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A Multi-Center Study of [−2]Pro-Prostate-Specific Antigen (PSA) in Combination with PSA and Free PSA for Prostate Cancer Detection in the 2.0 to 10.0 ng/mL PSA Range

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Mizrahi, Broyles, Shin and Cruz had full access to all of the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis.

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Abstract

Purpose—PSA and free PSA (fPSA) have limited specificity for detecting clinically significant, curable prostate cancer (PCa), leading to unnecessary biopsies and detection and treatment of some indolent tumors. [−2]proPSA (p2PSA) may improve specificity for detecting clinically significant PCa. Our objective was to evaluate p2PSA, fPSA, and PSA in a mathematical formula (prostate health index [*phi*] = [−2]proPSA / fPSA) × $PSA^{1/2}$) to enhance specificity for detecting overall and high-grade PCa.

Materials and Methods—We enrolled 892 men in a prospective multi-institutional trial with no history of PCa, normal rectal examination, a PSA of 2–10 ng/mL, and ≥6- core prostate biopsy. We examined the relationship of serum PSA, %fPSA and *phi* with biopsy results. The primary endpoints were the specificity and AUC using *phi* to detect overall and Gleason ≥7 prostate cancer on biopsy compared with %fPSA.

Results—For the 2–10 ng/mL PSA range, at 80–95% sensitivity, the specificity and AUC (0.703) of *phi* exceeded those of PSA and %fPSA. Increasing *phi* was associated with a 4.7-fold increased risk of PCa and 1.61-fold increased risk of Gleason ≥7 disease on biopsy. The AUC for *phi* (0.724) exceeded that of %fPSA (0.670) in discriminating between PCa with Gleason ≥ 4+3 vs. lower grade disease or negative biopsies. *Phi* results were not associated with age and prostate volume.

Conclusions—*Phi* may be useful in PCa screening to reduce unnecessary biopsies in men age ≥50 years with PSA 2–10 ng/mL and negative DRE, with minimal loss in sensitivity.

INTRODUCTION

PSA testing was approved by the FDA using a 4.0 ng/mL cutoff for recommending prostate biopsy. Lower cutoffs further enhance early prostate cancer (PCa) detection,[1] since PSA correlates with the risk of overall and high-grade PCa at PSA concentrations <4 ng/mL.[2] However, PSA testing may be confounded by benign conditions.

The low specificity at PSA <10.0 ng/mL has created a diagnostic gray zone in which PCa is found on biopsy in ~25% of patients. This is important, since most PCa is curable at PSA <10.0 ng/mL; whereas, PSA >10 ng/mL often portends advanced disease.[3]

PSA in serum is either complexed with proteins or in an unbound form called free PSA (fPSA).[4] At PSA levels of 4.0–10.0 ng/mL, the ratio of fPSA to PSA (%fPSA) significantly improves discrimination between PCa and benign conditions.[5]

Different regions of the prostate contain varying proportions of fPSA isoforms, including proPSA that is associated with PCa. [−2]proPSA (p2PSA) is the primary form in PCa tissue.[6–8] At PSA of 2.0–10.0 ng/mL, p2PSA further improves specificity for PCa detection relative to %fPSA.[9–13]

The utility of p2PSA at PSA <4.0 ng/mL and its relationship to PCa aggressiveness are relevant to the PCa screening debate, including concerns about overdiagnosis and overtreatment.[13–19] Preliminary evidence suggests that a higher percentage of p2PSA may be associated with more aggressive PCa.[10, 12, 13, 19]

Selecting thresholds for clinical use of p2PSA has received limited study. We evaluated the relationship of p2PSA** combined with fPSA and PSA in a mathematical formula called Prostate Health Index (*phi*) with prostate cancer detection and tumor features.

METHODS

Study Design

We conducted a multi-center, double-blind, case-control clinical trial to validate *phi* in the 2.0–10.0 ng/mL PSA range. This formula was developed from an independent dataset,[20] and is calculated as (p2PSA pg/mL / fPSA ng/mL) × (PSA ng/mL) ^{½}. Intuitively, higher [−2] proPSA and PSA with a lower fPSA has greater likelihood of PCa. The study protocol was approved by the IRB of each participating institution, and all participants provided informed consent.

