SUMMARY OF SAFETY AND EFFECTIVENESS DATA (SSED)

I. GENERAL INFORMATION

Device Generic Name:	Multi-analyte test system with algorithmic analysis as an aid in detection of prostate cancer
Device Trade Name:	IsoPSA Assay
Device Procode:	QRF
Applicant's Name and Address:	Cleveland Diagnostics, Inc. 3615 Superior Avenue, Suite 4406A Cleveland, OH 44114

Premarket Approval Application (PMA) Number: P200048

Date of FDA Notice of Approval:	November 28, 2025
Breakthrough Device:	Granted breakthrough device status on October 4, 2019

II. INDICATIONS FOR USE

The IsoPSA Assay is an in vitro test system that combines the results of total Prostate Specific Antigen (PSA) (Elecsys, Roche Diagnostics) and free PSA (Elecsys, Roche Diagnostics) from partitioned heparinized plasma into a single numerical index. The IsoPSA Assay is indicated for use in conjunction with other patient information to aid in detection of high-grade prostate cancer (Grade Group ≥ 2; Gleason score ≥ 7) in men ≥ 50 years of age, with total PSA levels ≥ 4.0 to ≤ 10.0 ng/mL for whom a biopsy is being considered by a urologist based on current standard of care, before consideration of IsoPSA Assay results.

Prostatic biopsy is required for the diagnosis of cancer.

III. CONTRAINDICATIONS

See limitations in the IsoPSA Assay Instruction for Use.

IV. WARNINGS AND PRECAUTIONS

The IsoPSA Assay's Instruction for Use contains the following boxed warning:

- For patients with an IsoPSA index ≤ 6.0, the clinician should also consider obtaining mpMRI results to reduce the risk of missing high-grade prostate cancer (Grade Group ≥ 2; Gleason score ≥ 7). Patients with an IsoPSA Index of ≤ 6.0 and prostate mpMRI results with PI-RADS score of 1-3 have decreased likelihood of high-grade prostate cancer (Grade Group ≥ 2; Gleason score ≥ 7) on biopsy.

- The IsoPSA Assay should not be used in patients with prostate mpMRI results with PI-RADS score of 4 – 5.

The other warnings, limitations and precautions can be found in the IsoPSA Assay Instruction for Use.

V. DEVICE DESCRIPTION

The IsoPSA Assay is an integrated test system that generates a numerical index based on the results of total Prostate Specific Antigen (PSA) (Elecsys, Roche Diagnostics) and free PSA (Elecsys, Roche Diagnostics) from partitioned heparinized plasma:

A. Device Components:

The IsoPSA Assay consists of the following components:

Materials provided:

- IsoPSA Reagent: The IsoPSA Reagent vial contains a mixture of polymers and salts which form an aqueous two-phase (biphasic) system. A dye has been added to the reagent vial to enhance visualization of the phase separation and facilitate removal of a top-phase fraction aliquot.

Materials required but not provided:

- IsoPSA Controls (Sold separately): The IsoPSA Controls (set of two levels) are formulated antigens titrated in a human serum matrix to represent two different IsoPSA Index target values (Control Level 1: 2.0 – 4.5; Control Level 2: 7.0 – 9.5). These controls are used as a Quality Control (QC) check for monitoring the performance of the IsoPSA Assay testing system. Controls are stored frozen (≤-20°C) until use, equilibrated to ambient temperature before use, and assayed in the same manner as clinical samples.

- Centrifuge-Mixer Device (Sold separately): The Centrifuge-Mixer Device (Private Label, ELMI North America, CA), is an IsoPSA Assay dedicated, small, semi-automated benchtop unit that is preprogrammed to mix the plasma sample with the IsoPSA Reagent and to centrifuge the reagent vial to separate into two equilibrated phases. The centrifuge-mixer processes up to six IsoPSA Reagent vials simultaneously.

Third-party Components and Accessories:

Total and free PSA measurements are performed using Roche Diagnostics (Indianapolis, IN) Elecsys total PSA Reagents (P990056) and Elecsys free PSA Reagents (P000027) in the native plasma sample and the processed IsoPSA top-phase fraction. These tests are performed following the manufacturer's instructions for use on the Roche cobas e 411 analyzer.

B. Test Procedure:

The IsoPSA Assay is performed in two steps.

- The first step (Partitioning): 200 µL of patient's plasma (Li-heparin) sample is added to the IsoPSA Reagent vial; the reagents are mixed to partition the PSA structural variants based on their structural and physicochemical properties. The vial is then placed into the IsoPSA Assay specific Centrifuge-Mixer Device; the dedicated centrifuge is preprogrammed for three minutes at speed 9 (10,000 RPM) to accelerate separation into two distinct phases.

- The second step: An aliquot is removed from the top phase (partitioned) fraction for quantitation. Total PSA and free PSA levels are measured in both the top-phase (partitioned) fraction and the native (not partitioned) plasma sample using Elecsys total PSA and Elecsys free PSA assays on Roche cobas e 411 analyzer. These measurements are checked for acceptability as follows:

  a. If the Top-phase total PSA is outside the analytical measuring interval (0.065 - 2.720 ng/mL), then the results are invalid. If the Top-phase free PSA is outside the analytical measuring interval (0.040 - 2.024 ng/mL), then the results are invalid.
  b. If both Top-phase free PSA and the Top-phase total PSA are inside their respective analytical measuring intervals, then compute the ratio of Top-phase free PSA to Top-phase total PSA (in the IsoPSA Reagent top-phase fraction).
  c. If the Top-phase free PSA/Top-phase total PSA ratio is outside 0.36-0.70, then repeat IsoPSA test procedure once (refer to section "Test Procedure" above).
  d. If the repeated result for the Top-phase free PSA/Top-phase total PSA ratio remains outside the range (0.36-0.70), classify the specimen as Invalid.
  e. If the Top-phase free PSA/Top-phase total PSA ratio is within 0.36-0.70, then proceed to calculate IsoPSA Index by the equation below:

$$IsoPSA\ Index = -0.2461 + 1.335 * \left(\frac{\frac{[free\ PSA]_{top}}{[free\ PSA]_{native}}}{\frac{[total\ PSA]_{top} - [free\ PSA]_{top}}{[total\ PSA]_{native} - [free\ PSA]_{native}}} \right)$$

It is the responsibility of the laboratory to perform the calculations based on the equation above.

C. Result Interpretation:

IsoPSA Assay is used in men ≥ 50 years of age, with total PSA levels ≥ 4.0 to ≤ 10.0 ng/mL for whom a biopsy is being considered. The results (described as IsoPSA Index) and their interpretation is shown in Table 1 for patients; including those with or without available multiparametric-magnetic resonance imaging (mpMRI) Prostate Imaging Reporting and Data System (PI-RADS) information.

Table 1: Result Interpretation

IsoPSA Index	Interpretation
≤6.00	The patient's score is associated with a decreased likelihood of high-grade prostate cancer (Grade Group ≥ 2; Gleason score ≥ 7) if a biopsy were performed. Patients with an IsoPSA Index of ≤ 6.0 and prostate mpMRI results with PI-RADS score of 1–3 have decreased likelihood of high-grade prostate cancer (Grade Group ≥ 2; Gleason score ≥ 7) on biopsy. The clinician should consider obtaining mpMRI results to reduce the risk of missing high-grade prostate cancer (Grade Group ≥ 2; Gleason score ≥ 7).
>6.00 to 10.00	The patient's score is associated with no change in likelihood of high-grade prostate cancer if a biopsy were performed.
>10.00	The patient's score is associated with an increased likelihood of high-grade prostate cancer if a biopsy were performed.

- The result should not be used in patients with prostate mpMRI results with PI-RADS score of 4–5.
- The result should be interpreted in the context of other clinical, laboratory, and radiology findings. Prostatic biopsy is required to confirm the presence or absence of prostate cancer.
- IsoPSA Assay performance has been evaluated with a limited number of African American or Black subjects.

VI. ALTERNATIVE PRACTICES AND PROCEDURES

Prostate biopsy with an appropriate number of cores is considered the "gold standard" and is required for a diagnosis of prostate cancer. There are several other alternatives and procedures that aid in the detection of prostate cancer, including one or more of the following: digital rectal examination (DRE), histological examination such as needle biopsy or transurethral resection, mpMRI and diagnostic imaging by transrectal ultrasound (TRUS). Other IVD devices for measuring total PSA, %free PSA and Prostate Health Index (*phi*) with venous serum or plasma samples are currently available to aid in the detection of prostate cancer in conjunction with DRE in men aged 50 and older. The 4Kscore Test has been previously approved for use as an aid in the decision for prostate biopsy and the detection of aggressive prostate cancer (Grade Group ≥ 2, Gleason score ≥

7) for whom a biopsy would be recommended by a urologist in men aged 45 and older who have an abnormal age-specific total PSA or who have normal age-specific total PSA with abnormal/suspicious DRE results. Each alternative has its own advantages and disadvantages. A patient should fully discuss these alternatives with his physician to select the method that best meets expectations and lifestyle.

VII. MARKETING HISTORY

The IsoPSA Assay has not been marketed in the United States or any foreign country as an in vitro diagnostic (IVD) test.

VIII. POTENTIAL ADVERSE EFFECTS OF THE DEVICE ON HEALTH

The IsoPSA Assay is intended for patients who are ≥ 50 years of age, with total PSA levels ≥ 4.0 to ≤ 10.0 ng/mL for whom a biopsy is being considered by a urologist based on current standard of care before consideration of the IsoPSA Assay. Patients with an IsoPSA Index of ≤ 6.0 and prostate mpMRI results with PI-RADS score of 1–3 have decreased likelihood of high-grade prostate cancer (Grade Group ≥ 2; Gleason score ≥ 7) on biopsy.

