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MOLECULAR CORRELATES OF INTERMEDIATE AND HIGH RISK LOCALIZED PROSTATE CANCER

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Abstract

Clinicopathologic parameters, including Gleason score, remain the most validated prognostic factors for patients diagnosed with localized prostate cancer. However, patients of the same risk groups have exhibited heterogeneity of disease outcomes. To improve risk classification, multiple molecular risk classifiers have been developed, which were designed to inform beyond existing clinicopathologic classifiers. Alterations affecting tumor suppressors and oncogenes, such as *PTEN*, *MYC*, *BRCA2*, and *TP53*, which have been long associated with aggressive prostate cancer, demonstrated grade-dependent frequency of alterations in localized prostate cancers. In addition to these genetic hallmarks, several RNA-based commercial tests have been recently developed to help identify men who would benefit from earlier interventions. Large genomic studies also correlate germline genetic alterations and epigenetic features with adverse outcomes, further strengthening the link between the risk of metastasis and a stepwise accumulation of driver molecular lesions.

I. INTRODUCTION

Despite being the most common visceral malignancy in American men, with approximately 161,000 new diagnoses in 2017, the fact that less than 20% of men diagnosed with prostate cancer (PCa) die from this disease [1] emphasizes the dual contradictions that while PCa screening and effective interventions are saving lives [2], a substantial proportion of the remaining 80% were never at risk of PCa-specific mortality. For both the patient and his physician, determining which factors to take into account when attempting to predict the likelihood of developing aggressive disease can be a daunting prospect. The challenge of

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classifying the tumor correctly and determining the risk of disease progression requires adequate tissue sampling as well as applying the appropriate risk-stratification tools.

The frequent and extensive intratumoral heterogeneity of PCa represents a major confounding consideration when assessing an initial diagnosis [3, 4]. 85–90% of prostate cancers are multifocal [5]. Branching morphogenesis that occurs during the development of the embryonic prostate organ remains at play during development of each individual prostate tumor [6]. Extensive divergence from a common cancer cell ancestor can result in multiple distinct yet co-existing tumor subclones, and prognostic features identified in one of these subclones may not be representative of others. Consequently, disease aggressiveness predicted by the gain of one or more molecular alterations must be considered in the context of what may have occurred truncally (*i.e.* shared by all subpopulations) thus being captured by biopsy, or what is subclonal and may be only present in the unsampled portions of the tumor. Therefore, a deeper understanding of the heterogeneous nature of PCa and mechanisms of molecular progression can improve the application of molecular ancillary tests for risk stratification and patient management.

All prostate biopsies, whether templated or MRI/ultrasound-fusion guided, suffers from the intrinsic challenge of tumor heterogeneity in that a high grade component might be under- or over- represented, due to limited sampling [7, 8]. Historically, the greatest value of biopsy was in the positive predictive power of the pathology (tumor grade and volume) for disease aggressiveness, guiding patient counseling and decision making. Historic undersampling is expected to decrease by application of advanced imaging modalities such as multi-parametric MRI, which has an increased sensitivity in detecting higher grade disease [9–11]. Based on the findings that the adjacent Gleason grade 3 and grade 4 components in intermediate to high risk PCa are clonally related and share some common molecular features, commercial molecular tests and in-house ancillary tests have been developed and increasingly used to predict final pathology on radical prostatectomy (RP). Moreover, alterations of a handful of driver genes may indicate molecular progression, which we define here as the stepwise accumulation of genomic or genetic alterations that may accompany changes in histopathology grades and are closely associated with disease progression. Finally, molecular alterations detected in the RP tumor tissue can offer prognostic value in determining which patients would benefit from adjuvant radiation therapy.

II. GLEASON GRADE 3 AND 4 COMPONENTS IN INTERMEDIATE TO HIGH RISK PROSTATE CANCERS ARE CLONALLY RELATED

Intratumoral heterogeneity was the basis for establishing a pathologic grading system (the Gleason score, GS) fifty years ago [12]. Indeed, the Gleason score’s consideration of multiple simultaneous cancer differentiation statuses remains the single best, and most validated, parameter for assessing PCa prognosis [4]. Both the original Gleason scoring system and the modified grade grouping system [13] assign an overall score by adding the most predominant and the second predominant architectural growth pattern, each reflecting distinct underlying molecular alterations. Following a 2014 International Society of Urological Pathology (ISUP) consensus, GS of 3+3=6 was reclassified into the lowest Grade

