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ORIGINAL REPORT

Can Urinary PCA3 Supplement PSA in the Early Detection of Prostate Cancer?

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See accompanying editorial on page 4033

A B S T R A C T

Purpose
Given the limited sensitivity and specificity of prostate-specific antigen (PSA), its widespread use as a screening tool has raised concerns for the overdiagnosis of low-risk and the underdiagnosis of high-grade prostate cancer. To improve early-detection biopsy decisions, the National Cancer Institute conducted a prospective validation trial to assess the diagnostic performance of the prostate cancer antigen 3 (PCA3) urinary assay for the detection of prostate cancer among men screened with PSA.

Patients and Methods
In all, 859 men (mean age, 62 years) from 11 centers scheduled for a diagnostic prostate biopsy between December 2009 and June 2011 were enrolled. The primary outcomes were to assess whether PCA3 could improve the positive predictive value (PPV) for an initial biopsy (at a score > 60) and the negative predictive value (NPV) for a repeat biopsy (at a score < 20).

Results
For the detection of any cancer, PPV was 80% (95% CI, 72% to 86%) in the initial biopsy group, and NPV was 88% (95% CI, 81% to 93%) in the repeat biopsy group. The addition of PCA3 to individual risk estimation models (which included age, race/ethnicity, prior biopsy, PSA, and digital rectal examination) improved the stratification of cancer and of high-grade cancer.

Conclusion
These data independently support the role of PCA3 in reducing the burden of prostate biopsies among men undergoing a repeat prostate biopsy. For biopsy-naive patients, a high PCA3 score (> 60) significantly increases the probability that an initial prostate biopsy will identify cancer.

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INTRODUCTION

Early detection of prostate cancer is in need of new clinical assays to reduce overdetection and improve detection of aggressive disease. Despite randomized trials examining the role of prostate-specific antigen (PSA)[1] as a screening test, controversy regarding its widespread use persists.[2,3] In addition, overdiagnosis of low-risk disease has contributed to overtreatment of prostate cancer, thus leading to excess morbidity.[4] These results have led to conflicting guidance from professional organizations, including the United States Preventive Services Task Force, the American Urological Association, the American Society of Clinical Oncology, and the American Cancer Society.[5-8]

The suboptimal performance of PSA in prostate cancer detection stems from the fact that PSA is not cancer specific; other common conditions such as benign prostatic hyperplasia, prostatitis, and urinary tract infections may lead to elevated PSA levels.[9] In practice, a PSA of more than 4 ng/mL often triggers a prostate biopsy, which is costly, invasive, and associated with the risk of bleeding and sepsis.[10,11] Attempts to improve performance of PSA have led to the use of several PSA derivative tests such as percent free PSA,[12,13] age-specific PSA ranges,[14] PSA velocity,[15] and a novel clipped form of the precursor form of PSA (-2proPSA),[16,17] but these are constrained by the same limitations as PSA itself—namely, confounding by benign prostatic conditions. One way to improve the value of early PSA-based detection would be to supplement PSA with a test that can improve cancer risk stratification, especially for high-grade disease.

In contrast to PSA, prostate cancer antigen 3 (PCA3) is a noncoding, large-chain RNA that is highly overexpressed in prostate cancer compared with noncancerous prostate tissue.[18,19] PCA3 is detectable in the urine of men with prostate cancer and has been found to be independent of prostate size and serum PSA level.[20-24] In February 2012, the US Food and Drug Administration registered PCA3 as a risk assessment for prostate cancer with an indicated use to facilitate biopsy decision making among men with prior negative prostate biopsies. To date, several studies[25-27] have examined the performance of PCA3 in determining the probability of cancer detection. Although high sensitivity (52% to 58%) and specificity (72% to 87%)[25,28] have been reported, its ability to improve the performance of PSA-based detection of prostate cancer (including high-grade disease) has not been examined in an independent, hypothesis-driven validation trial. Therefore, in this multicenter trial, we evaluated the performance of a urinary PCA3 assay for the detection of prostate cancer and examined the improvement in diagnostic accuracy above and beyond standard clinical risk factors.