Potential adverse effects (e.g., complications) associated with the use of the device include:

- Patient with a falsely low IsoPSA Index ≤ 6.0 may not receive a necessary biopsy and therefore, could delay recognition of the presence of prostate cancer by the clinician, and could adversely delay the initiation of treatment. For patients with an IsoPSA index ≤ 6.0, the clinician should also consider obtaining mpMRI results to reduce the risk of missing high-grade prostate cancer (Grade Group ≥ 2; Gleason score ≥ 7). The IsoPSA Assay should not be used in patients with prostate mpMRI results with PI-RADS score of 4–5.

- Patient with a falsely elevated IsoPSA Index could lead to a medical decision causing unnecessary biopsy.

The IsoPSA Assay results should be interpreted in conjunction with the patient's medical history, clinical presentation, and laboratory and radiology findings.

IX. SUMMARY OF NON-CLINICAL STUDIES

The performance for the two analytes, total PSA and free PSA in the native patient's plasma sample, relied on the already approved analytical performance in P990056 and P000027, respectively.

Analytical performance of total PSA and free PSA in the IsoPSA Reagent top-phase fraction, and the composite the IsoPSA Index value is summarized below:

A. Precision and Reproducibility Studies:

Precision testing was performed in accordance with Clinical and Laboratory Standards Institute (CLSI) guideline EP05-A3 (*Evaluation of Precision of Quantitative Measurement Procedures; Approved Guideline—Third Edition*).

a) *Within Laboratory precision*:
The studies were performed at a single site using one lot of the reagent on three Roche cobas e 411 analyzers by two operators per analyzer. Seven levels of human Li-heparinized plasma samples were run in two replicates per run, two runs daily (two operators per analyzer; each operator performing one run per day), over the course of five days, resulting in a total of 60 datapoints for each sample. The data were analyzed for repeatability (within-run), between-run, between-day, between analyzers and within-laboratory precision. The mean (ng/mL), standard deviation (SD) (ng/mL) and percent coefficient of variation (%CV) for Top-phase total PSA and for Top-phase free PSA are summarized in Table 2.

Table 2: Within-Lab Precision of Top-phase total PSA and Top-phase free PSA

Analyte	N	Mean (ng/mL)	Within-Run (Repeatability)		Between-Run/Operator		Between-Day		Between-Analyzer		Total	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV
Top-phase total PSA	60	0.13	0.00	1.6	0.00	2.4	0.00	0.0	0.01	8.2	0.01	8.7
	60	0.18	0.01	2.8	0.00	1.9	0.00	1.3	0.01	7.3	0.02	8.2
	60	0.28	0.01	2.7	0.01	2.5	0.00	1.1	0.02	8.3	0.03	9.1
	60	0.55	0.01	2.1	0.01	2.6	0.00	0.0	0.05	8.2	0.05	8.8
	60	0.94	0.01	1.5	0.02	1.7	0.01	1.0	0.05	5.0	0.05	5.6
	60	1.60	0.02	1.4	0.02	1.4	0.02	1.4	0.09	5.3	0.09	5.9
	60	2.38	0.03	1.2	0.04	1.8	0.00	0.0	0.12	5.1	0.13	5.5
Top-phase free PSA	60	0.08	0.00	2.7	0.00	2.4	0.00	0.0	0.00	4.9	0.01	6.1
	60	0.10	0.00	2.6	0.00	1.7	0.00	0.2	0.00	4.5	0.01	5.5
	60	0.17	0.00	2.4	0.00	1.3	0.00	0.2	0.01	4.8	0.01	5.5
	60	0.30	0.01	2.4	0.00	1.4	0.00	0.0	0.01	4.0	0.02	4.8
	60	0.60	0.01	2.3	0.01	2.3	0.01	1.3	0.03	4.9	0.04	6.0
	60	1.05	0.03	2.5	0.02	1.5	0.01	1.0	0.05	4.3	0.06	5.3
	60	1.54	0.05	3.3	0.03	1.6	0.00	0.0	0.06	3.9	0.08	5.3

b) *Lot-to-Lot Precision*:
The lot-to-lot precision was performed at a single site using three reagent lots on one Roche cobas e 411 analyzer. Three levels of plasma samples were run in three replicates per run, two runs per day over the course of five days, resulting in a total of 90 datapoints for each sample on each instrument. The data were analyzed for repeatability (within-run), between-run, between-day, between-reagent lot, and total precision. The results are summarized in Table 3:

Table 3: Lot-to-Lot Precision of Top-phase total PSA and Top-phase free PSA												
Analyte	**N**	**Mean (ng/mL)**	**Within-Run**		**Between-Run/Operator**		**Between-Day**		**Between-Lot**		**Total**	
			SD	**%CV**	**SD**	**%CV**	**SD**	**%CV**	**SD**	**%CV**	**SD**	**%CV**
Top-phase total PSA	90	0.88	0.02	2.1	0.01	1.5	0.01	1.4	0.03	2.8	0.04	4.0
	90	1.50	0.04	2.3	0.02	1.5	0.03	2.1	0.04	2.6	0.07	4.4
	90	2.23	0.05	2.2	0.04	1.6	0.03	1.2	0.07	3.0	0.09	4.2
Top-phase free PSA	90	0.57	0.02	3.3	0.01	1.2	0.00	0.8	0.02	3.1	0.03	4.7
	90	1.00	0.03	3.1	0.01	1.1	0.00	0.4	0.03	2.8	0.04	4.3
	90	1.48	0.05	3.4	0.02	1.4	0.00	0.0	0.04	2.4	0.07	4.4

c) *Site-to-Site Reproducibility*:
The study was performed at three sites (laboratories and/or instruments) using single lot of kit reagents. Seven levels of plasma samples were run in three replicates per run, two runs daily over the course of five days on Roche cobas e 411 analyzer, resulting in a total of 90 datapoints for each sample. The results are summarized in Table 4:

Table 4: Site-to-Site Reproducibility of Top-phase total PSA and Top-phase free PSA												
Analyte	**N**	**Mean (ng/mL)**	**Within-Run**		**Between-Run/Operator**		**Between-Day**		**Between-Site/Analyzer**		**Total**	
			SD	**%CV**	**SD**	**%CV**	**SD**	**%CV**	**SD**	**%CV**	**SD**	**%CV**
Top-phase total PSA	90	0.11	0.00	2.4	0.00	2.8	0.00	1.5	0.00	3.3	0.01	5.2
	90	0.16	0.00	1.8	0.00	1.2	0.00	2.3	0.00	2.7	0.01	4.1
	90	0.30	0.01	2.1	0.00	1.3	0.00	1.4	0.01	2.7	0.01	3.9
	90	1.00	0.03	2.5	0.02	1.6	0.02	1.9	0.03	3.4	0.05	4.9
	90	1.06	0.04	3.4	0.00	0.0	0.02	1.7	0.03	2.4	0.05	4.5
	90	1.80	0.04	2.4	0.02	0.8	0.03	1.7	0.04	2.3	0.06	3.8
	90	2.52	0.04	1.5	0.06	1.2	0.05	2.0	0.08	3.1	0.11	4.2
Top-phase free PSA	90	0.07	0.00	3.9	0.00	2.2	0.00	3.0	0.00	3.0	0.00	6.2
	90	0.10	0.00	3.1	0.00	1.9	0.00	1.1	0.00	1.8	0.00	4.2
	90	0.20	0.01	2.9	0.00	2.2	0.00	1.9	0.00	1.9	0.01	4.6
	90	0.55	0.02	2.8	0.01	2.0	0.01	2.0	0.00	0.0	0.02	4.0
	90	0.74	0.02	2.7	0.01	1.4	0.01	1.9	0.00	0.0	0.03	3.6
	90	1.29	0.04	3.4	0.02	1.8	0.02	1.5	0.00	0.0	0.05	4.2
	90	1.82	0.06	3.0	0.05	2.5	0.04	2.0	0.03	1.8	0.09	4.7

d) *Simulation Precision for IsoPSA Index*:
Precision profiles for the standard deviation of each analyte were generated using the N=7 samples with a total of 90 replicates as described in the reproducibility study above. IsoPSA Index values based on quartiles for each, the range, mean of measured IsoPSA Index values, minimum and maximum simulated SD and %CV for both Repeatability (Within-Run) and Within-Laboratory were determined. Simulation Precision for IsoPSA Index (by IsoPSA level cutoffs) are presented in Table 5.

Table 5: Simulation Precision for IsoPSA Index					
IsoPSA Index	Mean	Within-Run (Repeatability)		Within-Laboratory Precision	
		SD	%CV	SD	%CV
≤6.0	4.40	0.53	9.3	0.81	13.5
>6 to ≤ 10.0	7.74	0.86	9.4	1.19	13.4
>10	15.18	2.88	12.1	4.01	16.8

B. Linearity:

Linearity of Top-phase total PSA and Top-phase free PSA was evaluated in accordance with CLSI guideline EP06, 2nd Edition (*Evaluation of Linearity of Quantitative Measurement Procedures*). Two high-level sample pools were prepared using plasma samples with high native total PSA and free PSA concentrations which will yield Top-phase total PSA ≈ 2.5 ng/mL and Top-phase free PSA ≈ 1.5 ng/mL after partitioning. Additional two low-level sample pools were prepared to yield Top-phase total PSA ≈ 0.06 ng/mL and Top-phase free PSA 0.04 ng/mL after partitioning. Two dilution series (sets) of 11 levels were prepared by combining plasma sample pools with low- and high-analyte concentrations in varying amounts to achieve 11 equally spaced levels. Each resulting level was partitioned in IsoPSA Reagent. Each Top-phase total PSA and Top-phase free PSA concentrations were tested in five replicates. The expected values were calculated based on a dilution scheme. Deviation from linearity for each sample was calculated as a difference between the observed Mean value and Predicted value. %Deviation was calculated as Deviation/Predicted value. The results are summarized in Table 6.