Study population

We evaluated 1372 men from October 2003 through June 2009 from 8 medical centers. The study cohort included men age ≥50 years of all ethnic backgrounds who met the following criteria: (1) no history of PCa, (2) non-suspicious digital rectal examination (DRE) findings, (3) pre-study PSA of 1.5–11.0 ng/mL (all PSA concentrations were re-tested in the Access Hybritech assay, and only those 2–10 ng/mL were included), (4) ≥6 core biopsy within 6 months of blood draw, and (5) a histologic diagnosis from prostate biopsy.

Exclusion criteria were: (1) treatment with medications that alter PSA levels or interventions such as transurethral resection of the prostate prior to blood draw, (2) acute prostatitis or urinary infection at blood draw, (3) a final Access Hybritech PSA value outside the 2.0–10.0 ng/mL range, (4) no blood draw or biopsy at the appropriate time interval, or (5) prior androgen-replacement therapy.

Seven men were excluded due to unevaluable tests from hemolyzed or lipemic samples or p2PSA duplicate results with >15% coefficient of variation at p2PSA concentrations ≤ 20 pg/mL, for which samples could not be retested. Finally, one site enrolled only men aged 55–75 years (our study enrolled men aged ≥ 50 years), and our study-specific sample storage limit (≤ 5 years) further limited the evaluable population to men aged 62–74. Because the age distribution from this site may not be representative of the target population, we performed separate analyses excluding and including these men.

The final study population of 892 men included: (1) 121 (13.6%) prospectively enrolled, (2) 743 (83.3%) prospectively enrolled under separate protocols, and (3) 28 (3.1%) retrospective samples. The study population included 706 (79.2%) initial biopsies, 159 (17.8%) repeat biopsies, and 27 (3%) with unknown history of prior biopsy. Each institution enrolled an approximately equal number of men with or without PCa, for a total of 430 (48.2%) men with PCa and 462 (51.8%) without. Participants and investigators were blinded to p2PSA results, and testing sites were blinded to individual clinical information.

Test Methods

Access Hybritech p2PSA, PSA, and fPSA assays were measured on the Beckman Coulter Access 2 Immunoassay Analyzer***. Serum samples were collected and processed within 8 hours, then stored frozen at ≤−70°C prior to testing (≤5 years from the date of blood draw), conditions that allowed accurate measurement of *phi*.[21] Samples were tested at one of 3

**Pending FDA approval.
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laboratories. PSA and fPSA assays were run using one-sample replicate. The p2PSA assay was run in duplicate (first replicate used for data analysis, consistent with the proposed product labeling) according to the testing protocol. Evaluation of the first replicate compared to the mean of duplicates using Passing-Bablock regression analyses showed no difference (Spearman R=0.9985). The p2PSA assay is a two-site immunoenzymatic sandwich assay using specific monoclonal antibodies and 6 calibrators from 0- 5000 pg/mL.

Statistical Methods

The minimum sample size was estimated as 295 patients without cancer to detect a 10% difference in specificity between *phi* and % fPSA at $\alpha = 0.05$ and $\beta = 0.10$. In addition, a minimum sample size of 350 cancer patients was determined to accurately estimate sensitivity at 95% with a 95% confidence interval of ± <3%. The target sample size was then increased to 400 participants in each group.

The primary null hypothesis was that *phi* has no greater specificity than %fPSA at 95% sensitivity. This hypothesis was tested using bootstrap-based receiver operating characteristic (ROC) analysis.[22] Briefly, 1000 datasets of benign and PCa patients were generated to repetitively sample the study population.[23–25] Differences in the specificity between phi and %fPSA at 95% sensitivity were calculated for the 1000 pairs of replicate datasets. The standard error of the difference in specificities was then estimated with adjustment for correlation between the results of the two tests. Finally, the bootstrap-estimated standard error was used to evaluate whether the difference in specificities is >0 assuming normal distribution of the differences. A one-sided statistical test was performed for this analysis. This method was also used to compare the specificities of *phi* and %fPSA at 90%, 85%, and 80% sensitivities.

The secondary null hypothesis was that the area under the ROC curve (AUC) for *phi* equals that of %fPSA. This hypothesis was tested by evaluating whether the difference between the estimated AUCs for the two tests equals 0 using empirical methods.[26, 27] The standard error of the difference was calculated accounting for the correlation in AUCs as appropriate for comparison of paired data. The difference between the two estimated AUCs has been shown to have a Chi-square distribution with one degree of freedom. The AUCs for *phi* and %fPSA were also estimated for each prostate volume tertile to determine whether the observed trend in AUCs differed by prostate volume.

