

The Clinical Applications of Tissue Biomarkers in Prostate Cancer

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Abstract

The clinical course of localized prostate cancer varies widely, from indolent disease unlikely to need treatment to aggressive disease requiring intensive, multimodal therapy. Traditionally, treatment decisions have been based on clinical and pathologic factors, including serum prostate specific antigen (PSA), clinical stage, and Gleason score. However, these factors have limited ability to describe the underlying tumor biology. Tissue-based genomic tests have emerged as a promising tool to more accurately characterize prostate cancer biology and predict clinical course. Using prostate cancer tissue obtained at pre-treatment biopsy or radical prostatectomy, these tests exploit the expression of specific genes involved in key biological pathways and, as a result, have the potential to aid clinical decision-making. The current review summarizes available data describing the clinical use of 5 commercially available tissue-based genomic assays in a number of clinical scenarios.

Introduction

Over the last decade, numerous tissue-based genomic classifiers have been developed, providing additional diagnostic, prognostic, and predictive information in the management of prostate cancer (PCa). Using prostate tissue obtained at pre-treatment biopsy or radical prostatectomy, these tests exploit expression of specific genes involved in key biological pathways. Gene expression profiling therefore has the potential to aid clinical decision-making. In this review, we summarize 5 commercially available tissue-based genomic classifiers and summarize the currently available data on their use in a number of clinical scenarios ([Table 1](#)).

ConfirmMDX

Description of assay

ConfirmMDx (MdxHealth, Irvine, United States) for Prostate Cancer is an epigenetic assay, which uses multiplex methylation-specific polymerase chain reaction (PCR) to measure the epigenetic status of the PCa-associated genes *GSTP1*, *APC*, and *RASSF1* in residual cancer-negative prostate biopsy tissue samples [1].

Prediction of cancer after negative biopsy

The diagnostic performance of ConfirmMDx for predicting cancer on subsequent biopsies after an initial negative biopsy has been assessed in 2 retrospective studies: (1) Methylation Analysis to Locate Occult Cancer (MATLOC) [2] and (2) Detection Of Cancer Using Methylated Events in Negative Tissue (DOCUMENT) [3]. The MATLOC study generated a model with the methylation levels of the 3 genes and clinical parameters (age, PSA, DRE, initial biopsy pathology) in 483 men from the United Kingdom and Belgium. The studies found that this model resulted in a negative predictive value (NPV) of 90% [2]. The DOCUMENT validation cohort of 350 men from 5 centers across the

Key Words

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Competing Interests

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Abbreviations

AP	adverse pathology
AS	active surveillance
BCR	biochemical recurrence
CAPRA	Cancer of the Prostate Risk Assessment
CCP	cell cycle progression
CCR	cell cycle risk
DRE	digital rectal examination
GPS	genomic prostate score
NCCN	National Comprehensive Cancer Network
NPV	negative predictive value
PCa	prostate cancer
PCR	polymerase chain reaction
RP	radical prostatectomy

United States found similar results in which the model incorporating the methylation levels had an NPV of 88% [3]. The validity of the assay has also been assessed in a cohort of 211 African American men from 7 centers in the United States, in which methylation status had a NPV of 94.2% for the detection of Gleason score ≥ 7 cancer on repeat biopsy [4]. However, prospective validation studies have yet to be performed for this assay, and its performance compared with other biomarkers in men with a prior negative biopsy remains to be determined.

Oncotype Dx Genomic Prostate Score (GPS)

Description of assay

Oncotype Dx Genomic Prostate Score (Genomic Health, Inc., Redwood City, United States) consists of a panel of 12 PCa-related genes representing 4 distinct biological pathways (androgen signaling, cellular organization, stromal response, and cellular organization) relative to 5 reference genes, based on measured RNA expression levels by reverse transcriptase-PCR [5].

