Exploiting Myeloid-Stromal IL1RAP as a Therapeutic Vulnerability to Improve Chemoimmunotherapy Sensitivity in Pancreatic Cancer

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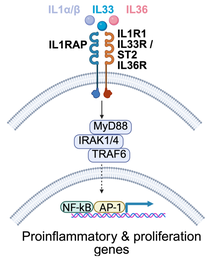
Background

- Pancreatic ductal adenocarcinoma (PDAC) remains highly resistant to chemotherapy and immunotherapy due to a tumor microenvironment (TME) dominated by myeloid-stromal immunosuppressive circuitries and T-cell dysfunction.
- Interleukin-1 (IL-1) family cytokines (IL-1α/β, IL-33, IL-36) converge on **IL-1 receptor accessory protein (IL1RAP)** signaling to reinforce a tolerogenic, pro-inflammatory TME which mediates chemoimmunoresistance.



Hypothesis: IL1RAP-expressing myeloid-stromal compartments sustain a therapeutic barrier in PDAC; disrupting this via pharmacologic IL1RAP inhibition could reprogram the TME to enhance chemoimmunotherapy efficacy.

Methods

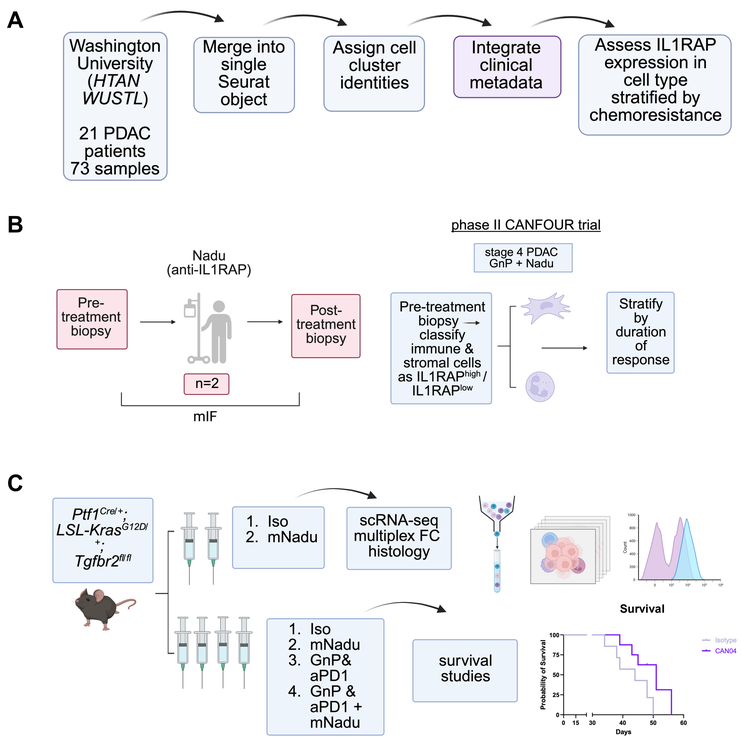


Figure 1: A. Workflow depicting curation of Seurat object from human PDAC samples with integration of metadata to assess IL1RAP expression stratified by chemoresistance. **B.** Schematic of monotherapy regime where pre-and post-treatment biopsies were examined by multiplex IF (n=2) (*left*) and diagram of stage II CANFOUR trial where pre-treatment tumor biopsies were collected and stratified by IL1RAP expression ("high" vs "low"). Patients then received combination therapy with gemcitabine + nab-paclitaxel (GnP) plus nadunolimab (Nadu). Duration of response was assessed, and pre-treatment IL1RAP tumor levels were correlated with overall survival. **C.** Diagram for treatment of PKT mice with mNadu monotherapy/combination therapy and downstream analyses.

① High IL1RAP expression in myeloid & stromal compartments is associated with increased duration of response following Nadu + GnP in PDAC

