# Biomarkers for improved management of prostate cancer

*by Dr Claire Tonry, Dr James C. Waddington and Prof. Stephen R. Pennington*

Prostate cancer is an extremely complex and heterogenous disease. A significant problem in the clinical management of PCa is being able to accurately stratify patients based on their risk for aggressive disease. Traditional biomarkers and clinical methods for personalized treatment are insufficient. Appropriate stratification of patients based on their risk for aggressive disease is required to optimize the balance between treatment efficacy and quality of life. This is an extremely active field of research and several promising new biomarker assays are already on the market, with more advancing through the biomarker development pipeline.

## Challenges in clinical management of prostate cancer

Prostate cancer (PCa) is a very common type of cancer. It is likely that many readers will know someone who is currently living with this disease. At the outset, the prognosis for PCa is generally good; it is a cancer that is usually diagnosed early and, in most cases, progresses very slowly such that most men will die with the disease, rather than from it. However, this is only true for men who respond well to treatment. For those who do not and who, after initial treatment, suffer cancer recurrence the outlook is not so good. Sadly, recurrent PCa is highly likely to spread (metastasize) to other sites and the average survival rate for metastatic PCa is just 3–5 years (Fig. 1). Herein lies the main challenge in effective clinical management of PCa: identifying and then selecting appropriate treatment regimens for patients based on their likelihood of developing aggressive disease or maintaining a more indolent (slowly progressing) disease.

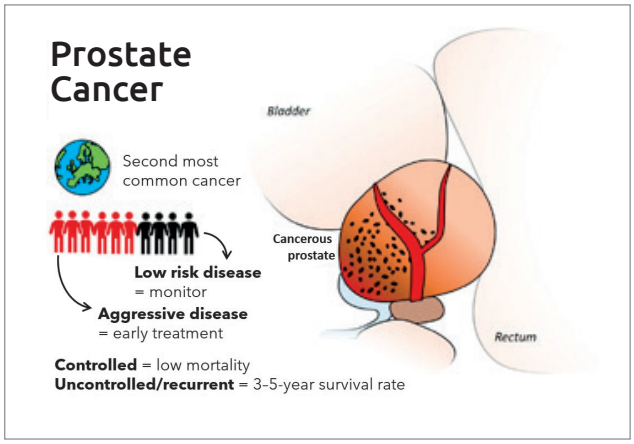


**Figure 1. Overview of prostate cancer** The prostate is located between the bladder and the rectum. Cancer of the prostate a large proportion of the male population. Patients will require different treatment, depending on their risk for uncontrolled disease, which is associated with only a 3–5-year survival rate.

There are currently several effective first-line treatment options for PCa; removal of the prostate gland by radical prostatectomy, inhibition of prostate tumour growth with hormone (androgen) deprivation therapy, and radiation therapy. However, although effective (in most cases) these are all associated with quite severe side effects that can have a negative impact upon the patient's quality of life. Considering that most men will live for a long time with the disease, implications for quality of life need due consideration, especially given that combined treatment with radiation and hormone therapy is considered to be the most effective treatment, but also increases the associated toxicity. For men with indolent disease, the side effects from such harsh treatments, which include impotence, loss of libido, erectile dysfunction and pulmonary embolism, can outweigh the benefit of removing the tumour and/or depleting androgen levels at a very early stage. Hence, the less invasive approach of 'active surveillance' is now advocated in place of surgery and radio/hormone therapy for men with slow-growing prostate-confined tumours. During active surveillance men regularly undergo PCa testing, digital rectal examination (DRE) and repeat biopsies to closely monitor the progress of the disease and determine if/when curative intervention will be required. On the other hand, men who have a more active form of PCa, which is likely to become resistant to standard first-line therapies and spread beyond the prostate, will need aggressive clinical intervention at the earliest possible stage. For example, chemotherapy can be combined with androgen-deprivation therapy, although the combined toxicity