Prediction of disease progression in active surveillance

The GPS score was developed to determine the likelihood of adverse pathology in men undergoing immediate radical prostatectomy. Although they were developed in the surgical context, studies have also shown that the GPS correlates with disease progression in men on active surveillance (AS), and the current National Comprehensive Cancer Network (NCCN) [6] and American Association of Clinical Oncology (ASCO) [7] guidelines recommend considering such genomic tests for men qualifying for AS in situations in which the assay results are likely to impact clinical decision-making.

In the first such study in an AS cohort of 271 men who had GPS testing, 144 experienced biopsy upgrading, and GPS was associated with an increased risk of upgrading [8]. A further study from the same AS cohort assessed the ability of GPS to predict adverse pathology (AP) as well as the risk of biochemical recurrence (BCR) after radical prostatectomy (RP). In 215 men on AS who underwent delayed RP, GPS was associated with an increased risk of adverse pathology at RP and BCR [9].

GPS as a predictor of AP at RP has recently been reported in the multicenter Canary Prostate Active Surveillance Study (PASS) cohort [10]. Overall, 101 men on AS underwent RP after a median of 2.1 years, and 52 men were found to have AP. GPS was significantly associated with AP when adjusted for diagnostic Gleason score, but not when also adjusted for PSA density [10]. Although this was a prospective multicenter AS cohort study with a defined AS protocol using tissue from original diagnostic biopsy tissue, the sample size for the AP endpoint was small and further validation is warranted.

The University of California San Francisco (UCSF) AS cohort has also reported the value of serial GPS testing in the context of AS [11]. In 111 men on AS who underwent serial GPS testing, the GPS at first biopsy was associated with an upgrade at second biopsy; however, the second GPS was not [11]. A further study in 1031 men from the UCSF cohort found that a composite score of 3 tissue genomic markers (including GPS) predicted biopsy reclassification at first surveillance biopsy and up to 3 years after enrollment but was not associated with reclassification at 5 and 10 years [12]. These early validation studies of GPS in the context of AS are limited by their retrospective nature and the potential selection bias of those who underwent genomics testing as part of treatment.

A recently published study of a cohort of 296 men with very low-risk (37.8%) or low-risk (62.2%) disease who underwent GPS testing found that GPS score did not differ significantly across quartiles of disease volume (defined as percent of positive cores, number of cores with > 50% involvement, largest involvement of any single core, and PSA density) [13]. However, the median likelihood of favorable pathology at RP was statistically different between volume quartiles. Seven of the 105 men (6.3%) with very low-risk disease were reclassified to low-risk, and 13 of 181 (7.2%) with low-risk disease were reclassified to intermediate-risk. On univariate analysis, disease volume did not correlate with reclassification on GPS testing [13]. A further study to clarify the clinical utility of GPS in determining eligibility for AS found that no men with NCCN very low-risk disease were reclassified [14]. In this cohort, nearly one-third of men with low- or intermediate-risk

