Multigene Testing in Prostate Cancer Risk Stratification

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Prostate Cancer

INITIAL PROSTATE CANCER DIAGNOSIS

Life expectancy ≤ 55 y and asymptomatic

DRE
PSA
Gleason primary and secondary grade

INITIAL CLINICAL ASSESSMENT

No further workup or treatment until symptomatic, except in high- or very-high-risk groups

Bone scan if any of these:
- T1 and PSA >20 ng/mL
- T2 and PSA >10 ng/mL
- Gleason score ≥ 8
- T3, T4
- Symptomatic

Suspicious nodes
Consider biopsy

Pelvic CT or MRI if any of these:
- T3, T4
- T1-T2 and nomogram indicated probability of lymph node involvement > 10%

All others: no additional imaging

*Men with clinically localized disease may consider the use of tumor-based molecular assays. Retrospective case cohort studies have shown that molecular assays performed on biopsy or prostatectomy specimens provide prognostic information independent of NCCN-risk groups. These include, but are not limited to, likelihood of death with conservative management, likelihood of biochemical progression after radical prostatectomy or external beam therapy, and likelihood of developing metastasis after radical prostatectomy or salvage radiotherapy.
Objectives

• Briefly review relevant molecular biology of localized prostate cancer

• Review currently available molecular tests for prostate cancer and the potential risks and benefit of their use

Prostate Cancer – Landscape and Decision Uncertainty

Decision Making Complexity
• Broad Spectrum of Localized Disease
• Relatively Long Natural History (Competing Risks)
• Morbidity of Treatment

Screening and Diagnosis → Surveillance → Treatment → Prostatectomy, Radiation, Other → Surveillance → Adjuvant/Salvage Tx → Metastatic Disease → mCRPC
Advances

- Molecular understanding of localized prostate cancer has increased

- We have increased facility to obtain molecular information from routinely collected and stored formalin-fixed paraffin embedded tissue

Molecular Tissue Testing – Issues

- Confusion among patients and providers
  - Tests assess similar molecular phenotypes
  - Aggressive marketing
  - Lack of head-to-head comparisons

- Existing risk stratification tools for localized disease perform well

- Cost
RELEVANT MOLECULAR BIOLOGY OF LOCALIZED PROSTATE CANCER

- “Prostate cancer is characterized by extraordinary genomic complexity”
  - Copy number alterations
    - Deletions and amplifications
  - Chromosomal rearrangements
  - Point mutations

Schoenborn et al Clin Cancer Res 2013

PTEN Loss, An Important Early Event

Baca et al. Cell 2013
PTEN Loss, An Important Early Event

- PTEN is a tumor suppressor
  - Normally halts the PI3K/AKT pathway
  - PI3K pathway members are altered in up to 40% of all primary prostate cancer and 100% of metastasis

PTEN Loss, An Important Early Event

- PTEN loss is a key component of animal models for prostate cancer
- PTEN loss can be detected in ~15-40% of primary cases and ~50% of metastasis
- Loss correlates with stage and grade

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Tests Based on Cell Cycle Proliferation

- Cell cycle abnormalities are common in localized prostate cancer
- Ki-67 IHC has independent prognostic significance after radiation (XRT) or radical prostatectomy (RP) (Khor et al JCO 2004, Tollefson et al Mayo Clin Proc 2014)
- Prolaris
  - qRT-PCR
  - 31 cell cycle genes normalized to 15 housekeeping genes
  - Currently marketed for use in men diagnosed with very-low-risk and low-risk prostate cancer
Prolaris Scores Correlate with Clinical Outcomes Post RP / Watchful Waiting

Prolaris Scores Correlate with Clinical Outcomes Post RP

Cuzick et al Lancet Onc 2011

Cooperberg et al JCO 2013
Prolaris / Cell Cycle Progression (CCP) Score – Needle Biopsy Cohorts (post RP)

- 582 men undergoing RP, Needle Biopsy
- Primarily low- and intermediate-risk disease
- Relatively few metastatic events
- Hazard ratio (HR) per unit increase in score, 4.2 (2.1-8.5) on multi-variate analysis (MVA)

Bishoff J Urol 2014

Prolaris / CCP score – Needle Biopsy Cohorts (post radiation)

- 141 men undergoing XRT, Needle Biopsy
- 73% intermediate- or high-risk
- HR biochemical recurrence (BCR; Phoenix definition) per unit increase in score, 2.1 (1.05-4.25) MVA