Table 6: Linearity of Top-phase total PSA and Top-phase free PSA						
Analyte	Series*	Range (ng/mL)	Slope (95% CI)	Intercept (95% CI)	R^2	% Deviation**
Top-phase total PSA	1	0.06 to 2.72	1.017 (1.009 to 1.025)	-0.002 (-0.007 to 0.004)	1.000	-1.6 to 1.9
	2	0.07 to 2.86	1.016 (0.999 to 1.034)	-0.002 (-0.008 to 0.004)	0.999	-3.7 to 2.4
Top-phase free PSA	1	0.04 to 2.02	1.031 (1.019 to 1.042)	-0.001 (-0.006 to 0.004)	1.000	-3.0 to 2.4
	2	0.04 to 2.16	1.004 (0.985 to 1.023)	-0.001 (-0.005 to 0.003)	0.999	-3.6 to 4.9

*Dilution Series

**Minimum and maximum deviation from linearity

The data show the linearity interval of 0.06 to 2.86 ng/mL for Top-phase total PSA and 0.04 to 2.16 ng/mL for Top-phase free PSA with the deviations from linearity within ±10.0%.

The study results indicate the linearity of the claimed analytical measuring interval (AMI):

- Total-phase total PSA: 0.065 to 2.720 ng/mL
- Top-phase free PSA: 0.040 to 2.024 ng/mL.

C. Detection Capability:

The detection capability studies for Top-phase total PSA and Top-phase free PSA using the IsoPSA Reagent, were conducted for Limit of Blank (LoB), Limit of Detection (LoD) or Limit of Quantitation (LoQ) in accordance with CLSI guideline EP17-A2 (*Evaluation of Detection Capability for Clinical Laboratory Measurement Procedures; Approved Guideline, 2nd Edition*). For determining the LoB and LoD, two replicates per sample were tested for four days resulting in a total of 64 (4 x 2 x 4 x 2) measurements for each analyte. The LoQ was tested in three replicates per sample for three days resulting in a total of 72 (4 x 3 x 3 x 2) measurements for each analyte.

a) *LoB*:
Four analyte free samples of Top-phase total PSA and Top-phase free PSA prepared after partitioning using the IsoPSA Reagent were tested in two replicates per run per sample, one run per day for four days, with two reagent lots on one instrument (N=64 per analyte). The LoB estimates were calculated for each lot using nonparametric (as the upper 95% point of the numerically recorded readings) option. The LoB for each analyte was determined to be the highest observed LoB across two IsoPSA Reagent lots.

b) *LoD*:
Four low level samples of Top-phase total PSA and Top-phase free PSA prepared after partitioning using the IsoPSA Reagent were tested in two replicates per run per sample, one run per day for four days, with two reagent lots on one instrument (N=64 per analyte). The LoD estimates were calculated using the LoB estimate; the SD_{pooled}, pooled across all LoD test samples for the Top-phase total PSA or Top-phase free PSA were used to calculate LoD estimates. The LoD for each analyte was determined to be the highest LoD across two IsoPSA Reagent lots.

c) *LoQ*:
Four low level samples of Top-phase total PSA and Top-phase free PSA prepared after partitioning using the IsoPSA Reagent were tested in three replicates per run per sample, one run per day for three days, with two reagent lots on one instrument (N=72 per analyte). The LoQ is determined to be the highest value across two IsoPSA Reagent lots meeting a goal of %CV of 20% of imprecision.

The claimed LoB/LoD/LoQ for the Top-phase total PSA and Top-phase free PSA assays are summarized in Table 7:

Table 7: Detection Capability for Top-phase total PSA and Top-phase free PSA

	LoB	LoD	LoQ
Top-phase total PSA	0.023 ng/mL	0.027 ng/mL	0.044 ng/mL
Top-phase free PSA	0.022 ng/mL	0.025 ng/mL	0.030 ng/mL

D. Assay Reportable Range:

The reportable range is the same as the claimed analytical measuring interval (AMI):

- The AMI for Roche Elecsys total PSA and Elecsys free PSA is approved under P990056 and P000027, respectively.

- The AMI for Top-phase total PSA is 0.065 to 2.720 ng/mL and for Top-phase free PSA is 0.040 to 2.024 ng/mL.

E. Analytical Specificity/Interference:

The interference study was performed according to CLSI guidelines EP07, 3rd Edition (*Interference Testing in Clinical Chemistry*) and EP37, 1st Edition (*Supplemental Tables for Interference Testing in Clinical Chemistry*) to determine the effect of various endogenous and exogenous substances on the IsoPSA Assay. A two level panel of lithium-heparin plasma samples were targeted for IsoPSA Index of <6.0 and >6.0. The resulting panel-1 with Top-phase total PSA concentrations of 0.321 to 0.725 ng/mL and Top-phase free PSA concentrations of 0.160 to 0.471 ng/mL and panel 2 with Top-phase total PSA concentrations of 0.151 to 0.285 ng/mL and Top-phase free PSA concentrations of 0.093 to 0.171 ng/mL were used for the study. For each interfering substance, both spiked (with interfering substance) and non-spiked (control) samples were tested in quadruplicate by two operators using the IsoPSA Assay in conjunction with the Roche reagents. Total PSA and free PSA concentrations in the native plasma sample and top phase fraction were measured and IsoPSA Index values were calculated for each replicate. The percent interference (% Interference) for each analyte in the native sample matrix and IsoPSA Reagent top phase fraction was calculated as the observed interference divided by the mean measurand value of the control sample and multiplied by 100. Non-significant interference was defined as % interference within ±10% of IsoPSA Index.

a) *Endogenous Substance Interference*:
The following endogenous substances were tested using IsoPSA Assay. Non-significant interference was found for each substance at the concentrations listed in Table 8.

Table 8: Endogenous Interference for the IsoPSA Assay

Interfering Substance	Concentration
Albumin	7.4 g/dL
Bilirubin (Conjugated)	2 mg/dL
Bilirubin (Unconjugated)	15 mg/dL
Calcium Chloride	30 mg/dL
Cholesterol	220 mg/dL
Ferritin	300 ng/mL
γ-globulin	20 g/L
Hemolysate	500 mg/dL
Human Anti-Mouse Antibodies	52.5 ng/mL
Human Chorionic Gonadotropin	50 IU/L

Table 8: Endogenous Interference for the IsoPSA Assay	
Interfering Substance	**Concentration**
Prolactin	50 ng/mL
Rheumatoid Factor	161 U/mL
Triglyceride-rich Lipoproteins	750 mg/dL
Urea	30 mg/dL
Uric Acid	7 mg/dL

b) *Exogenous Substance Interference*:
The potential interference of the performance of IsoPSA Assay in presence of 39 commonly used drugs (including those used for cancer treatment) and dietary supplements was evaluated. The results are summarized in Table 9.

Table 9: Exogenous Interference for the IsoPSA Assay			
Interfering Substance	**Concentration**	**Interfering Substance**	**Concentration**
5'-Fluorouracil	9 mg/dL	Nizoral (Ketoconazole)	15 ug/mL
Aleve (Naproxen Sodium)	36 mg/dL	Norvasc (Amlodipine Besylate)	0.008 mg/dL
Aspirin (Acetylsalicylic Acid)	3 mg/dL	Oxybutynin Chloride	30 ng/mL
Avodart (Dutasteride)	120 ng/mL	Prazosin HCl	240 ng/mL
Azulfidine (Sulfasalazine)	7.5 mg/dL	Prilosec (Omeprazole)*	17.4 µmol/L
Benadryl (Diphenhydramine HCl)	0.077 mg/dL	Prinivil (Lisinopril)	0.025 mg/dL
Biotin	1,200 ng/mL	Prozac (Fluoxetine HCl)	0.142 mg/dL
Cardura (Doxazosin)	0.030 mg/dL	Riomet (Metformin HCl)	1.2 mg/dL
Cyclosporine	0.18 mg/dL	Saw Palmetto (Extract)	1125 mg/dL
Deltasone (Prednisone)	0.02 mg/dL	Selenium	450 ng/mL
Doxycyline Hyclate	1.8 mg/dL	Tagamet HB (Cimetidine)*	3 mg/dL
Finasteride	150 ng/mL	Tenormin (Atenolol)	0.9 mg/dL
Flomax (Tamsulosin HCl)	124.8 ng/mL	Terazosin HCl*	0.027 mg/mL
Flutamide	234 ng/mL	Testosterone	1200 ng/dL
Gadobutrol*	0.9 mmol/L	Theophylline	25 µg/mL
Heparin	330 U/dL	Tylenol (Acetaminophen)	31.2 µg/mL
Ibuprofen*	0.219 mg/mL	Uroxatral (Alfuzosin HCl)*	40 ng/mL
Jantoven (Warfarin HCl)	7.5 mg/dL	Viagra (Sildenafil Citrate)	12.9 pmol/L
Lipitor (Atorvastatin Calcium)	0.075 mg/dL	Xanax (Alprazolam)	0.25 mg/dL
Microzide (Hydrochlorothiazide)	0.113 mg/dL	Zyloprim (Allopurinol)	6 mg/dL
Moxatag (Amoxicillin)	5.4 mg/dL		

*% interference was >10.0 to <16.0%

c) *Cross-Reactivity*:
The potential cross-reactivity of the following substances was evaluated using the IsoPSA Assay. Non-significant interference was found for each substance at the concentrations listed in Table 10.

Table 10: Cross-Reactivities for the IsoPSA Assay	
Cross-Reactive Substance	**Concentration**
Alpha-Fetoprotein	20 ng/mL
Carcinoembryonic Antigen	15 ng/mL
Prostatic Acid Phosphate	250 ng/mL

F. Traceability:

The IsoPSA Assay uses Roche total PSA and free PSA approved under P990056 and P000027.