TABLE 1.
Summary of tissue genomic biomarkers used in prostate cancer

Genomic Test	Tissue Source	Clinical Requirements	Marker Measurement	Reported Outcomes
ConfirmMDx	Negative biopsy tissue	Prior negative biopsy, no ASAP	DNA methylation (<i>GSTP1</i> , <i>APC</i> , <i>RASSF1</i> , <i>ACTB</i>)	<ul style="list-style-type: none"> Any cancer [2,3] Gleason score ≥ 7 [4]
Oncotype Dx GPS	Positive biopsy tissue	Gleason 3+3 or 3+4, NCCN very low to intermediate risk	RNA (17 gene expression: <i>AZGP1</i> , <i>FAM13C</i> , <i>KLK2</i> , <i>SRD5A2</i> , <i>FLNC</i> , <i>GSN</i> , <i>GSTM2</i> , <i>TPM2</i> , <i>BGN</i> , <i>COL1A1</i> , <i>SFRP4</i> , <i>TPX2</i> , <i>ARF1</i> , <i>ATP5E</i> , <i>CLTC</i> , <i>GPS1</i> , <i>PGK1</i>)	<ul style="list-style-type: none"> Adverse pathology at RP (Gleason $\geq 4+3$ or \geq pT3a) [8,15,16] Metastasis after RP [17] Prostate cancer-specific death after RP [17] Biopsy upgrade on active surveillance [8,11] Adverse pathology at RP while on AS [10]
ProMark	Positive biopsy tissue	Gleason 3+3 or 3+4	Protein (<i>DERL1</i> , <i>CUL2</i> , <i>SMAD4</i> , <i>PDSS2</i> , <i>HSPA9</i> , <i>FUS</i> , <i>pS6</i> , <i>YBOX1</i>)	<ul style="list-style-type: none"> Unfavorable pathology at RP (Gleason $\geq 3+4$ or \geq pT3a), pN1, pM1 [18,20]
Prolaris	Positive biopsy tissue or RP	None	RNA (46 gene expression: 31 cell cycle progression genes, 15 housekeeping genes)	<ul style="list-style-type: none"> Biochemical recurrence after RP [21,23–25] Prostate cancer-specific death after RP [22] Selection for active surveillance [22] Metastasis after RP [23] Risk stratification [25] Metastasis after definitive treatment [27]
Decipher	Positive biopsy tissue or RP	None	RNA (22 gene expression: <i>NFIB</i> , <i>NUSAP1</i> , <i>ZWILCH</i> , <i>ANO7</i> , <i>PCAT-32</i> , <i>UBE2C</i> , <i>CAMK2N1</i> , <i>MYBPC1</i> , <i>PBX1</i> , <i>THBS2</i> , <i>EPPK1</i> , <i>IQGAP3</i> , <i>LASP1</i> , <i>PCDH7</i> , <i>RABGAP1</i> , <i>GLYATL1P4</i> , <i>S1PR4</i> , <i>TNFRSF19</i> , <i>TSBP</i> , 3 RNA markers not associated with genes)	<ul style="list-style-type: none"> Metastasis after RP [28-32] Prostate cancer-specific death after RP [28] Biochemical recurrence after RP [29] Prostate cancer-specific death after RP [33,34] Adverse pathology at RP [35,36]

disease were reclassified, but management decisions did not always reflect the change [14]. Thus, GPS provides additional risk stratification for patients, but is likely best reserved for patients whose clinical risk does not indicate a clear clinical recommendation for or against definitive treatment.

Prediction of adverse pathology at surgery

The panel of genes used in GPS was initially identified from 732 candidate genes selected through a meta-

analysis of publicly available microarray datasets, based on the ability to predict cancer-specific mortality across multiple foci of cancer within RP specimens. It has been validated for predicting AP on RP specimens using biopsy specimens [15]. GPS has also been validated prospectively as an independent predictor of AP at RP [16]. On multivariable analysis in models that included clinical variables (PSA, clinical stage, and biopsy grade), GPS was a significant predictor of AP at RP in both intermediate-risk and favorable

intermediate-risk groups. The AUC of the adverse pathology predictive model was 0.726 when combining CAPRA and GPS versus 0.633 with CAPRA alone [16]. This study confirms prior validations that employed a prospective-retrospective design in men who underwent immediate RP.

Prediction of post-operative outcomes

In a retrospective cohort study of 279 men treated with RP from 1995 to 2010 for localized prostate cancer spanning low to high NCCN risk groups, GPS from biopsy tissue was found to be independently associated with BCR, metastasis, and death after adjusting for CAPRA score and NCCN risk category [17]. The c-index for PCa metastasis at 10 years increased from 0.65 for CAPRA alone to 0.73 with the addition of GPS. For prostate cancer death, a similar increase of 0.78 to 0.84 was found with the addition of GPS to CAPRA [17]. However, it is unclear what impact GPS has on clinical decisions, particularly as models in the study were not adjusted for receipt of any adjuvant post-RP treatment.