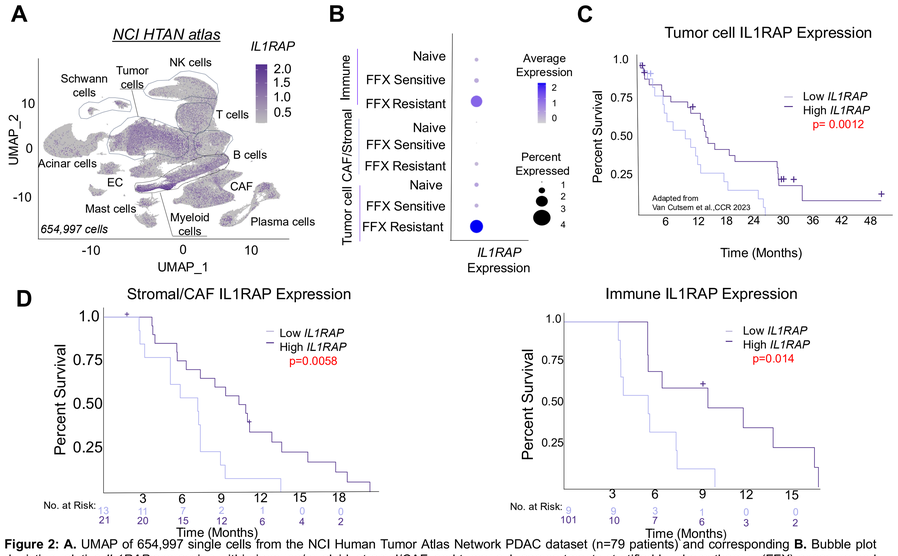


Figure 2: A. UMAP of 654,997 single cells from the NCI Human Tumor Atlas Network PDAC dataset (n=79 patients) and corresponding **B.** Bubble plot depicting relative *IL1RAP* expression within immune/myeloid, stromal/CAF, and tumor sub-compartments stratified by chemotherapy (FFX) exposure and response status. **C.** Schema of CANFOUR trial (NCT03267316) evaluating nadunolimab—human IgG1 anti-IL1RAP mAb) plus gemcitabine/nab-paclitaxel (GnP) in patients with metastatic PDAC. Kaplan–Meier analysis of overall survival stratified by *tumor cell*-level IL1RAP expression by IHC, demonstrates inferior outcomes in the IL1RAP-high subgroup **D.** Kaplan-Meier curves show duration of response on nadunolimab plus GnP in CANFOUR PDAC patients with available baseline biopsies for analysis of stromal/CAF (n=24; *left*) or immune (n=19; *right*) IL1RAP expression by IHC.

② IL1RAP inhibition reprograms myeloid and T-cell states to enhance immune responsiveness; mNadu + chemoimmunotherapy drives durable survival benefit

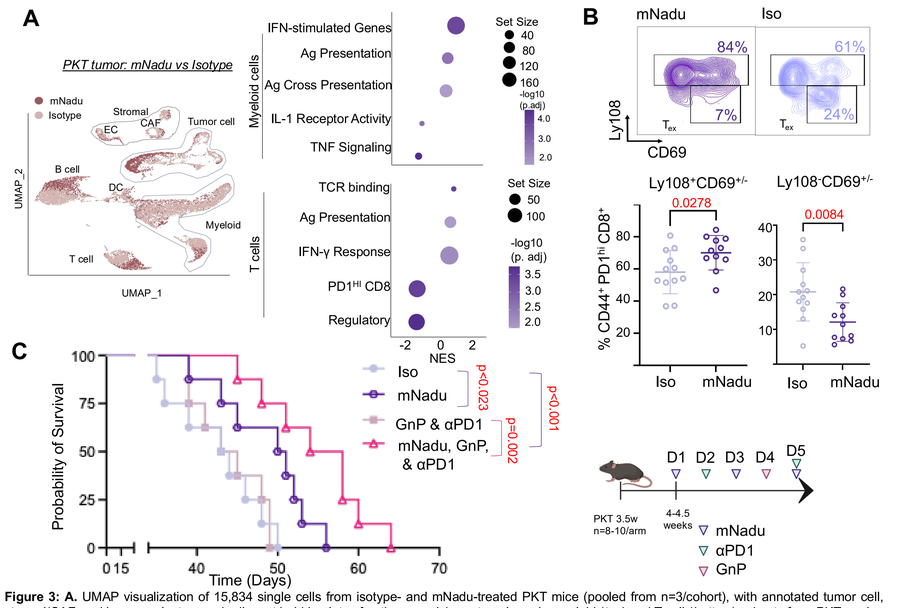


Figure 3: A. UMAP visualization of 15,834 single cells from isotype- and mNadu-treated PKT mice (pooled from n=3/cohort), with annotated tumor cell, stromal/CAF, and immune clusters and adjacent bubble plots of pathway enrichment analyses in myeloid (*top*) and T-cell (*bottom*) subsets from PKT murine PDAC tumors treated with isotype control or mNadu. Selected pathways depicted were identified using KEGG, GO, HALLMARK, and MSigDB knowledgebases. **B** Flow cytometry analysis of intratumoral CD8⁺ T cells from PKT tumors treated with isotype control or mNadu. Representative contour plots and quantification of Ly108⁺CD69⁺/⁻ (memory progenitor exhausted) and Ly108⁻CD69⁺ (terminally exhausted) CD8⁺ T-cell populations are shown (n=11 mice/group) **C**. *In vivo* treatment schedule (right) and Kaplan-Meier survival curves (left) of PKT cohorts treated with isotype control, mNadu monotherapy, GnP plus anti-PD1 Ab, or mNadu combined with GnP plus anti-PD1 (n=8 mice/group). P-values reflect comparisons of median overall survival for each cohort.