The validity of pooling data across sites was evaluated by fitting a logistic regression model with cancer status as the dependent variable, with *phi* (dichotomized at the estimated cutoff for 95% sensitivity) and site as independent predictors including interaction terms for site and *phi*. A statistically significant parameter estimates for this interaction terms was considered evidence of heterogeneity in *phi* performance by site.

Comparisons between participant subgroups were performed using the Wilcoxon Rank-Sum test for continuous variables and the $\chi 2$ test for categorical variables. Two-sided statistical tests were used on all analyses except as noted above, and statistical significance was defined as $p<0.05$. All analyses were performed using SAS version 9.2 (SAS Institute, Cary, North Carolina).

Individual Patient Risk Assessment

A 25% PCa detection rate has been previously reported in men with PSA of 2.0–10.0 ng/mL.[3] For this study, cancer patients were over-sampled by design, resulting in 48.2% of study participants with PCa. Since the proportion of PCa was determined by design, direct calculation of PCa probability would result in inflated estimates for detecting PCa. Therefore, to obtain more accurate risk estimates for PCa, we adjusted the proportion of PCa

to 25% by repetitively sampling the study population 1000 times with each replicate dataset consisting of 462 (75%) benign and 154 (25%) cancer participants.[23–25] The mean probability of cancer in the bootstrapped datasets for each *phi* range was used as the point estimate, and bootstrap-estimated standard errors were used to calculate 95% confidence intervals. Likewise, relative risk estimates were calculated for each replicate dataset by dividing the probability of PCa in each *phi* range to that of *phi* 0–24.9. The mean relative risk and bootstrap-estimated standard errors were used to calculate the risk estimate and 95% confidence intervals. In addition, age-stratified probability estimates for PCa were calculated to determine whether observed trends persist in all age groups.

Association of phi with Gleason Score

Among participants with PCa, the probability of a Gleason score ≥7 was calculated directly from the proportion of participants in each *phi* range with Gleason score ≥7. Risk ratios were estimated by dividing the probability of Gleason score ≥7 in each *phi* range to that of *phi* 0–24.9. Confidence intervals were calculated using the normal approximation of the binomial distribution. The Cochrane-Armitage test for trend was used to determine whether increasing *phi* ranges corresponds to increasing probability of PCa with Gleason score ≥7. ROC analysis was used to evaluate the clinical utility of *phi* in detecting PCa with Gleason scores 4+3 or higher.

RESULTS

Participants

Table 1 shows the demographics and results for each assay. Both *phi* and p2PSA were significantly higher in PCa than controls; whereas, fPSA and %fPSA were lower in PCa than controls. Total PSA and age were comparable between groups.

Of the participants, 89.8% had ≥12-core biopsy, and 98% had ≥10 cores. Overall, 30.6%, 49.9%, and 19.6% of participants were aged 50–59, 60–69 and 70–84 years, respectively. Mean age and PSA were similar across the 7 clinical sites. In addition, none of the interaction terms in the statistical model for evaluating heterogeneity by site was significant, supporting data pooling across sites. There were no significant differences in age (P=0.123), PSA (P=0.106), p2PSA (P=0.088), %fPSA (P=0.125), or *phi* (P=0.848) between Caucasians and African-Americans.

Receiver Operating Characteristic (ROC) Results

Figure 1 shows the sensitivity and specificity for all observed PSA, fPSA, p2PSA, %fPSA, and *phi* cutoffs in the 2.0–10.0 ng/mL PSA range. At a given sensitivity, *phi* demonstrated greater specificity than the other analytes (Table 2). At 95% sensitivity, the specificity of *phi* was 16.0% compared to 8.4% for %fPSA (P=0.015), 7.6% for p2PSA, 6.5% for PSA, and 3.5% for fPSA, rejecting the primary null hypothesis. Moreover, at lower sensitivities (90%, 85%, and 80%) for PCa detection, the specificity of *phi* was significantly greater than %fPSA (i.e., unnecessary biopsies possibly avoided: 26% vs. 18%, P= 0.036; 39% vs. 28%, P= 0.006; 45% vs. 37%, P= 0.031, respectively).

The AUC for PCa detection was significantly greater for *phi* (AUC=0.703) than for %fPSA (0.648, P=0.004), fPSA (0.615), p2PSA (0.557), or PSA (0.525), rejecting the secondary null hypothesis.

