The Role of Biomarkers in Prostate Cancer Management

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Since the introduction of serum prostate specific antigen (PSA) thirty years ago, prostate cancer diagnosis and management have been guided by this biomarker. Yet, PSA has proven controversial as a diagnostic assay due to its limitations.

PSA has persisted in clinical practice owing in large part to the public's demand for prostate cancer screening. Indeed, PSA remains an inexpensive and sensitive biomarker for disease detection, monitoring progression and recurrence following curative therapy of local disease. Furthermore, because PSA screening is so common, the clinical evaluation of new biomarkers has only occurred in patient populations previously screened for PSA. Thus, future iterations of prostate cancer biomarkers will most likely retain PSA as a primary clinical tool in conjunction with other tests, unless new biomarkers are shown to be superior to PSA in head-to-head comparisons. In this regard, new biomarker assays will likely complement PSA-based detection of prostate cancer [1, 2].

In 1986, the U.S. Food and Drug Administration (FDA) approved PSA as an adjunctive test to the digital rectal exam (DRE) for the detection of prostate cancer in men over the age of 50. In 1991, Catalona and colleagues demonstrated that the combination of a serum PSA measurement of more than ≥4.0 ng/mL with other clinical findings, such as the results of a DRE, improved detection of prostate cancer in a prospective study of 1653 healthy men with no history of cancer [3]. Numerous groups confirmed that PSA was useful as a diagnostic test for prostate cancer.

There have been numerous efforts to improve the performance of the PSA test, such as normalizing PSA to the size of the gland (the PSA 'density') [4–6] or monitoring the dynamics of PSA change in serum (the PSA velocity and doubling time) [7–11]. In addition, assays measuring alternative molecular traits of PSA have also gained attention, including free and complexed PSA (fPSA and cPSA, respectively) [12–15], and isoforms of the PSA protein (proPSA, most commonly). Among these, cPSA and fPSA have been considered adjunctive tests to total serum PSA rather than replacement assays (Fig. 1). cPSA measurements exploit the molecular interactions of PSA mainly with α-1-antichymotrypsin (ACT) in the blood [13]. Conversely,

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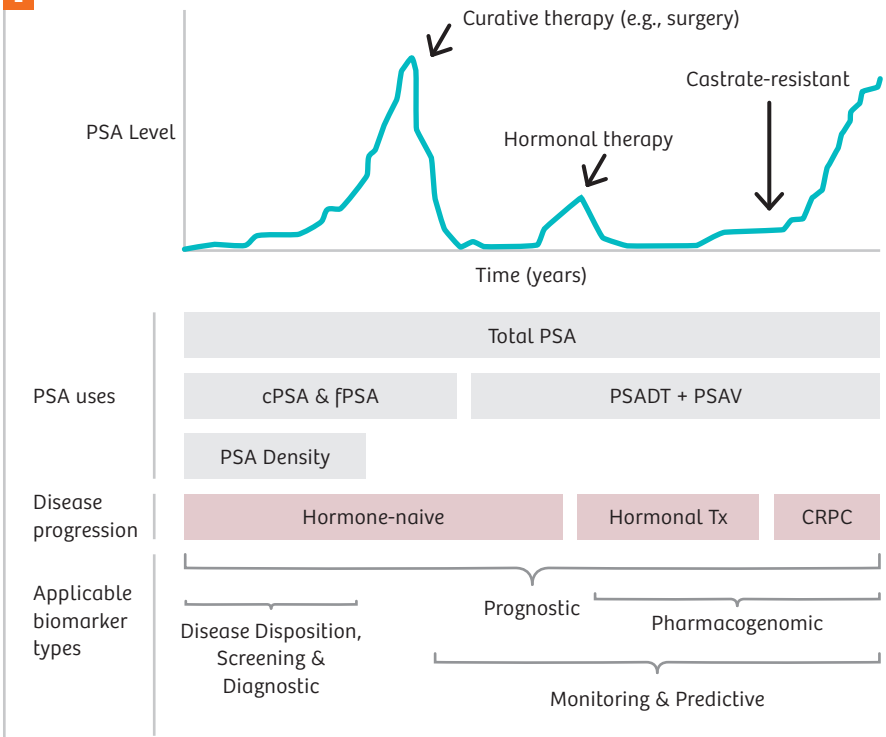