Freedland et al IJROBP 2013
Prolaris/CCP Score - Needle Biopsy
Conservative Management

- Transatlantic Prostate Group
- HR death per unit CCP increase ~2 (bivariate analysis with CAPRA)

Tests Based on Molecular Features More Specific to Prostate Cancer

- PTEN/ERG
- Oncotype Dx Prostate
- ProMark
- Decipher
PTEN Testing

- IHC
- FISH

Shipitsin et al. Proteome Science 2014

PTEN Testing – Watchful Waiting, Transatlantic Prostate Group

- 675 men on watchful waiting (WW)
- PTEN IHC & FISH (IHC better) [cheaper too, $95 at JHH]
- TURP specimens; 18% PTEN loss; 3% among low risk

Shpitsin et al. Proteome Science 2014

Cuzick et al. BJC 2013
PTEN Testing – Upgrading

- 174 men with Gleason score (GS) 6 disease undergoing RP
- PTEN IHC on biopsy tissue
- PTEN lost in 18% of upgraded cases with 7% of cases that did not upgrade
- OR 3 (1.1-8.6) for upgrading

Lotan et al Mod Path 2015

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PTEN/ERG – Outcomes after Prostatectomy (Health Professionals Follow-Up Study and Physicians Health Study)

<table>
<thead>
<tr>
<th>PTEN Status</th>
<th>N</th>
<th>Lethal</th>
<th>HR for lethal prostate cancer</th>
</tr>
</thead>
<tbody>
<tr>
<td>PTEN positive</td>
<td>735</td>
<td>46</td>
<td>1</td>
</tr>
<tr>
<td>PTEN mixed</td>
<td>90</td>
<td>7</td>
<td>1.2 (0.6-3.0)</td>
</tr>
<tr>
<td>PTEN negative</td>
<td>139</td>
<td>28</td>
<td>1.9 (1.2-3.0)</td>
</tr>
</tbody>
</table>

Adjusted for age at diagnosis, BMI at diagnosis, Gleason grade, and cTNM

Ahearn et al JNCI 2016
**Tests on Multiple Features of Prostate Cancer Oncotype DX Prostate**

- qRT-PCR of 12 genes (derived from 732 genes which correlated with poor oncologic outcome) and 5 housekeeping genes

  - Run on biopsies
  - Each 20-point increase (~IQR) in Genomic Prostate Score (GPS) equals ~2 fold risk of ≥ 4+3 or pT3 disease at RP
  - Marketed for men with low- to low intermediate-risk considering active surveillance
  - Not reported in active surveillance populations

**PTEN /ERG – Outcomes after RRP (Health Professionals Follow-Up Study and Physicians Health Study)**

<table>
<thead>
<tr>
<th>PTEN / ERG status</th>
<th>N</th>
<th>Lethal</th>
<th>MVA HR</th>
</tr>
</thead>
<tbody>
<tr>
<td>PTEN+ and ERG-</td>
<td>306</td>
<td>20</td>
<td>REF</td>
</tr>
<tr>
<td>PTEN+ and ERG+</td>
<td>260</td>
<td>12</td>
<td>0.9 (0.4-1.8)</td>
</tr>
<tr>
<td>PTEN- and ERG-</td>
<td>48</td>
<td>13</td>
<td>3.1 (1.5-6.3)</td>
</tr>
<tr>
<td>PTEN- and ERG+</td>
<td>132</td>
<td>12</td>
<td>0.7 (0.4-1.5)</td>
</tr>
</tbody>
</table>

Klein et al Eur Urol 2014
Cullen et al Eur Urol 2014

Ahearn et al JNCI 2016
Tests on Multiple Features of Prostate Cancer

Oncotype DX Prostate

- Meta-analysis 732 patients (2 studies, UCSF, CPDR) for prediction of favorable pathology (pT2 and GS 3+4=7 or less)

**Tests on Multiple Features of Prostate Cancer

ProMark**

- Multiplexed proteomics assay
- Run on biopsies
- Score ranges form 0-1, each 0.25-point increase equals ~3 fold risk of unfavorable pathology
- Thresholds of <0.33 and >0.8 appear useful and include ~40% of patients
- Marketed for men with low- to low intermediate-risk considering AS
- Not yet tested in AS populations