Individual Patient Risk Assessment

Higher *phi* values were associated with an increased risk of PCa detection based upon the adjusted 25% proportion of PCa cases (Table 3). Of the study population, 25%, 33%, 30%,

and 13% had *phi* values of 0–24.9, 25.0–34.9, 35.0–54.9, and ≥ 55.0, respectively. Compared to *phi* < 25.0, the relative risk of PCa detection on biopsy was 1.6-, 3.0-, and 4.7-fold higher at *phi* values of 25.0–34.9, 35.0–54.9, and ≥ 55.0, respectively. Overall, a *phi*≥ 55.0 was associated with a 52.1% probability of PCa.

Age and Probability of PCa

Higher *phi* values were also associated with higher bootstrapped risk estimates of PCa within each age group. The probability (and relative risk [RR]) of PCa ranged from 10.9% (*phi* 0–24.9) to 53.4% (*phi* ≥ 55) (RR 4.9) for the 50–59 age group, 12.5% (*phi* 0–24.9) to 54.5% (*phi* ≥ 55) (RR 4.4) for the 60–69 age group, and 5.8% (*phi* 0–24.9) to 44.8% (*phi* ≥ 55) (RR 7.7) for the > 70 age group.

Association of phi with Gleason Score

Phi also had a significant relationship with biopsy Gleason score (r=0.138, P=0.004). Among participants with PCa, biopsy Gleason score was <7 in 290 (67.6%) and ≥7 in 139 (32.4%) Compared to *phi* < 25.0, the relative risk of Gleason ≥ 7 PCa increased to 1.08 for *phi* values from 25.0–34.9, 1.15 for *phi* values from 35.0–54.9, and 1.61 for *phi* ≥ 55.0. The corresponding proportion of cancers with a Gleason score ≥ 7 increased from 26.2% to 28.2%, 30.1%, and 42.1% at *phi* values of 0–24.9, 25.0–34.9, 35.0–54.9, and ≥ 55.0, respectively (Cochran-Armitage test for trend, P=0.013) (Table 4). The AUC for *phi* (0.724) exceeded that of %fPSA (0.670) in discriminating between Gleason ≥ 4+3 vs. lower Gleason grade PCa or negative biopsies.

Relationship of TRUS volume and phi

The AUCs for *phi* exceeded those of %fPSA in all three prostate volume tertiles (≤38, 39–53, and ≥54cc): 1st tertile: AUC 0.693 for *phi* vs. 0.614 for %fPSA; 2nd tertile: 0.707 vs. 0.593; 3rd tertile: 0.642 vs. 0.559.

Evaluation of Excluded Participants

AUCs for *phi* with and without the excluded site were 0.696 and 0.703, respectively. Similarly, AUCs for %fPSA were 0.634 and 0.648, respectively.

COMMENT

Prostate biopsy is routinely recommended for suspicious DRE results regardless of PSA.[3] Biopsy is also recommended using PSA thresholds ranging from 2.5 to 4.0 ng/mL.[1, 2, 15] However, this has led to unnecessary biopsies and possible over-detection of some cancers.[15–17] To elucidate whether *phi* PSA-isoform measurement can improve PCa early detection, we examined a large, prospective cohort to predict biopsy findings in patients with moderate PSA elevations (2.0–10.0 ng/mL) and benign DRE findings. Such men are at higher risk of PCa (25% cancer detection rate compared with 4% in the general male population aged ≥50 years).[3] Our bootstrapped population was designed to mirror this 25% incidence of PCa on biopsy.

Prostate biopsy may be associated with discomfort, anxiety, and financial costs. Minor complications occur frequently, and major complications are possible, underscoring the need for more specific markers to reduce unnecessary biopsies. We sought to determine the utility of p2PSA and *phi* for this clinical goal.