PATIENTS AND METHODS

Study Design

This prospective randomized open blinded end point–compliant study[29] was designed to validate the use of PCA3 to complement PSA-based detection of prostate cancer. The targeted population included men who had previously been screened for prostate cancer, primarily with a PSA test, some of whom had undergone prior prostate biopsy. This National Cancer Institute Early Detection Research Network validation trial was conducted at 11 centers in the United States; institutional review board approval was obtained at each site, and patients provided written informed consent.

Study Populations

From December 2009 to June 2011, 928 men scheduled for a prostate biopsy were invited to participate in this validation trial (Fig 1). They were required to have one or more of the following indications: elevated or increasing PSA, less than 15% free PSA, positive family history, prior atypical small acinar proliferation or high-grade prostate intraepithelial neoplasia, or abnormal digital rectal examination (DRE). Men who had a history of prostate cancer, were participating in an intervention trial for prostate disease, had prior prostate surgery, had a prior saturation biopsy or any prostate biopsy within 6 months of consenting, and had prior exposure to PCA3 testing were excluded.

Specimen Collection and Assays

Urine specimens for PCA3 measurement were collected after an attentive DRE in which the examiner performed a minimum of 6 pressed strokes on the prostate from lateral to medial and before undergoing the prostate biopsy. Each urine sample was processed by transferring to the Progensa PCA3 urine specimen transport tube, stored locally at ≤ -70°C, and batch-shipped to the National Cancer Institute's central repository (Frederick, MD) and then to the Johns Hopkins Early Detection Research Network Biomarker Reference Laboratory. PCA3 was assayed in duplicate (Progensa PCA3; Gen-Probe, San Diego, CA). PCA3 scores were reported as a ratio of urinary PCA3 mRNA to PSA mRNA.[30] Clinical sites were blinded to the results of PCA3 tests. During the course of the study, a random 10% of serum specimens were selected for independent quality assurance testing for PCA3 at the Gen-Probe laboratory.

Serum specimens were collected before prostate examination and biopsy, processed within 4 hours, and frozen at ≤ -70°C. Total PSA was

