Regence

Medical Policy Manual

**Topic:** Protein Biomarkers for Screening, Detection, and/or Management of Prostate Cancer  
**Date of Origin:** October 2015

**Section:** Laboratory  
**Last Reviewed Date:** December 2016

**Policy No:** 69  
**Effective Date:** January 1, 2017

**IMPORTANT REMINDER**

Medical Policies are developed to provide guidance for members and providers regarding coverage in accordance with contract terms. Benefit determinations are based in all cases on the applicable contract language. To the extent there may be any conflict between the Medical Policy and contract language, the contract language takes precedence.

PLEASE NOTE: Contracts exclude from coverage, among other things, services or procedures that are considered investigational or cosmetic. Providers may bill members for services or procedures that are considered investigational or cosmetic. Providers are encouraged to inform members before rendering such services that the members are likely to be financially responsible for the cost of these services.

**DESCRIPTION**

There are a variety of protein biomarkers that have been associated with prostate cancer. These tests have the potential to improve the accuracy of risk prediction, diagnosis, staging, prognosis or management of prostate cancer.

Prostate cancer is a complex, heterogeneous disease. At the extremes of the spectrum, if left untreated, some prostate cancers behave aggressively, metastasize quickly, and cause mortality, while others are indolent and never progress to cause harm. Current challenges in prostate cancer care are risk assessment; early and accurate detection; monitoring low-risk patients undergoing surveillance only; prediction of recurrence after initial treatment; detection of recurrence after treatment; and assessing efficacy of treatment for advanced disease.

In response to the need for better biomarkers for risk assessment, diagnosis, prognosis and management, a variety of exploratory research is ongoing. Some products of this work have already been translated or are in the process of being translated into commercially available tests, including:

- 4Kscore Test (OPKO Lab), a blood test that measures four prostate specific kallikreins which are combined into an algorithm to decide whether a patient should proceed to prostate biopsy.
• Prostarix™ (Metabolon/Bostwick Laboratories), a post-DRE urine test based on several metabolites and an algorithm to decide whether a patient should proceed to prostate biopsy or undergo repeat biopsy after an initial negative biopsy.
• Promark™, a protein biomarker test that uses immunofluorescence and automated quantitative images in intact biopsy tissue to risk stratify patients to active surveillance or therapeutic intervention.

While studies using these tests generate information that may help elucidate the biologic mechanisms of prostate cancer and eventually help design treatments, the above-mentioned tests are currently in a developmental phase, with insufficient evidence of clinical utility.

**Regulatory Status**

None of the tests addressed in this policy have been submitted to the U.S. Food and Drug Administration (FDA) for marketing clearance but, if available, are offered as laboratory-developed tests by Clinical Laboratory Improvement Amendments (CLIA) licensed laboratories.

**MEDICAL POLICY CRITERIA**

Protein biomarkers for the screening, detection, and management of prostate cancer are considered investigational. These include, but are not limited to the following:

A. Kallikrein markers (eg, 4Kscore™ Test);
B. Metabolomic profiles (eg, Prostarix™);
C. Immunofluorescence markers (eg, Promark™)

**SCIENTIFIC EVIDENCE**

In general, the evidence for biomarker tests related to prostate cancer screening, detection, and management addresses either preliminary clinical associations between protein expression and disease states or, in some cases, the clinical validity of these tests, i.e., the association of the test result with outcomes of interest, expressed in terms of clinical performance characteristics such as sensitivity, specificity, predictive value, and comparisons to current standards using receiver-operating curve (ROC) analysis and/or logistic regression. There is limited evidence of clinical utility, i.e., that using a protein biomarker test will change treatment decisions and improve subsequent outcomes that matter to the patient such as mortality, morbidity, or quality of life.