Precursor forms of PSA have been shown to improve the accuracy of PSA for detecting PCa.[5, 6, 9–12, 28, 29] Specifically, preliminary reports suggest that p2PSA may be useful at PSA concentrations from 2.0–10.0 ng/mL.[6, 9–12, 28, 29] Some, but not all, studies have

suggested an association between proPSA and PCa aggressiveness.[10, 12, 20] Thus, p2PSA and *phi* are being investigated in active surveillance programs to help overtreatment of insignificant PCa.[19, 30]

Catalona et al. previously reported in the PSA range of 2.0–10.0 ng/mL, the proPSA-to-fPSA ratio (%proPSA) yielded a higher specificity than %fPSA.[9] Results from a separate multi-site study also supported the role of p2PSA, in combination with PSA and fPSA, in reducing unnecessary biopsies.[12, 13]

In the current study, the specificity for *phi* was higher than %fPSA at all pre-specified sensitivities, and PCa risk increased directly with increasing *phi* values. This suggests a role for *phi* as a patient monitoring tool, since increasing *phi* values reflect PCa risk.[19] For example, at 95% sensitivity, the specificity of *phi* was 16.0% compared to 8.4% for %fPSA. Moreover, at lower sensitivities (90%, 85%, and 80%) for PCa detection that might be preferred to reduce the detection of possibly “insignificant” tumors, *phi* had a significantly greater specificity than %fPSA. These results were consistent across age groups, PSA concentrations, and ethnic groups, suggesting that they are representative of the intended-use population.

For individual risk assessment, the probability of PCa varied considerably based upon *phi* values. For example, a man with a *phi* ≥ 55 (13% of the study population) had a > 52% probability of PCa and 4.7-fold increased relative risk of positive biopsy. In contrast, at approximately 90% sensitivity, a patient with a *phi* < 25 had an 11% probability of PCa.

For the PCa group, higher *phi* values were also significantly associated with a higher percentage of biopsy Gleason grade ≥ 7, ranging from 26% to 42% for *phi* concentrations < 25 and ≥ 55, respectively. For the entire study population, the AUC for *phi* (0.724) exceeded that of %fPSA (0.670) in discriminating Gleason ≥ 4+3 PCa vs. lower Gleason grade PCa or negative biopsies. Using a *phi* cutoff of 21.3 (95% sensitivity), 25% of missed cancers were Gleason score ≥7; therefore, careful surveillance is necessary. The AUCs for *phi* also exceeded those of %fPSA in all three prostate volume tertiles, suggesting that *phi* provides better discrimination of PCa from benign disease than %fPSA across the spectrum of prostate volumes. Because *phi* did not differ by age and race these results suggest that *phi* may be applicable to a broad spectrum of men as an adjunct to predict clinically-significant PCa.

The large number of subjects in the present validation study provides confidence in the *phi* cutoffs determined. *Phi* is highly effective when used in patients with moderately elevated PSA concentrations who may be most likely to benefit from early diagnosis and curative PCa treatment. A physician might recommend biopsy for a patient with a *phi* ≥ 55.0 (risk = 52.1%) and surveillance for some men with a *phi* <25.0 (risk = 11.0%). For patients reluctant to undergo prostatic biopsy, a high *phi* might increase compliance with the appropriate follow-up.

We conclude that the *phi* measurement ([−2]proPSA / fPSA) × $PSA^{1/2}$) may be useful to reduce unnecessary biopsies with improved specificity at various sensitivities for PCa detection in men age ≥50 years with PSA concentrations from 2.0–10.0 ng/mL, and negative DRE findings.****

****Our results apply to the Access Hybritech p2PSA, PSA and fPSA assays on the Beckman Coulter Access Immunoassay Systems, as studies have shown that results differ when assays from different manufacturers or standardization are used.[31]

