SUMMARY OF SAFETY AND EFFECTIVENESS DATA

I. GENERAL INFORMATION

Device Generic Name:	free (non-complexed) prostate specific antigen test to distinguish prostate cancer from benign conditions
Device Trade Name:	VIDAS Free PSA rt (FPSA) Assay
Applicant's Name and Address	Biomerieux Inc. 595 Anglum Road Hazelwood, MO 63042
Date(s) of Panel Recommendation:	None
Premarket Approval Application (PMA) Number	P080008
Date of FDA Notice of Approval:	October 8, 2009
Expedited:	Not Applicable

II. INDICATIONS FOR USE

VIDAS FPSA rt is an automated quantitative test for use on the VIDAS instruments, for the quantitative measurement of the free fraction of prostate specific antigen (PSA) in human serum using the ELFA technique (Enzyme linked fluorescent assay). The VIDAS fPSA rt is intended to be used in conjunction with the VIDAS TPSA assay in men age 50 years or older who have digital rectal examination (DRE) that is not suspicious for prostate cancer and VIDAS TPSA values between 4 and 10 ng/mL to determine the %free PSA value. The VIDAS %FPSA value can be used as an aid in discriminating between prostate cancer and benign disease. Prostate biopsy is required for diagnosis of prostate cancer.

III. CONTRAINDICATIONS

There are no known contradictions for this device.

IV. WARNINGS AND PRECAUTIONS

The warnings and precautions can be found in the VIDAS FPSA rt Assay labeling.

V. DEVICE DESCRIPTION

The VIDAS Free PSA rt (fPSA) assay device is a two step sandwich enzyme immunoassay producing a fluorescence signal to detect the non-complexed (free) fraction of prostate specific antigen in human serum.

The VIDAS® FPSA rt kit is configured for 30 tests. The components of the kit are as follows:

30 FPSA Strips	Ready-to-use.
30 fPSA SPRs 1 x30	Ready-to-use. Interior of SPR coated with mouse monoclonal anti-free PSA specific antibodies.
FPSA Control 1 x2 mL (lyophilized)	Human serum* + human PSA (free fraction) + preservatives: 1 g/L Methylisothiazolone (MIT) and 0.2 g/L Bromo Nitro Dioxane (BND) The confidence interval in ng/mL is indicated on the MLE card after the following mention: "Control C1 Dose Value Range".
FPSA Calibrator 1 x2.5mL	Bovine albumin + human PSA (free fraction) + 0.9 g/L sodium azide. The concentration in ng/mL is indicated on the MLE card after the following mention: "Calibrator (S1) Dose Value". The confidence interval in "Relative Fluorescence Value" is indicated on the MLE card after the following mention: "Calibrator (S1) RFV Range".
1 MLE card	Specifications sheet containing the factory master calibration data required to calibrate the test.
1 Package insert	

The Solid Phase Receptacle (SPR) serves as the solid phase as well as the pipetting device for the assay. Reagents for the assay are ready-to-use and pre-dispensed in the sealed reagent strips. All of the assay steps are performed automatically by the instrument. The sample is cycled in and out of the SPR several times. This operation enables the antibody fixed onto the interior wall of the SPR to capture the free fraction of the prostate specific antigen present in the sample. Unbound components are eliminated during the washing steps. The alkaline phosphatase labeled monoclonal antibody is then incubated in the SPR where it binds with the prostate specific antigen. Unbound conjugate is then eliminated during the washing steps. During the final detection step, the substrate (4-Methyl-umbelliferyl phosphate) is cycled in and out of the SPR. The conjugate enzyme catalyzes the hydrolysis of this substrate into a fluorescent product (4-Methyl-umbelliferone) the fluorescence of which is measured at 450 nm. The intensity of the fluorescence is proportional to the concentration the free fraction of prostate specific antigen present in the sample. At the end of the assay, results are automatically calculated by the VIDAS® instrument in relation to the calibration curve stored in memory, and then printed. The measurement range of the VIDAS Free PSA rt assay is: 0.05 - 10 ng/mL. Calibration, using the calibrator provided in the kit, is performed each time a new lot of reagents is opened, when troubleshooting the assay, or under appropriate conditions. Calibration should then be performed every 14 days.

