Proteomic profiling of gemcitabine-resistant pancreatic cancer cells unravels microtubule-associated protein 2 overexpression, that correlated to poorer survival, but also to increased sensitivity to nab-paclitaxel.

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Gemcitabine chemoresistance

Extrinsic factors

Desmoplastic response

Intrinsic factors

Epithelial-mesenchymal transition

Cancer Stem Cells

Current cancer treatments
In vitro characterization of gemcitabine-resistant PDAC cells

- PANC1
- PANC1R

Graph showing cell growth vs control with IC50 values:
- PANC1R (IC50=157nM)
- PANC1 (IC50=25nM)
In vivo characterization of gemcitabine-resistant PDAC cells
Up-regulated proteins in gemcitabine–resistant PANC-1 cells versus sensitive cells

<table>
<thead>
<tr>
<th>gene name</th>
<th>protein name</th>
<th>p value</th>
<th>FC</th>
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<tbody>
<tr>
<td>NAP2</td>
<td>Microtubule-associated protein 2</td>
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<td>ANK3</td>
<td>Ankyrin-3</td>
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<td>NES</td>
<td>Nesin</td>
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<td>CAT</td>
<td>Catalase</td>
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<td>KIF1A</td>
<td>Kinesin-like protein KIF1A</td>
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<tr>
<td>STAT6</td>
<td>Signal transducer and activator of transcription 5</td>
<td>0.0019</td>
<td>NA</td>
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<td>HTT</td>
<td>Huntingtin</td>
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<td>GSTM2</td>
<td>Glutathione-S-transferase Mu 2</td>
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<td>CACHD1</td>
<td>VWFA and cache domain-containing protein 1</td>
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<td>MAPRE2</td>
<td>Isoform 4 of microtubule-associated protein RP/EB family member 2</td>
<td>0.0028</td>
<td>NA</td>
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FC, fold change; PANC1, gemcitabine-sensitive cell line.
Up-regulated proteins in gemcitabine–resistant PANC-1 cells versus sensitive cells.
Validation of ANK3 as prognostic biomarker in gemcitabine treated patients

ANK3 was expressed at equal levels in all tumours and scoring on high versus low expression did not predict gemcitabine resistance

Metastatic cohort  \( n = 36 \)

Adjuvant cohort  \( n = 86 \)
Validation of MAP2 as prognostic biomarker in gemcitabine treated patients

- **MAP2**
- **Gemcitabine-resistant phenotype**

**Metastatic cohort**  
$n = 36$

**Adjuvant cohort**  
$n = 86$
Validation of MAP2 as predictive biomarker of gemcitabine sensitivity
Differential protein expression and pathway analysis

- positive regulation of microtubule binding
  - modulation by virus of host process
    - primary neural tube formation
      - modulation by virus of host process
        - morphogenesis of an epithelial sheet
          - regulation of RNA export from nucleus
            - peptidyl-lysine trimethylation
              - wound healing, spreading of cells
                - positive regulation of sprouting angiogenesis
                  - Sertoli cell development
Exploration of microtubule inhibitors as a therapeutic option

PANC1 vs. PANC1R

- Cell growth vs control (%)
  - Dosage: paclitaxel [nM]
  - Comparison: PANC1 vs. PANC1R

- Survival (%)
  - Comparison: nab-paclitaxel (n=7) vs. vehicle, NaCl (n=7)

- Tumour volume mm³
  - Days (after injection)
  - Comparison: vehicle, NaCl vs. nab-paclitaxel

- Images: Day 20 (after treatment)
Conclusions

- We unravelled new differentially expressed proteins in a gemcitabine-resistant model of PDAC, including MAP2.

- MAP2-positive staining was validated as a prognostic biomarker in two patients cohorts treated with gemcitabine monotherapy, either in palliative or adjuvant setting. High expression of MAP2 was correlated with poorer survival.

- Phosphorilation of MAP2 is an important regulator of its function, thus guiding microtubule dynamics. Changing microtubules dynamics by higher expression and phosphorilation of MAP2 might result in changed drug transport.
Conclusions

- We obtained preclinical data, in vitro and in vivo, showing that (nab-)/paclitaxel was effective against resistant/MAP2-overexpressing cells.

- Our findings support the current therapy with gemcitabine and nab-paclitaxel. Part of the success of this combination therapy might be due to cytotoxic effect on resistant cells.