Figure 1: There have been numerous efforts to improve the performance of the PSA test.
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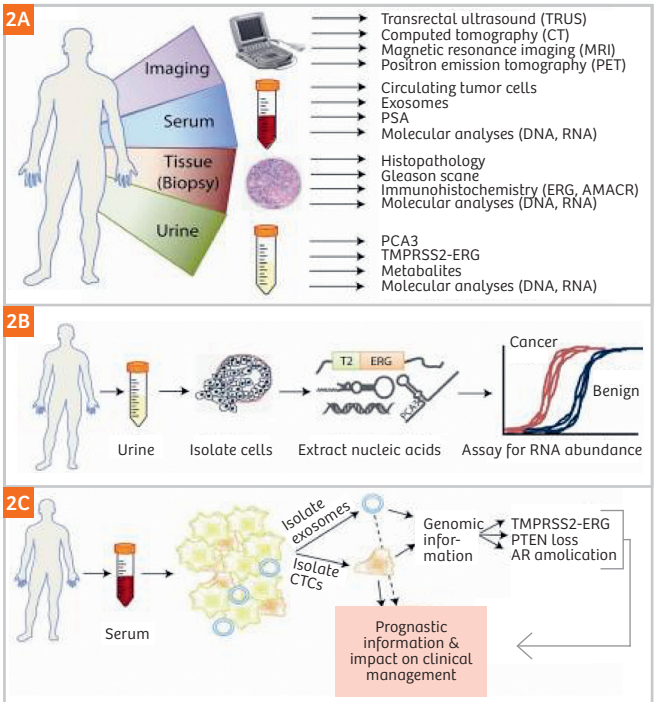

Figure 2: (2A) Biomarker research also includes advances in tissue and imaging-based tools. **(2B)** Promising future directions for biomarker research. **(2C)** One area of expanding investigation is circulating tumor cells.
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fPSA measures the percentage of total serum PSA not bound to ACT. This %fPSA decreases in prostate cancer, making it useful in distinguishing men with benign prostatic hyperplasia (BPH) from men with cancer. A %fPSA of less than 25% has been shown to improve the sensitivity and specificity of a total PSA test and to reduce unnecessary biopsies [12, 15]. %fPSA has thus gained FDA approval for use when patients have a total PSA in the 4–10 ng/mL 'gray zone'. Furthermore, combined measurement of pro-PSA (a peptide precursor to mature PSA) with fPSA may help diagnose early prostate cancers with a PSA of 2 to 10 ng/mL [16, 17]. fPSA has several drawbacks, such as the potential instability of the fPSA measurement if sample processing occurs after 24 hours of collection [18]. The %fPSA may also increase following DRE or biopsy procedures [19], confounding its use in those settings.

PSA velocity (PSAV) and doubling time (PSADT) have prognostic value [20]. PSAV is defined as the change in PSA concentration per year, with a high PSAV being strongly associated with prostate cancer and a 9-fold elevated risk of cancer-death follows prostatectomy [7, 8, 21]. PSADT is defined as the time necessary for the serum PSA level to double. PSADT is most commonly used to monitor disease progression following curative therapy for organ-confined disease, as an increasing PSA level following radiotherapy or prostatectomy indicates the presence of residual tumor cells. Numerous studies have demonstrated that a more rapid PSADT (<10 months) is associated with reduced survival [9, 10]. In rare cases, disease may recur in the absence of an elevated PSA [22]. Nevertheless, neither test has been shown to improve over a standard PSA measurement for prostate cancer screening [11]. Taken together, measurement of PSA isoforms and dynamics have modestly improved care but are largely hindered by the same issues confounding PSA itself.

The 30 years since the widespread adoption of PSA have witnessed a remarkable maturation of genomic technologies, such as microarrays and whole-genome sequencing [23]. These advances in DNA sequence and RNA transcriptome profiling have enabled detailed dissections of cancer biology at a level previously unattainable [23, 24]. As a result, the world of biomarker research has shifted to use these '-omics' methods, populating the prostate cancer literature with discoveries based on profiling prostate tumors for aberrations in DNA, RNA, or epigenetic DNA methylation states. The discovery and characterization of emerging urine assays for prostate cancer, including prostate cancer antigen 3 (*PCA3*), the most prominent biomarker emerging as a non-PSA-based diagnostic test for prostate cancer. *PCA3* is a long noncoding RNA (lncRNA) that has been shown to be elevated in >90% of prostate cancer tissues, but not to normal or BPH tissues, an important distinction to serum PSA [25, 26] and the *TMPRSS2-ERG* gene fusion product arising from a translocation of the androgen-induced transmembrane protease, serine 2 (*TMPRSS2*) gene with the transcription factor v-ets erythroblastosis virus E26 oncogene homolog (*ERG*) is one of the most common genetic events in prostate cancer, present in approximately 50% of all cases and accounting for 90% of prostate cancer fusions [27]. *TMPRSS2-ERG* fusions are specific for prostate cancer, and can even be detected in precursor lesions, such as prostate intraepithelial neoplasia (PIN), if these lesions are proximal to, or contiguous with, regions of cancer [28] although the biomarker research also includes advances in tissue and imaging-based tools as well (Fig. 2A).