VI. ALTERNATIVE PRACTICES AND PROCEDURES

Other than in vitro diagnostic devices for free PSA available from other manufacturers, there are currently no alternative practices or procedures to discriminate prostate cancer from benign prostatic diseases. For detection of prostate cancer, alternative practices and procedures include digital rectal examination, transrectal ultrasound, and total PSA testing to provide additional information about prostate abnormalities and guide prostate biopsy.

Other devices for measuring serum free PSA are commercially available to aid in the discrimination of prostate cancer from benign prostatic disease in men aged 50 years and older whose total PSA values are between 4.0 and 10.0 ng/mL and whose DRE is not suspicious for cancer.

VII. MARKETING HISTORY

The BioMerieux VIDAS® FPSA assay is CE marked and is currently marketed in the following countries:

Subsidiaries:

Austria, Belgium, France, Germany, Greece, Italy, The Netherlands, Norway, Poland, Portugal, Russia, Spain, Switzerland, Turkey, United Kingdom, Denmark, Argentina, Brazil, Chile, Colombia, Mexico, China, India, Korea, Thailand, Australia

Distributors:

Andorra, United Arab Emirates, Albania, Netherlands Antilles, Angola, Azerbaijan, Bosnia-Herzegovina, Bangladesh, Burkina-Faso, Bulgaria, Bahrain, Benin, Bolivia, Belarus, Democratic Republic of Congo, Congo, Ivory Coast, Cameroon,' China, Costa Rica, Cape Verde, Cyprus, Czech Republic, Djibouti, Dominican Republic, Algeria, Estonia, Egypt, Ethiopia, Gabon, Georgia, Ghana, Guatemala, Honduras, Croatia, Hungary, Indonesia, Israel, Iraq, Iran, Island, Jordan, Kenya, Kosovo, Kuwait, Lebanon, Sri Lanka, Libya, Morocco, Moldova, Macedonia, Mali, Myanmar, Mauritania, Malta, Malaysia, Mozambique, Nigeria, Oman, Panama, Peru, Philippines, Paraguay, Palestine, Qatar, Romania, Saudi Arabia, Singapore, Slovenia, Slovakia, Senegal, El Salvador, Serbia, Arabic Syrian Republic, Togo, Tunisia, Taiwan, Tanzania, Uganda, Uruguay, Venezuela, Vietnam, Yugoslavia, South Africa, Serbia & Montenegro

Since the launch of the VIDAS® FPSA assay in the above listed countries, no withdrawals for reasons of safety or effectiveness have occurred.

VIII. POTENTIAL ADVERSE EFFECTS OF THE DEVICE ON HEALTH

Since the VIDAS Free PSA rt (FPSA) assay kit is for in vitro diagnostic use, there is no direct adverse effect on the health of a patient. However, failure of the product to perform as intended or errors in the use of the product could lead to a false result. Low ratios of FPSA to total PSA can occur in patients with benign prostatic disorders and elevated ratios are not always associated with absence of disease. Patient status must not be assessed entirely by FPSA or %FPSA results but in conjunction with information from a complete clinical evaluation including DRE or other diagnostic tests. Potential adverse effects are:

1. A falsely elevated FPSA or %FPSA result (false negative) may lead to a delay beneficial treatment (biopsy and needed therapy).
2. A falsely low FPSA or %FPSA result (false positive) may cause unnecessary biopsy and needless therapy.

The concentration of free PSA in a given specimen determined with different assays can vary due to the differences in assay methods and reagent specificity. The results reported by the

laboratory to the physician must include the identity of the assay used. Free and total PSA values should be obtained using assays from the same manufacturer

IX. SUMMARY OF PRECLINICAL STUDIES

A. Non-clinical Studies

1. Precision

Studies sought to determine total imprecision, between-day component, between-run component, and within-run (repeatability) imprecision. Three serum pools, each of different fPSA concentration were tested in 4 replicates 2 runs per day for 10 days for each of 2 reagent lots using a single instrument. The serum pools were 0.52 ng/mL fPSA (pool 1), 1.92 ng/mL fPSA (pool 2), and 4.97 ng/mL fPSA (pool 3). The testing was performed at 3 sites. The percent coefficient of variation was calculated for each component of imprecision. The calculated results are summarized as follows:

%CV total imprecision: < 5.3%

%CV between-day imprecision: < 4.4%

%CV between-run imprecision: < 2.8%

%CV within-run precision (repeatability): < 4.6%

A lot-to-lot variation study was performed at three locations for ten days, running four runs each day with two different reagent lots of the assay. Three serum pools of different concentration, a commercially available quality control material of three different concentrations, and the kit control were utilized as samples. The results are summarized as follows:

	Mean level	Lot-to-lot SD	Lot-to-lot %CV
Kit control	0.694	0.177	25.5%
Control 1	0.515	0.015	2.9%
Control 2	1.684	0.024	1.4%
Control 3	6.387	0.108	1.7%
Serum pool 1	0.509	0.012	2.4%
Serum pool 2	1.897	0.028	1.5%
Serum pool 3	4.894	0.075	1.5%

2. Performance at Low Levels (Analytical and Functional sensitivity (Limit of Blank and Limit of Detection))

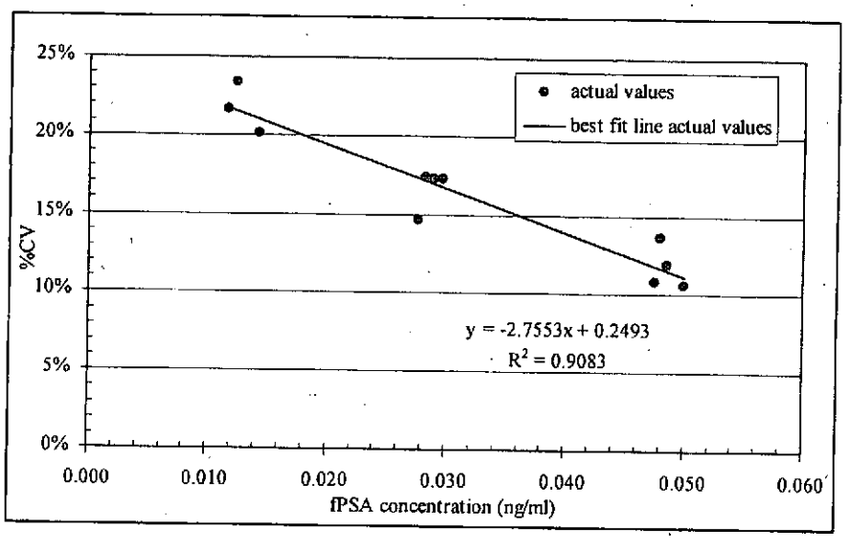
The Limit of Blank defined as the highest value that is likely to be observed (with 95% probability) for a sample with no analyte, was calculated as the value two standard deviations above the value of replicates of the zero calibrator. For the determination, 20 replicates of the 0 ng/mL calibrator for each of three kit lots were utilized. The relative fluorescence unit (RFU) for each replicate was converted to

ng/mL using the slope between the lowest quality control and mean 0 calibrator RFU. The mean fPSA value for each kit lot was 0.006 ng/mL, 0.000 ng/mL, and 0.005 ng/mL fPSA. The overall calculated limit of blank, as calculated in CLSI EP17-A "Protocols for Determination of Limits of Detection and Limits of Quantitation; Approved Guideline", 2004, is 0.0094 ng/mL (n = 60). This value, approximately 0.01 ng/mL, is below the sponsor's specification of < 0.05 ng/mL.

The Limit of Quantitation (functional sensitivity) is defined as the concentration when samples are serially diluted at which the coefficient of variation of multiple replicates equals 20%. The sponsor utilized 3 lots of assay reagent on 1 instrument. A patient sample, initially 9.5 ng/mL, was diluted 40-fold with assay diluent to prepare a pool from which 4 serial 2-fold dilutions were made. Replicates of 4 from each serial 2-fold dilution were tested with each reagent lot. The relative fluorescence unit at each dilution was converted to the concentration based on the dilution for a particular sample. The summarized results are as follows:

	Lot 1	Lot2	Lot3
Mean fPSA 1:4 dilution	0.053	0.050	0.043
%CV 1:4 dilution	2.1%	6.5%	2.1%
Mean fPSA 1:8 dilution	0.025	0.033	0.028
%CV 1:8 dilution	4.4%	2.7%	8.3%
Mean fPSA 1:16 dilution	0.010	0.014	0.015
%CV 1:16 dilution	20.2%	12.4%	6.1%
Limit of Quantitation	< 0.025	< 0.014	< 0.015

In order to more precisely calculate the concentration giving 20% CV at low fPSA concentrations, a linear model was utilized for 3 lots across 3 fPSA concentrations. The mean fPSA concentration and %CV for 3 lots at a particular concentration were calculated. The equation of the best fitting straight line was $y = 2.7553x + 0.2493$, $n = 33$. The graph of the best fitting line and values at each point are shown in the following graph.