Group (GG) of 1, GS 3+4=7 to GG2, 4+3=7 to GG3, 4+4=8 to GG4, and any amount of Gleason pattern (GP) 5 (4+5, 5+4, or 5+5), into the highest GG of 5 [13, 14]. These grade groupings reflect increased recognition that the quantity of GP4 present (and further distinguishing 3+4 from 4+3) is highly significant, stratifying risk of recurrence free survival 5 years after RP from ~90% to ~60%. For example, what may be a small group of cancer cells buried within a tumor of GS7 (or their unsampled progenitor) could potentially be the precursor for a distant metastasis; while morphologically identical to their sister cells, these cells could harbor molecular alterations that predispose the patient to an adverse outcome.

It was the Gleason grade's prognostic power that prompted several research groups to assess patterns of mutation and gene expression to ask what makes GP4 behave worse than GP3, and if found together, whether they were clonally related. Next-generation sequencing permitted single-base resolution of the *TMPRSS2-ERG* breakpoint to be used as a definitive clonal marker to demonstrate a common somatic ancestry in ERG-positive GP3 and GP4 foci, in a series of four GS7 cancers [15]. Moreover, *PTEN* copy number was noted as a potential mechanism of Gleason progression in two cases with clonal *ERG* breakpoints. The finding of clonal relationship between adjacent GP3 and GP4 was confirmed by a study by Kovtun and colleagues, examining global genomic breakpoints in 14 cases of GS7 PCa [16].

Two independent studies subsequently revealed that the GP3 and GP4 components diverge very early in the development of intermediate to high risk PCa. VanderWeele et al. examined the lineage relationship of GP3 and GP4 tumors using whole exome sequencing for higher resolution of shared (and distinct) somatic variants and performing phylogenetic reconstruction in four cases of GS7 cancer, including lymph node metastases in two patients [17]. In addition to tumors showing evidence of common ancestry, fewer mutations were shared between low-grade and high-grade foci versus high-grade and metastastic foci, indicating early divergence. A similar conclusion was made in a separate study of 12 GS≥7 cases [18], demonstrating GP3 cancer diverged early from a common ancestor that also had given rise to GP4 disease. Furthermore, the authors identified that increased MYC activity mediates progression from GP3 to GP4 in a subset of tumors. With regard to the precursor lesions of adjacent GP3 and GP4, deeper resolution of data from multi-region sequencing studies suggested that the true origin of these tumors is morphologically normal tissue, representing the "field effect" or somatic mosaicism in the earliest stages of PCa evolution [19, 20]. In addition, somatic alterations in high-grade prostatic intraepithelial neoplasia (PIN) have been shown to indicate a distant clonal relationship between PIN and adjacent GP3 and GP4 disease [21, 22]. However, whether alterations present in an aggressive precursor can set a fate of the tumor to progress rapidly is yet to be determined.

Approximately 95% of men with GS6 cancer are cured after undergoing RP [14]. From the perspective of molecular progression, the 5% that do recur may harbor some volume of aggressive disease that establishes a micrometastasis prior to treatment. These tumor cells may have taken the form of either (a) an unsampled GP4 component or (b) morphologically GP3 tumors that had underwent further occult oncogenic alterations as the source of potentially lethal disease. With the latter being exceedingly rare, the converse argument may also be made about those men with GS≥7 (4+3) PCa upon RP who never experience relapse: (a) either their tumor had aggressive potential but was resected early enough to prevent

micrometastastic spread, or (b) despite being morphologically high grade, it had not yet undergone oncogenic transformations required for metastasis and thus resection could have been delayed. These examples, which are not uncommon, demand investigation to identify distinct molecular signatures of aggressive GP3 and indolent GP4.

III. MOLECULAR CLASSIFIERS THAT PREDICT UPGRADING FROM BIOPSY TO RADICAL PROSTATECTOMY

Intratumoral heterogeneity and sampling limitations by biopsy are significant hurdles when determining which men are the best candidates for surveillance. On the basis of pathology alone, numerous studies show that a high volume of GS6 is more likely to result in a GS upgrade upon RP, thus the finding may sway patients from surveillance to surgery [23]. Indeed, there is excellent concordance between biopsy tumor volume and RP tumor volume, as well as between RP tumor volume and RP GS [23]. However, the many cases of high volume GS6 on biopsy which do not upgrade on RP beg the question whether molecular alterations in a GP3 tumor could predict the presence of unsampled higher grade disease in the prostate. A similar question is whether molecular features of a small amount of GP4 tumor detected on biopsy could predict the presence of a high-volume GP4 or its capacity to quickly develop into lethal disease.