G. Stability:

Stability for the IsoPSA Assay is based on reagent stability for the individual component assays. The stability of Roche Elecsys total PSA and Elecsys free PSA was established under P990056 and P000027, respectively. The stability study for the IsoPSA Reagent was performed according to CLSI EP25, 2nd edition (*Evaluation of Stability of In Vitro Medical Laboratory Test Reagents).*

a) *Shelf-life Stability*:
The real-time stability of the IsoPSA Reagent was evaluated using a three sample panel containing three levels of the IsoPSA Index: 3.2, 4.7 and 7.0 and two lots of reagents (stored at 2-8°C and at 18-24°C). The samples were assayed soon after the dates of reagent manufacture, two runs per day, one replicate per run to establish the baselines for each sample for each lot. Each individual IsoPSA Index result was plotted on the y-scale against timescale (testing days on the x-scale). The results show IsoPSA Reagent is stable up to 18 months when stored at 2-8°C and up to 90 days when stored at 18-24°C.

b) *Reagent Kit shipping stability*:
The stability of the IsoPSA Reagent and IsoPSA Controls after packaging and transport using a commercially available shipping system under simulated real-world transport conditions. The shipping stability of the IsoPSA Reagent packaging and shipping process has met the pre-determined acceptance criteria for use in routine 72-hour transport.

c) *Sample Stability*:
Plasma samples (N=at least 30) were collected at the hospital lab and transported to the testing site under ambient or cold pack (2-8°C) conditions within four hours of collection. Upon arrival at the testing site, the plasma specimens were aliquoted, and aliquots was placed in an ambient incubator (18-24°C), in the refrigerator (2-8°C), at -20°C and at -80°C. Each temperature condition included its own T0 measurement to establish baselines. Additionally, a total of ten plasma samples stored at -80°C were subjected to a total of five freeze–thaw cycles. Top-phase total PSA, Top-phase free PSA and IsoPSA Index were measured in samples stored in an ambient incubator, in the refrigerator, at -20°C and at -80°C, at T=0 and at three additional time points, and

samples from freeze-thaw study. Passing-Bablok regression was used in data analysis. The results indicate that plasma samples are stable:

- up to 48 hours if stored refrigerated (2-8°C)
- up to 24 hours at ambient temperature (18-24°C)
- up to 14 days when stored frozen at -20°C
- up to 90 days when stored frozen at -80°C for long term storage
- up to 5 freeze-thaw cycles

X. SUMMARY OF PRIMARY CLINICAL PERFORMANCE STUDY

A clinical study was conducted to establish a reasonable assurance of safety and effectiveness of the IsoPSA Assay as an aid in detection of high-grade prostate cancer (Grade Group ≥ 2; Gleason score ≥ 7) in men ≥ 50 years of age, with total PSA levels ≥ 4.0 to ≤ 10.0 ng/mL for whom a biopsy is being considered by a urologist based on current standard of care, before consideration of IsoPSA Assay results. Data from this clinical study were the basis for the PMA approval decision. A summary of the clinical study is presented below.

A. Study Design

The study enrolled a total of 1422 subjects between June 14, 2023, and March 1, 2024, from 14 community-based urology practices in the United States. Following the exclusion criteria, a total of 792 subjects were available as the clinically relevant study population. Blood samples were obtained from study participants for the IsoPSA Assay within 30 days prior to standard of care prostate biopsy (at least 12 cores) by (1) transrectal and/or transperineal ultrasound guided systematic biopsy (TRUS BX), (2) mpMRI ultrasound fusion guided systematic biopsy with/without targeting (MRI-Fusion BX) according to local institutional standards and investigator preferences. All laboratory test results were performed and reported masked to the biopsy pathology results.

a) *Clinical Inclusion and Exclusion Criteria*:
Enrollment in the pivotal clinical performance study was limited to patients who met the following inclusion criteria.

- Age: ≥ 50 years of age
- Gender: Male
- Diagnosis:
  - Scheduled for standard of care prostate biopsy
  - Most current total PSA test ≥ 4 ng/mL to ≤ 10ng/mL
- Consent: Demonstrates understanding of study procedures, restrictions, and willingness to participate as evidenced by voluntary written informed consent and has received a copy of the consent document
- Compliance: Understands and is willing, able, and likely to comply with all study procedures and restrictions

Patients were not permitted to enroll in the pivotal clinical performance study if they met any of the following exclusion criteria:

- Previous prostate biopsy (≤ 3 months)
- Recent prostate manipulation (≤ 3 days)
- Recent urinary tract infection and/or prostatitis (< 2 weeks)
- Recent prostate surgery, urinary catheterization, prostate infarction, urinary endoscopy (≤ 3 months)
- Hormonal ablation surgery (≤ 90 days)
- Documentation of any other urinary tract malignancy
- Previous IsoPSA Test (≤ 1 year)

b) *Follow-up Schedule*:

Not applicable

c) *Clinical Endpoints*:

The objective of the study was to validate clinical performance of the IsoPSA Assay against biopsy as an aid in the detection of high-grade prostate cancer (defined as Gleason Score ≥ 7) from low-grade prostate cancer (defined as Gleason Score = 6 or lower) or benign prostate conditions. The clinical diagnosis information for each subject was obtained by TRUS biopsy (≥ 12 cores), transperineal biopsy, or MRI-TRUS Fusion biopsy to collect tissue for histopathologic examination. Each subject was assigned a binary histopathologic classification for each indication using the primary diagnosis reported on the case report form according to the description provided in Table 11.

Table 11: Histopathologic Classification based on Primary Diagnosis Information

HG-PCa* Binary Classification	**Primary Diagnosis from Case Report Form**
HG-PCa (Actionable/Significant)	Adenocarcinoma of the prostate At least one core with (3+4), (4+3), (4+4), (4+5) or (5+5)
Negative for HG-PCa [low-grade Cancer or Benign (No Cancer)]	Adenocarcinoma of the prostate At least one core with Gleason (3+3) and no cores with Gleason (3+4), (4+3), (4+4), (4+5) or (5+5)
	Benign Prostate Tissue
	Benign Prostate Tissue with chronic inflammation
	Atypical Intraductal proliferation
	Hyperplasia
	Atypical Small Acinar Proliferation
	PIN**

	PIN-ATYP***
	High-Grade PIN Chronic Inflammation, Chronic Prostatitis
	Nonspecific Granulomatous Prostatitis

*HG-PCa=high-grade prostate cancer
**PIN=Prostatic Intraepithelial Neoplasia
***PIN-ATYP=PIN with adjacent atypical glands

Performance of the IsoPSA Assay was analyzed by sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), probability (predictive value, PV), likelihood ratio (LR) based on the IsoPSA Index points of 6.0, and 10.0. In addition, data was analyzed for clinically relevant subgroups including:

- Age (50-59 yrs. 60-69 yrs. and > 70 yrs.);
- Race (African American and Non-African American men);
- Patients with a prior prostate biopsy;
- Digital Rectal Examination (DRE) status;
- Presence or absence of prostate MRI in the medical record (MRI+ vs. MRI-); and various groupings of PI-RADS scores in combination with and separate from patients that did not receive MRI.

B. Accountability of PMA Cohort:

The study enrolled a total of 1422 subjects. A total of 630 samples were excluded after applying the exclusion criteria, resulting a total of 792 eligible subjects in align with the Intended Use population of the IsoPSA Assay. Among these 792 patients, 430 patients with mpMRI results and 362 patients with no MRI.

C. Study Population Demographics and Baseline Parameters:

From a total of 792 subjects in the study, the distributions of demographic and clinical variables are described in Table 12.

Table 12: Distribution of Demographic, Clinical Variables in Clinical Performance Study (N=792)				
Demographic or Clinical Variable		**With mpMRI (N=430)**		**No mpMRI (N=362)**
		PI-RADS 1-3 (N=204)	**PI-RADS 4-5 (N=226)**	
Age, Median (Years)		66.5	68.0	66.0
Total PSA, Median (ng/mL)		6.6	6.9	6.1
% free PSA, Median (%)		17.0	15.2	17.5
Race, n (%)	White	183 (92.9%)	205 (94.5%)	310 (92.3%)
	African American	12 (6.1%)	7 (3.2%)	16 (4.8%)
	Asian	2 (1.0%)	5 (2.3%)	9 (2.7%)
	Other	7 (3.4%)	9 (4.0%)	27 (7.5%)
Biopsy Type[a], n (%)	Biopsy Naïve	142 (69.6%)	177 (78.3%)	338 (93.4%)
	Repeat Biopsy	62 (30.4%)	49 (21.7%)	24 (6.6%)
DRE, n (%)	Abnormal	53 (26.0%)	47 (20.8%)	78 (21.5%)
	Normal	122 (59.8%)	126 (55.8%)	239 (66.0%)
	Not Performed	29 (14.2%)	53 (23.5%)	45 (12.4%)
BPH Medication Treatment, n (%)	No	147 (72.1%)	182 (80.5%)	267 (73.8%)
	Yes	57 (27.9%)	44 (19.5%)	95 (26.2%)
Cancer Status by Biopsy n (%)	Negative (Benign)	104 (51.0%)	39 (17.3%)	175 (48.3%)
	low-grade PCa[b] (GG[c]=1; GS[d]=6)	61 (29.9%)	52 (23.0%)	73 (20.2%)
	high-grade PCa (GG ≥2, GS ≥7)	39 (19.1%)	135 (59.7%)	114 (31.5%)

[a] Note: Study did not collect the number of previous or prior biopsies.
[b] PCa=Prostate Cancer
[c] GG=Gleason Grade Group
[d] GS=Gleason Score

D. Safety and Effectiveness Results:

a) *Safety Results*:
The IsoPSA Assay involves testing of Li-Heparin plasma from venous blood using venipuncture. These specimens are routinely taken as part of the practice of medicine for a typical laboratory test and, therefore, sample collection presents no additional safety hazard to the patient being tested.