ProMark

Description of assay

ProMark (Metamark Genetics, Inc., Waltham, United States) is a test that uses quantitative tissue proteomics [18] to generate a risk score from the levels of 8 target proteins, derived from a candidate biomarker study [19], for men with Gleason score 3+3 and 3+4 cancer on biopsy tissue. The score gives the probability of having AP at RP.

Prediction of adverse pathology following surgery

A clinical prognostic risk model was developed in 381 patients with biopsy and RP specimens and then validated in a separate cohort of 276 patients, assessing their proteomic panel risk against NCCN risk category and D'Amico classification [20]. In the validation cohort, the authors reported an AUC for predicting AP at RP for the proteomic panel alone was 0.68 compared with the NCCN model of 0.69 and D'Amico model of 0.65. However, when the proteomic panel was added to both the NCCN and D'Amico models, the AUCs increased to 0.75. Additionally, when the authors used clinical cutoffs for the protein panel combined with NCCN risk categories, they reported the positive predictive value for favorable pathology was 82% for NCCN intermediate-risk, 82% for low-risk, and 95% for very low-risk. Finally, a decision curve analysis was performed that found a net benefit with the addition of the protein panel for patients in NCCN intermediate- and high-risk groups, which may change treatment decision-making [20]. However, for patients with lower clinical risk disease, the ProMark proteomic panel may be more suitable to reassure and confirm that men are indeed low risk. To

date, the ProMark genomic panel has been validated only for predicting AP at RP. Although AP is associated with an overall less favorable prognosis, no study to date has validated ProMark for predicting survival or post-treatment disease recurrence.

Prolaris

Description of assay

Prolaris (Myriad Genetics, Inc., Salt Lake City, United States) is a panel that measures the RNA expression levels of 31 genes involved in cell cycle progression (CCP) relative to the expression of 15 housekeeping genes in cancerous tissue [21].

Identification of men suitable for active surveillance

A model combining CCP scores and clinical parameters, termed cell cycle risk (CCR) score, has been developed and applied to the identification of men with low-risk disease who are appropriate for AS. In a validation cohort of 585 men conservatively managed with CCR scores below the threshold, the predicted mean 10-year PCa mortality was 2.7%, and the CCR significantly dichotomized low- and high-risk disease. The potential clinical benefit of selecting men for AS using the CCR score was evaluated in a sequential, unselected cohort of 19 215 men. The proportion of men identified as candidates was substantially higher when the CCR score was used than when clinicopathologic features alone were used, while the mean 10-year predicted PCa mortality risks remained identical (1.9% versus 2.0%) [22]. However, the validation cohort in this study was composed of men who deferred curative therapy and were therefore not a "true" AS cohort as there was minimal scheduled surveillance in the absence of symptoms of clinical progression.

Prediction of post-operative outcomes

The CCP score has also been assessed on biopsy specimens of men who later underwent definitive treatment. In a cohort of 582 men who were treated with RP, biopsy CCP scores were significantly associated with BCR and metastases on multivariable analyses; it should be noted that only 2% of men developed metastases [23]. The CCP score was not significantly associated with clinical variables in any models, suggesting that it provides additional predictive information beyond traditional clinical parameters. However, the prognostic value of CCP was not directly compared with any established clinical risk calculators. In a further study using the same cohort, the CCP score provided additional prognostic value for BCR after RP in men with NCCN low-risk disease beyond the CAPRA score, with 35% of men with high CCP score experiencing BCR at 5 years. The c-index was 0.56 for the CAPRA score alone and 0.66 for CAPRA combined with CCP score.

A decision curve analysis which found that the CCP score provided net clinical benefit beyond CAPRA [24]. Therefore, using only pre-treatment data, the CCP score allows for prognostic stratification within the low-risk category, and this information could inform decision-making regarding AS and local treatments in certain patients.