**4Kscore Test (OPKO Lab)**

The 4Kscore test, also referenced in the literature as the four-kallikrein panel, is a blood test that generates a risk score for the probability for finding high-grade prostate cancer (defined as Gleason score ≥7) if a prostate biopsy were performed. The intended use of the test is to aid in the decision of whether or not to proceed with a prostate biopsy. The test algorithm combines the measurement of four prostate specific kallikreins (total prostate-specific antigen [tPSA], free PSA [fPSA], intact PSA [iPSA] and human kallikrein 2 [hK2]), which are combined in an algorithm with patient age, digital rectal exam...
(DRE) (nodules or no nodules), and whether the patient has had a prior negative prostate biopsy. A kallikrein is a subgroup of enzymes that cleave peptide bonds in proteins. The iPSA and hK2 tests are immunoassays that employ distinct mouse monoclonal antibodies. The test is not intended to be used in patients with a previous diagnosis of prostate cancer, a patient who has had a DRE in the previous four days, a patient who has received 5-alpha reductase inhibitor therapy in the previous six months, or a patient who has undergone any procedure or therapy to treat symptomatic benign prostatic hypertrophy in the previous six months.

A recent study by the 4Kscore investigators assessed the ability of the four-kallikrein panel to predict high-grade cancer at ten-core prostate biopsy in 4765 men in the ProtecT study.[1] Cryopreserved blood from men with elevated PSA (≥3.0ng/mL) was tested to predict any-grade or high-grade (Gleason score≥7) prostate cancer. Area under the curve (AUC) for the four kallikreins was 0.72 (95% confidence interval [CI]: 0.70 to 0.73) vs 0.63 (95% CI: 0.62 to 0.65, \( p < 0.001 \)) for PSA and age alone for any-grade cancer, and 0.82 (95% CI: 0.80 to 0.84) vs 0.74 (95% CI: 0.72 to 0.76, \( p < 0.001 \)) for high-grade cancer. Using a cutoff of 6% risk of high-grade cancer, the study determined that 428 out of 1000 men could avoid biopsy. The clinical utility of this test, i.e. if the results influenced treatment decisions was not addressed. The authors concede that further evaluation of this panel is needed in prospective studies that test fresh samples in a clinical setting (as opposed to a research lab as performed in this study). Other recent European retrospective studies have found similar results in terms of the ability of the 4Kscore test to predict high grade cancer when the referral criteria were expanded to men who had elevated PSA (≥3 ng/ml), low %free PSA (<20%), or suspicious DRE.[2]

The performance of the 4Kscore test was validated in a total of 1012 patients who were enrolled from October 2013 to April 2014 in a blinded, prospective study at 26 urology centers in the United States.[3] Enrollment into the study was open to all men who were scheduled for a prostate biopsy, regardless of age, PSA level, DRE or prior prostate biopsy. Each patient underwent a TRUS-guided prostate biopsy of at least 10 cores. A blinded blood sample that was collected prior to biopsy was sent to OPKO Lab for measurement of the four kallikrein markers. The results of the kallikrein markers, prostate biopsy histopathology, patient age, DRE and prior biopsy status were unblinded and analyzed. The biopsy was negative in 54% of cases (n=542), showed low-grade (all Gleason grade 6) prostatic cancer in 24% (n=239) and high-grade cancer in 23% (n=231). The statistical analysis of the 4Kscore test clinical data had an AUC of 0.82 for the detection of high-grade prostate cancer; the AUC for all patients using tPSA, age, DRE and prior biopsy was 0.76. Limitations of the study include lack of standard criteria for biopsy referral and lack of central laboratory used for histopathology.

Based on the US trial, OPKO has established assay specifications, available on the company website, for two of the four proteins, intact PSA (iPSA) and Kallikrein 2 (hK2) for biopsy negative (median: 0.416ng/mL and 0.069ng/mL, respectively), low-grade disease (Gleason = 6) (median: 0.469ng/mL and 0.081ng/mL, respectively) and high-grade disease (Gleason ≥7) (median: 0.511 and 0.107ng/mL, respectively). They also have published precision values for iPSA (0.01 - 0.10 ng/mL CV ≤ 15%, 0.11 - 1.0 ng/mL CV ≤ 8%, 1.1 - 15 ng/mL CV ≤ 5%) and hK2 (0.01 - 0.10 ng/mL CV ≤ 10%, 0.11 - 1.0 ng/mL CV ≤ 8%, 1.1 - 8 ng/mL CV ≤ 10%), thereby demonstrating the analytic validity of the test. These values have been previously determined for the other two prostate specific kallikreins, tPSA and fPSA, with commercial assays approved for use in human diagnostics by the FDA. Based on the US prospective trial and several retrospective European trials, the test has demonstrated the ability to detect high-grade cancer in specific populations (i.e. men with high PSA and/or men already scheduled for biopsy). The potential of the 4Kscore test to reduce biopsy in patients whose biopsy samples did not indicate high-grade cancer was also evaluated. The investigators reported sensitivity, specificity, positive and negative predictive value for four different thresholds investigated for biopsy reduction:

≥6.0%, 9%, 12% and 15% probability of high-grade cancer, thereby demonstrating the test’s clinical validity. Additional prospective studies are needed to establish the clinical utility of this test.