The fPSA value at which the %CV is exactly 20% was 0.018 ng/mL, a value just above the initially estimated range. The parametric limit of detection calculated is 0.016 ng/mL with a %CV of 20.5%. This value is below the sponsor's claim as a limit of detection < 0.05 ng/mL).

3. Analytical specificity

To assess effects of interfering substances on the assay a variety of substances (endogeneous substances present in serum, medications, and potential cross-reacting substances in serum) in known concentrations were added to 2 serum pools of different concentrations (0.5 ng/mL and 5.0 ng/mL). Four or five replicates of each serum pool were tested using a single lot of assay reagents and a single instrument. For testing various interfering substances present in serum, the substances were added to the serum pools at 3-10 times the normal expected value (or therapeutic levels). The mean concentration of the spiked sample was compared with the mean concentration of serum pool without added substance. Samples whose concentrations were within ± 10% of the mean value of samples without added substance were accepted as without significant interference at the highest concentration of interfering substance tested without effect. The calculated percentage of assay response with interfering substance compared with the assay response without interfering substance was determined. The following table summarizes the assay response to the tested substances.

		free PSA with interferent / free PSA without interferent in %	
Substance tested	Concentration of tested substance	0.5 ng/mL pool	5.0 ng/mL pool
Triglycerides (lipemia)	30 mg/mL	91%	98%

Hemoglobin	12 mg/mL	91%	92%
Bilirubin	0.6 mg/mL	94%	97%
Total protein (albumin)	120 mg/mL	105%	104%
Transferrin	5.0 mg/mL	96%	100%
Urea	5.0 mg/mL	96%	99%
Rheumatoid factor	183 U/mL	97%	101%
Human anti-mouse antibodies (HAMA)	0.11 ug/mL	98%	99%
Prostate specific antigen complexed with alpha-1-antichymotrypsin	50 ng/mL	106%	104%
Acetaminophen (tylenol)	25 ug/mL	94%	101%
Aspirin	0.60 mg/mL	94%	95%
Amikacin	15 ug/mL	96%	99%
Cortisol/hydrocortisone	1.0 mg/mL	96%	100%
Coumarin	1.4 mg/mL	97%	99%
Cyclosporin A	3.0 ng/mL	98%	99%
Digoxin	50 ng/mL	96%	96%
Gentamicin	0.12 mg/mL	96%	94%
Heparin	3.0 U/mL	95%	100%
Lithium carbonate	0.12 mg/mL	102%	102%
Mitomycin C	60 ug/mL	96%	97%
Paclitaxel	4.0 ng/mL	97%	102%
Phenytoin	0.1 mg/mL	96%	98%
Propanolol	5.0 ug/mL	96%	95%
Salicylate/salicylic acid	0.5 mg/mL	100%	97%
Theophylline/aminophylline	40 ug/mL	96%	94%

Caffeine	0.1 mg/mL	99%	100%
5'-fluorouracil	1.0 mg/mL	96%	95%
Adriamycin	0.1 mg/mL	96%	95%
Amethopterin/methotrexate	1.0 mg/mL	99%	100%
Cisplatin dichloride	0.1 mg/mL	96%	97%
Cyclophosphamide	0.25 mg/mL	93%	93%
Leucovin	1.1 mg/mL	95%	94%
Levamisole	5.0 mg/mL	96%	96%
Novatrone	0.5 mg/mL	97%	91%
Oxiplatin	0.25 mg/mL	96%	98%

The results for cross-reacting substances are noted as follows:

	fPSA test results (ng/mL)	
Cross-reacting substance	0.5 ng/mL fPSA pool	5.0 ng/mL fPSA pool
CEA	< 0.05	<0.05
Prostatic acid phosphatase	< 0.05	< 0.05
Alpha-fetoprotein	< 0.05	< 0.05
CA15-3	< 0.05	< 0.05
CA125	< 0.05	< 0.05
Prostate-specific antigen-alpha-1-antichymotrypsin complex	< 0.5 at 0 ng/mL fPSA	

Based on the supplied information, none of the substances tested (endogeneous serum constituents, medications potentially present in serum, or cross-reacting substances) caused significant interference in the fPSA assay at the indicated concentrations. Regarding the presence of signal in the fPSA assay from complexed PSA (Prostate-specific antigen- alpha-1-antichymotrypsin complex), the percentage cross-reactivity in the assay due to complexed PSA would be at most 1%.