Use in risk prediction

In addition to biopsy tissue, the CCP score has also been derived from RP tissue to prognosticate and stratify according to risk of BCR. In a study of 413 men who underwent RP, CCP score was able to effectively stratify patients within CAPRA-S risk groups [25]. Regardless of CAPRA-S risk group, men with a very low CCP score did not experience cancer recurrence within 5 years of RP, whereas recurrence was almost 50% in men with a high CCP score. In the overall recurrence predictive model, the c-index of CAPRA-S alone was 0.73 and increased to 0.77 with the addition of the CCP score. Therefore, the CCP score provides some additional prognostic information, beyond pathologic factors, on the risk of BCR following RP. This additional information may aid decision-making regarding adjuvant treatments.

Risk stratification is often more challenging in certain cohorts such as African American men, for whom non-clinical social determinants of health may influence receipt of treatment and ultimate oncologic outcomes [26]. In a retrospective cohort of 767 men of whom 281 (37%) were African American, the CCP score was a significant predictor of metastatic disease on multivariable analysis including clinical parameters; however, there was no interaction with ancestry or treatment [27]. Of note, ancestry was self-reported, and this may have introduced error thereby limiting the generalizability of the conclusions, as population-based genetic heterogeneity was not addressed.

Decipher

Description of assay

The Decipher genomic classifier (Decipher Biosciences, San Diego, United States) is a genomic assay based on a full transcriptome microarray, including both protein coding and non-coding RNA, and measures RNA expression levels from 22 genes that were initially selected from 1.4 million candidate RNA probes [28].

Prediction of post-operative outcomes

Decipher was initially validated for predicting metastases in a cohort of 545 patients who underwent RP at the Mayo Clinic, of whom 213 subsequently developed metastases. Decipher has been further validated in multiple studies for predicting outcomes after RP [28]. In a study of 85 men who developed BCR following RP, Decipher predicted metastases with an AUC of 0.82, and

in modeling with clinicopathologic variables, Decipher was the only significant predictor of metastasis [29]. In a larger cohort of 260 men with intermediate- and high-risk disease at the time of RP who did not receive adjuvant treatment, Decipher added prognostic accuracy to the CAPRA-S for estimating metastatic disease at 10 years. The c-index of Decipher and CAPRA-S alone was 0.76 and 0.77, respectively; when Decipher and CAPRA-S were combined, the c-index improved to 0.87. The greater net benefit of the combination was also confirmed with decision curve analysis [30]. A similar finding was observed in a cohort of 169 men that included 15 who had metastatic progression within 5 years of RP. In multivariable analysis, after adjusting for clinical risk factors, Decipher predicted metastases and also had the highest c-index (0.77), compared with CAPRA-S and Stephenson nomogram, as well as a panel of previously reported, unrelated prostate cancer biomarkers [31].

Use in risk stratification

Decipher has also been assessed in determining risk in men with a persistently detectable PSA after RP, as a subset of these patients likely harbor metastatic disease. In a cohort of 477 men who underwent RP at 3 academic centers, 150 had an immediately detectable post-operative PSA. The 5-year metastasis rate was 0.90% for Decipher low/intermediate and 18% for Decipher high-risk [32]. On multivariable analysis, only Decipher high-risk, detectable PSA, and lymph node invasion remained prognostic factors for metastasis [32]. The sample size of men with a detectable PSA post-RP in this study may have limited the power to detect differences in various prognostic variables.

Decipher has also been shown to prognosticate prostate cancer death. Combining CAPRA-S and Decipher predicted prostate cancer death at 5 years after RP in 185 men with high-risk disease. The combination of CAPRA-S and Decipher had greater clinical benefit on decision curve analysis (DCA) than either alone [33]. Supporting this finding, a multicenter study of 561 men with adverse pathologic features at RP, the combination of Decipher and CAPRA-S improved the prediction of 10-year prostate cancer death following RP (c-index 0.73) compared with either test alone (c-index 0.73 for each) [34]. DCA also confirmed the net benefit of Decipher combined with CAPRA-S for 10-year prostate cancer mortality [34].