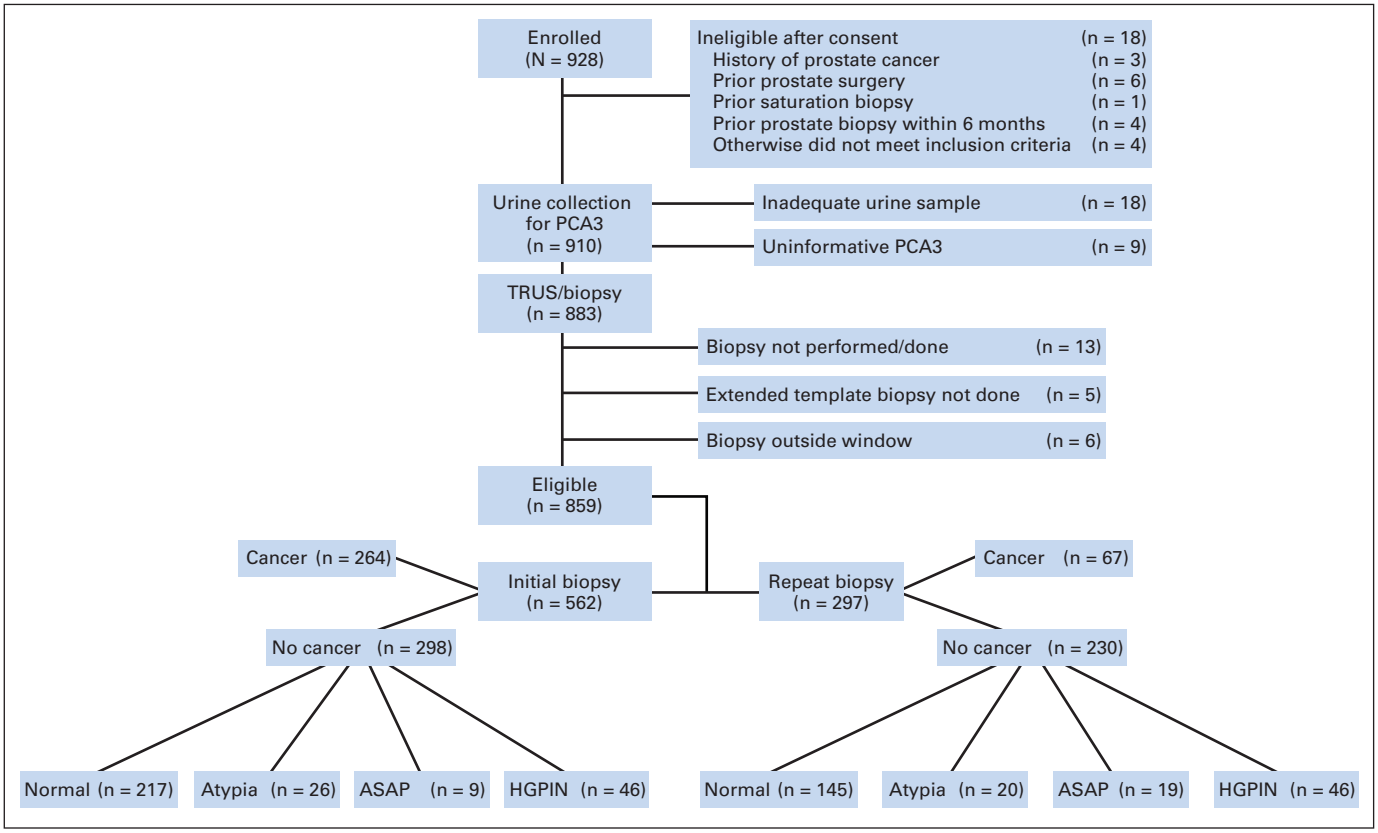

Fig 1. Standards for Reporting of Diagnostic Accuracy flow diagram. ASAP, atypical small acinar proliferation; HGPIN, high-grade prostate intraepithelial neoplasia; PCA3, prostate cancer antigen 3; TRUS, transrectal ultrasound.

measured by using the Hybritech assays on the Access II Immunoassay analyzer (Beckman Coulter, Fullerton, CA).

Prostate Biopsy and Pathologic Review

Prostate biopsies were performed under transrectal guidance by using a standard template,[31] and prostate size was measured by sonography. Pathologists at each clinical site interpreted their respective biopsy specimens; a random 10% of specimens were selected by the data coordinating center to be independently rereviewed by a central pathologist for a diagnosis of cancer.

Statistical Analyses

The primary end point of the study was diagnosis of prostate cancer on biopsy and the secondary end point was diagnosis of high-grade prostate cancer, defined as a Gleason score greater than 6. Patients were stratified into two distinct groups for the purposes of analyses: men presenting for initial prostate biopsy and those presenting for repeat prostate biopsy. Descriptive statistics summarized clinical factors; χ^2 and t tests were used to compare the initial and repeat biopsy groups.