4. Linearity estimation

Two types of linearity, both using CLSI guidance EP6-A, were assessed: i) linearity in the reportable range; and ii) linearity on dilution in sample diluent. A total of 10 samples were utilized, 4 for linearity in the reportable range and 6 for linearity on dilution in sample diluent. Linearity was calculated using a weighted regression analysis since the assay precision is not constant over the assay measurement range. For linearity in the reportable range, 4 serum samples were diluted with a serum pool of low fPSA value to obtain 7 dilutions. Each dilution was tested in 4 replicates on 1 instrument using 1 reagent lot. The second or third order polynomials in the evaluation were not significant. The assay is best fit using a linear model for the reportable range. The VIDAS fPSA is linear from 0.05 ng/mL to 10 ng/mL.

For linearity using assay sample diluent rather than a serum pool, 6 serum samples were diluted to obtain 9 dilutions. Each dilution was tested in 3 or 4 replicates on 1 instrument using 1 reagent lot. For all samples the data are linear or display acceptable deviation from linearity.

5. High Dose Hook Effect

The effect of high dose hook effect was assessed by dilution of a single very high free PSA sample, ~300,000 ng/mL, tested using a series of dilutions until concentrations could be read in the reportable range. The concentrations of fPSA above the reportable range were determined from actual assay values corrected with the corresponding dilution. The result is as follows:

Sample fPSA concentration (ng/mL)	Assay readout (ng/mL)
0.00	<0.05
3.75	3.75
7.72	7.72
19.0	>10
38.1	>10
76.2	>10
381	>10
3810	>10
38100	>10
190500	>10

381000	>10

There is no high dose hook effect up to 381,000 ng/mL.

6. Accuracy Determination

A spiked recovery experiment was utilized to assess accuracy of the assay. Free PSA was added to normal serum and sample diluent. Samples were tested in 4 replicates on the same instrument using a single lot of assay reagents. The percent free PSA recovered was calculated and compared with the expected concentration. The overall mean recovery for 3 concentrations of fPSA spiked into sample diluent (approximately 3, 6, or 10 ng/mL) was 98.6%. The overall mean recovery for 3 concentrations of fPSA spiked into serum (approximately 2.5, 5 and 9 ng/mL) was 96.4%. The overall percentage recovery was specified to be 100% ± 10%. All individual replicate recoveries were between 90% to 110%.

The acceptance criteria appear to represent both bias from a true value (inaccuracy) as well as random assay imprecision (dispersion of repeated values). The total imprecision of the assay as described in the imprecision studies is 5% coefficient of variation. The remaining 5% in the acceptance criteria represent an estimated 5% accuracy bias.

The standard deviation of the observed from true value was 0.045 ng/mL (accuracy bias). The standard deviation of the observed from the true value represents a mean coefficient of variation for accuracy of 3.8% (95% confidence interval of variance 3.0% to 5.0%). This calculated bias from the true value of 3.8% (at most 5%) is similar to the specified value of 5% as described above. Therefore, the data is acceptable.

8. Upper Limit of Reference Interval

Serum specimens were obtained for a normal healthy male cohort who met criteria as described below. Serum specimens were obtained from 216 men recruited from 6 geographically-divers sites in the U. S. using a generic collection protocol and IRB approval. The normal cohort consisted of 83% Caucasian, 14% Hispanic and 3% African-American or Asian men. The sites are listed as follows:

SITE ID	n	% of total subjects
06484	82	38.0%
33134	20	9.3%
33324	30	13.9%
33442	37	17.1%
78130	46	21.3%
01002	1	0.5%
	216	

Inclusion criteria – Normal Healthy cohort

All men, regardless of race, who voluntarily donate a blood sample:

1. are age 50 or older
2. have no history of prostate disease
3. are free of signs and symptoms of active disease in Red Cross Blood criteria for blood collection
4. can understand and sign informed consent
5. feel healthy on day of study visit
6. have no history of malignancy, basal cell skin cancer allowed

Exclusion criteria – Normal Healthy cohort

1. men younger than 50 years
2. men with a history of chronic prostate disease
3. men with any active disease or history of malignancy, other than basal cell skin cancer

The upper limit of reference interval of the fPSA/tPSA ratio was determined based upon the CLSI document C28-A3 "Defining, Establishing, and Verifying Reference Intervals in the Clinical Laboratory: Approved Guideline – Third Edition", 2008.