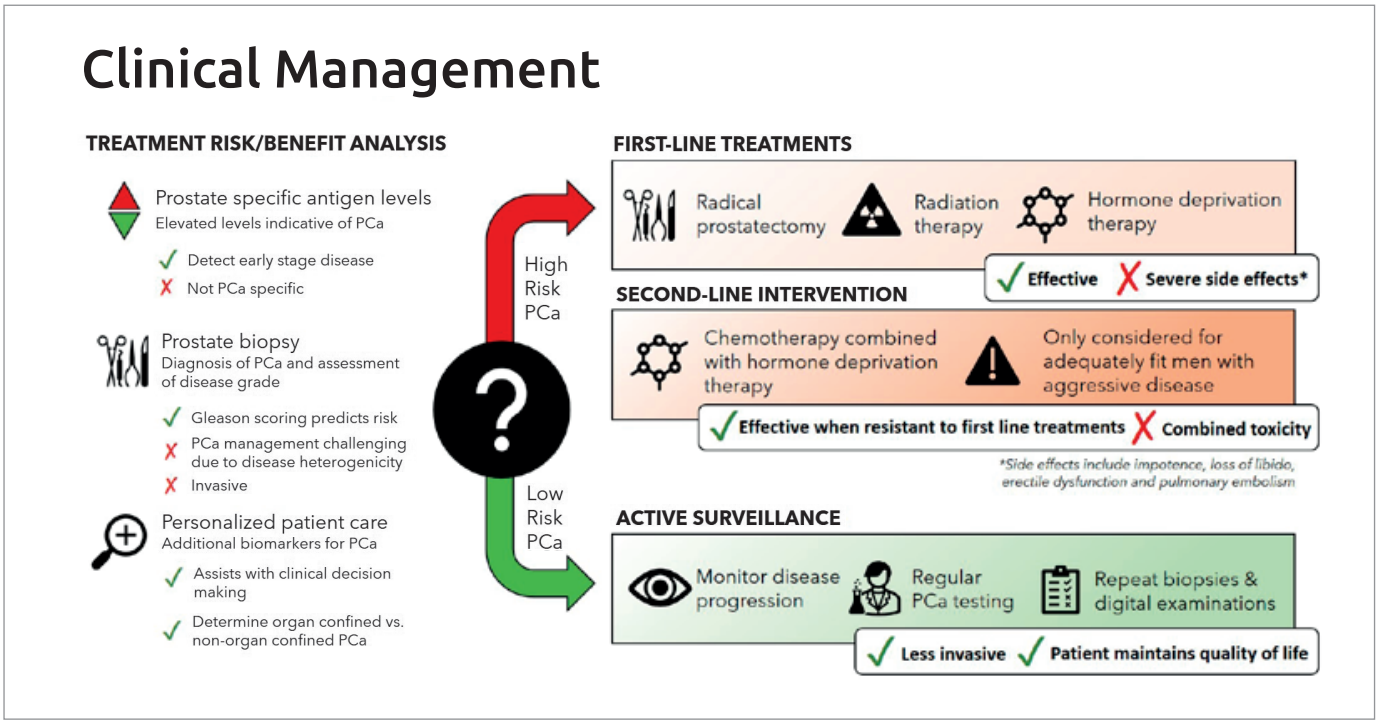


**Figure 2. Clinical management of prostate cancer** PCa is detected by elevated levels of PSA in the blood. Diagnosis is confirmed following a digital rectal exam and tissue biopsy. Low-risk PCa can be managed without treatment and close monitoring of disease progression. There are a number of treatment options for high-risk PCa, however, each are associated with toxic side effects. Personalized patient care will benefit from novel biomarker assays to give more accurate information on patients risk for aggressive PCa (and need for treatment).

from such a treatment approach means that it is only considered for adequately fit men with very aggressive disease (Fig. 2). Although PCa can be diagnosed easily, reducing the associated mortality from metastatic PCa and avoiding over-treatment of indolent disease relies on appropriate stratification of patients based on the severity of their disease.