The primary analyses, including PCA3 thresholds, were specified a priori, and statistical power was based on independent analyses of prevalidation data obtained from similar cohorts.[32,33] From preliminary data, the observed positive predictive value (PPV; PCA3 $>$ 60) was 77% for the initial biopsy group, and the observed negative predictive value (NPV; PCA3 $<$ 20) was 86% for the repeat biopsy group. At PPV of 75% or NPV of 85% for the respective populations, 305 repeat biopsies (assuming 44% with PCA3 $<$ 20) and 273 initial biopsies (assuming 18% with PCA3 $>$ 60) were needed to achieve 90% power. Adjusting for the ratio of initial versus repeat biopsies on the basis of the preliminary data, the minimum total sample size necessary would be 610 men who present for biopsy. However, the actual composition of two groups and the prevalence of men with PCA3 score of more than 60 or less than 20 may differ from the preliminary data; therefore, we inflated the calculated sample size by 40% to 850. The clinical application for a diagnostic test in the initial

Table 1. Baseline Characteristics of Study Population

Characteristic	Total				Initial Biopsy				Repeat Biopsy				P*
	No.	%	Mean	SD	No.	%	Mean	SD	No.	%	Mean	SD	
No. of patients	859				562	65			297	35			
Age, years			62	8			62	8			64	8	$<$ .001†
Race													.34‡
White	683	80			442	79			241	82			
Black	110	13			79	14			31	11			
Other	63	7			41	7			22	7			
Hispanic ethnicity	70	8			43	8			27	9			.47‡
Family history of prostate cancer	242	29			153	28			89	31			.28‡
Indication for biopsy													$<$ .001‡
Abnormal DRE	141	16			109	19			32	11			
PSA $>$ 2.0 ng/mL	503	59			332	59			171	58			
Elevated PSA velocity	170	20			108	19			62	21			
Lower PSA value with other risk factors	3	$<$ 1			2	$<$ 1			1	$<$ 1			
Prior ASAP or HGPIN	20	2			0	0			20	7			
% free PSA $<$ 15%	12	1			9	2			3	1			
Other	10	1			2	$<$ 1			8	3			
Study PSA			8	14			7	15			10	10	.003†
Study free PSA			1.1	1.5			1.0	1.5			1.4	1.4	$<$ .001†
PCA3 score			44.4	56.9			46.9	59.7			39.9	51.1	.07†
5ARI use	107	12			46	8			61	20			$<$ .001‡
Prostate size on TRUS	51	31			44	22			65	38			$<$ .001†
No. of cores taken			13	3			12	1			13	5	$<$ .001†
Prostate cancer found on index biopsy	331	38			264	47			67	23			$<$ .001‡
Clinical T stage among positive biopsies													.02‡
T1	225	68			175	67			50	76			
T2	72	22			66	25			6	9			
T3	2	1			1	$<$ 1			1	2			
T4	0	0			0	0			0	0			
Tx	30	9			21	8			9	14			
Gleason score among positive biopsies													.06‡
6	157	47			116	44			41	61			
7	131	40			114	43			17	25			
8	23	7			17	6			6	9			
9	17	5			15	6			2	3			
10	3	1			2	1			1	1			

NOTE. There were incomplete or missing data for race (n = 3), Hispanic ethnicity (n = 5), family history of prostate cancer (n = 18), central measurement of prostate-specific antigen (PSA) and free PSA (n = 2), prostate size from transrectal ultrasound (TRUS; n = 32), and clinical T stage (n = 2).

Abbreviations: 5ARI, 5-alpha-reductase inhibitor; ASAP, atypical small acinar proliferation; DRE, digital rectal examination; HGPIN, high-grade prostate intraepithelial neoplasia; IQR, interquartile range; PCA3, prostate cancer antigen 3; SD, standard deviation.

*P value comparing initial and repeat biopsy groups.

†t test.

‡χ^2 test.

biopsy setting is generally for the detection of cancer; thus, we emphasized optimizing the PPV in the trial design. We calculated the PPV (at a PCA3 > 60) and its 95% CI and tested the null hypothesis that PPV at this PCA3 threshold would not be significantly higher than 55% (55% was selected as a prebiopsy probability for a positive biopsy in clinical cohorts; the PPVs were reported to be approximately 44% to 45%, and a 10% improvement in PPV was considered clinically meaningful). In contrast, men presenting for follow-up after a prior negative biopsy were generally seeking to avoid repeating the biopsy; therefore, in the design phase, we optimized for the NPV for the repeat biopsy group. For this group, we calculated the NPV (at a PCA3 < 20) and its 95% CI and tested the null hypothesis that NPV at this threshold would not be significantly larger than 75%. The primary analysis results adjusted for interim analysis by using the uniformly minimal variance unbiased estimator[34] did not differ from the unadjusted primary analysis results. PPV and NPV were also calculated in repeat biopsy and initial biopsy groups, respectively, although these were not a priori hypotheses.