Decipher has also been applied to prostate biopsies from patients to predict AP at RP [35]. In a retrospective multicenter cohort of 266 men with NCCN very low-, low- and favorable intermediate-risk PCa, Decipher from diagnostic biopsies was an independent predictor of AP at RP. This study did not have long-term follow-up to evaluate survival outcomes, and the sample

size and low number of events did not allow Decipher to be assessed in the individual NCCN risk (eg, favorable intermediate only) groups. A further study using this cohort also found that NCCN favorable intermediate-risk disease with Decipher low or intermediate score was not associated with significantly higher odds of AP compared to very low-risk/low-risk disease [36].

Taken together, these studies demonstrate that the Decipher has a prognostic value for both metastatic disease and PCa mortality following RP. This can potentially assist patients and clinicians in decision-making around adjuvant therapy—if a patient's Decipher risk for metastasis and prostate cancer death is low, then he could potentially forgo adjuvant treatments, even if his pathological characteristics are less favorable.

Emerging tissue biomarkers

A benefit of the technology from GenomeDx, is the ability to study other RNA expression signatures, which may be associated with outcomes beyond the scope of the original gene expression panel. Retrospectively, GenomeDx has used their technology to identify signatures related to other treatment modalities such as radiation, androgen deprivation therapy, and chemotherapy sensitivity. Zhao et al. identified a 24-gene molecular signature that predicts patient response to adjuvant radiotherapy following radical prostatectomy. The gene signature, termed the Post-Operative Radiation Therapy Outcomes Score (PORTOS) was identified from patients included in 1 of 5 published studies of men with prostate cancer who had radical prostatectomy (with or without

post-operative radiotherapy) and had gene expression analysis of the tumor, with long-term follow-up and for whom complete clinicopathological data were available. The authors demonstrated that in those patients who had a high PORTOS score, those who had radiotherapy had a lower 10 year metastases rate than those who did not (5% versus 63%, HR 0.12, $P < 0.001$) [37]. PORTOS remains to be validated prospectively, and its association with RT sensitivity has been shown only in post-RP patients. Therefore, further investigation is required to validate the gene panel prospectively and in the setting of RT for primary treatment of prostate cancer.

Future Challenges and Directions

Many of the growing number of tissue-based genomic tests are clinically validated to improve on clinical parameters alone by more accurately determining prognosis and specific oncologic outcomes. However, whether a genomic test may be predictive of a clinically significant response to a particular management strategy is a more complex question. Many of the genomic tests outlined have been shown to predict multiple endpoints, including AP, BCR, metastasis-free survival, and/or cancer-specific mortality, but one endpoint is not a surrogate for another. The temporal associations of many tissue genomic tests and outcomes remain obscure, but there are current prospective trials designed to address this issue.

Tissue-based genomic tests have the potential to optimize the care for men with prostate cancer, but determining the optimal genomic test for each patient to ensure the best outcome requires continued study.

