Modeling prostate cancer in mice

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Modeling molecular pathways of prostate cancer progression

Normal epithelium → Prostatic intraepithelial neoplasia (PIN) → Adenocarcinoma (latent) → Adenocarcinoma (clinical) → Metastasis

<table>
<thead>
<tr>
<th>Processes</th>
<th>Initiation</th>
<th>Progression</th>
<th>Treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Inflammation</td>
<td>Oxidative/DNA damage</td>
<td>Senescence</td>
<td>Re-activation of developmental signaling pathways</td>
</tr>
<tr>
<td>Telomere shortening</td>
<td></td>
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</tbody>
</table>

<table>
<thead>
<tr>
<th>Genes</th>
<th>Initiation</th>
<th>Progression</th>
<th>Treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td>NMX3.1 down-regulation</td>
<td>MYC overexpression</td>
<td>PTEN inactivation</td>
<td>ERK/MAPK activation</td>
</tr>
<tr>
<td>TMPRSS2-ERG fusion</td>
<td>p53</td>
<td>EZH2 overexpression</td>
<td></td>
</tr>
</tbody>
</table>
Translational opportunities for mouse models of prostate cancer

Distinguish indolent from aggressive cancer

- Avoid over-treating men with low risk cancer

Identify molecular processes of malignancy

- Improve diagnosis and treatment of lethal cancer
Modeling molecular pathways of prostate cancer progression

(Shen and Abate-Shen, 2010)
“Next-generation” mouse models of prostate cancer

(Wang et al., Nature 2009)
Mouse models of prostate cancer

Low grade PIN (N model)

Cancer/CRPC with no mets (NP model)

Cancer/CRPC with infrequent metastases (NPB model)

Cancer/CRPC with frequent metastases (NPK model)
Pten loss and Kras activation cooperate in aggressive prostate cancer in mice

Wild-type  NP  NPK

Aytes, et al. PNAS (2013)
NPK mice display high penetrance of metastases

Aytes, et al. PNAS (2013)
Lineage-tracing of metastases in NPK mice

Aytes, et al. PNAS (2013)
Temporal relationship of tumors and metastases in NPK mice

![Diagram showing temporal relationship and analysis of tumors and metastases in NPK mice.]
ETV4 promotes metastases \textit{in vivo}
Pathways of lethal prostate cancer

A. Taylor et al.

B. Taylor et al.

Sbone et al.

Spearman correlation ρ = 0.42
P value = 1.2 × 10^{-7}

Kendall correlation z = 5.0
P value = 5.3 × 10^{-7}

Low PI3-K and RAS

High PI3-K and RAS

Biochemical recurrence free survival

Prostate cancer specific survival

Log Rank p value = 0.003

Log Rank p value = 0.014

Months

Months
Conservation of molecular pathways of NPK mouse model with human prostate cancer

Aytes, et al. PNAS (2013)
Summary:
Modeling metastatic prostate cancer in mice

• Modeling lethal metastatic prostate cancer
• Spatial and temporal relationship of tumor and metastasis
• Mechanisms of metastasis
• Molecular pathways are well-conserved from mouse to man
From mice to man:
Systems approaches for cross species analyses

(Collaboration with Michael Shen and Andrea Califano)
From mice to man: Cross species analyses of malignant cancer

Strategy:

• Mouse and human prostate cancer interactomes
• Demonstrate conservation of their activities
• Identify synergistic master regulators
• Functional and clinical validation

Assemble mouse and human prostate cancer interactomes

**Human Interactome**
- Human prostate cancer
  - Normal
  - Primary tumors
  - Metastases
- Dataset: 185 GEPs

**Mouse Interactome**
- Prostate cancer GEMMs
  - Normal
  - LG-PIN
  - HG-PIN/Cancer
  - CRPC
  - Metastases
- Dataset: 384 GEPs

**Drug perturbations**
- Hormone signaling
- Signaling pathways
- Kinase inhibitors
- Other pathways

Mice from: Sawyers, Van Dyke, Foster, Williams
Cross-species integration of prostate cancer regulatory networks
Identification of conserved master regulators
Conserved master regulators of malignancy

**Human**
- Indolent
- Aggressive

**Mouse**
- Indolent
- Aggressive

### Genes
- **DA DE**
  - Human: CHAF1A
  - Mouse: 445
- **13323**
- **411**
- **511**
- **578**
- **5867**
- **3173**
- **3060**
- **13359**
- **9114**
- **19222**
- **411**
- **1267**
- **717**
- **15953**
FOXM1 and CENPF are predicted to be synergistic master regulators
Functional synergy of FOXM1 and CENPF
FOXM1 and CENPF synergize to promote tumor growth
Clinical synergy of FOXM1 and CENPF: Kaplan Meier based on protein expression

TMA with >900 primary tumors each with extensive clinical outcome data (MSKCC)
Summary: Cross species analysis of master regulators of malignancy

- Cross-species interactomes to integrate data from mouse and man to identify master regulators of malignancy
- Broadly relevant for informing cancer phenotypes and therapeutic response
From mice to man:
Cross species analyses of drug response