Summary Analysis of Normal Healthy Cohort

This group of men comprised 216 men aged 50 and older with no evidence of prostate disease meeting Red Cross Criteria for blood donation. The men ranged in age from 50 to 86 years of age (mean 59 years with standard deviation 7.1 years). In this group of normal ostensibly healthy men, 83% of men were Caucasian, 14% Hispanic, and 3% African-American or Asian. In this cohort, 95% of normal healthy men had a free PSA concentration at or below 0.86 ng/mL (2.5^{th} percentile = 0.08 ng/mL, median = 0.28 ng/mL, 97.5^{th} percentile = 1.37 ng/mL). A summary of the empirical distribution of %free PSA in normal healthy men is as follows:

percentile	%fPSA
0.025	7.8%
0.05	12.1%
0.1	14.3%
0.2	17.1%
0.3	19.4%
0.4	21.1%
0.5	23.3%

0.6	25.5%
0.7	27.8%
0.8	30.1%
0.9	33.9%
0.95	37.7%
0.975	41.6%
0.99	43.2%

The median %free PSA of normal healthy men is 23% free PSA. Note further that 95% of apparently normal healthy men have %free PSA values at or below 38% free PSA.

B. Summary of Primary Effectiveness Study

A clinical study was undertaken to assess the clinical effectiveness of the VIDAS fPSA for differentiating between benign prostatic conditions and prostate cancer using the ratio of fPSA to tPSA in patients with tPSA levels in the range of 4.0 to 10.0 ng/mL and with DRE results that are not suspicious of cancer.

1. Study Design

The serum samples utilized in this study are part of a larger sample collection study by a contract research organization. At the time enrollment closed for this study, a total of 1963 subjects/samples were available. All men, regardless of race, presenting to a practicing urologist with symptoms resulting in an evaluation for prostate cancer and sufficiently collected serum volume were eligible for inclusion. From this pool, further inclusion/exclusion criteria were applied to obtain 1673 evaluable subjects from 29 geographically-diverse sites in the U. S. The following inclusion/exclusion criteria were applied to 1673 enrolled subjects to determine the target population for the proposed intended use.

a) Clinical Inclusion and Exclusion Criteria

Enrolled in the study were all men, regardless of race, presenting to a practicing urologist with symptoms resulting in an evaluation for prostate cancer, including trans-rectal prostate biopsy containing at least 6 cores, who have total PSA results using the proposed device between 4.0 and 10.0 ng/mL inclusive and whose DRE results are not suspicious for cancer who:

1. Have not been treated for benign prostate disease with hormone-based therapy, surgery, or radiation within 90 days prior to visit
2. Have no history of an evaluation for prostate cancer prior to visit
3. Are 50 years of age or older
4. Can understand and sign an informed consent for prior to study procedures

5. Have appropriate medical records available for verification of medical history
6. Will have had a DRE either as part of the study procedures after blood sample collection or have had a DRE not more than 30 days prior the initial visit but not within 5 days prior to the blood sample collection
7. have a biopsy of at least 6 cores
8. can give a blood sample of at least 5 ml not more than 15 days prior to the study biopsy
9. have available results of biopsy report
10. if diagnosed with cancer, have Gleason score available, individual Gleason grades if possible, and surgical stage and grade if prostatectomy is performed

Patients were <u>not</u> enrolled in the study if they were:

1. Men younger than 50 years of age
2. Men with a history of prostate cancer prior to study visit
3. Men with a prior history of benign prostate disease treated within 90 days of study visit with hormone therapy, surgery, or radiation
4. Men having a DRE or other form of prostate manipulation less than 5 days prior to blood collection
5. Men failing to meet other inclusion criteria

Serum samples taken from subjects were stored at -70°C or lower for less than 5 years from date of collection in order to be included. Serum samples were tested in 2005 using the VIDAS total PSA assay in order to select subjects with tPSA values between 4.0 and 10.0 ng/mL, inclusive. Due to re-calibration of the free PSA assay, re-testing in the total PSA assay and selection for subjects with re-tested tPSA values between 4.0 and 10.0 ng/mL yielded 679 subjects for final analysis. Subjects were obtained from 28 geographically-diverse collection sites. Free PSA and Total PSA testing for the bioMeriux study was performed at a contract research organization using a single lot of total PSA reagents and a single lot of Free PSA reagents. Three separate instruments were used in the laboratory at the contract research organization.

