EPIGENETIC DRUG DECITABINE EFFICIENTLY REACTIVATES METHYLATION-SILENCED GENES IN HUMAN PANCREATIC DUCTAL ADENOCARCINOMA CELL LINES

Maria Urbanova[1], Verona Buocikova[1], Eliska Svastova[2], Jozef Bizik[1], Sona Ciernikova[1], Bozena Smolkova[1]

[1]Department of Molecular Oncology, Cancer Research Institute, Biomedical Research Center, Slovak Academy of Sciences, Dubravska Cesta 9, 845 05 Bratislava, Slovakia
[2]Department of Tumor Biology, Institute of Virology, Biomedical Research Center, Slovak Academy of Sciences, Dubravska Cesta 9, 845 05 Bratislava, Slovakia

Introduction: Epigenetic regulation is one of the mechanisms driving epithelial-to-mesenchymal transition (EMT), responsible for aggressiveness and dismal prognosis of pancreatic ductal adenocarcinoma (PDAC). The hypomethylation drug decitabine (DAC) is a deoxycytidine analog approved to treat hematologic malignancies. However, the success of DAC used by default as a cytotoxic agent for solid tumors treatment has been limited in clinical trials due to unwanted toxicity. Recently, epigenetic drugs have demonstrated the potential to modulate cancer cells' sensitivity to other forms of anticancer therapy. Therefore, we hypothesize that they could help overcome PDAC resistance to treatment, the major obstacle in the clinical management of this deadly disease.

Aims: This study aims to clarify the role of DNA methylation in regulating the expression of selected genes associated with PDAC progression *in vitro*.

Materials and Methods: Four PDAC cell lines (BxPC-3, MIA PaCa-2, PANC-1, and SU.86.86) cultivated in 2D conditions were exposed to subcytotoxic concentrations (IC_{20}) of DAC. Promoter DNA methylation of 15 EMT-associated genes was assessed before and for highly methylated genes after exposure by quantitative pyrosequencing method. In addition, DNA methylation levels were correlated with relevant gene expression changes evaluated by quantitative PCR.

Results: DAC efficiently decreased DNA methylation in all highly methylated genes. DNA methylation changes correlated inversely with gene expression. However, upregulation was also found in the genes with low methylation levels, whose methylation did not change significantly, suggesting a possible indirect DAC effect. Importantly, DAC reactivated silenced gene expression in all cell lines, mainly in MIA PaCa-2 and PANC-1, characterized by frequent hypermethylation of most studied genes.

Conclusion: These results confirm the critical role of DNA methylation in gene expression regulation of several genes involved in PDAC progression and thus uncover new possibilities for combination therapy. However, further research is needed to evaluate the efficacy and safety of this approach.

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