In this study, all enrolled subjects were men presenting to a urologist with symptoms that would lead to an evaluation for prostate cancer and who were being considered to receive a prostate needle biopsy. Although the most significant safety concern with respect to biopsy is often associated with infectious complications following the procedure, a false positive IsoPSA Assay result for these study subjects did not alter the medical decision for these patients, therefore, present no additional safety hazard to the subjects being tested.

b) *Effectiveness Results*:

The clinical performance evaluation of the IsoPSA Assay was based on a total of 792 patients of the clinical performance study. Each patient in the study had the IsoPSA Assay result (i.e., IsoPSA Index) and biopsy findings. The IsoPSA Index were divided into three intervals: ≤6.0, >6.0 to ≤10.0 and >10.0. Biopsy findings were divided into two groups: Gleason Score ≥7 (GG ≥2) and Gleason Score <7. Clinical performance data were summarized below:

i) *Clinical performance of the IsoPSA Assay for all patients*:

The clinical performance of IsoPSA Assay for all 792 patients was evaluated in accordance with CLSI guideline EP12, 3rd Edition, (*Evaluation of Qualitative, Binary Output Examination Performance*) and described for each of three intervals of the IsoPSA Index by probability of having high-grade prostate cancer (Grade Group ≥ 2, Gleason Score ≥ 7) by the IsoPSA Assay (Predictive Value, PV), likelihood ratio (LR) and frequency (percent) of the results as summarized in Table 13.

Table 13: Clinical Performance of the IsoPSA Assay: Probability and Likelihood Ratio (All patients, N=792, Prevalence=36.4%)

IsoPSA Index	GS[a] ≥7 (GG[b] ≥2)		Total	Probability of GG ≥2, PV (95% CI)[c]	LR (95% CI)[d]	Frequency (n/N)[e]
	Yes	No				
≤6.0	18	130	148	12.20% (8.0%, 18.0%)	0.242 (0.151, 0.384)	18.70%
>6.0 to ≤10.0	70	189	259	27.00% (22.6%, 31.8%)	0.648 (0.512, 0.814)	32.70%
>10.0	200	185	385	51.90% (48.5%, 55.4%)	1.892 (1.649, 2.173)	48.60%
Total	**288**	**504**	**792**	**Prevalence = 36.4% (288/792)**		

[a] GS=Gleason Score; [b] GG=Gleason Grade Group
[c] 95% CIs are calculated using 95% CI for the corresponding likelihood ratio and prevalence.
[d] 95% CIs are calculated using an asymptotic method for ratios of two independent binomial proportions.
[e] Frequency within all subjects shown (out of N=792).

- Among patients with IsoPSA Index ≤ 6.0, the probability of high-grade prostate cancer if biopsied was 12.2% (18/148) (95% CI: 8.0%, 18.0%), demonstrating a statistically significant decreased likelihood of GG ≥2 (the upper bound of 95% CI of 18.0% is less than prevalence of 36.4%).
- Among patients with IsoPSA Index > 10.0, the probability of high-grade prostate cancer if biopsied was 51.9% (95% CI: 48.5%, 55.4%), demonstrating a statistically and clinically significant increased likelihood of GG ≥2 (the lower bound of 95% CI of 48.5% is more than prevalence of 36.4%).

The clinical performance for 792 patients was analyzed based on sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) with corresponding 95% CI for two IsoPSA Assay cut points: 6.0 and 10.0 as summarized in Table 14.

Table 14: Performance of the IsoPSA Assay: Sensitivity, Specificity and Predictive Values (All patients, N=792, Prevalence=36.4%)

IsoPSA Index cutoff	Sensitivity (n/N) (95% CI)	Specificity (n/N) (95% CI)	PPV (n/N) (95% CI)	NPV (n/N) (95% CI)
			Prevalence = 36.4%	
6.0	93.8% (270/288) (90.3%; 96.0%)	25.8% (130/504) 22.2%; 29.8%	41.9% (270/644) (40.5%; 43.4%)	87.8% (130/148) (82.0%; 92.0%)
10.0	69.4% (200/288) (63.9%; 74.5%)	63.3% (319/504) (59.0%; 67.4%)	51.9% (200/385) (48.5%; 55.4%)	78.4% (319/407) (76.2%; 81.4%)

ii) *Clinical performance of the IsoPSA Assay for patients with mpMRI results*:

Among 430 patients who had undergone mpMRI before taking the IsoPSA Assay test, 204 patients were with PI-RADS 1–3 and 226 patients with PI-RADS 4–5. The clinical performance of IsoPSA Assay for these patients are summarized in the tables 15 and 16.

Table 15: Clinical Performance of the IsoPSA Assay: Probability and Likelihood Ratio (Patients with mpMRI, N=430, Prevalence = 40.5%)

mpMRI	IsoPSA Index	GS[a] ≥7 (GG[b] ≥2)		Total	Probability of GG ≥2, PV (95% CI)[c]	LR (95% CI)[d]	Frequency (n/N)[e]
		Yes	No				
PI-RADS 1–3	≤6.0	2	37	39	5.1% (1.4%, 15.6%)	0.229 (0.062, 0.782)	19.1%
	>6.0 to ≤10.0	10	68	78	12.8% (7.5%, 19.7%)	0.622 (0.344, 1.039)	38.2%
	>10.0	27	60	87	31.0% (24.7%, 37.3%)	1.904 (1.388, 2.512)	42.6%
	Total	**39**	**165**	**204**	**Prevalence = 19.1% (39/204)**		
PI-RADS 4–5	≤6.0	11	14	25	44.0% (27.5%; 62.0%)	0.530 (0.256; 1.099)	11.1%
	>6.0 and ≤10.0	31	36	67	46.3% (36.6%; 56.2%)	0.580 (0.389; 0.866)	29.6%
	>10.0	93	41	134	69.4% (64.1%; 74.8%)	1.529 (1.202; 1.999)	59.3%
	Total	**135**	**91**	**226**	**Prevalence = 59.7% (135/226)**		

[a] GS=Gleason Score, [b] GG=Gleason Grade Group

[c] 95% CI are calculated using 95% CI for the corresponding likelihood ratio and prevalence.

[d] 95% CI are calculated using an asymptotic method for ratios of two independent binomial proportions.
[e] Frequency within all subjects shown (out of N=204).

Patients with mpMRI PI-RADS 1–3

- Among subjects with IsoPSA Index ≤ 6.0, the probability of high-grade prostate cancer if biopsied was 5.1% (2/39), (95% CI: 1.4%, 15.6%), demonstrating a statistically and clinically significant decreased likelihood of GG ≥2 (the upper bound of 95% CI of 15.6% is less than prevalence of 19.1%).
- Among subjects with IsoPSA Index > 10.0, the probability of high-grade prostate cancer if biopsied was 31.0% (95% CI: 24.7%, 37.3%), demonstrating a statistically and clinically significant increased likelihood of GG ≥2 (the lower bound of 95% CI of 24.7% is more than prevalence of 19.1%).

Patients with mpMRI PI-RADS 4–5

- Among subjects with IsoPSA Index ≤ 6.0, the probability of high-grade prostate cancer if biopsied was 44.0% (11/25), (95% CI: 27.5%, 62.0%). Because the upper bound of the 95% CI (62.0%) exceeds the observed prevalence (59.7%), the IsoPSA Assay is not statistically informative for the IsoPSA Index cutoff of 6.0, in patients with PI-RADS 4–5.
- Among subjects with IsoPSA Index > 10.0, the probability of high-grade prostate cancer if biopsied was 69.4% (95% CI: 64.1%, 74.8%).

Table 16: Performance of the IsoPSA Assay: Sensitivity, Specificity and Predictive Values (Patients with mpMRI, N=430, Prevalence=40.5%)

mpMRI	IsoPSA Index Cutoff	Sensitivity (n/N) (95% CI)	Specificity (n/N) (95% CI)	PPV (n/N) (95% CI)	NPV (n/N) (95% CI)
PI-RADS 1–3	6.0	94.9% (37/39) (83.1%; 98.6%)	22.4% (37/165) (16.7%; 29.4%)	22.4% (37/165) (20.0%; 24.3%)	94.9% (37/39) (84.4%; 98.6%)
	10.0	69.2% (27/39) (53.6%; 81.4%)	63.6% (105/165) (56.1%; 70.6%)	31.0% (27/87) (24.7%; 37.3%)	89.7% (105/117) (85.0%; 93.6%)
	Prevalence=19.1% (39/204)				
PI-RADS 4–5	6.0	91.9% (124/135) (86.0%; 95.4%)	15.4% (14/91) (9.4%; 24.2%)	61.7% (124/201) (59.4%; 4.4%)	56.0% (14/25) (38.0%; 72.5%)
	10.0	68.9% (93/135) (60.6%; 76.1%)	54.9% (50/91) (44.7%; 64.8%)	69.4% (93/1340) (64.1%; 74.8%)	54.3% (50/92) (46.6%; 62.0%)
	Prevalence=59.7% (135/226)				

Patients with mpMRI PI-RADS 1–3

For the IsoPSA Index cutoff of 6.0, sensitivity was 94.9% with 95% CI: (83.1%; 98.6%) and NPV was 94.9% with 95% CI: (84.4%; 98.6%). For the IsoPSA Index

cutoff of 10.0, specificity was 63.6% with 95% CI: (56.1%; 70.6%) and PPV was 31.0% with 95% CI: (24.7%; 37.3%).

Patients with mpMRI PI-RADS 4–5
For the IsoPSA Index cutoff of 6.0, sensitivity was 91.9% with 95% CI: (86.0%; 95.4%) and NPV was 56.0% with 95% CI: (38.0%; 72.5%). An in vitro test is statistically informative if NPV is larger than 1-prevalence. Among patients with mpMRI PI-RADS 4-5, 1- prevalence was 40.3% and the 95% CI for the NPV was (38.0%; 72.5%). The data did not demonstrate that NPV is larger than 40.3% because the 95% CI for the NPV includes 40.3%. Therefore, for the IsoPSA Index cutoff of 6.0, the IsoPSA Assay is not statistically informative.

