The NF–Y splicing signature controls hybrid EMT and ECM-related pathways to promote aggressiveness of colon cancer

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The NF-Y-binding site is one of the most representative regulatory elements in the promoters of eukaryotic genes. NF-YA, the DNA binding subunit of the NF-Y transcription factor, undergoes alternative splicing producing two isoforms, NF-YAs and NF-YAl that differ in the transactivation domain. The expression of the splicing variants of NF-YA is deregulated in many cancers compared to healthy tissues. Consistently, a different ratio in NF-YAs/NF-YAl transcripts is observed throughout different tumor subtypes.

Colorectal cancer (CRC) is the third most common type among tumors. Based on comprehensive gene expression profiles, CRC is classified into 4 consensus molecular subtypes (CMSs). In this study, we demonstrated that the lower level of NF-YAs/NF-YAl ratio is associated with the mesenchymal CMS4 subtype, characterized by poor response to therapies and the worst prognosis compared to other CMSs. Moreover, high NF-YAl expression predicts lower patients' survival. Using in vitro cellular models, we determined the predominant expression of NF-YAs in all CRC cell lines, with NF-YAl level increasing from epithelial to mesenchymal cells. NF-YA isoforms oppositely regulate the transcription of the E-cadherin gene, through direct binding to its promoter. Compared to NF-YAs^{high}, NF-YAl^{high} cells show alterations of genes associated with extracellular matrix and epithelial-mesenchymal transition. 2D and 3D assays identified different migratory and invasive behavior of transduced cells: NF-YAl^{high} cells are characterized by single cell ameboid-like migration and form irregular spheroids with low levels of cell-to-cell adhesion, consistently with low E-Cadherin expression. We further confirmed the increased metastatic potential of NF-YAl^{high} CRC cells by injection of transduced cells in the zebrafish xenograft model.

Altogether, our data highlight the direct role of the NF-YAl in cell dissemination and aggressiveness of CRC. NF-YAl represents a new CRC prognostic factor and splice-switching strategies may reduce metastatic CRC progression.