To identify differences between indolent versus lethal GS7 tumors, Penney and colleagues harnessed mRNA from the Swedish Watchful Waiting cohort (n=358) and the Physicians' Health Study (n=109) [24]. Comparing GS6 to GS8–10, a 157-gene signature was derived to predict lethal disease, with predictive power to reclassify ambiguous GS7 cancers as lethal or indolent (odds ratio of 1.47 ±0.47). Surprisingly this signature did not contain any of the expected genes historically associated with PCa, such as *MYC*, *PTEN*, or *AR*, but gene set enrichment analysis identified the cell cycle pathway and NOTCH pathway, nominated in part due to differential expression of genes such *CDKN3* and *NOTCH3* [24].

Nonetheless, the recurrent identification of a set of genes that enabled the tumor cells undergo cell division and withstand the terminally-differentiating effects of the Androgen Receptor (AR) in aggressive tumors led to the development of the RNA-based Cell Cycle Progression (CCP) score, marketed as Prolaris by Myriad Genetics, [25]. Informing on an underlying proliferative biology of a potentially-aggressive tumor, this 45-gene assay consists of 31 CCP genes, including *RAD51*, *BUB1B*, and *TOP2A*, plus 15 housekeeping genes [26], and has been extensively evaluated as a risk assessment tool for guiding enrollment into active surveillance. When biopsied tumors are subjected to this qRT-PCR assay, each unit of increase to the CCP (aggressiveness) score doubles the risk of progression [26]. Importantly, the CCP score was developed using multivariate analyses independent of clinicopathologic prognosticators such as GP [26]. Thus, applying CCP as evidence of molecular progression in conjunction with other clinical variable nomograms such as the CAPRA score [27] adds prognostic value to all risk categories [26].

The likelihood of upgrading from biopsy to RP, or having pT3 disease on RP, is addressed by the Oncotype DX commercial test from Genomic Health. The RNA, RT-PCR-based Oncotype DX score is based on the expression of 17 genes (12 cancer plus 5 housekeeping)

that account for stromal content cellular organization, AR activity, and proliferation [28]. These genes were selected based on their expression being predictive of clinical recurrence independent of GP, suggesting that molecular progression involves altered expression of genes in co-dependent pathways reflecting reorganization of the tumor biology beyond proliferation [29]. The resulting Genomic Prostate Score (GPS) presents a risk, from 0% to 100%, of harboring unsampled higher grade disease that may warrant further biopsy, imaging, or resection [28]. In separate studies, when the genes that drive the GPS were applied to RNA sampled from different patients, the GPS showed minimal intraprostatic heterogeneity across different Gleason patterns in the same individual [18, 30]. A plausible explanation for these findings is that the selected genes are independent of histology and are influenced by the underlying biology of the tumor, resulting in similar GPS yielded from clonally-related GP3 and GP4 foci. Although prospective studies are still in progress, interim analysis after at least one year supports that men with low GPS, who show no evidence of molecular progression, can be enrolled and managed safely in surveillance programs [31].

Although gene expression tests may show homogeneity amongst pathologically diverse foci, examination of somatic genomic (DNA-level) changes paints a very different picture. Despite a common clonal relationship, GP3 and GP4 foci in the same individual diverge extensively from their common progenitor, acquiring private somatic mutations and copy number changes [17, 18, 30]. While clonal progression from GP3 to GP4 in some cases was associated with single copy loss of *PTEN, PTEN* loss itself was not sufficient for GP4 development, as GP3 foci also harbored single copy losses of *PTEN* in other cases [15]. Extensive evolutionary branching not surprisingly results in alterations to other cancer-related genes, including *MYC*, *BRCA1*, *BRCA2*, *NKX3-1*, *TP53*, and *HRAS* – in some cases these are shared by clonally-related GP3 and GP4 foci, and in some cases these alterations are private to GP3 foci, despite similar alterations are often found in higher grade disease [18]. Therefore, if single copy deleterious mutations in classical tumor suppressors are observed in GP3 foci, multiple region sampling is necessary to verify whether those deletions are truncal and thus likely clinically significant [19]. Nonetheless, PTEN loss is probably the most reliable single genetic or protein-level marker of Gleason upgrading, as a study of 7,813 RP cases has shown that 14% of GS3+3=6 tumors harbor PTEN loss by immunohistochemical (IHC) staining, in contrast to 21% in GS7 (3+4), 38% in GS7 (4+3), and 41% in GS≥8 tumors [32]. This large-scale study result was consistent with prior work from the same investigators, showing that PTEN protein loss by IHC predicts upgrading from GS6 on biopsy to any GS7 on RP, with an age-adjusted odds ratio of 2.93 (P=0.034) [33].