Because the probability of GG ≥2 for patients with IsoPSA Index ≤ 6 is 44%, the IsoPSA Assay should not be used for patients with mpMRI PI-RADS 4-5. The package insert of the IsoPSA Assay has this warning statement.

iii) *Clinical performance of the IsoPSA Assay for patients with no mpMRI results*: Among 792 patients, 362 patients had not undergone mpMRI results before taking the IsoPSA Assay test. The clinical performance of IsoPSA Assay for these patients is presented in tables 17 and 18:

Table 17: Clinical Performance of the IsoPSA Assay: Probability and Likelihood Ratio (Patients with no mpMRI, N=362, Prevalence=31.5%)

IsoPSA Index	GS[a] ≥7 (GG[b] ≥2)		Total	Probability of GG ≥2, PV (95% CI)[c]	LR (95% CI)[d]	Frequency (n/N)[e]
	Yes	No				
≤6.0	5	79	84	6.0% (2.6%; 12.7%)	0.138 (0.058; 0.316)	23.2%
>6.0 to ≤10.0	29	85	114	25.4% (19.1%; 32.5%)	0.742 (0.514; 1.049)	31.5%
>10.0	80	84	164	48.8% (43.5%; 54.1%)	2.072 (1.675; 2.561)	45.3%
Total	**114**	**248**	**362**	**Prevalence = 31.5% (114/362)**		

[a] GS=Gleason Score, [b] GG=Gleason Grade Group
[c] 95% CI are calculated using 95% CI for the corresponding likelihood ratio and prevalence.
[d] 95% CI are calculated using an asymptotic method for ratios of two independent binomial proportions.
[e] Frequency within all subjects shown (out of N=362).

- Among patients with IsoPSA Index ≤ 6.0, the probability of high-grade prostate cancer if biopsied was 6.0% (5/84) (95% CI: 2.6%, 12.7%), demonstrating a statistically and clinically significant decreased likelihood of GG ≥2 (the upper bound of 95% CI of 12.7% is less than prevalence of 31.5%).
- Among patients with IsoPSA Index > 10.0, the probability of high-grade prostate cancer if biopsied was 48.8% (84/164) (95% CI: 43.5%, 54.1%), demonstrating a statistically and clinically significant increased likelihood of

GG ≥2 (the lower bound of 95% CI of 43.5% is more than prevalence of 31.5%).

Table 18: Performance of the IsoPSA Assay: Sensitivity, Specificity and Predictive Values (Patients with No mpMRI, N=362, Prevalence = 31.5%)				
IsoPSA Assay Cutoff	**Sensitivity (n/N) (95% CI)**	**Specificity (n/N) (95% CI)**	**PPV (n/N) (95% CI)**	**NPV (n/N) (95% CI)**
			Prevalence =31.5%	
6.0	95.6% (109/114) (90.1%; 98.1%)	31.9% (79/248) (26.4%; 37.9%)	39.2% (109/278) (37.0%; 41.6%)	94.0% (79/84) (87.3%; 97.4%)
10.0	70.2% (80/114) (61.2%; 77.8%)	66.1% (164/248) (60.0%; 71.7%)	48.8% (80/164) (43.5%; 54.1%)	82.8% (164/198) (78.5%; 86.8%)

In the clinical performance study, group of 362 patients who have not undergone mpMRI was different from 430 patients with mpMRI results: prevalence of GG ≥2 was 31.5% (114/362) vs 40.5% [(39+135)/430] and probability of GG ≥2 among patients with IsoPSA Index ≤ 6 was 6.0% (5/84) vs 20.3% [(2+11)/(39+25)]. Among 430 patients who underwent mpMRI, 53% (226 out of 430) had PI-RAD 4-5. Multiple Independent studies have reported that at least 33% of men who undergo biopsy following mpMRI in the PSA zone (4–10 ng/mL) display PI-RADS 4–5 lesions on pre-biopsy mpMRI.

Because of this, *for patients with an IsoPSA Index ≤ 6.0, the clinician should also consider obtaining mpMRI results to reduce the risk of missing high-grade prostate cancer (Grade Group ≥ 2; Gleason Score ≥ 7).* The package insert of the IsoPSA Assay has this warning statement.

iv) Subgroup Analyses:
Clinical performance of the IsoPSA Assay was analyzed based on 792 subjects stratified by different demographics and other clinical characteristics. In addition, similar subgroup analyses were performed based on 204 patients with mpMRI PI-PADS 1-3 because the IsoPSA Assay should not be used for patients with mpMRI PI-RADS 4-5.

A) Age
Prevalence of prostate cancer is well-known to increase with advancing age. The performance of the IsoPSA Assay was analyzed for the different age groups: 50-59 years old, 60–69 years old, and 70-88 years old. The results are summarized in Table 19:

Table 19: Performance of the IsoPSA Assay Stratified by Age (All Patients, N=792)						
Age Group	**IsoPSA Index range**	**GG ≥2**		**Total**	**Probability of GG ≥2 (Predictive Value, PV)**	
		Yes	**No**		**Estimate**	**95% CI**
50–59	≤6.0	2	11	13	15.4%	(4.4%; 40.4%)
	>6.0 to ≤10.0	7	34	41	17.1%	(8.9%; 28.7%)
	>10.0	30	57	87	34.5%	(28.7%; 40.0%)
	Total	**39**	**102**	**141**	Prevalence=27.7%	
60–69	≤6.0	6	55	61	9.8%	(4.7%; 19.2%)
	>6.0 to ≤10.0	27	108	135	20.0%	(14.7%; 26.2%)
	>10.0	97	82	179	54.2%	(49.2%; 59.2%)
	Total	**130**	**245**	**375**	Prevalence=34.7%	
70–88	≤6.0	10	64	74	13.5%	(7.7%; 22.1%)
	>6.0 to ≤10.0	36	47	83	43.4%	(34.7%; 52.3%)
	>10.0	73	46	119	61.3%	(54.6%; 67.9%)
	Total	**119**	**157**	**276**	Prevalence=43.1%	

- For age groups 60-69 and 70-88, among patients with IsoPSA Index ≤ 6.0, the probability of high-grade prostate cancer if biopsied were 9.8% and 13.5% correspondingly and it was demonstrated statistically significant decreased likelihood of GG vs corresponding prevalences of 34.7% and 43.1%.
- For age group 50-59, among patients with IsoPSA Index ≤ 6.0, the probability of high-grade prostate cancer if biopsied was 15.4% with 95% CI: (4.4%; 40.4%). The data did not demonstrate a statistically significant decrease of likelihood of GG ≥2 because the upper bound of the 95% CI, 40.4%, is not lower than the prevalence of 27.7%.

The performance of the IsoPSA Assay stratified by age for 204 patients with mpMRI PI-RADS 1–3 is summarized in Table 20.

Table 20: Performance of the IsoPSA Assay Stratified by Age (Patients with mpMRI PI-RADS 1–3, N=204)						
Age Group	**IsoPSA Index range**	**GG ≥2**		**Total**	**Probability of GG ≥2 (Predictive Value, PV)**	
		Yes	**No**		**Estimate**	**95% CI**
50–59	≤6.0	0	4	4	0.0%	(0.0%; 32.8%)
	>6.0 to ≤10.0	1	15	16	6.3%	(1.2%; 13.2%)
	>10.0	1	16	17	5.9%	(1.1%; 12.3%)
	Total	**2**	**35**	**37**	Prevalence=5.4%	
60–69	≤6.0	0	16	16	0.0%	(0.0%; 16.9%)
	>6.0 to ≤10.0	4	34	39	10.5%	(4.4%; 20.1%)
	>10.0	13	30	43	30.2%	(21.7%; 38.5%)
	Total	**17**	**80**	**97**	Prevalence=17.5%	
70–88	≤6.0	2	17	19	10.5%	(3.0%; 27.9%)
	>6.0 to ≤10.0	5	19	24	20.8%	(9.9%; 35.9%)
	>10.0	13	14	27	48.1%	(34.4%; 61.6%)
	Total	**20**	**50**	**70**	Prevalence=28.6%	

B) Race and Ethnicity:
The performance of the IsoPSA Assay for 792 patients was evaluated by stratifying into two subgroups: African American (N=35) and non-African American (N =757) subpopulations. The results are summarized in Table 21.

Table 21: Performance of the IsoPSA Assay Stratified by Race and Ethnicity (All Patients, N=792)						
Race	**IsoPSA range**	**GG ≥2**		**Total**	**Probability of GG ≥2 (Predictive Value, PV)**	
		Yes	**No**		**Estimate**	**95% CI**
Non-African American	≤6.0	17	124	141	12.1%	(7.8%; 18.0%)
	>6.0 to ≤10.0	68	183	251	27.1%	(22.6%; 31.9%)
	>10.0	190	175	365	52.1%	(48.5%; 55.6%)
	Total	**275**	**482**	**757**	Prevalence=36.3%	
African American	≤6.0	1	6	7	14.3%	(2.6%: 47.1%)
	>6.0 to ≤10.0	2	6	8	25.0%	(7.5%; 54.8%)
	>10.0	10	10	20	50.0%	(35.9%; 64.2%)
	Total	**13**	**22**	**35**	Prevalence=37.1%	

Data did not demonstrate that IsoPSA Assay for African American patients is statistically informative: all three predictive values are not statistically different from the prevalence (all three 95% CIs includes the estimate of prevalence of 37.1%). The uncertainty about clinical performance of the IsoPSA Assay for African American patients is mitigated by a requirement to collect more data in the **Post-Approval Study**.

The performance of the IsoPSA Assay stratified by race and ethnicity for 204 patients with mpMRI PI-RADS 1–3 is summarized in Table 22.