## Limitations to current screening and diagnostic strategies

The level of prostate specific antigen (PSA) in the blood has become the most common molecular marker (biomarker) for the presence of PCa. In fact, this protein is the most commonly used screening biomarker for any cancer. PSA levels greater than *4.0 ng/mL* are indicative of possible PCa. Although PSA screening is beneficial in that it can detect even low-risk PCa at a very early stage, PSA is not a specific biomarker. PSA levels can be increased as result of non-cancer associated inflammation and/or enlargement of the prostate caused by chronic prostatitis or benign prostate hyperplasia. Historically PSA screening was recommended for men over the age of 50, but there is now debate around whether it is beneficial to test for PCa in men who do not have symptoms. PSA screening has been associated with overdiagnosis and over-treatment. In the UK there is no national prostate cancer screening programme because the PSA test is not deemed reliable enough.

Currently, definitive diagnosis of PCa and assessment of disease grade and stage requires a prostate biopsy. This is where difficulties in PCa management really become evident. PCa is an inherently heterogeneous disease; its pathogenesis differs greatly between individuals (as evidenced by the slow-growing or aggressive nature of the disease) but can also differ histopathologically between the multiple tumour foci found within an individual's prostate. Grading of PCa is based on the Gleason scoring system, which describes different tumour growth patterns. The scoring pattern is a sum of the two most prominent Gleason scores (1–10 based on histopathologic evidence of the degree of tumour differentiation) from multiple samples of tumour. In 2014 this scoring system was modified to categorize tumours into 'grade groups' based on their Gleason score. Patients with a combined Gleason score ≤6 are considered to have 'low-risk' disease (grade group 1). Patients with a combined Gleason score of 7 are considered to be of intermediate risk, with more favourable outcomes if their tumour is predominantly Gleason score 3 (grade group 2) as opposed to if their tumour is predominantly Gleason score 4 (grade group 3). Patients with a combined Gleason score of 8 and above are defined as high/very high risk (grade groups 4 and 5). The Gleason scoring system is summarized in Table 1.

**Table 1. Gleason scores and grading system** (Source: Prostate Cancer Foundation website: https://www.pcf.org/about-prostate-cancer/diagnosis-staging-prostate-cancer/gleason-score-isup-grade/)

| Risk group | Gleason score | Grade group |
|---|---|---|
| Very low/low | Gleason score ≤6 | Grade group 1 |
| Intermediate (favourable/ unfavourable) | Gleason score 7 (3+4) | Grade group 2 |
| | Gleason score 7 (4+3) | Grade group 3 |
| High/very high | Gleason score 8 | Grade group 4 |
| | Gleason score 9–10 | Grade group 5 |

However, this system cannot fully account for the multifocal nature of PCa; because only a small proportion of the prostate is sampled during biopsy, the most aggressive areas of tumour are frequently either oversampled or undersampled. Thus, there is currently no way of predicting which patients will have indolent disease and will respond well to standard treatments, or which patients will have a much more aggressive form of disease that will become resistant to standard first-line therapies and extremely difficult to control beyond that.

## Biomarker tests for personalized patient care

The current 'gold-standard' biomarker for PCa is PSA which, as described above, has some significant limitations as a clinical decision-making tool. Development of novel biomarker assays for clinical management of PCa is an active area in PCa research. Some novel biomarker assays for assessment of PCa risk have emerged for analysis of prostate tumour tissue. These tests have been developed in a bid to overcome limitations with PCa biopsy and Gleason grading, namely the inherent variations between regions of individual tumours and the limited tumour material acquired by needle biopsy. One example is the Decipher® test offered by GenomeDx Biosciences. This test measures the expression of 22 non-coding RNA sequences to calculate the probability of clinical metastasis. Similarly, the Oncotype DX® offered by Exact Sciences measures a 17-gene signature as an independent predictor of high-risk pathology for men newly diagnosed with very low-risk PCa (in selected cases, men with intermediate risk PCa may also benefit from this test). The Prolaris® test (Myriad Genetics Inc.), is an RNA-expression-based assay that directly measures tumour cell growth characteristics. It can be used in conjunction with clinical parameters such as Gleason score and PSA to identify low-risk patients who can be safely managed with active surveillance. The ProMark® assay (Metamark Genetics) measures eight protein markers of PCa pathophysiology using a multiplexed *in situ* imaging system. However, the challenge of PCa heterogeneity is not completely circumvented with these tissue-based assays. Less-invasive tests that are amenable to routine sampling would be more suitable for monitoring PCa risk and improving personalized clinical management of PCa (Fig. 3).