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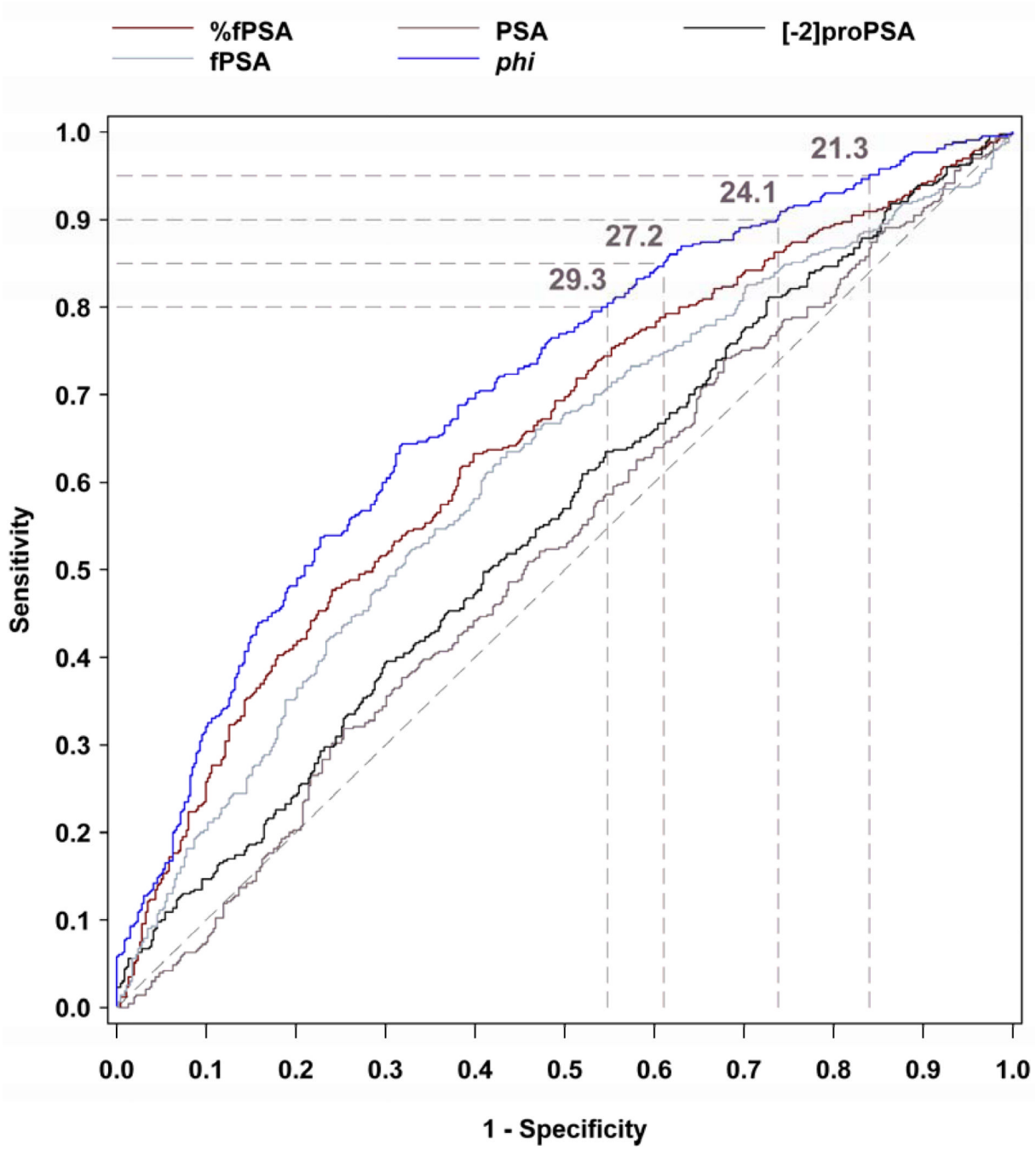

Figure 1.
PSA, fPSA, [−2]proPSA, %fPSA, and *Phi* ROC Curves in the 2–10 ng/mL PSA Range
Sensitivity × 1-Specificity for Sequential Cutpoints

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TABLE 1

Clinical Characteristics of the Study Population

Characteristic		Benign N=462	Cancer N=430	p-value	Total N=892
Age	**Median**	63.0	63.0		63.0
	Mean ± SD	62.6 ± 7.0	63.0 ± 7.1		62.8 (7.0)
	Range	50 – 84	50 – 84		50 – 84
				0.477	
Race, n(%)	**Caucasian**	361 (78.1)	365 (84.9)		726 (81.4)
	African-American	24 (5.2)	22 (5.1)		46 (5.2)
	Other	22 (4.8)	9 (2.1)		31 (3.5)
	Unknown	55 (11.9)	34 (7.9)		89 (10.0)
				0.025	
Ethnicity, n(%)	**Hispanic**	14 (3.0)	6 (1.4)		20 (2.2)
	Not Hispanic	187 (40.5)	153 (35.6)		340 (38.1)
	Unknown	261 (56.5)	271 (63.0)		532 (59.6)
				0.059	
Prostate Volume	**Median**	51.0	40.0		45.0
	Mean ± SD	55.1 ± 23.2	44.3 ± 19.4		50.1 ± 22.2
	Range	16 – 209	14 – 120		14 – 209
				<0.001	
Prior Biopsy, n(%)	**No prior biopsy**	345 (74.7)	361 (84.0)		706 (79.2)
	Prior biopsy	105 (22.7)	54 (12.6)		159 (17.8)
	Unknown	12 (2.6)	15 (3.5)		27 (3.0)
				<0.001	
Gleason Score, n(%)	**5**	Not Applicable	1 (0.2)		1 (0.2)
	6		289 (67.2)		289 (67.2)
	7		119 (27.7)		119 (27.7)
	8		9 (2.0)		9 (2.0)
	9		11 (2.6)		11 (2.6)