**Brand et al Urology 2016**

Tests on Multiple Features of Prostate Cancer Decipher

- High density array performed on FFPE prostate tissue
  - 1.4 million probes (coding and non-coding)
  - CLIA certified
- 22 genomic markers selected for ability to predict rapid mets after RP
  - Outputs a genomic classifier (GC) score (0-1)
  - Marketed for post RP testing and recently available for use on biopsy tissue

Tests on Multiple Features of Prostate Cancer Decipher – Post RP

- Metastasis signature validation in 219 high-risk men after RRP at Mayo Clinic
- Categorical cut-offs at <0.4 (60%), 0.4-0.6 (20%) and >0.6 (20%) associated with HR for mets on MVA of 1, 2.4 (1.1-5.2) and 7.3 (3.5-15.1)
- Mayo cohort with high amount of adjuvant and salvage treatment post-RP
Tests on Multiple Features of Prostate Cancer
Decipher – Post RP

- JHH cohort, 260 NCCN intermediate- or high-risk men, 99 with metastatic progression
  - Natural history (no treatment post-RP until mets)
- Median GC score 0.34 (IQR 0.22-0.52)
- HR met MVA 1.5 (1.3-1.7) per 0.1 increase in score
- Similarly predictive in RP and biopsy tissue from a Cleveland Clinic cohort managed without adjuvant radiation

Tests on Multiple Features of Prostate Cancer
Decipher – Independent Predictor Cancer Specific Mortality

- HR 1.8 per 0.1 unit increase on MVA

<table>
<thead>
<tr>
<th></th>
<th>HR PCSM</th>
<th>P val</th>
</tr>
</thead>
<tbody>
<tr>
<td>GC</td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.4–0.6 (ref: &lt;0.4)</td>
<td>1.09 (0.26–3.77)</td>
<td>0.9</td>
</tr>
<tr>
<td>&gt;0.6 (ref: &lt;0.4)</td>
<td>11.26 (4.69–30.37)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>CAPRA-S &gt;5</td>
<td>2.36 (1.06–5.68)</td>
<td>0.04</td>
</tr>
<tr>
<td>Adjuvant therapy</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Radiation</td>
<td>0.56 (0.11–1.80)</td>
<td>0.36</td>
</tr>
<tr>
<td>Androgen deprivation</td>
<td>1.55 (0.72–3.36)</td>
<td>0.26</td>
</tr>
</tbody>
</table>

Ross et al Eur Urol 2015
Klein et al Eur Urol 2015
Klein et al Urology 2015

Cooperberg et al Eur Urol 2015
Tests on Multiple Features of Prostate Cancer Decipher – Post RP Adjuvant and Salvage Radiation

- 170 men with BCR undergoing salvage XRT
- MVA HR for metastasis 1.6 per 0.1 unit increase p=0.002
- 12% metastasis rate
- 39% patients in the upper 2 tertiles of Briganti Risk and 49% of CAPRA-S intermediate/high-risk patients reclassified as GC low risk (97% and 96% MFS)

Freedland et al Eur Urol 2016

Decipher – Post RP Radiation Therapy

Kames et al. (2013), N=219
Design: Case-control

Den et al. (2014), N=139
Design: Case-control

Ross et al. (2015), N=290
Design: Case-control

Freedland et al. (2015), N=117
Design: Case-control

Exclusion:
1) Ne (n=4)
2) Pretreatment or post-treatment prostate cancer treatment prior to surgery (n=50)
3) Residual tumor or metastasis treatment prior to clinical evidence of metastasis (n=36)
4) Pretreatment PSA greater than 10 ng/mL (n=4)

Indication:
1) Patients in the randomly selected sub-cohort of case-control studies
2) Achieved PSA nadir after surgery
3) European clinical data
4) Residual tumor or metastasis treatment prior to clinical evidence of metastasis
5) pT3 or BPH patients

applying inclusion and exclusion criteria

Kames et al. (2013), N=219
Den et al. (2014), N=139
Ross et al. (2015), N=290
Freedland et al. (2015), N=117

Final Cohort (N=22)

Ross and Den et al PCAN 2016 in press
Den et al JCO 2015
Den et al Int J Rad Onc Biol Phys 2015
Decipher – Post RP Radiation Therapy

Predictive of Treatment Response - Metastatic Castration-Resistant Prostate Cancer (mCRPC) Failing Multiple Agents

Surveillance of Very-Low-Risk (VLR) Men – Consider No Testing

- NO molecular testing recommended in VLR >65 years
Surveillance of Low-Risk Men – PTEN/ERG, Oncotype Dx, ProMark, Prolaris

- Most testing will be confirmatory (non-informative)
  - Cost Considerations
- Limited published data from active surveillance cohorts

Intensity/Use of Primary Radiation Therapy (RT) for all NCCN Risk Groups: ?

