Integrative analysis of the genomic and transcriptomic landscape identifies novel key genes as a therapeutic target in bile duct cancer

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BACKGROUND

Bile duct cancer (BDC) is a malignancy thought to be derived from cholangiocytes, the epithelial cells lining the biliary tree. BDC is a highly aggressive tumour whose incidence has been increasing worldwide over the past two decades, now accounting for 10–15% of all hepato-biliary malignancies. The mechanisms underlying cholangiocyte malignant transformation and BDC progression is still not completely understood. Genomic profiling can offer a clearer understanding of their carcinogenesis, classification and treatment strategy. We performed large-scale genome sequencing analyses on BDCs to identify novel key-genes driving BDC and drug-resistance.

METHODS

We analyzed 100 BDC samples from an Italian Cohort, 40 by whole-exome sequencing, 80 by RNA sequencing, and a further 30 samples by SmallRNA sequencing. By using a bio-informatic pipeline, we integrated somatic mutation patterns and epigenetic features defined at the spatial level to identify novel target genes in the tumour microenvironment.

RESULTS

A total of 3392 and 6315 DEGs (Differentially expressed genes) were respectively observed in BDC comparing tumour (T), normal (N) and stromal (ST) areas with the criterion of false discovery rate <0.05. In top-ranked differentially regulated gene sets, we identified primary cilium-associated genes (PC). OFD1, CNGB1, AURKA, CENPF, STIL, STK39, RAB23 and OSR1 were found based on the criteria of fold change >2.5 and P<0.01. We started also to clarify at molecular level the role of PC in BDC pathogenesis and progression. A therapeutic approach targeting OFD1 in BDC cells was also investigated.

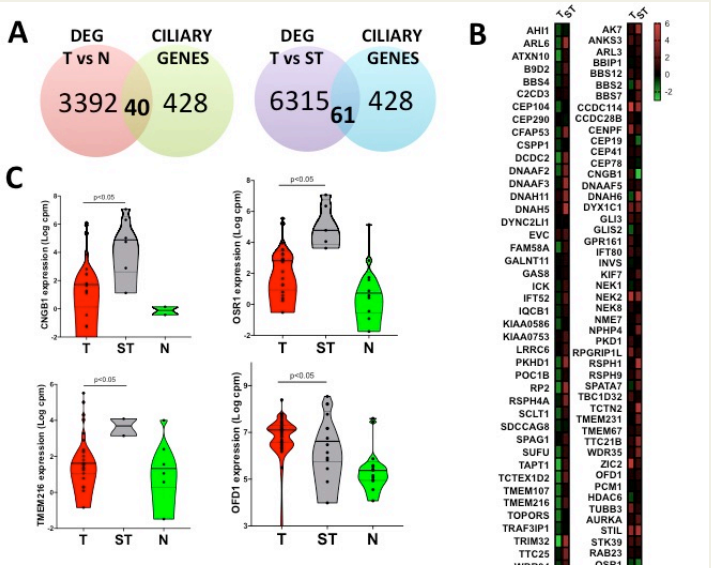

Figure 1. *Identification of mRNAs associated with BDC using next-generation RNA sequencing from laser micro-dissected archival FFPE tissue specimens*

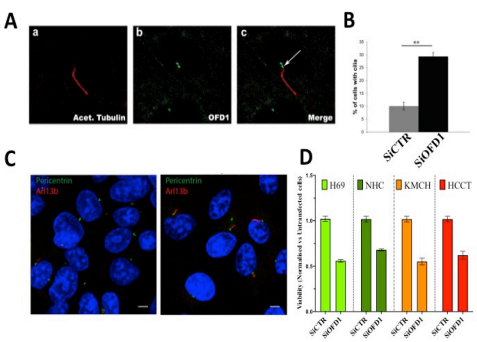

Figure 2. *OFD1 depletion in cholangiocyte and BDC cells induces ciliogenesis and suppresses tumour growth in vitro. A) IF analysis of OFD1 and Acetylated-Tubulin expression. B-C) Effect of OFD1 KD on cilia formation. D) Effect of OFD1 KD on viability of cholangiocyte (H69, NHC) and BDC (KMCH, HCCT) cells.*

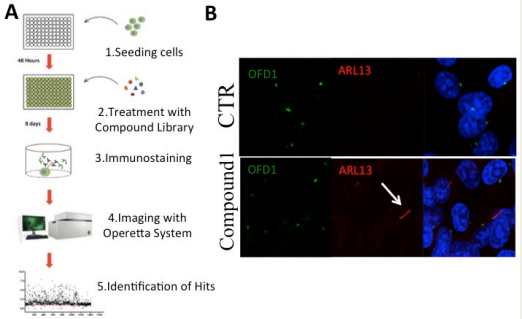

Figure 3. *A) Schematic representation of the screening strategy of the HCS using the human H69 cell line model. B) Representative confocal images of cilium marker acetylated ARL13 of H69 cells subjected to serum starvation and 24 h treatment with vehicle CTR and novel identified compound targeting ciliogenesis.*

CONCLUSIONS

Loss of PC is frequently observed in BDC, suggesting that the absence of this organelle may promote tumorigenesis through aberrant signal transduction and the inability to exit the cell cycle. We investigated the molecular mechanisms underlying the cilia loss and test whether may be potential therapeutic target. These findings could be useful to establish treatment and diagnostic strategies for BDCs based on genetic information.

CONTACTS

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