Prior to the US trial, this group had conducted multiple studies predicting the use of the test in patient cohorts from the European Randomized Study of Prostate Cancer (ERSPC).\[^{4-8}\] The majority of these studies were retrospective in nature, mainly assaying cryopreserved blood samples previously collected. In one of the studies, 392 men with high PSA (≥3.0 ng/mL) who underwent radical prostatectomy were screened for the four kallikrein markers to see if the test could distinguish between pathologically insignificant and aggressive disease when used in conjunction with clinical predictors (age, stage, PSA, biopsy findings). The AUC for the clinical predictors alone was 0.81, while using the clinical predictors in conjunction with the 4Kscore test improved the AUC to 0.84. Both of which are significantly better at predicting aggressive cancer than total PSA alone (AUC = 0.68).\[^{9}\] The limitations of this study are mainly in its design: retrospective in nature, using cryopreserved sample and relying on 6-core biopsies, and not the 10-12 core currently recommended for grading accuracy.

Recently, small retrospective study in Spain has also evaluated the 4Kscore, along with the Prostate Cancer Prevention Trial Risk Calculator 2.0 and the European Research Screening Prostate Cancer Risk Calculator in 51 patients undergoing a prostate biopsy.\[^{10}\] According to the authors, all of the models assessed showed good discriminative ability for high-grade prostate cancer, but this study was limited by the retrospective design and small sample size.

Another recent study examined the use of the 4Kscore in higher-risk patients with either a positive DRE or PSA 10-25 ng/ml.\[^{11}\] This was a meta-analysis of individual patient data from 2,891 subjects, collected from 8 cohorts. The authors reported that the addition of the kallikrein test added to the discriminative power of their model, but this has not been replicated and the clinical utility of using the test in this manner has not been assessed prospectively.

**Prostarix™ (Metabolon/Bostwick Laboratories)**

Prostarix™ is a post-DRE urine test which is based on a panel of biomarkers and is used in the early detection of prostate cancer. The results are intended to aid in clinical decision-making as to whether to biopsy or repeat biopsy the prostate, particularly in patients who have a suspicious DRE and modestly elevated PSA (2.5-10 ng/mL). The test addresses metabolic abnormalities that have been associated with prostate cancer. Prostarix measures the concentration of several metabolites: sarcosine, alanine, glycine, and glutamate, and these quantitative measurements are combined in a logistic regression algorithm to generate a Prostarix Risk Score. If PSA level and TRUS-determined prostate volume are available, they can be used along with the metabolite measurements to generate the Prostarix-PLUS Risk Score. The test claims to have increased sensitivity and specificity over standard assessment tools to predict the likelihood of a positive prostate biopsy.

Sarcosine, a derivative of glycine, is believed to be a mechanistic biomarker of aggressive prostate cancer. Two studies, described below, correlated the level of sarcosine in urine of prostate biopsy positive and negative patients, and found increased levels of sarcosine in the urine of patients with prostate cancer, however, is not clear in which patient population a test measuring urine sarcosine would be used, or what level of sarcosine would warrant a prostate biopsy. In their initial study of the potential role of metabolomic profiles to delineate the role of sarcosine in prostate cancer progression, Sreekumar et al. profiled 1126 metabolites across 262 prostate-derived clinical samples (42 tissue samples and 110 matched specimens of plasma and post-DRE urine from biopsy positive cancer patients [n=59] and
biopsy negative control patients \([n=51]\)). The authors reported that levels of sarcosine increased progressively during prostate cancer progression to metastasis.

