 7.435 ± 0.43, respectively, $p = 0.047$) and the Validation set (10.549 ± 0.97 and 7.975 ± 0.79, respectively, $p \leq 0.0001$). Moreover, most of the DNA repair genes correlating with mtDNA damage showed higher expression in tumor tissue ($p \leq 0.0001$). We identified a significant association between low expression of the *BRCA1* gene in tumors ($p = 0.029$) and improved patient survival. However, mtDNA content did not significantly differ between the tumor and adjacent mucosa in either the Pilot set (6.858 ± 1.00 and 6.235 ± 0.61, respectively, $p = 0.071$) or the Validation set (7.125 ± 1.02 and 7.072 ± 0.62, respectively, $p = 0.728$).

In summary, our results suggest that tumors exhibit less mtDNA damage than adjacent mucosa, possibly due to the upregulation of DNA repair genes in tumors facilitating efficient mtDNA damage repair. This underscores the role of mtDNA changes in CRC. Further research is warranted to elucidate whether these changes are causal or consequential in CRC development.