The most promising novel fluid-based biomarker for PCa is a protein called prostate cancer antigen 3 (PCA3). It is included in the Progensa assay, which compares the concentration of PCA3 messenger RNA (mRNA) levels to PSA mRNA levels to produce a urinary PCA3 score. Because PCA3 mRNA is not expressed in normal prostate tissue and expressed at very low levels in benign prostatic hyperplasia specimens, urinary PCA3 scores (PCA3-mRNA/PSA-mRNA) are superior to serum PSA levels for ruling out non-cancerous causes for prostate enlargement. The measurement of PCA3 has also been combined with another well-known biomarker of PCa – the TMPRSS2:ERG gene fusion – as part of the Mi-Prostate Score, which is intended for stratification of PCa tumours. PCA3 has been incorporated into a newer test called the ExoDx™ Prostate Intelliscore (ExosomeDx). This test analyses exosomal RNA for three biomarkers – PCA3, TMPRSS:ERG and SAM pointed domain containing ETS transcription factor (SPDEF). When combined with standard clinical variables (PSA, age, race and family history of PCa), this test improves discrimination between low-grade (Gleason 6) and high-grade (Gleason ≥7) PCa and is now available in the USA as a Clinical Laboratory Improvement Amendments (CLIA)-based clinical laboratory-developed test (LDT). The Prostarix™ test (Metabolon Inc.), uses metabolomics technology to measure levels of four amino acids associated with PCa. This test can be used to distinguish between benign prostate, clinically localized PCa and metastatic disease. A newly available urine test from MDxHealth (SelectMDx®) measures expression of *HOXC6* and *DLX1* genes in urine using the *KLK3* gene (PSA) as an internal reference. The risk score derived from combining these gene markers with information on PSA density, DRE and PSA has been shown to accurately detect high-grade PCa upon biopsy. Cost effectiveness studies have revealed that incorporation of the SelectDx® test into clinical assessment of PCa resulted in a saving of €128 (US$143) and a gain of 0.25 in patient quality of life years, compared to using only PSA to select patients for prostate biopsy. The uptake of this test in the USA is high, with recent figures from 2020 showing a 98% increase in billable tests since 2018.

Although these tests are promising, there are some limitations with urine-based tests; some of the urine-based assays require urine that is passed immediately following DRE, and results will not be valid otherwise. Moreover, the collection of urine is performed privately by the patients themselves and so it is difficult to standardize. This variability may influence results. Blood, on the other hand, is also easily accessible and collected under much more controlled conditions such that there is a more effective 'chain of custody'. The 4Kscore® is a serum test combining measurement of total PSA, free PSA (fPSA), intact PSA and human kallikrein-related peptide 2 (hK2). The serum 4Kscore® assay has been shown to accurately predict the risk of biopsy-detectable high-grade PCa in men who have not undergone a prostate biopsy. Although not currently FDA approved it is commercially available in the USA as a CLIA-approved LDT and appears to have some clinical utility.