Characteristic		Benign N=462	Cancer N=430	p-value	Total N=892
	Unknown		1 (0.2)		1 (0.2)
				Not Applicable	
PSA (ng/mL)	**Median**	5.1	5.3		5.1
	Mean ± SD	5.3 ± 1.9	5.4 ± 1.9		5.4 ± 1.9
	Range	2.0 – 10.0	2.0 – 9.8		2.0 – 10.0
				0.199	
fPSA (ng/mL)	**Median**	1.0	0.7		0.9
	Mean ± SD	1.0 ± 0.5	0.9 ± 0.5		1.0 ± 0.5
	Range	0.1 – 4.3	0.2 – 3.9		0.1 – 4.3
				<0.001	
[−2]proPSA (pg/mL)	**Median**	12.9	14.1		13.3
	Mean ± SD	14.4 ± 7.1	16.8 ± 11.1		15.5 ± 9.3
	Range	2.9 – 43.5	2.9 – 93.5		2.9 – 93.5
				0.003	
%fPSA	**Median**	18.8	15.1		17.0
	Mean ± SD	20.0 ± 8.0	16.4 ± 7.6		18.3 ± 8.0
	Range	3.1 – 53.2	3.7 – 51.1		3.1 – 53.2
				<0.001	
phi	**Median**	30.3	42.2		34.7
	Mean ± SD	33.9 ± 15.0	49.2 ± 31.3		41.3 ± 25.5
	Range	13.7 – 98.2	10.2 – 325.8		10.2 – 325.8
				<0.001	

TABLE 2

Sensitivity and Specificity for PCa Using Various *phi* Cutoffs in Men with Non-Suspicious DRE

% Sensitivity	*phi* Cutoff	% Specificity (n)
99	**17.2**	5.2 (24)
98	**18.4**	8.4 (39)
95	**21.3**	16.0 (74)
90	**24.1**	26.2 (121)
89.1	**25.0**	29.4 (136)
85	**27.2**	39.0 (180)
80	**29.3**	45.2 (209)
75	**31.1**	52.6 (243)
70	**33.4**	60.0 (277)
65	**35.0**	65.2 (301)
60	**37.5**	70.3 (325)
55	**39.1**	74.2 (343)
50	**42.2**	79.0 (365)
45	**44.3**	82.7 (382)
40	**46.7**	85.7 (396)
35	**49.3**	87.4 (404)
30	**52.6**	90.7 (419)
25	**55.9**	91.8 (424)
20	**61.9**	93.7 (433)
15	**67.6**	95.2 (440)
10	**78.1**	97.6 (451)
5	**104.2**	100 (462)

TABLE 3

Risk Assessment Probability of PCa using *phi*

phi **Range**	**Probability of Cancer (95% Confidence Interval)**	**Relative Risk (95% Confidence Interval)**	**Percent of patients in *phi* range**
0–24.9	11.0% (6.5% – 15.8%)	1.0	24.9%
25.0–34.9	18.1% (13.7% – 22.6%)	1.6 (1.0 – 3.1)	32.8%
35.0–54.9	32.7% (27.3% – 38.0%)	3.0 (1.9 – 5.3)	29.5%
55.0+	52.1% (42.0% – 62.1%)	4.7 (3.0 – 8.3)	12.8%

TABLE 4

Relationship of *phi* with Biopsy Gleason Score

	Gleason Score on Biopsy		
***phi* Range**	**Less than 7 n (%)**	**≥7 n (%)**	**Risk Ratio (95% CI)**
0–24.9	34 (73.9)	12 (26.1)	1.0
25.0–34.9	74 (71.8)	29 (28.2)	1.08 (0.61, 1.92)
35.0–54.9	116 (69.9)	50 (30.1)	1.15 (0.67, 1.98)
55.0+	66 (57.9)	48 (42.1)	1.61 (0.95, 2.75)

Note: One participant excluded with missing Gleason score.
Cochran-Armitage test for trend, p=0.01