References

1. Van Neste L., Bigley J, Toll A, et al. A tissue biopsy-based epigenetic multiplex PCR assay for prostate cancer detection. *BMC Urol*. 2012;12:16.
2. Stewart GD, Van Neste L, Delvenne P, et al., Clinical utility of an epigenetic assay to detect occult prostate cancer in histopathologically negative biopsies: results of the MATLOC study. *J Urol*. 2013; 189(3):1110-6.
3. Partin AW, Van Neste L, Klein EA, et al. Clinical validation of an epigenetic assay to predict negative histopathological results in repeat prostate biopsies. *J Urol*. 2014;192(4):1081-7.
4. Waterhouse RL Jr, Van Neste L, Moses KA, et al. Evaluation of an epigenetic assay for predicting repeat prostate biopsy outcome in African American men. *Urology*. 2019;128:62-65.
5. Knezevic D, Goddard AD, Natraj N, et al. Analytical validation of the Oncotype DX prostate cancer assay - a clinical RT-PCR assay optimized for prostate needle biopsies. *BMC Genomics*. 2013;14: 690.
6. Mohler JL, Antonarakis ES, Armstrong AJ, et al. Prostate Cancer, Version 2.2019, NCCN Clinical Practice Guidelines in Oncology. *J Natl Compr Canc Netw*. 2019;17(5):479-505.
7. Eggner SE, Rumble RB, Armstrong AJ, et al. Molecular biomarkers in localized prostate cancer: ASCO guideline. *J Clin Oncol*. 2020;38(13):1474-1494.
8. Kornberg Z, Cowan JE, Westphalen AC, et al. Genomic Prostate Score, PI-RADS version 2 and progression in men with prostate cancer on active surveillance. *J Urol*. 2019;201(2): 300-307.
9. Kornberg Z, Cooperberg MR, Cowan JE, et al. A 17-gene genomic prostate score as a predictor of adverse pathology for men on active surveillance. *J Urol*. 2019; 202(4):702-709. doi: 101097/ju0000000000000290
10. Lin DW, Zheng Y, McKenney JK, et al. 17-Gene Genomic prostate score test results in the Canary Prostate Active Surveillance Study (PASS) Cohort. *J Clin Oncol*. 2020; 10;38(14):1549-1557. doi: 10.1200/JCO.19.02267