One area of expanding investigation is circulating tumor cells (CTCs). CTCs are found in the bloodstream and are particularly prevalent in locally aggressive or metastatic disease. CTCs can be both a biomarker for cancer detection and a source of molecular information, such as *TMPRSS2-ERG*, androgen receptor (AR) and phosphatase and tensin homolog (*PTEN*) copy number status (Fig. 2C) [32]. Similar effort has recently focused on developing assays to detect prostate-derived exosomes (also called prostatosomes). Exosomes are small vesicles (50–150 nm in diameter) generated from internalized parts of the cellular membrane that are subsequently secreted into the blood, semen, or urine (Fig. 2B, C) [33]. Prostate cancer patients exhibit increased numbers of exosomes in their serum compared to men with no disease, and elevated levels of exosomes may also correlate with increasing Gleason score (rates cases of prostate cancer on a scale of 2 to 10, with higher scores

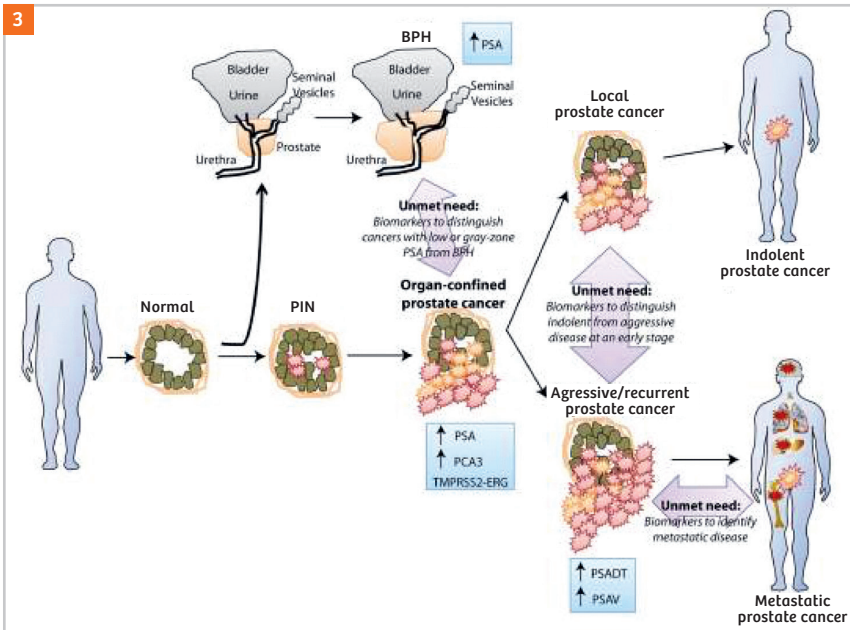

Figure 3: The most critical biomarker studies will focus on biomarker candidates that address the current gaps in prostate cancer biomarker development, including prognostic and predictive biomarkers. *Figure reproduced with permission from Sci Transl Med. 2012 Mar 28; 4(127): 127rv3.*

assigned to cancers that are growing more quickly and score of 7 indicates a moderate growth rate. While doctors are likely to watch and wait when a cancer has a low score of 2 to 6) [34]. Prostate cancer RNA biomarkers, including *PCA3* and *TMPRSS2-ERG*, can also be detected in urine-derived exosomes from prostate cancer patients [35]. Although these efforts remain mainly research-oriented at this time, they provide promising future directions for biomarker research (Fig. 2B).

The most critical biomarker studies will focus on biomarker candidates that address the current gaps in prostate cancer biomarker development, including prognostic and predictive biomarkers (Fig. 3). The utility of PSA as a diagnostic biomarker for prostate cancer is limited by the fact that only about 3% of PSA-screened men with prostate cancer have lethal disease, thus leading to overtreatment of indolent disease [29]. Development of new biomarkers that only identify more prostate cancer cases does not address this discrepancy. It follows, then, that the identification and validation of novel biomarkers to 'rule out' lethal prostate cancer at the point of screening is the greatest unmet clinical need, as this may reduce unnecessary interventions that may cause more harm than good.

A common theme in prostate cancer biomarker development is the desirability of non-invasive assays to replace biopsy as the diagnostic gold standard. Biopsy procedures are associated with increased risk of adverse events, such as bleeding and sepsis, owing to their invasive nature. Studies have routinely shown that biopsies are associated with a 23–25% false negative rate, perhaps owing to inefficient sampling, where normal tissue is biopsied in addition to diseased tissue. Non-invasive bio-markers in serum and urine have the potential to improve the standard tissue biopsy procedure, although they cannot provide direct histopathological or spatiotemporal information. As such, supplementing PSA measurements with urine biomarker analyses may become standard practice in the near future [30, 31].

Finally, these developments also need to be considered in conjunction with tissue biomarkers and imaging technologies, such as transrectal ultrasound (TRUS), computed tomography (CT), magnetic resonance imaging (MRI), and positron emission tomography (PET). Indeed, the role of imaging is crucial to patient management for visualizing and staging both localized prostate cancers and metastatic disease, especially in the bone.

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