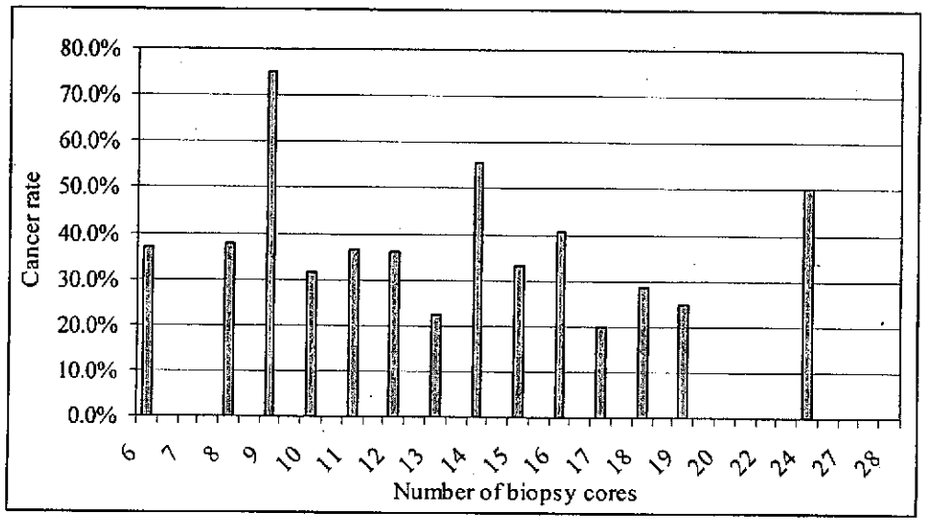
2. Clinical Reference Standard

The clinical status of men in the clinical study was determined by biopsy diagnosis. With regards to effectiveness, analysis will determine the significance of the association of the ratio of FPSA/TPSA at a cutoff determined from ROC analysis with the presence or absence of cancer determined by biopsy.

3. Study Population Demographics and Baseline Parameters

In the target intended use population of 679 subjects collected from 28 geographically different sites, mean age was 64.3 years (standard deviation 7.9 years). The racial composition of this cohort was 82% Caucasian, 8.6% African American, 1 % Asian descent, 7.9% Hispanics, the remainder (0.5%) other ethnicity. The overall cancer rate was 35.6%. The cancer rate by site ranged from 0% to 67%.

Of all biopsies, 63% were 12 core biopsies and 75% of all biopsies were 12 or more biopsy cores. The cancer rate is not significantly different by number of biopsy cores ($p > 0.05$). This is indicated graphically by the following:



The cancer rate appears relatively stable (centered at 35.6%) across all biopsy cores taken.

Among the subjects of the clinical study, several characteristics were examined to assess if data from subjects at the 28 geographic sites can be pooled for final performance evaluation. The mean age by site was 64.2 years and ranged from 60.3 years to 68.8 years. A subject characteristic assessed was the percentage of men with a normal DRE result by site. In this case, a DRE result can be classified by the evaluating physician as normal or abnormal but not suspicious for cancer. The overall rate of a normal DRE result was 77%. The range of mean percentages of normal DRE result by site was from 22% to 100%. Another characteristic assessed by site was mean total PSA value. The overall mean tPSA value was 6.4 ng/mL. The mean tPSA by site ranged from 5.4 ng/mL to 7.1 ng/mL.

The sponsor has required, as part of the inclusion criteria, subjects who were biopsied. Men without biopsy result, who form a proportion of subjects in the target population of subjects with total PSA between 4.0 and 10.0 ng/mL and whose DRE is not suspicious for cancer, were not included in the sampled population. It is not possible currently to assess if the biopsied subjects are representative of the target population – biopsied and

non-biopsied men – whose total PSA values are between 4.0 and 10.0 and whose DRE results are not suspicious for cancer. Therefore, there is the potential for verification bias – only subjects with certain characteristics are selected for verification of the disease status. The lack of verification of disease status in some proportion of the sampled population can lead to performance estimates that possibly are not representative of the total target population. If biopsied subjects are selected from the target population at random and the non-biopsied subjects have equivalent clinical and intrinsic subject characteristics as the biopsied subjects, then the assay performance will be representative and likely unbiased. Information has not been provided on sampled subjects within the potential target population who did not have biopsy. Therefore, there is no available method to assess if the sampled population is a random representative sample of the target population.