A secondary objective of this validation study was to examine PCA3 as a panel member to improve prediction of prostate cancer risk in addition to known risk factors—age, race, PSA, DRE abnormalities, prior negative biopsy, and family history of prostate cancer. These variables are expressed as a risk value based on the Prostate Cancer Prevention Trial's (PCPT's) risk calculator.[35,36] We plotted receiver operating characteristic curves across two groups—PCPT risk and PCPT risk with PCA3 score combined—in logistic regression models predicting a positive biopsy for cancer. To test whether PCPT (with and without PCA3) has diagnostic value for discriminating aggressive cancer, we repeated these analyses for the prediction of high-grade prostate cancer (defined as Gleason score > 6). The area under the curve (AUC) derived from the receiver operating characteristic curve was calculated by the average of AUCs from 10-fold cross-validation sets. The likelihood ratio test was used to test the statistical significance of the added contribution of PCA3 to the PCPT model.[37] Tables examining the individual risk for prostate cancer and high-grade prostate cancer risks were generated from logistic regression models for PCA3 score (< 20, 20 to 60, > 60) and PSA level (< 4, 4 to 10, > 10 ng/mL). Statistical analyses were performed by the data coordinating center using SAS version 9.2 (SAS Institute, Cary, NC).

RESULTS

Among 928 patients who were enrolled, 18 were deemed ineligible, 18 did not have an adequate urine sample for PCA3 assay, nine had an uninformative PCA3 test result, and 24 did not undergo a biopsy as required by protocol (Fig 1), leaving 859 evaluable patients (mean age, 62 years) for analyses. Baseline demographic and clinical characteristics are summarized in Table 1 and are stratified by initial and repeat biopsy status. Of these, 562 patients were presenting for their initial prostate biopsy. Thirteen percent of the sample was African American, and 8% reported a Hispanic ethnicity. The mean PSA was 8 ng/mL and was higher among men presenting for repeat biopsy ($P = .003$). Family history for prostate cancer was reported by 29%, and an abnormal DRE was present in 16%.

An informative PCA3 test with values ranging from 0.9 to 555.6 was achieved in 99% of consenting patients. The distribution of PCA3 scores in men undergoing an initial or repeat biopsy stratified by biopsy findings is summarized in Table 2. PCA3 values were not correlated with prostate size ($r_p = -0.06$; $P = .10$), but they were associated with PSA ($r_p = 0.08$; $P = .01$), albeit with a small r value. Random 10% samples were selected for measurement of PCA3 by the manufacturer (Gen-Probe; Spearman $r = 0.95$; $P < .001$) and for centralized pathology review for the presence of prostate cancer (98% concordance).

Validation of PCA3 Among Men Presenting for an Initial Prostate Biopsy

In this scenario, the PPV was 80% (95% CI, 72% to 86%) in the initial biopsy group. By using a PCA3 score of more than 60, diagnostic sensitivity and specificity of PCA3 in the initial biopsy group was 0.42 (95% CI, 0.36 to 0.48) and 0.91 (95% CI, 0.87 to 0.94), respectively (Table 2).

Validation of PCA3 Among Men Presenting for a Repeat Prostate Biopsy

In this scenario, NPV was 88% (95% CI, 81% to 93%) in the repeat biopsy group. By using a PCA3 score of less than 20 among men with prior biopsies, sensitivity and specificity were 0.76 (95% CI, 0.64 to 0.86) and 0.52 (95% CI, 0.45 to 0.58), respectively (Table 2).