Table 22: Performance of the IsoPSA Assay Stratified by Race and Ethnicity (Patients with mpMRI PI-RADS 1–3, N=204)

Race	IsoPSA Index range	GG ≥2		Total	Probability of GG ≥2 (Predictive Value, PV)	
		Yes	No		Estimate	95% CI
Non-African American	≤6.0	2	35	37	5.4%	(1.5%; 16.3%)
	>6.0 to ≤10.0	10	65	75	13.3%	(7.9%; 20.3%)
	>10.0	24	56	80	30.0%	(23.4%; 36.5%)
	Total	**36**	**156**	**192**	Prevalence=18.8%	
African American	≤6.0	0	2	2	0.0%	(0.0%; 58.7%)
	>6.0 to ≤10.0	0	3	3	0.0%	(0.0%; 46.0%)
	>10.0	3	4	7	42.9%	(20.9%; 64.8%)
	Total	**3**	**9**	**12**	Prevalence=25.0%	

C) Biopsy History

The performance of the IsoPSA Assay was analyzed for 792 patients based on history of biopsy: naïve (657 patients) and "repeat" (135 patients). The results are summarized in in Table 23.

Table 23: Performance of the IsoPSA Assay Stratified by Biopsy History (All Patients, N=792)

Biopsy History	IsoPSA Index range	GG ≥2		Total	Probability of GG ≥2 (Predictive Value, PV)	
		Yes	No		Estimate	95% CI
Naive	≤6.0	15	111	126	11.9%	(7.5%; 18.3%)
	>6.0 to ≤10.0	61	153	214	28.5%	(23.6%; 33.8%)
	>10.0	171	146	317	53.9%	(50.1%; 57.8%)
	Total	**247**	**410**	**657**	Prevalence=37.6%	
Repeat	≤6.0	3	19	22	13.6%	(4.9%; 31.4%)
	>6.0 to ≤10.0	9	36	45	20.0%	(11.5%; 31.0%)
	>10.0	29	39	68	42.6%	(35.0%; 50.3%)
	Total	**41**	**94**	**135**	Prevalence=30.4%	

Data did not demonstrate that IsoPSA Assay for patients with repeat biopsies is statistically informative: all three predictive values are not statistically different from the prevalence (all three 95% CIs includes the estimate of prevalence of 30.4%).

The performance of the IsoPSA Assay stratified by biopsy history for 204 patients with mpMRI PI-RADS 1–3 is shown in Table 24:

Table 24: Performance of the IsoPSA Assay Stratified by Biopsy History (Patients with mpMRI PI-RADS 1–3, N=204)						
Type of Biopsy	**IsoPSA Index range**	**GG ≥2**		**Total**	**Probability of GG ≥2 (Predictive Value, PV)**	
		Yes	**No**		**Estimate**	**95% CI**
Naive	≤6.0	1	25	26	3.8%	(0.7%; 17.3%)
	>6.0 to ≤10.0	8	47	55	14.5%	(8.0%; 23.2%)
	>10.0	22	39	61	36.1%	(28.1%; 43.9%)
	Total	**31**	**111**	**142**	Prevalence=21.8%	
Repeat	≤6.0	1	12	13	7.7%	(1.4%; 26.7%)
	>6.0 to ≤10.0	2	21	23	8.7%	(2.5%; 20.0%)
	>10.0	5	21	26	19.2%	(9.8%; 28.8%)
	Total	**8**	**54**	**62**	Prevalence=12.9%	

D) DRE status

The performance of the IsoPSA Assay for 665 patients was analyzed based on the DRE status: 178 patients with abnormal DRE findings, 487 patients with normal DRE findings (DRE was not performed for 127 patients). The results are summarized in Table 25:

Table 25: Performance of the IsoPSA Assay Stratified by DRE status (All Patients, N=792)						
DRE Status	**IsoPSA Index range**	**GG ≥2**		**Total**	**Probability of GG ≥2 (Predictive Value, PV)**	
		Yes	**No**		**Estimate**	**95% CI**
Abnormal	≤6.0	1	37	38	2.6%	(0.5%; 12.9%)
	>6.0 to ≤10.0	22	37	59	37.3%	(27.6%; 47.6%)
	>10.0	46	35	81	56.8%	(48.9%; 64.6%)
	Total	**69**	**109**	**178**	Prevalence=38.8%	
Normal	≤6.0	13	79	92	14.1%	(8.6%; 22.0%)
	>6.0 to ≤10.0	38	118	156	24.4%	(19.0%; 30.4%)
	>10.0	112	127	239	46.9%	(42.6%; 51.1%)
	Total	**163**	**324**	**487**	Prevalence=33.5%	

- For patients with abnormal DRE and normal DRE, among patients in the with IsoPSA Index ≤ 6.0, the probability of high-grade prostate cancer if biopsied were 2.6% and 14.1% correspondingly and it was demonstrated statistically significant decreased likelihood of GG ≥2 vs corresponding prevalences of 38.8% and 33.5%.
- For patients with abnormal DRE and normal DRE, among patients with IsoPSA Index >10, the probability of high-grade prostate cancer if biopsied were 56.8% and 46.9% correspondingly and it was demonstrated statistically significant increased likelihood of GG ≥2 vs corresponding prevalences of 38.8% and 33.5%.

The performance of the IsoPSA Assay stratified by DRE status for 175 patients with mpMRI PI-RADS 1–3 is summarized in Table 26.

Table 26: Performance of the IsoPSA Assay Stratified by DRE Status (Patients with mpMRI PI-RADS 1–3, N=204)

DRE Status	IsoPSA Index range	GG ≥2		Total	Probability of GG ≥2 (Predictive Value, PV)	
		Yes	No		Estimate	95% CI
Abnormal	≤6.0	0	15	15	0.0%	(0.0%; 16.4%)
	>6.0 to ≤10.0	4	15	19	21.1%	(9.4%; 35.9%)
	>10.0	6	13	19	31.6%	(17.6%; 46.4%)
	Total	**10**	**43**	**53**	Prevalence=18.9%	
Normal	≤6.0	1	19	20	5.0%	(0.9%; 21.0%)
	>6.0 to ≤10.0	4	44	48	8.3%	(3.4%; 16.5%)
	>10.0	15	39	54	27.8%	(20.3%; 34.9%)
	Total	**20**	**102**	**122**	Prevalence=16.4%	

c) *Pediatric Extrapolation*:
In this premarket application, existing clinical data was not leveraged to support approval of a pediatric patient population. The device is indicated to be used in the population of men 50 years and older.

XI. FINANCIAL DISCLOSURE

The Financial Disclosure by Clinical Investigators regulation (21 CFR 54) requires applicants who submit a marketing application to include certain information concerning the compensation to, and financial interests and arrangement of, any clinical investigator conducting clinical studies covered by the regulation. The pivotal clinical study included 14 investigators. None of the clinical investigators had disclosable financial interests/arrangements as defined in sections 54.2(a), (b), (c), and (f). The information provided does not raise any questions about the reliability of the data.

XII. SUMMARY OF SUPPLEMENTAL CLINICAL INFORMATION

Not applicable.

XIII. PANEL MEETING RECOMMENDATION AND FDA'S POST-PANEL ACTION

In accordance with the provisions of section 515(c)(3) of the act as amended by the Safe Medical Devices Act of 1990, this PMA was not referred to the panel, an FDA advisory committee, for review and recommendation because the information in the PMA substantially duplicates information previously reviewed by this panel.

XIV. CONCLUSIONS DRAWN FROM PRECLINICAL AND CLINICAL STUDIES

A. Effectiveness Conclusions

Analytical studies demonstrate acceptable analytical performance of Top-phase total PSA assay from 0.065 to 2.720 ng/mL and Top-phase free PSA assay is 0.040 to 2.024 ng/mL.

The clinical effectiveness of the IsoPSA Assay as an aid in the detection of high-grade prostate cancer (Grade Group ≥ 2, Gleason Score ≥ 7) was demonstrated by testing 792 men ≥ 50 years of age, with total PSA levels ≥ 4.0 to ≤ 10.0 ng/mL for whom a biopsy is being considered by a urologist, at 14 community-based urology practices in the United States. The study results indicated that at a cut-off of IsoPSA Index ≤ 6.0, the probability of high-grade prostate cancer (Grade Group ≥ 2, Gleason Score ≥ 7) in subjects with mpMRI PI-RADS 1–3 was 5.1% (95% CI: 1.4%, 15.6%) if biopsied. Therefore, the studies support the effective use of the device to aid in detection of high-grade prostate cancer (Grade Group ≥ 2; Gleason score ≥ 7).

B. Safety Conclusions

The risks of the device are based on nonclinical laboratory testing as well as data collected in a clinical study conducted to support PMA approval as described above. As an in vitro diagnostic (IVD) test, the IsoPSA Assay requires routine blood collection to be performed by trained healthcare professionals in a clinical setting. Sample collection by venipuncture presents no greater safety risk than standard blood collection procedures used for other IVD Tests. Potential downstream risks are associated with clinical decisions based on false positive IsoPSA Assay results, including complications related to prostate biopsy such as infection, fever, rectal bleeding, hematuria, hematospermia, urinary retention, and, in some cases, hospitalization.

The results of the analytical and clinical performance studies demonstrated the acceptable safety profile of the IsoPSA Assay when used in accordance with its Instructions for Use, including boxed warnings, warnings, precautions, and limitations, in conjunction with the clinician's overall assessment and applicable professional guidelines.