Subsequently, the investigators used benign prostate and localized prostate cancer tissues obtained from a radical prostatectomy series from one University’s hospitals. Urine specimens were collected from patients who were being screened for prostate cancer with PSA levels considered clinically significant \((8.59 \pm 6.30)\). Urine was collected post-DRE but before prostate biopsy. Urine collected from patients undergoing prostatectomy was collected before surgery and used as a positive control. In total, 211 biopsy-positive and 134 biopsy-negative urine sediments were used. Using a logistic regression model, sarcosine levels were elevated in prostate cancer urine sediments compared with controls, with an AUC of 0.71.

Other studies done by different authors not affiliated with Metabolon have shown conflicting results with regards to the use of sarcosine as a biomarker for prostate cancer from urine. A case control study published However, a case control study reported by Cao et al. used multiple algorithms to clarify and reevaluate the potential value of sarcosine in prostate cancer. The authors reported that neither biopsy Gleason score nor clinical T-stage were correlated with sarcosine, regardless of the algorithm used, and ROC analysis indicated that the diagnostic power of any of sarcosine algorithm tested was not significantly better than urine PSA, percent free PSA orPCA3. Prior to that Jentzmik et al. evaluated urine sarcosine levels after digital rectal examination in 106 prostate cancer patients and 33 healthy controls. While the authors reported that sarcosine values were not associated with tumour stage \(pT2 \text{ vs } pT3\) or grade \((\text{Gleason score } <7 \text{ vs } \geq 7)\), and that ROC analyses determined that sarcosine levels were no different than total PSA in terms of diagnostic performance; they acknowledged that the disproportionate number of cases to controls was a substantial study limitation.

**ProMark™ (Metamark Genetics)**

The protein biomarker test, ProMark™ (Metamark Genetics, Cambridge, MA), is an automated quantitative imaging method to measure protein biomarkers by immunofluorescent staining in defined areas in intact formalin-fixed paraffin-embedded biopsy tissue, in order to provide independent prognostic information to aid in the stratification of patients with prostate cancer to active surveillance or therapy.

**Analytic Validity**

Shipitsin et al. reported on the analytic validity of the automated quantitative multiplex immunofluorescence in situ imaging approach assessing: the ability of the test to quantitate markers in a defined region of interest (tumor vs surrounding benign), tissue quality control, assay staining format and reproducibility. To evaluate tissue sample quality, they assessed the staining intensities of several protein markers in benign tissue and using these, categorized prostate cancer tissue blocks into four quality groups, of which the best two groups were used to generate tumor microarray blocks; 508 prostatectomy specimens were used and of these, 418 passed quality testing and were used for the tumor microarray blocks. For intra-experiment reproducibility, two consecutive sections from a prostate tumor test microarray block were stained in the same experiment and scatter plots compared the mean values of the staining intensities; signals from consecutive sections showed \(R^2\) correlation values above 0.9 and differences in absolute values typically less than 10%.

**Clinical Validity**
Blume-Jensen et al. reported on a study of 381 biopsies matched to prostatectomy specimens which were used to develop an 8-biomarker proteomic assay to predict prostate final pathology on prostatectomy specimen using risk scores.[17] Biomarker risk scores were defined as favorable if less than or equal to 0.33 and nonfavorable if greater than 0.80 with a possible range between 0 and 1 based on false negative and false-positive rates of 10% and 5%, respectively. The risk score generated for each patient was compared with two current risk stratification systems, National Comprehensive Cancer Network (NCCN) guideline categories and the D’Amico system. Results from the study showed that, at a risk score of less than or equal to 0.33, the predictive value of the assay for favorable pathology in very low- and low-risk NCCN and low-risk D’Amico groups were 95%, 81.5%, and 87.2%, respectively, while the NCCN and D’Amico risk classification groups alone had predictive values of 80.3%, 63.8%, and 70.6%, respectively. The positive predictive value for identifying favorable disease with a risk score of less than or equal to 0.33 was 83.6% (specificity, 90%). At a risk score of greater than 0.80, 77% had nonfavorable disease. Overall, 39% of the patients in the study had risk scores less than or equal to 0.33 or greater than 0.8, 81% or which were correctly identified with the 8-biomarker assay. Of the patients with intermediate risk scores (>0.33 to ≤0.8), 58.3% had favorable disease. The performance of the assay was evaluated on a second blinded study of 276 cases to validate the assay’s ability to distinguish “favorable” pathology (defined as Gleason score on prostatectomy less than or equal to 3+4 and organ-confined disease) versus “nonfavorable” pathology (defined as Gleason score on prostatectomy greater than or equal to 4+3 or non-organ-defined disease). The second validation study separated favorable from nonfavorable pathology (AUC 0.68, p<0.001; odds ratio, 20.9).