- Unlike RP, RT is biologic and modified based on predicted cancer aggression
- Androgen deprivation therapy (ADT) adds morbidity
- RTOG 0815: RT +/-ADT for intermediate-risk men
- Short term ADT (SADT) vs long-term ADT (LADT) for high-risk men
- Addition of chemotherapy for very-high-risk and locally advanced men
- Ongoing research
After RP – Adjuvant Radiation Example 2 Step Process – Nomogram First

• Most men with adverse pathological features (APF) after RP do not benefit from adjuvant XRT

NNT, number needed to treat; ART, adjuvant radiation therapy; OBS, observation

After RP – Adjuvant Radiation Example 2 Step Process – Nomogram First

<table>
<thead>
<tr>
<th>Overall Mortality</th>
<th>Radiation</th>
<th>Wait and see</th>
<th>HR (95% CI)</th>
<th>Heterogeneity test p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>Number of events/number of patients</td>
<td>Number of events/number of patients</td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;65</td>
<td>427/380</td>
<td>647/365</td>
<td>0.91 (0.60-1.39)</td>
<td>0.0008</td>
</tr>
<tr>
<td>65-69</td>
<td>481/230</td>
<td>52/105</td>
<td>0.97 (0.45-1.34)</td>
<td></td>
</tr>
<tr>
<td>≥70</td>
<td>43/584</td>
<td>20/292</td>
<td>2.94 (1.75-5.03)</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>130/502 (35.9%)</td>
<td>135/502 (22.9%)</td>
<td>0.61</td>
<td>1.00 (p = 0.95-5.51)</td>
</tr>
</tbody>
</table>

• Men with APF after RP may be harmed by adjuvant XRT (long term results EORTC 22911, Bolla et al. Lancet 2012)
After RP – Adjuvant Radiation Example 2 Step Process – Nomogram First

- Natural history cohort of men undergoing RP at JHH

<table>
<thead>
<tr>
<th>Nomogram Cut-point</th>
<th>Cumulative Incidence of Metastasis</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No. of APF Patients (%)</td>
</tr>
<tr>
<td>CAPRA-S &lt;3</td>
<td>200 (26%)</td>
</tr>
<tr>
<td>CAPRA-S 3-5</td>
<td>435 (56%)</td>
</tr>
<tr>
<td>CAPRA-S &gt;5</td>
<td>139 (18%)</td>
</tr>
<tr>
<td>Eggener &lt;2.5%</td>
<td>302 (39%)</td>
</tr>
<tr>
<td>Eggener 2.5-5.0%</td>
<td>264 (34%)</td>
</tr>
<tr>
<td>Eggener 5-15%</td>
<td>113 (17%)</td>
</tr>
<tr>
<td>Eggener 15-25%</td>
<td>48 (6%)</td>
</tr>
<tr>
<td>Eggener &gt;25%</td>
<td>27 (4%)</td>
</tr>
</tbody>
</table>

(B) pT3 or SM+ Patients

Adjuvant Radiation after RP – 2 Step Process
Decipher if Eggener >2.5% <25%

- Strong consideration to treat with adjuvant XRT vs early salvage if Decipher >0.45, particularly if <70 years old

Eggener 0.025 to 0.25

26% metastasize

42% metastasize

34% metastasize

57% metastasize
Summary

- Our molecular understanding of prostate cancer is increasing exponentially
  - PTEN loss appears to be an early and important event for the development of aggressive prostate cancer
- Tests have been developed that may help treatment decisions
- Current evidence suggests tests are prognostic and selective use of these tests may be beneficial
- Tests should be put into context of current practice
  - Multiparametric MRI (mpMRI) / fusion biopsy
  - PHI / PCA3 / 4Kscore
  - Use of nomograms for decision making
- Head-to-head comparisons are needed to inform providers
- Largest impact will come in areas of greatest decision uncertainty
  - RP vs RT
  - RT +/- ADT
  - Early use of docetaxel

Thanks and Questions