# Lack of L1CAM increases tumorigenicity, stemness and tumor fibrosis in pancreatic ductal adenocarcinoma

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Pancreatic ductal adenocarcinoma (PDAC) is a devastating disease with a 5-year survival rate of <8%, characterized by extensive fibrosis, which causes chemotherapy failure and tumor progression. Pancreatic stellate cells (PSCs) are the major cellular stromal component in PDAC, which contributes to extracellular matrix (ECM) deposition and tumor development.

L1 cell adhesion molecule (L1CAM) expression is generally associated with metastases and poor prognosis in different human tumors, while its role in PDAC remains debated.

We found that the majority of PDAC biopsies have reduced L1 expression that involves a more aggressive phenotype. We also found, *in vitro*, that the L1low cells had enhanced CSC features, including self-renewal, migration and chemoresistance. *In vivo* the L1low cells display augmented tumorigenic potential and were able to recapitulate the tumor heterogeneity compared to their L1high counterpart. Conversely, ectopic overexpression of L1 resulted in a consistent decrease in cell proliferation and reduction in the stem properties of tumor cells. This data indicates that restoration of L1 expression could counteracts the malignant behavior of the tumor cells, thus leading to a less aggressive phenotype.

Mechanistically we found that the TGF-β1 secreted by the PSC increased *in vitro* the migratory potential and the resistance to gemcitabine of the PDAC cells through the reduction of L1 expression. PSC silenced for TGF-β1 were not able anymore to modulate the L1 expression, and *in vivo* when co-injected with PDAC cells do not sustain tumor formation as observed in the PSC control.

The transcriptome profiling (by mRNA sequencing) of L1low cells revealed pathways associated with collagen secretion and degradation, suggesting that the absence of L1 in cancer cells concurs to the remodeling of the surrounding microenvironment. *In vitro* experiments with cancer cells knocked down for *L1* showed an increase in collagen deposition, which contribute to enhance extracellular matrix (ECM) stiffness and tumor fibrosis.

Interestingly, we demonstrated in co-culture experiments that the L1low PDAC cells stimulate the collagen production in PSC. By contrary, the overexpression of L1 has no significant effects on collagen production by PSC cells.

Altogether these data demonstrate an intrigued cross-talk between PSC and PDAC cells, in which the PSCs represent a supportive niche for PDAC cells promoting their aggressiveness and stemness through the downregulation of L1CAM mediated by TGF-β1 and the secretion of collagen.