11. Cedars BE, Washington SL 3rd, Cowan JE, et al. Stability of a 17-gene genomic prostate score in serial testing of men on active surveillance of early stage prostate cancer. *J Urol.* 2020(4):696-701. doi: 10.1097/JU.0000000000000271
12. Lonergan PE, Washington SL 3rd, Cowan JE, et al. Risk factors for biopsy reclassification over time in men on active surveillance for early stage prostate cancer. *J Urol.* 2020: 101097JU00000000000001186.
13. Nyame YA, Grimberg DC, Greene DJ, et al. Genomic scores are independent of disease volume in men with favorable risk prostate cancer: implications for choosing men for active surveillance. *J Urol.* 2018;199(2):438-444.
14. Gaffney C, Golan R, Cantu MD, et al. The clinical utility of the genomic prostate score in men with very low to intermediate risk prostate cancer. *J Urol.* 2019; 202(1): 96-101.
15. Klein EA, Cooperberg MR, Magi-Galluzzi C, et al. A 17-gene assay to predict prostate cancer aggressiveness in the context of Gleason grade heterogeneity, tumor multifocality, and biopsy undersampling. *Eur Urol.* 2014; 66(3):550-60.
16. Eggener S, Karsh LI, Richardson T, et al. A 17-gene panel for prediction of adverse prostate cancer pathologic features: prospective clinical validation and utility. *Urology.* 2019;126:76-82.
17. Van Den Eeden SK, Lu R, Zhang N, et al. A biopsy-based 17-gene genomic prostate score as a predictor of metastases and prostate cancer death in surgically treated men with clinically localized disease. *Eur Urol.* 2018;73(1):129-138.
18. Shipitsin M, Small C, Giladi E, et al. Automated quantitative multiplex immunofluorescence in situ imaging identifies phospho-S6 and phospho-PRAS40 as predictive protein biomarkers for prostate cancer lethality. *Proteome Sci.* 2014; 12:40.
19. Shipitsin M, Small C, Choudhury S, et al. Identification of proteomic biomarkers predicting prostate cancer aggressiveness and lethality despite biopsy-sampling error. *Br J Cancer.* 2014;111(6): 1201-12.
20. Blume-Jensen P, Berman DM, Rimm DL, et al. Development and clinical validation of an in situ biopsy-based multimarker assay for risk stratification in prostate cancer. *Clin Cancer Res.* 2015; 21(11):2591-600.
21. Cuzick J, Swanson GP, Fisher G, et al. Prognostic value of an RNA expression signature derived from cell cycle proliferation genes in patients with prostate cancer: a retrospective study. *Lancet Oncol.* 2011;12(3):245-55.
22. Lin DW, Crawford ED, Keane T, et al. Identification of men with low-risk biopsy-confirmed prostate cancer as candidates for active surveillance. *Urol Oncol.* 2018;36(6):310 e7-310 e13.
23. Bishoff JT, et al. Prognostic utility of the cell cycle progression score generated from biopsy in men treated with prostatectomy. *J Urol.* 2014;192(2):409-14.
24. Tosoian JJ, Chappidi MR, Bishoff JT, et al. Prognostic utility of biopsy-derived cell cycle progression score in patients with National Comprehensive Cancer Network low-risk prostate cancer undergoing radical prostatectomy: implications for treatment guidance. *BJU Int.* 2017; 120(6):808-814.
25. Cooperberg MR, Simko JP, Cowan JE, et al. Validation of a cell-cycle progression gene panel to improve risk stratification in a contemporary prostatectomy cohort. *J Clin Oncol.* 2013;31(11):1428-34.
26. Kagawa-Singer M, Valdez Dadia A, Yu MC, et al. Cancer, culture, and health disparities: time to chart a new course? *CA Cancer J Clin.* 2010; 60(1):12-39.
27. Canter DJ, Reid J, Latsis M, et al. Comparison of the prognostic utility of the cell cycle progression score for predicting clinical outcomes in African American and Non-African American men with localized prostate cancer. *Eur Urol.* 2019;75(3):515-522.
28. Erho N, Crisan A, Vergara IA, et al. Discovery and validation of a prostate cancer genomic classifier that predicts early metastasis following radical prostatectomy. *PLoS One.* 2013;8(6):e66855.
29. Ross AE, Feng FY, Ghadessi M, et al. A genomic classifier predicting metastatic disease progression in men with biochemical recurrence after prostatectomy. *Prostate Cancer Prostatic Dis.* 2014; 17(1):64-9.
30. Ross AE, Johnson MH, Yousefi K, et al. Tissue-based genomics augments post-prostatectomy risk stratification in a natural history cohort of intermediate- and high-risk men. *Eur Urol.* 2016;69(1):157-65.
31. Klein EA, Yousefi K, Haddad Z, et al. A genomic classifier improves prediction of metastatic disease within 5 years after surgery in node-negative high-risk prostate cancer patients managed by radical prostatectomy without adjuvant therapy. *Eur Urol.* 2015;67(4):778-86.
32. Spratt DE, Dai DLY, Den RB, et al. Performance of a prostate cancer genomic classifier in predicting metastasis in men with prostate-specific antigen persistence postprostatectomy. *Eur Urol.* 2018;74(1):107-114.
33. Cooperberg MR, Davicioni E, Anamaria Crisan A, et al. Combined value of validated clinical and genomic risk stratification tools for predicting prostate cancer mortality in a high-risk prostatectomy cohort. *Eur Urol.* 2015;67(2):326-33.
34. Karnes RJ, Choeurng V, Ross AE, et al. Validation of a genomic risk classifier to predict prostate cancer-specific mortality in men with adverse pathologic features. *Eur Urol.* 2018;73(2):168-175.
35. Kim HL, Li P, Huang H-C, et al. Validation of the Decipher test for predicting adverse pathology in candidates for prostate cancer active surveillance. *Prostate Cancer Prostatic Dis.* 2019; 22(3):399-405.
36. Herlemann A, Huang H-C, Alam R, et al. Decipher identifies men with otherwise clinically favorable-intermediate risk disease who may not be good candidates for active surveillance. *Prostate Cancer Prostatic Dis.* 2020;23(1):136-143. doi: 10.1038/s41391-019-0167-9
37. Zhao SG, Chang SL, Spratt DE, et al. Development and validation of a 24-gene predictor of response to postoperative radiotherapy in prostate cancer: a matched, retrospective analysis. *Lancet Oncol.* 2016;17(11):1612-1620.