Impact of PCA3 on Risk Assessment

We divided the initial and repeat biopsy patients into groups on the basis of serum PSA level and then determined the risk of cancer or high-grade cancer for men with PCA3 scores of less than 20, 20 to 60, and more than 60 (Table 3). Increasing PCA3 score correlates with a higher probability of any cancer as well as high-grade cancer regardless of PSA cutoff. Roughly two thirds of participants had a PCA3 score of either more than 60 or less than 20 and could have benefited by this risk stratification.

We then examined PCA3 performance in combination with the PCPT risk calculator with regard to the detection of any prostate cancer and high-grade prostate cancer (Table 4). In the setting of an initial prostate biopsy, classification was improved by the addition of PCA3 to the PCPT risk calculator factors with an AUC increasing

Table 2. PCA3 Distribution and Cancer Detection on Biopsy (unadjusted)

	Initial Biopsy (n = 562)				Repeat Biopsy (n = 297)					
	Benign		Cancer		Benign		Cancer		Total	
PCA3	No.	%	No.	%	No.	%	No.	%	No.	%
< 20	175	59	56	21	119	52	16	24	366	43
20-35	64	21	39	15	48	21	15	22	166	19
35.1-60	31	11	57	22	35	15	8	12	131	15
> 60	28	9	112	42	28	12	28	42	196	23
All	298	100	264	100	230	100	67	100	859	100

Abbreviation: PCA3, prostate cancer antigen 3.

Table 3. Impact of Supplementing PSA Cutoffs With PCA3 Scores in Probability of Detecting Any Cancer and HG Cancer

PCA3 Score	Initial Biopsy PSA (ng/mL)				Repeat Biopsy PSA (ng/mL)			
	< 4	4 to 10	> 10	Total	< 4	4 to 10	> 10	Total
PCA3 score < 20								
Any cancer	31	44	67	41	15	18	22	19
Any cancer (plus PCA3)	17	26	49	25	7	10	11	9
Any cancer (observed)	15	26	53	24	9	13	12	12
HG cancer	11	20	53	19	4	6	11	7
HG cancer (plus PCA3)	6	12	39	12	2	3	6	4
HG cancer (observed)	5	15	41	13	0	3	6	2
PCA3 score 20 to 60								
Any cancer	34	52	68	49	15	18	31	21
Any cancer (plus PCA3)	39	56	69	53	16	19	33	23
Any cancer (observed)	35	56	59	50	21	22	21	22
HG cancer	13	29	54	28	3	7	16	9
HG cancer (plus PCA3)	14	29	53	28	3	6	16	9
HG cancer (observed)	15	28	55	28	0	7	15	8
PCA3 score > 60								
Any cancer	41	55	85	56	32	24	41	30
Any cancer (plus PCA3)	65	77	94	76	47	42	63	49
Any cancer (observed)	65	82	100	80	33	52	50	50
HG cancer	18	30	77	33	10	8	23	13
HG cancer (plus PCA3)	27	44	87	45	14	14	34	21
HG cancer (observed)	23	42	100	45	0	21	30	23
Total (any cancer)	34	50	72	47	16	19	30	22
Total (HG cancer)	13	26	60	26	4	7	16	9

NOTE. "Any cancer" denotes the probability of detection of prostate cancer based on a multivariable logistic regression risk prediction model that includes prostate-specific antigen (PSA), first-degree family history, digital rectal examination, prior biopsy, age, and prostate size. "High-grade (HG) cancer" denotes the probability of detection of high-grade prostate cancer based on a multivariable logistic regression risk prediction model that includes PSA, digital rectal examination, prior biopsy, age, African American race, prostate size, and free PSA. "Plus prostate cancer antigen 2 (plus PCA3)" denotes the probability of detection of prostate cancer when a risk prediction model is supplemented with PCA3 score. "Observed" denotes actual cancer or high-grade cancer detection in the cohort.

