PROTEIN X is a Super-Enhancer Tagged Human Cancer Gene and a Novel Therapeutic Target in Pancreatic Cancer

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Disclosures

Kathryn Austgen PhD, David Orlando PhD, John Carulli PhD, Chris Fiore PhD, Brian Johnston MS, Sofija Miljovska MS, Cindy Collins MS, and Tracey Lodie PhD are current or former employees of Syros Pharmaceuticals, Inc. and may have an equity interest in the company.
Background

Super-Enhancers

- Large non-coding regions made up of clusters of transcription regulators
- Drive expression of genes that
  - define cell type differentiation
  - affect immune cell functional states
- Play a role in regulation of tumor promoting or inhibiting genes

PROTEIN X

- Regulates actin cytoskeleton dynamics
- Shown to play a role in epithelial to mesenchymal transition and cancer progression in gastric and breast cancer (not yet studied in pancreatic cancer)
- Functions in axon guidance, synaptogenesis, and other morphologic cell changes
  - Axon guidance is a major area of gene dysregulation in pancreatic cancer
- Highly differentially SE-tagged between normal pancreas and PDAC tissue

Hypothesis:
- Down-regulation of PROTEIN X will affect basic cellular functions, leading to decreased proliferation and migration of human pancreatic cancer cells
Super-Enhancer Analysis

- Human normal pancreas and primary PDAC tumors dissociated into single-cell suspension
- ChipSeq used to identify SE regions (rich in H3K27 acetylation)
- SE regions linked to the specific genes they regulate

Investigation of PROTEIN X

- AsPC1 (human pancreatic cancer cell line with high baseline PROTEIN X expression) PROTEIN X knockdown line created using Lenti shRNA
- Small molecule inhibitor of PROTEIN X, non-toxic at high concentrations/doses
- Colony formation assays were used to assess cell proliferation
- Wound healing assays were used to assess cell migration
Results: Primary PDAC samples have unique epigenetic landscapes compared to Normal Pancreas

More acetylated in PDAC than Normal

Define “Pancreas”

More acetylated in Normal than PDAC
Results: PROTEIN X controls cell proliferation and migration in AsPC1 pancreatic cancer cells

Colony formation assay demonstrates decreased cell proliferation associated with PROTEIN X inhibition or knockdown (p < 0.005).

Wound healing assay demonstrates decreased cell migration associated with PROTEIN X inhibition or knockdown (p < 0.001).
Conclusions

• Super-enhancer analysis can define key genes that may drive pancreatic cancer oncogenesis

• PROTEIN X is a strongly super-enhancer-tagged gene in pancreatic cancer

• PROTEIN X plays a role in regulation of basic cellular functions, including cell proliferation and cell migration

• PROTEIN X represents a novel potential therapeutic target in pancreatic cancer with implications in cancer progression and metastases. Further validation is ongoing
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