Current Topics in Prostate Cancer Research

RPN532 (Tang, Dean G.; Pharmacol & Therap)/04/06/2017

Paper for students:
Points to discuss:

* Why do current treatments (ADT) fail – Cellular heterogeneity?
* Prostate cancer genetics: AR & beyond
* Normal cell lineage: stem cells & their progeny
* Cancer cell plasticity: Genetic, epigenetic, & treatment-induced
Prostate Cancer Treatments

Charles Huggins in 1941: Castration (Lupron [leuprolide acetate]/Casodex [bicalutamide])

- **2004**: Taxotere (docetaxel) for advanced metastatic patients who no longer respond to castration
- **2010**: Cabazitaxel (Jevtana) for tumors that have stopped responding to docetaxel
- **2010**: Sipuleucel-T (Provenge) as vaccine (for advanced/met patients who no longer respond to castration)
- **2012**: Zytiga (abiraterone acetate) + Xtandi (enzalutamide)
- **2013**: Radium 223 (Xofigo) for metastases that have spread to the bone

**Patient survival:**

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Survival Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>Docetaxel</td>
<td>3.3 months (vs. mitoxantrone)</td>
</tr>
<tr>
<td>Cabazitaxel</td>
<td>2.4 months (vs. mitoxantrone)</td>
</tr>
<tr>
<td>Sipuleucel-T (Provenge)</td>
<td>4.1 months (vs. placebo)</td>
</tr>
<tr>
<td>Abiraterone</td>
<td>3.9 months (vs. placebo)</td>
</tr>
<tr>
<td>Enzalutamide</td>
<td>4.8 months (vs. placebo)</td>
</tr>
<tr>
<td>Radium-223</td>
<td>2.8 months (vs. placebo)</td>
</tr>
<tr>
<td>Ipilimumab</td>
<td>No (vs. placebo after radiotherapy; toxic – more patients died)</td>
</tr>
<tr>
<td>Prostvac</td>
<td>PROSPECT Phase III trial (PSA-targeted IT)/second half of 2017</td>
</tr>
<tr>
<td>ARN-509</td>
<td>SPARTAN &amp; ATLAS trials??(2019)</td>
</tr>
</tbody>
</table>
Clinical treatment of PCa patients by chemical castration and anti-AR drugs (anti-androgens)
New anti-PCa drug development
Points to discuss:

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*Normal cell lineage: stem cells & their progeny
*Cancer cell plasticity: Genetic, epigenetic, & treatment-induced
AR gene, mRNA, and protein

AR and drug targets

Sadar MD. Cancer Res. 71, 1208
2011
Genomic heterogeneity within localized, multifocal prostate cancer

Boutrous PC et al., Nat Genet 2015, May 25
Mutational landscape of lethal metastatic CRPC

Grasso CS et al.,
Nature 2012
Integrative Clinical Genomics of mCRPC

Robinson D et al., Cell, 161, 1215-28, 2015
## AR splice variants

<table>
<thead>
<tr>
<th>Variants found in PCa</th>
<th>Domain excluded</th>
<th>Domain-disrupted</th>
<th>Protein MW</th>
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<tbody>
<tr>
<td>AR23</td>
<td>Exon 1 2 2b 3 4 5 6 7 8</td>
<td>DBD</td>
<td>106kDa</td>
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<tr>
<td>AR-V14</td>
<td>Exon 1 2 3 4 5 6 7 9</td>
<td></td>
<td>85kDa</td>
</tr>
<tr>
<td>AR-V13</td>
<td>Exon 1 2 3 4 5 6 9</td>
<td>LBD</td>
<td>85kDa</td>
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<tr>
<td>AR-V12</td>
<td>Exon 1 2 3 4 8 9</td>
<td>LBD</td>
<td>84kDa</td>
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<tr>
<td>AR\textsuperscript{v567es}</td>
<td>Exon 1 2 3 4 8</td>
<td>LBD</td>
<td>80kDa</td>
</tr>
<tr>
<td>AR-V7</td>
<td>Exon 1 2 3 3e</td>
<td>HD to LBD</td>
<td>80kDa</td>
</tr>
<tr>
<td>AR-V9</td>
<td>Exon 1 2 3 3d</td>
<td>HD to LBD</td>
<td>80kDa</td>
</tr>
<tr>
<td>AR-V5</td>
<td>Exon 1 2 3 3c</td>
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</tr>
<tr>
<td>AR-V1</td>
<td>Exon 1 2 3 3b</td>
<td>HD to LBD</td>
<td>80kDa</td>
</tr>
<tr>
<td>AR-V3</td>
<td>Exon 1 2 2b</td>
<td>ZF2 to LBD</td>
<td>75kDa</td>
</tr>
<tr>
<td>AR\textsuperscript{Ex1/2b}</td>
<td>Exon 1 2b</td>
<td>ZF1 to LBD</td>
<td>75kDa</td>
</tr>
</tbody>
</table>
A global view of advanced PCa genome