from 0.68 for PCPT to 0.79 for PCPT plus PCA3 ($P < .001$). Similarly, classification in the repeat biopsy setting was improved by the addition of PCA3 to the PCPT risk calculator factors with an AUC increasing from 0.64 for PCPT to 0.69 for PCPT plus PCA3 ($P < .001$). For the detection of high-grade cancer, AUC improvements for initial and repeat biopsies were 0.74 to 0.78 and 0.74 to 0.79, respectively ($P \leq .003$).

By plotting model risk for cancer detection against the actual percentile cancer risk (predictiveness curve[38,39]), one can compare cancer risk in a continuous fashion (Data Supplement).

Table 4. ROC AUC for Initial and Repeat Biopsies Comparing PCPT Model with PCPT Model Plus PCA3 After Correction by 10-Fold Cross Validation

Model	ROC AUC			
	Any Cancer		HG Cancer	
	Initial	Repeat	Initial	Repeat
PCPT	0.68	0.64	0.74	0.74
PCPT plus PCA3	0.79	0.69	0.78	0.79
Likelihood ratio test	< .001	< .001	< .001	.003

Abbreviations: AUC, area under the curve; HG, high-grade; PCA3, prostate cancer antigen 3; PCPT, Prostate Cancer Prevention Trial; ROC, receiver operating characteristic.

Biopsies Avoided and Potential for Underdiagnosis of Cancer

An important potential benefit of modifying the current screening policy by the addition of PCA3 would be to decrease the need to undergo a repeat prostate biopsy. In this trial, if a group with a PCA3 score of less than 20 and a PSA of less than 4 ng/mL is considered to be low risk, then 23 of 297 men (8%) would have avoided a biopsy among the repeat biopsy population (Appendix Table A1, online only). Keeping in mind that the benefit of reducing biopsies may be outweighed by an increased risk for underdetection of cancer, two of 23 patients with cancer (9%) would have been underdiagnosed, neither of which had high-grade cancers. Focusing only on men with PCA3 scores of less than 20, regardless of their PSA, 46% would have avoided a biopsy; however, 12% would have had undiagnosed cancer and 3% would have had undiagnosed high-grade cancer. Likewise, thresholds can be applied to those undergoing an initial biopsy; however, there is a higher rate of underdiagnosis of aggressive cancers at a PCA3 score of less than 20 (13%).

Role of PCA3

In this comprehensive validation trial, the test performance of PCA3 exceeded a priori thresholds typical of PSA-based detection of prostate cancer. Moreover, the addition of PCA3 to individual risk estimation models, which included age, race, prior biopsy, PSA, and DRE, improved the stratification of cancer and high-grade cancer

risks. These results empirically demonstrate that overdetection of low-grade cancer in the repeat biopsy setting and underdetection of high-grade cancer in the initial biopsy setting can be improved through the adoption of new biomarkers such as PCA3.

PSA-based prostate cancer screening, although born of good intentions, has become a flashpoint in health policy, with the US Preventive Services Task Force facing off against specialists, patient advocates, and professional societies. First, preventive approaches to care promote early detection of curable cancers to avoid the agonizing, painful path to metastatic cancer death. Conversely, when some evidence fails to support the policy, it is difficult to retract an established clinical practice, but this is precisely where the issue of PSA screening is now. How best to address this is more than a mere policy issue, because countless men are routinely confronted with having to decide whether to be screened for prostate cancer or not, and if they decide to be screened, how best to do that.