While the aforementioned studies were not designed to assess the power of molecular alterations in sampled tissue to predict pathology of unsampled tissue, this key issue was addressed by Trock and colleagues [34], who generated tissue microarrays from RP specimens with 3+3=6, 3+4=7, and 4+3=7 tumors, with tissue cores only taken from the GP3 regions. Because the prostate TCGA study showed recurrent losses in chromosome 8p22 (*LPL*) and 10q23 (*PTEN*) and gain in 8q24 (*MYC*), expression of these three genes was assayed by IHC or *in situ* hybridization (ISH) staining, and analyzed for correlation with the final GS of each RPs. In univariate models, any amount of GP3 harboring PTEN

loss by IHC, *MYC* gain by ISH, or *LPL* loss by ISH, predicted a GS of 7 versus 6 on RP with $P<0.05$ and odds ratios of 4.99, 5.36, and 3.96, respectively [34]. PTEN/*MYC* and *MYC*/*LPL* multivariate models were also predictive of GS7, and univariate modeling showed statistical significance to predict 4+3=7 but not 3+4=7, with odds ratios of 5.50, 6.00, and 9.00, respectively [34]. Taken together, detection of molecular alterations in PTEN, *MYC*, and *LPL* can be used to predict Gleason upgrading for patients with GS6 detected on biopsy.

Conversely, are there any histologic or molecular features of GP4 detected on biopsy can predict favorable pathology on RP? Indeed, there is increasing evidence that patients with small volume GS 3+4=7 prostate cancers can be managed safely by active surveillance [35]. In the largest study to date, Siadat and colleagues assessed 320 autopsy prostate specimens and 248 cystoprostatectomy specimens, all with incidental findings of PCa [36]. In this retrospective series, 11% of the autopsy and 7% of the cystoprostatectomy cases harbored GS7 or higher disease, with the major contributing GP4 component being small fused glands (8% and 2%, respectively). While overall upstaging has been reported in 30% of men who harbor Gleason 3+4=7 on biopsy, attention should be particularly given to patients who harbor large cribriform architecture GP4 on biopsy as it has been recognized as a highly aggressive variant of GP4 more likely to lead toward adverse outcomes [37]. In another study of 1,275 patients, poorly-formed GP4 was associated with a lower risk of recurrence, indicating this variant is not as aggressive as variants like cribriform GP4 [38]. Notably, approximately 10% of the GS7 patients in the prostate TCGA cohort [39] harbored minimal genomic changes, raising the question whether a more silent genomic landscape, independent of GS, may represent an indolent behavior, and if “good” GP4 such as fused and poorly-formed variants harbor molecular features that are distinct from the “bad” (cribriform architecture) GP4.

IV. MOLECULAR CLASSIFIERS THAT PREDICT POST-PROSTATECTOMY RECURRENCE

Biochemical recurrence (BCR), rising serum PSA after definitive therapy, indicates the presence of castration-sensitive occult tumor metastases that are invisible to most imaging modalities. To provide a window into the molecular landscape of recurrent, potentially lethal PCa, investigators have analyzed molecular features of primary PCa tissue from patients who had regional lymph node metastases, or who developed BCR or biopsy-proven metastases after receiving definite treatment.

When examining the spectrum of alterations shared between localized and metastatic PCas, genomic losses and/or deleterious mutations of classical tumor suppressors including *TP53*, *RB1*, and *PTEN* are present in both groups [39, 40]. However, these mutations are significantly enriched in the metastatic cohort [39, 40]. More strikingly, alterations to genes mediating DNA homologous recombination (HR), namely *BRCA1*, *BRCA2*, and *ATM,* are enriched in the metastatic tumors [40, 41]. While single-copy losses to these genes or loss-of-function germline mutations are frequently observed in men presenting with high-grade localized disease, metastatic tumors more frequently harbor secondary somatic events to

these genes in addition to germline alterations [40, 41]. These findings indicate that prostate cancers that select for HR defects early are predisposed to disease progression along a stepwise path.