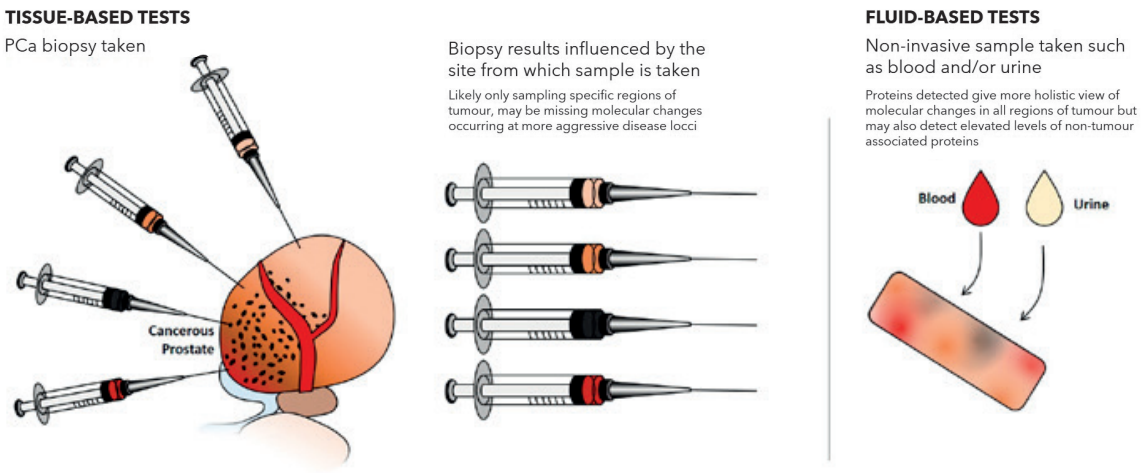


**Figure 3. Considerations for novel tissue and fluid-based biomarker tests** Novel biomarker assays have been designed to improve accuracy of risk stratification. These can be applied to biopsy samples, however, tissue sampling is limited by the heterogeneity of prostate tumours. Minimally-invasive assays, such as those applied to blood and urine, can overcome this, provided that specificity for the disease is not compromised.

## Benefit to the patient

The emerging tests show great potential to significantly improve patient care. Clinicians and researchers often do not recognize the challenge of overdiagnosis in cancer care as underdiagnosis is deemed too risky. Recent tests on the market (and under investigation) have all been designed with the objective of providing clinicians (and patients) with the information they need to decide on the most appropriate treatment course for patients, based on their unique tumour patho-physiology. The National Institute for Health and Care Excellence (NICE) recommends that patients who wish to be involved in clinical decisions regarding their treatment must be well informed on their risk of aggressive PCa and the side effects associated with the various treatment options. This personalized approach to patient care is intended to reduce over-treatment of men and spare them unnecessary side effects for as long as possible. Concurrently, patients who are at high risk of developing aggressive disease can be identified earlier and treated more proactively from the outset, with the hope that tumour growth can be impeded before it has a chance to spread and become independent to androgen signalling (and thereby resistant to androgen-deprivation therapy). As well as benefitting the patients, there is data to suggest that the introduction of such tests would reduce the economic burden associated with clinical management of PCa. This is likely due to the anticipated avoidance of unnecessary biopsies, increased use of active surveillance and reduced hospitalizations from uncontrolled PCa.

## Future perspectives

The primary motivation in PCa research is to optimize clinical management of the disease based on what is now understood about its complex and highly variable underlying pathology. Improving quality of life for men with PCa is important due to the longevity of the disease. Therefore, routine monitoring of PCa progression is the ultimate goal for clinicians. A lot of progress has been made, with urine-based assays such as the Progensa and SelectDx® showing clinical utility. Blood-based tests would be more appropriate for routine monitoring of PCa during active surveillance (Fig. 3). So far, however, only the serum 4kscore® assay has made an impact. There is room to improve on this with assays that may better reflect specific disease-relevant molecular changes in the tumour. One such blood-based test called OCProDx, which can differentiate between organ-confined and non-organ confined PCa with high accuracy, is being developed by Atturos. The test is based on multiplexed measurement of a panel of proteins associated with PCa pathology. This could be a clinically useful tool in the earlier stages of PCa diagnosis, as it can highlight situations in which radical prostatectomy will not be sufficient to ameliorate tumour progression. Tests such as OCProDX, which measure PCa-related changes to the prostate in blood, evolve from ongoing research into the complex PCa tumour micro-environment. For example, elucidating the molecular changes that occur in response to common 'enablers' of PCa tumour progression (androgen signalling, hypoxia and nutrient deprivation) has led to the identification of proteins and molecular pathways, which have potential to be incorporated into clinical tests or leveraged for the development of novel therapeutic strategies. Hence, ongoing and ever-more insightful research outputs are continuing to advance efforts for improved, patient-centred, clinical management of PCa.


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