A reasonable middle ground in this debate may be to allow PSA-based cancer screening[8] but include supplementary information and tests to refine the risk of having high-grade prostate cancer. Given that the basis for this controversy is the limited performance characteristics of PSA, we examined PCA3 as a supplementary, noninvasive diagnostic test to improve test performance. In this multicenter validation study of men who were prescreened with PSA, PCA3 significantly improved the prebiopsy probability for cancer detection. This incremental effect of PCA3 was observed for both initial and repeat prostate biopsy groups, and there is also evidence to suggest that PCA3 may aid in the detection of high-grade prostate cancer. Moreover, at the individual patient level, using PCA3 to augment PSA-based screening not only improved diagnostic accuracy over conventional clinical risk factors but also significantly enhanced diagnostic certainty.

The underdiagnosis of high-grade cancer could be considered a potential trade-off when supplementing PSA early detection with PCA3. At a PCA3 score of 20 or lower, we found that the rate of underdiagnosis of high-grade cancers was low in the setting of repeat prostate biopsy; however, a high rate of underdiagnosis was observed in the initial biopsy setting. This latter finding suggests that a lower PCA3 threshold or other biomarkers may be necessary to rule out a biopsy in the initial setting but does not detract from its ability to rule in a biopsy at a PCA3 of more than 60.

By including men undergoing initial and repeat biopsies, we were able to validate the use of PCA3 beyond the narrow indication that the US Food and Drug Administration subsequently approved. With more than 1 million prostate biopsies per year in the United States, the addition of PCA3 may decrease morbidity, which is an area of concern raised by the US Preventive Services Task Force. Conversely, one might criticize use of PCA3 testing because it may result in some high-grade prostate cancers not being detected. Nevertheless, such a policy seems preferable to one that does not screen at all and thus fails to diagnose all cases of high-grade cancers until they have reached advanced stages.

For PCA3 to be useful, it should add to the available clinical information that builds a case for or against recommending a biopsy. Our models not only validated the role for PCA3 in the detection of prostate cancer but also found that PCA3 enhanced risk determination as estimated by the patient-level PCPT risk models. From a patient's perspective, PCA3 may alter the need for an invasive prostate biopsy in a way that reduces uncertainly for a diagnosis of prostate cancer.

Several limitations should be noted in the context of these findings. Because the potential role of PCA3 as a screening test to replace PSA screening was not examined, these findings do not extend to men who have not been prescreened. We followed patients only through the index biopsy, and it is likely that some of the patients had false-negative prostate biopsies, given the known sampling error of a needle biopsy for the detection of cancer and the underdetection of high-grade prostate cancer.

In conclusion, in the repeat biopsy setting, a PCA3 score cutoff of 20 yields a high NPV and could help avoid unnecessary repeat biopsies. For biopsy-naive patients, a high PCA3 score (>60) increases the probability that cancer will be detected, but a PCA3 score of less than 20 will be necessary to rule out a biopsy in the initial setting.

AUTHORS' DISCLOSURES OF POTENTIAL CONFLICTS OF INTEREST

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Appendix

Table A1. Potential Thresholds for Performing a Biopsy and Risk for Underdiagnosis of Any Prostate Cancer and HG Prostate Cancer

	Initial Biopsy						Repeat Biopsy					
	Biopsies Avoided*		Cancers Missed†		HG Cancers Missed‡		Biopsies Avoided*		Cancers Missed‡		HG Cancers Missed	
Potential Thresholds	No./No.	%	No./No.	%	No./No.	%	No./No.	%	No./No.	%	No./No.	%
PCA3 < 20 and PSA < 4	84/562	15	13/84	15	4/84	5	23/297	8	2/23	9	0/23	0
PCA3 < 20 and PSA < 10	214/562	38	47/214	22	24/214	11	102/297	34	12/102	12	2/102	2
PCA3 < 20	231/562	41	56/231	24	31/231	13	135/297	46	16/135	12	4/135	3

Abbreviations: HG, high-grade; PCA3, prostate cancer antigen 3; PSA, prostate-specific antigen.
*Proportion of biopsies avoided out of all eligible biopsies.
†Proportion of cancers missed if biopsies are avoided.
‡Proportion of HG cancers missed if biopsies are avoided.