To further assess this genetic predisposition to metastasis, Taylor and colleagues [42] examined molecular features of extremely aggressive PCa, which harbored intraductal carcinoma (IDC), by multi-region sequencing. IDC is a histologic feature appreciated for its significant correlation with high recurrence rates independent of GS [43]. When examining a series of 10 cases, they found that the IDC component shared a common somatic ancestor with co-existing invasive carcinoma. Importantly, in the 4 cases that also harbored a germline *BRCA2* deficient allele, specific molecular events were found to occur earlier in tumorigenesis, including amplifications of *MED12*, *MED12L*, *MYC*, and chromosome 3q. In contrast, *TP53*, *PTEN*, and *RB1* were lost earlier if *BRCA2* alterations occurred sporadically [42]. Genomically, both classes of tumor resembled aggressive, metastatic disease even in their localized stage, including increased number of mutations overall and a greater percentage of the genome involved in somatic copy number alterations [42]. The contextual importance of these findings is highlighted by the IMPACT study's initial screening results: targeting PCa screening to men with *BRCA1* or *BRCA2* carrier status significantly enriched for aggressive high grade localized disease [44]. Therefore, the findings of localized PCa carrying mutations that are typically found in metastases, either in the form of germline or sporadic mutations, warrant better sampling and/or earlier aggressive treatment.

Chua and colleagues assessed the presence of IDC as well as cribriform architecture (CA) and biochemical recurrence rates in a Canadian cohort and a cohort from Memorial Sloan Kettering Cancer Center (MSKCC), in a total of 1,325 cases [45]. The presence of IDC/CA independently predicted increased risk of biochemical recurrence in both cohorts, with hazard ratios of 2.17 and 2.32 for the Canadian and MSKCC cohorts, respectively. Biological features distinguishing IDC/CA+ tumors from IDC/CA- tumors were tumor hypoxia (64.0% vs. 45.5%, although P=0.17), a greater percentage of the genome alteration (7.2% vs. 3.0%, P<0.001) and increased tumors with clustered chromosomal rearrangements, known as chromothripsis (34.2% vs. 16.0%, P=0.033). IDC/CA+ tumors expressed only one gene threefold greater than IDC/CA- tumors: the noncoding RNA *SChLAP1* [45]. Outlier high expression of *SChLAP1* was initially discovered in studies of metastatic versus primary disease [46]; its high expression by *in situ* hybridization in RP tissue is associated with poor outcome after RP (HR=2.343, P=0.005) [47]. Mechanistic investigations have revealed that *SChLAP1* functions to enhance Polycomb Repressive Complex 2 function, which in turn dysregulates genes maintaining AR-driven terminal differentiation [48].

In-depth genomic interrogation of localized prostatic adenocarcinomas with *a priori* known recurrence status by Fraser et al. [3] revealed a paucity of somatic single nucleotide variants, consistent with the reported low overall mutation rate of PCa [39, 49]. In those tumors from men who had recurred, there were only six genes mutated recurrently in at least 2% of patients, including *SPOP*, *MED12*, *FOXA1*, and *TP53*. Not surprisingly, this study confirmed that mutations to *AR* are extremely rare in castration-sensitive disease [40].

Across 200 whole genomes, 277 additional whole exomes, 73 transcriptomes and 104 methylomes, the authors defined 40 molecular predictors of recurrence, including mutation density, localized substitution hypermutation (kataegis), chromothripsis and methylation [3]. Their predictive model was dominated by point mutations to *ATM* (HR=5.62), intrachromosomal translocations in chromosome 7 (HR=3.09), *MYC* amplification (HR=2.79), and hypermethylation of *TCERG1L* at the 5′ end (HR=2.90). Interestingly, differential methylation overall was the factor most tightly-associated with recurrence, suggesting that likelihood of recurrence may be driven in part by epigenetic plasticity.