Strategy:
• Predict drugs that inhibit the FOXM1-CENPF in vivo
• Evaluate potential drug synergy
• Validate drug synergy/response in vivo
• Determine mechanism of drug synergy

Mitrofanova, Aytes... Shen, Abate-Shen, Califano. (in prep)
Predicting drugs that inhibit FOXM1-CENPF
Predicting drug synergy
Rapamycin and PD0325901 inhibit FOXM1-CENPF in vivo
Drug inhibition of tumor pathways I: Mouse to Mouse

Long-term treatment

Short-term treatment

Running Enrichment Score

Pathway enrichment

$p$ value

Biological pathways

NES = -8.58

$p$ value = 0.001

NES = -8.34

RAP+PD

Vehicle

WT

KEGG_P53_SIGNALING_PATHWAY

KEGG_ECM_RECEPTOR_INTERACTION

KEGG_CELL_ADHESION_MOLECULES_CAMS

BIOCARTA_GSK3_PATHWAY

REACTOME_CELL_CYCLE/MITOTIC

REACTOME_CELL_SURFACE_INTERACTIONS_AT_THE_VASCULAR_WALL

KEGG_STEROID_BIOSYNTHESIS

BIOCARTA_ARF_PATHWAY

BIOCARTA_P53_PATHWAY
Drug inhibition of tumor pathways II: Mouse to Human
Master regulators of drug response: Mouse to Human
Activity levels of treatment response master regulators stratify metastases

(Balk dataset)
Master regulators of the drug response are predictive of outcome in human cancer

<table>
<thead>
<tr>
<th>Protein</th>
<th>% Cases</th>
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<tbody>
<tr>
<td>TOP2A</td>
<td>43%</td>
</tr>
<tr>
<td>WHSC1</td>
<td>41%</td>
</tr>
<tr>
<td>MCM4</td>
<td>35%</td>
</tr>
<tr>
<td>CENPF</td>
<td>41%</td>
</tr>
<tr>
<td>BRCA1</td>
<td>41%</td>
</tr>
<tr>
<td>MCM2</td>
<td>30%</td>
</tr>
<tr>
<td>FOXM1</td>
<td>32%</td>
</tr>
<tr>
<td>UHRF1</td>
<td>16%</td>
</tr>
<tr>
<td>ASF1B</td>
<td>32%</td>
</tr>
<tr>
<td>BLM</td>
<td>11%</td>
</tr>
<tr>
<td>CCNA2</td>
<td>32%</td>
</tr>
<tr>
<td>E2F1</td>
<td>11%</td>
</tr>
<tr>
<td>SUV39H1</td>
<td>8%</td>
</tr>
<tr>
<td>MYBL2</td>
<td>19%</td>
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<tr>
<td>CRY2</td>
<td>14%</td>
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</table>

- mRNA Upregulation
- mRNA Downregulation

**Sboner dataset**

- LOW activity of MRs
- HIGH activity of MRs

Log Rank p-value = 2.63 X 10^-5

![Graph showing overall survival](image)
Summary:
Cross species interrogation of drug response

• Cross-species integration of preclinical data from mouse and man to study drug response \textit{in vivo}

• Valuable approach for predicting drug response \textit{in vivo} and for evaluating the mechanism of action
Translational opportunities for mouse models of prostate cancer

Distinguish indolent from aggressive cancer

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Identify molecular processes of malignancy

- Improve treatment for lethal prostate cancer
Modeling cancer initiation in mice

Loss of the Nkx3.1 leads to precancerous lesions (PIN)
Nkx3.1 mutant mice model indolent prostate cancer
Biomarkers of indolent prostate cancer

Strategy:

• Molecular signature of aging and senescence
• GSEA to identify an “indolence signature”
• Decision tree learning to identify 3-gene panel
• Validation on TMAs and biopsies

Irshad, Bansal... Benson, Shen, Califano, Abate-Shen. Cancer Cell (Sci Trans Med)
Indolent prostate cancer is enriched for a molecular signature of aging and senescence.
Validation on low Gleason score prostate cancer

Taylor dataset

Validates the hypothesis and the indolence signature
Decision tree learning model to identify 3-gene panel
3-gene panel distinguishes indolent lesions on biopsy samples from patients on active surveillance

<table>
<thead>
<tr>
<th>FGFR1</th>
<th>PMP22</th>
<th>CDKN1A</th>
</tr>
</thead>
<tbody>
<tr>
<td>Non-Failed Biopsy</td>
<td>Non-Failed Biopsy</td>
<td>Non-Failed Biopsy</td>
</tr>
<tr>
<td>Failed Biopsy</td>
<td>Failed Biopsy</td>
<td>Failed Biopsy</td>
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<table>
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<tr>
<th>Biopsy samples from Gleason 6 patients on surveillance</th>
</tr>
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<tbody>
<tr>
<td>N</td>
</tr>
<tr>
<td>----</td>
</tr>
<tr>
<td>Non-failures</td>
</tr>
<tr>
<td>Failures</td>
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p=1.54 x 10^-5
Summary II: Biomarkers of indolent prostate cancer

- Biomarker to help stratify low-Gleason score prostate cancer
- Now validating in retrospective and prospective studies
Acknowledgements

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