Taylor et al., Cancer Cell 18, 11-22, 2010.
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*Normal cell lineage: stem cells & their progeny
*Cancer cell plasticity: Genetic, epigenetic, & treatment-induced
Cell lineage development: Self-renewal, proliferation, & differentiation

Phenotypic and developmental plasticity in CSCs and their progeny

A) Early stages of tumor development
Under ‘normal’ conditions

CSCs → Progenitors → Non-CSCs; Differentiated tumor cells

B) ‘Intrinsic’ plasticities in undifferentiated tumor cells and CSCs

molecular mimicry

EC → GBM CSCs → EC

C) Accompanying tumor progression
(hypoxia; EMT, inflammation; microenvironment changes)

D) Experimental manipulations/treatment; therapies

Tang, Cell Res. 2012
Early phase: c-Myc the driving force
Intermediate phases: Oct-4 & Sox2
Late (&early) phase: Klf4

Sancho-Martinez I & Izpisua Belmonte JC
Cell-of-origin vs. CSCs

Rycaj K & Tang DG. *Cancer Res*, 2015
Functional Assays of Cancer Cell of Origin

Rycaj & Tang, *Cancer Res*, 2015
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Increasingly 'mature' PCa cells

PCSC pool (PSA⁻/lo)

CD44+
ALDHhi
α2β1+
SP
ABCG2+
Holoclones

(PSA⁺/hi)

Intrinsically tumorigenic (PSA⁺/hi) PCa cells

Phenotypic & Tumorigenic Heterogeneity of Human PCa cells
Heterogeneity of AR Expression in Untreated PCa & CRPC

Untreated HPCa57

CRPC-13553 (Huang)
Four CRPC models exhibit distinct AR heterogeneity

A
LNCaP, VCaP, LAPC4, LAPC9 tumors grown in intact male mice (AD tumors) → Serially passaged in castrated mice → Primary (1°) CRPC → Enzalutamide Tx in castrated mice → Secondary (2°) CRPC

B

<table>
<thead>
<tr>
<th></th>
<th>LNCaP</th>
<th>LAPC9</th>
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</thead>
<tbody>
<tr>
<td>AD</td>
<td>1° CRPC (P6)</td>
<td>AD</td>
</tr>
<tr>
<td>nuc</td>
<td>nuc</td>
<td>nuc</td>
</tr>
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</table>

<table>
<thead>
<tr>
<th></th>
<th>LAPC4</th>
<th>VCaP</th>
</tr>
</thead>
<tbody>
<tr>
<td>AD</td>
<td>1° CRPC (P7)</td>
<td>AD</td>
</tr>
<tr>
<td>nuc</td>
<td>nuc/cyto</td>
<td>nuc</td>
</tr>
</tbody>
</table>
Four CRPCs respond differently to Enzalutamide

- **LNCaP CRPC**
- **LAPC4 CRPC**
- **VCaP CRPC**
- **LAPC9 CRPC**
Prostate cancer cell plasticity: Reprogramming by NANOG

PCa stem cells (PSA⁻/lo)

- Holoclones
  - ALDH⁺
  - CD44+
- SP
- ABCG2⁺
- α3β1⁺

NANOG

(AR⁺PSA⁺)

Jeter/Liu
NANOG reprograms PSA+ PCa cells to PSA-/-lo, stem-like CRPC cells by dynamically repressing and engaging AR/FOXA1 signaling axis

Jeter C & Liu B et al., Cell Discovery, 2016
Understanding & Targeting Undifferentiated PCSCs

A) Untreated (AD)
- LNCaP cultures
- LAPC9 xenografts
- HPCa primary T.

B) Castration in vitro (CDSS/bica/MDV)
- LNCaP-abl (AR+PSA-)
- LNCaP-95
- LNCaP-CL1, CL1.1 (AR-PSA-)
- LNCaP-KR sublines (AR-PSA-)

C) Castration in vivo
- xenograft models (LNCaP/LAPC9/LAPC4/VCaP/CWR22......)

CRPC

- PSA⁺ PCa cells (fast proliferating; castration sensitive)
- PSA⁺ PCa cells - untreated (dormant; castration resistant)
- PSA⁻ PCa cells - treated (cycling; castration resistant?)

- Limited tumor propagating capability
- Tumors contain mostly PSA⁺ cells
- Sensitive to castration

- Unlimited tumor propagating capability
- Recreate parental tumor cell heterogeneity
- Resistant to castration

castration
dé-differentiation
Tx-induced reprogramming
Combinatorial therapies targeting both AR$^+$ bulk AND PCSCs to prevent cancer cell plasticity

PCSC pool
(PSA$^{-/lo}$)

Anti-PCSC Therapeutics

De-bulking agents
(Enzalutamide; Abiraterone, Chemo/radiation, Immunother.)

PREVENT RECURRENT & METASTASIS

Increasingly ‘mature’ PCa cells