An industry-sponsored simulation study published in 2015 modeled the effects of using the ProMark test on 60-year-old patients with early prostate cancer (Gleason 3+3 and 3+4).[18] This study projected that the use of the test in this population could improve patient outcomes and reduce costs, but this has not been replicated in actual patients.

No published prospective studies on the clinical utility of the ProMark™ test were identified, therefore the current data are insufficient to establish the analytic and clinical validity and clinical utility of the ProMark™ test.

Clinical Practice Guidelines

Current NCCN guidelines recommend that consideration may be given to the 4Kscore test as one of several tests to consider for patients who are thought to be at higher risk for clinically significant prostate cancer.[19] Guideline authors note:

“Biomarkers that improve the specificity of detection are not recommended as first-line screening tests. However, there may be some patients who meet either PSA standards for consideration of prostate biopsy, but for whom the patient and/or the physician wish to further define the probability of high-grade cancer. A percent free PSA <10%, PHI >35 or 4Kscore (which provides an estimate of the probability of high-grade prostate cancer) are potentially informative in patients who have never undergone biopsy;”

This recommendation is based on lower level evidence (Category 2A), but despite the absence of higher level studies, there is uniform consensus for this recommendation.

Summary
The research on how protein biomarkers related to prostate cancer can be used to improve health outcomes for patients is variable and incomplete. Some tests may be useful to predict risk in the diagnosis or prognosis of prostate cancer, however, more research is needed to show how much these tests can add to the currently available tests, and what effects they have on treatment decisions and outcomes. Therefore, use of protein biomarker testing for risk assessment, diagnosis, prognosis, and management of prostate cancer is considered investigational.

REFERENCES

18. Roth, JA, Ramsey, SD, Carlson, JJ. Cost-Effectiveness of a Biopsy-Based 8-Protein Prostate Cancer Prognostic Assay to Optimize Treatment Decision Making in Gleason 3 + 3 and 3 + 4 Early Stage Prostate Cancer. The oncologist. 2015 Dec;20(12):1355-64. PMID: 26482553

CROSS REFERENCES

Gene-Based Tests for Screening, Detection, and/or Management of Prostate Cancer, Genetic Testing, Policy No. 17

Gene Expression Analysis for Prostate Cancer Management, Genetic Testing, Policy No. 71

<table>
<thead>
<tr>
<th>CODES</th>
<th>NUMBER</th>
<th>DESCRIPTION</th>
</tr>
</thead>
<tbody>
<tr>
<td>CPT</td>
<td>81313</td>
<td>PCA3/KLK3 (prostate cancer antigen 3 [non-protein coding]/kallikrein-related peptidase 3 [prostate specific antigen]) ratio (eg, prostate cancer)</td>
</tr>
<tr>
<td></td>
<td>81479</td>
<td>Unlisted molecular pathology procedure</td>
</tr>
<tr>
<td></td>
<td>81539</td>
<td>Oncology (high-grade prostate cancer), biochemical assay of four proteins (Total PSA, Free PSA, Intact PSA, and human kallikrein-2 [hK2]), utilizing plasma or serum, prognostic algorithm reported as a probability score</td>
</tr>
<tr>
<td></td>
<td>81599</td>
<td>Unlisted multianalyte assay with algorithmic analysis</td>
</tr>
<tr>
<td></td>
<td>84999</td>
<td>Unlisted chemistry code</td>
</tr>
<tr>
<td>0010M</td>
<td></td>
<td>Oncology (High-Grade Prostate Cancer), biochemical assay of four proteins (Total PSA, Free PSA, Intact PSA and human kallikrein-2 [hK2]) utilizing plasma, prognostic algorithm reported as a probability score (Deleted 1/1/2017)</td>
</tr>
<tr>
<td>CODES</td>
<td>NUMBER</td>
<td>DESCRIPTION</td>
</tr>
<tr>
<td>-------</td>
<td>--------</td>
<td>-------------</td>
</tr>
<tr>
<td>HCPCS</td>
<td>None</td>
<td></td>
</tr>
</tbody>
</table>