C. Benefit-Risk Determination

The IsoPSA Assay clinical performance study included only men for whom a biopsy is being considered by a urologist based on current standard of care. Therefore, the performance of the IsoPSA Assay has not been established in men for whom a biopsy was not already recommended.

a) *Summary of Benefits*:
The probable benefits of the device are also based on data collected in clinical studies conducted to support PMA approval as described above. The decision to perform a prostate biopsy is complex and depends on multiple factors including patient age, family history, results of genetic tests, PSA level, results of digital rectal examination

(DRE), results of other laboratory tests, and, importantly, patient preferences. The benefits of performing prostate biopsy in the specified intended use population include identification of high-grade prostate cancers (Grade Group ≥ 2, Gleason score ≥ 7) for which the patient would benefit which may include prolongation of the lifespan due to the detection and subsequent treatment of a potentially fatal cancer, and may also reduce cancer complications and thereby result in a better quality of life. For the individual patient, patient preferences are a major component of the decision-making process to measure the PSA and/or to do a prostate biopsy. The benefits of not performing a prostate biopsy include not identifying a cancer that is expected to be indolent, and therefore not subjecting the patient to a diagnosis of cancer, as well as surgery, radiation and/or other therapies that would have significant morbidity without providing a concomitant benefit with respect to longevity. These factors are considered in the discussion between the urologist and patient in which the likelihood of various outcomes is estimated and accordingly the risks and benefits of doing the biopsy are discussed.

Before prescribing biopsy, the patients with elevated levels of PSA and/or abnormal DRE are prescribed mpMRI, and/or FDA approved diagnostic tests like free-PSA, p2PSA (*phi*; prostate health Index) or multianalyte 4Kscore. These tests identify subjects that could avoid undergoing biopsy. In the same space, the goal of use of IsoPSA Assay is to identify potentially high-grade prostate cancers (Grade Group ≥ 2, Gleason score ≥ 7) and to avoid biopsy in men ≥ 50 years of age, with total PSA levels ≥ 4.0 to ≤ 10.0 ng/mL specified in the Indications for Use. The clinical study population was limited to men for whom a prostate biopsy had already been recommended by a urologist.

The clinical benefit–risk profile in prostate cancer detection depends on a test's ability to accurately distinguish high-grade from indolent (low-grade) prostate cancer and benign disease. As is true with the other FDA approved tests, insufficient diagnostic accuracy of IsoPSA Assay can lead to unnecessary biopsies, missed high-grade cancers, over-diagnosis of indolent disease, and associated complications. These risks can be reduced by using a test with strong diagnostic performance to guide appropriate biopsy decisions. The clinical performance study was evaluated in a total of 792 patients, each of which had biopsy findings (clinical truth) with results reflecting the proportion of patients with high-grade prostate cancer (Grade Group ≥ 2, Gleason score ≥ 7), and IsoPSA Assay results at the two relevant cutoffs (IsoPSA Index 6.0 and 10.0). Out of the total of 792 patients, 430 subjects also had mpMRI results. The results indicate that among subjects with mpMRI PI-RADS 1–3, and IsoPSA Index ≤ 6.0, the probability of high-grade prostate cancer (Grade Group ≥ 2, Gleason Score ≥ 7.0) was 5.1% (95% CI: 1.4%, 15.6%) if biopsied. In summary, the results provided by the IsoPSA Assay in men ≥ 50 years of age, with total PSA levels ≥ 4.0 to ≤ 10.0 ng/mL, the patients with an IsoPSA Index of ≤ 6.0 and prostate mpMRI results with PI-RADS score of 1–3 have decreased likelihood of high-grade prostate cancer (Grade Group ≥ 2, Gleason score ≥ 7). These results when used in conjunction with other clinical factors and patient preferences, can contribute to a properly informed decision

as to whether or not to proceed with a prostate biopsy, with an expected outcome that is probabilistically more favorable for the individual patient.

b) *Summary of Risks*:

The probable risks of the device are also based on data collected in the clinical studies conducted to support PMA approval as described above. The risks of performance of a prostate biopsy in the specified intended use population include identification of an indolent prostate cancer for which the patient would not likely benefit from immediate treatment. This risk is mitigated by the current practice of active surveillance of such expected indolent tumors. The risks of not performing a prostate biopsy include failure to identify a high-grade prostate cancer (Grade Group ≥ 2, Gleason score ≥ 7), and therefore not treating the patient with surgery, radiation and/or other therapies that could have provided a benefit with respect to longevity and/or quality of life. The use of the IsoPSA Assay in men ≥ 50 years of age, with total PSA levels ≥ 4.0 to ≤ 10.0 ng/mL and with mpMRI PI-RADS 1–3 will result in a decision to perform a prostate biopsy. A false-positive risks infectious complications of the biopsy procedure in about 1-2% patients (based on the medical literature). In patients that have mpMRI PI-RADS 1–3, a false negative IsoPSA Assay results (IsoPSA Index ≤ 6.0) may result in missing a high-grade prostate cancer (Grade Group ≥ 2, Gleason Score ≥ 7) in 5.1% subjects (95% CI: 1.4%, 15.6%), which may or may not be potentially fatal. In patients that have mpMRI PI-RADS 4–5, the IsoPSA Assay should not be used as the test result (IsoPSA Index ≤ 6.0) misses a significantly high 44.0% (95% CI: 27.5%, 62.0%) high-grade prostate cancer (Grade Group ≥ 2, Gleason Score ≥ 7). In patients that have not performed mpMRI before taking the IsoPSA Assay, a false negative test result (IsoPSA Index ≤ 6.0) may result in missing a high-grade prostate cancer (Grade Group ≥ 2, Gleason Score ≥ 7) in 6.0% subjects (95% CI: 2.6%, 12.7%); however, multiple studies have reported that approximately 33-57% of men who undergo biopsy following mpMRI in the intended use population display PI-RADS 4–5 lesions on pre-biopsy mpMRI. The risk of missing high-grade prostate cancer (Grade Group ≥ 2, Gleason Score ≥ 7) in this group of patients (with no pre-biopsy mpMRI and with an IsoPSA Index ≤ 6.0) is mitigated as the clinician should also consider obtaining mpMRI results to confirm the absence of mpMRI PI-RADS 4–5.

The clinical benefit–risk profile also depends on whether sufficient number of subjects (representing the demographic distribution) were included in the clinical performance study. For the IsoPSA Assay, the pivotal clinical performance study included only 4.4% African American subjects (35 subjects out of total 792 subjects), and only 12 subjects with mpMRI PI-RADS 1–3. The evaluation of only 4.4% African American introduces uncertainty of the risk as the low number of subjects does not sufficiently represent the demographic presence of this sub-group. These risks are reduced when the clinician uses caution in interpretation of IsoPSA Assay results for patients in this population, and through a post-approval study.

c) *Benefit-Risk Conclusion*:
There is reasonable assurance that the use of this device, shown by the provision of probabilities of the occurrence of high-grade prostate cancer (Grade Group ≥ 2, Gleason Score ≥ 7) in the population of men in the specified age groups with specified total PSA levels, can provide important and probabilistically accurate information about the risk of Grade Group ≥ 2 prostate cancer and accordingly assist in the clinical assessment of whether performance of a prostate biopsy is in the best interests of the patient considering the benefits and risks of such a biopsy. This is provided that the test is not used alone but rather is used in conjunction with other available clinical information as per the standard of care for such patients who are considered at risk of prostate cancer, including results of digital rectal examination, results of other laboratory tests. Accordingly, based on the data provided, it appears likely that the benefits of use of the device outweigh the risks of use of the device.

Patient Perspective
This submission either did not include specific information on patient perspectives or the information did not serve as part of the basis of the decision to approve or deny the PMA for this device.

In conclusion, given the available information above and boxed warning, the data support that IsoPSA Assay, indicated for use in conjunction with other patient information to aid in detection of high-grade prostate cancer (Grade Group ≥ 2; Gleason score ≥ 7) in men ≥ 50 years of age, with total PSA levels ≥ 4.0 to ≤ 10.0 ng/mL for whom a biopsy is being considered by a urologist based on current standard of care, before consideration of IsoPSA Assay results, the probable benefits outweigh the probable risks.

D. Overall Conclusions

The data in this application support the reasonable assurance of safety and effectiveness of this device when used in accordance with the indications for use. Data from nonclinical and clinical studies support the use of the IsoPSA Assay as an aid in detection of high-grade prostate cancer (Grade Group ≥ 2; Gleason score ≥ 7) in men ≥ 50 years of age, with total PSA levels ≥ 4.0 to ≤ 10.0 ng/mL for whom a biopsy is being considered by a urologist based on current standard of care, before consideration of IsoPSA Assay results. The results indicate that the IsoPSA Assay should not be used in patients with prostate mpMRI results with PI-RADS score of 4–5. For patients with an IsoPSA index ≤ 6.0, the clinician should also consider obtaining mpMRI results to reduce the risk of missing high-grade prostate cancer (Grade Group ≥ 2; Gleason score ≥ 7).

XV. CDRH DECISION

CDRH issued an approval order on November 28, 2025.

The applicant's manufacturing facilities have been inspected and found to be in compliance with the device Quality System (QS) regulation (21 CFR 820).

XVI. APPROVAL SPECIFICATIONS

Directions for use: See device labeling.

Hazards to Health from Use of the Device: See Indications, Contraindications, Warnings, Precautions, and Adverse Events in the device labeling.

Post-approval Requirements and Restrictions: The IsoPSA Assay post-approval study (PAS) in an African American population, meeting the criteria described below:

- Study purpose/objectives: Demonstrate the effectiveness of IsoPSA Assay in an African American population (from intended use population) as per P200048 approval.
- Study design: A prospective, single-blind, multi-site post-approval study of the IsoPSA Assay in an African American population.
- Total number of subjects: At least 250 African American subjects resulting in at least 50 African American subjects with an IsoPSA Index ≤ 6.0.
- Length of follow-up and frequency of assessments: Provide complete data, analysis, and results for FDA's review within 59 months of the P200048 approval date.
- Endpoints: Provide IsoPSA Assay clinical performance for positive high-grade prostate cancer (Grade Group 2, Geason Score 7) – vs – negative for high-grade prostate cancer [low-grade cancer or benign (no cancer)] as per PAS.
- High-level description of the data analysis: As described in the clinical validation section of the IsoPSA Assay labeling.

XVII. REFERENCES

N/A