In contrast to deep genomic sequencing, RNA and protein-based markers are faster to analyze and more cost-efficient for risk prediction. While *SChLAP1* expression independently predicts recurrence, the 22-gene Decipher assay is a robust classifier was built by deep examination of the whole transcriptome based on expression of individual Affymetrix Human Exon 2.0 microarray probes. This assay was developed initially to predict early metastasis following RP by comparing transcriptomes of 545 men, of whom 213 developed early clinical metastasis [50]. However, there is no obvious mechanism implied by the Decipher probes (which include the coding region of *MYBPC1*, an intron of *TNFRSF19* and an untranslated region of *ZWILCH*) [50]. Applied clinically, the Decipher assay was recently validated to predict rates of PCa-specific mortality with an increased hazard ratio of 1.37 for each 10% increase in the Decipher score, using RNA extracted from earlier biopsy specimens in men who had already undergone RP or first line radiation therapy (RT) plus ADT [51]. Multi-institutional validation was employed retrospectively using archival tissue, and high Decipher scores were associated with a benefit from adjuvant RT, while lower scores saw minimal differences between salvage vs. adjuvant RT [52, 53]. Thus, the Decipher assay may be useful in determining which patients may benefit from earlier RT.

With the genomic-outcome correlation studies connecting biopsy findings, RP findings, and metastatic disease, can we infer metastatic potential by studying molecular features on biopsy? Guedes and colleagues assessed frequency of PTEN protein loss by IHC on a series of 260 archival cases with Gleason 3+4=7 biopsy, and analyzed its association with pathologic stage T3 on matched RP specimens, an indicator of increased metastatic potential [54]. Tumors showing PTEN loss were nearly two times (52% vs. 27%, $P<0.001$) more likely to have non-organ confined disease at the time of RP, versus those men who had intact PTEN. The same group of investigators also reported that P53 missense mutation on biopsy may predict an increase risk of metastasis [55]. Noticing that gain-of-function missense mutations to *TP53* are enriched in metastatic PCa [40] but relatively rare in unselected primary tumors [39], Guedes and colleagues first established that p53 nuclear accumulation on IHC was a surrogate marker for *TP53* missense mutations [55]. They then examined 40 high-risk biopsies with GS9-10 tumors and found that 13% had p53 nuclear accumulation on IHC. They also examined a cohort of 195 men who had undergone RP followed by BCR. Positive staining of p53 nuclear accumulation was enriched in GS9-10 tumors (20%) and predicted metastasis with a univeraiate HR of 4.14 (95% CI, 2.41–7.11) [55]. Taken together, *PTEN* loss and/or *TP53* mutation is an early event driving progression in high risk PCa and can be detected on prostatic core biopsies. PTEN loss and/or p53 nuclear accumulation should be considered amongst exclusion criteria for enrollment into active

surveillance for otherwise eligible patients, as staining of diagnostic tissues offers substantial prognostic value at minimal cost, while characterizing alterations to genes with known biological importance to PCa progression.

V. CONCLUSION

While decades of outcome studies have generated reproducible risk stratification algorithms based on conventional clinicopathologic parameters for men diagnosed with localized prostate cancer, our increased understanding of the molecular events driving tumor evolution and progression have improved PCa risk assessment. Consequently, when the appropriate molecular test is applied, risk of progression can be adjusted in either direction, offering the patient confidence in his individualized treatment plan. However, we are likely approaching a saturation point for addressing molecular progression via discovery research using a genomic approach, with some signatures surviving retrospective, multi-institutional validation. As ancillary tests performed by CLIA-regulated pathology laboratories and commercial genomic tests become eligible for Medicare or private insurance reimbursement, consideration should now be given to initiate prospective studies to assess whether morbidity or mortality of PCa will be improved by incorporating these assays. Finally, future studies of tumor epigenetics, the integration of germline and somatic alterations, and the interplay between tumor genomics and immunology, would be likely to provide us with new insights in PCa progression and novel tools to improve patient management.

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HIGHLIGHTS

- Despite intratumoral histologic heterogeneity confounding sampling, studies have revealed clonal relationships in intermediate to high risk prostate cancers via their underlying molecular alterations.
- Distinct molecular signatures of Gleason grade 4-associated Gleason grade 3 captured on biopsy can predict the presence of unsampled Gleason grade 4 tumors.
- Risk classifiers are most useful when they measure gene expression levels circumventing intratumoral heterogeneity, addressing issues of sampling limitation.
- Distinct molecular alterations detected in prostatectomy tissue can predict disease recurrence in patients with intermediate to high risk prostate cancer.