③ IL1RAP blockade with mNadu reduces tumor burden and myeloid infiltration while enhancing T-cell expansion

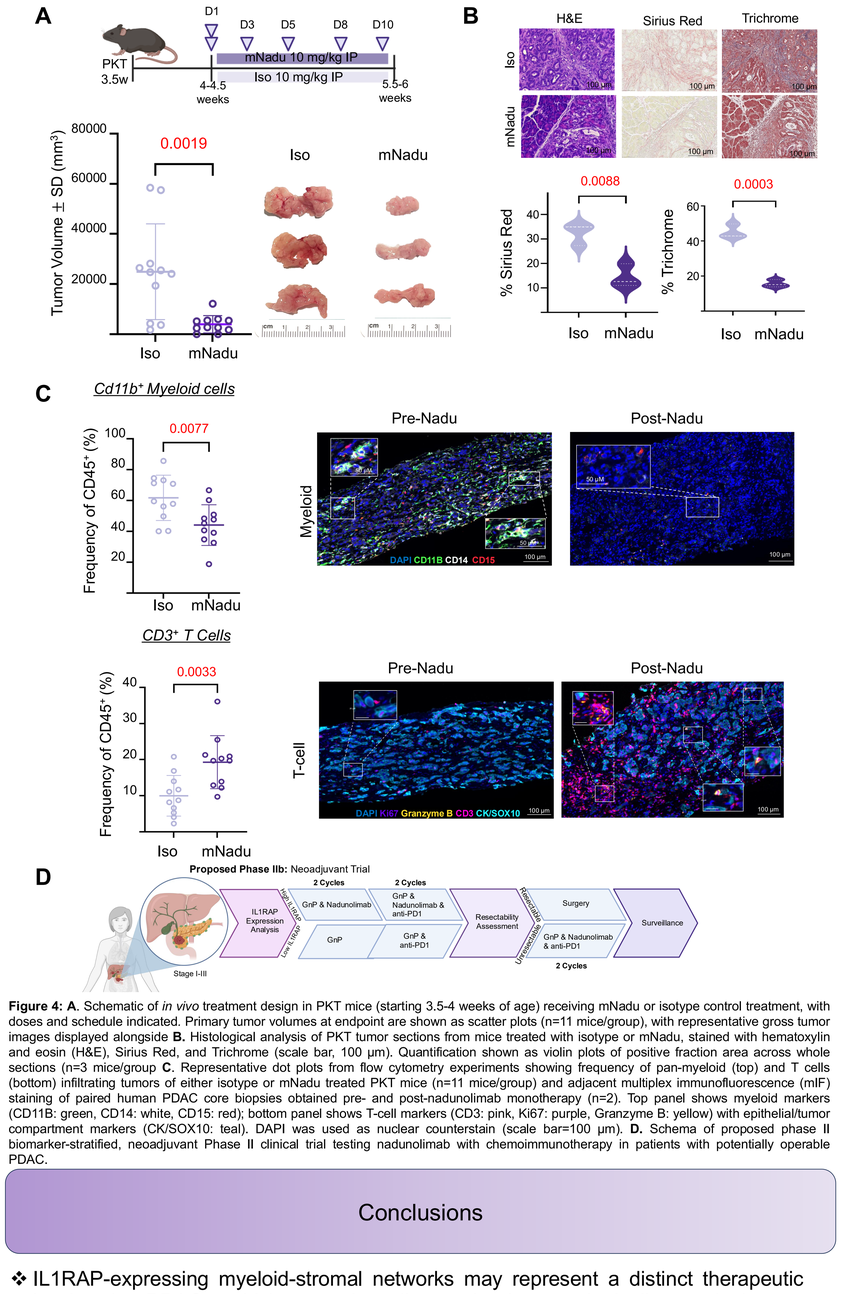


Figure 4: A. Schematic of *in vivo* treatment design in PKT mice (starting 3.5-4 weeks of age) receiving mNadu or isotype control treatment, with doses and schedule indicated. Primary tumor volumes at endpoint are shown as scatter plots (n=11 mice/group), with representative gross tumor images displayed alongside **B.** Histological analysis of PKT tumor sections from mice treated with isotype or mNadu, stained with hematoxylin and eosin (H&E), Sirius Red, and Trichrome (scale bar, 100 μm). Quantification shown as violin plots of positive fraction area across whole sections (n=3 mice/group **C**. Representative dot plots from flow cytometry experiments showing frequency of pan-myeloid (top) and T cells (bottom) infiltrating tumors of either isotype or mNadu treated PKT mice (n=11 mice/group) and adjacent multiplex immunofluorescence (mIF) staining of paired human PDAC core biopsies obtained pre- and post-nadunolimab monotherapy (n=2). Top panel shows myeloid markers (CD11B: green, CD14: white, CD15: red); bottom panel shows T-cell markers (CD3: pink, Ki67: purple, Granzyme B: yellow) with epithelial/tumor compartment markers (CK/SOX10: teal). DAPI was used as nuclear counterstain (scale bar=100 μm). **D.** Schema of proposed phase II biomarker-stratified, neoadjuvant Phase II clinical trial testing nadunolimab with chemoimmunotherapy in patients with potentially operable PDAC.

Conclusions

- IL1RAP-expressing myeloid-stromal networks may represent a distinct therapeutic barrier in PDAC, which can be disrupted pharmacologically to invigorate immunotherapy-permissive CD8⁺ T-cell subsets & improve chemoimmunotherapy sensitivity.
- These findings support an upcoming neoadjuvant trial combining nadunolimab with chemoimmunotherapy in patients with operable PDAC