4. Safety and Effectiveness Results

The empirical distribution of %fPSA value for both malignant and non-malignant subjects in the clinical study was calculated for 679 subjects meeting inclusion/exclusion criteria. Among benign subjects, the mean %fPSA was 19.4%, while the mean %fPSA in the malignant subjects was 14.8%. A table categorizing the %fPSA into 4 categories for benign disease and cancer subjects is as follows:

		%free PSA ranges			
Biopsy result	Number of subjects	≤10%	>10 to <19.9%	≥ 20 to < 25%	≥25%
negative	437	33	233	85	86
positive	242	66	133	26	17

The mean %fPSA value was significantly lower in subjects with prostate cancer (benign mean %fPSA = 19.4% vs. cancer mean %fPSA= 14.8%).

ROC analysis of the %free PSA among 679 subjects indicated that the area under the ROC curve was 0.698 (95% confidence interval 0.656 to 0.740).

At a cutoff of 23%, the sensitivity of %free PSA was 90.5% (219/242) and the specificity was 26.5% (116/437).

Based on a cutoff of 23% free PSA, the subjects can be categorized by assay result and biopsy result into 2 categories for each parameter. The following 2 x 2 table is analysis of the 679 subjects with total PSA between 4.0 and 10.0 and whose DRE is not suspicious for cancer (normal DRE and abnormal but not suspicious for cancer).

	Malignant	Benign	Total
%fPSA ≤ 23%	219	321	540
%fPSA > 23%	23	116	139
Total	242	437	679

Clinical performance with %fPSA cutoff=23%

	Estimate	Lower limit of 95% CI	Upper limit of 95% CI
sensitivity	90.5% (219/242)	86.1%	93.6%
specificity	26.5% (116/437)	22.6%	30.9%
PPV	40.6% (219/540)	38.9%	42.3%
NPV	83.5% (116/139)	77.0%	88.5%

Prevalence=35.6%

For the cutoff of 23%, sensitivity was 90.5% (219/242) and specificity was 26.5% (116/437). The true positive rate (sensitivity) was significantly higher than the false positive rate (1-specifcity = 73.5%). Difference between TPR and FPR was =17.0% with 95% CI: 11.3% to 22.5%. The PPV (40.6%) was statistically higher than the prevalence (35.6%) and the NPV (83.5%) was statistically higher than 1-prevalence (64.4%). The %free PSA assay was informative.

Note that if the biopsy procedure will be performed on every subject from the population of subjects with total PSA value of 4.0 – 10.0 ng/mL and DRE negative results, then sensitivity will be 100% and specificity will be 0%. Every subject from this population has quantitative value of total PSA and the quantitative value total PSA test values with cutoff higher than 4.0 will improve specificity (more than 0%) and decrease sensitivity (less than 100%). Therefore, in addition, it should be demonstrated an improvement in %fPSA specificity over the specificity of tPSA test when the sensitivities of %fPSA and the tPSA are the same.

At a cutoff of 4.53 ng/mL, the sensitivity of total PSA alone was 90.5% (equaling the sensitivity of %free PSA) and the specificity was 11.4% (95% confidence interval 8.6% to 14.8%). The difference in specificity of %free PSA compared with specificity of total PSA was 15.1% (95% confidence interval for difference 10.1% to 20.1%). The difference in specificities was significant ($p < 0.001$) since the confidence interval does not include 0.

Biopsy Negative			
%fPSA	tPSA >= 4.53	tPSA < 4.53	total
≤ 23%	287	34	321
> 23%	100	16	116

total	387	50	437

When the level of sensitivity was 90.5%, the specificity of %fPSA was 26.5% (116/437) and the specificity of tPSA was 11.4% (50/437). An improvement in specificity was +15.1% with 10.1% to 20.1%. Therefore, the specificity of %free PSA is significantly higher than the specificity of total PSA in the range 4.0 to 10.0 ng/mL.

Sensitivity and Specificity of the %fPSA for the Cutoff of 23% Stratified for Different Ranges of tPSA

In addition to a comparison of specificities of %free PSA and total PSA, the sensitivity and specificity of %free PSA in various total PSA ranges between 4.0 and 10.0 ng/mL was calculated from the dataset of 679 subjects:

	Total PSA (ng/mL)				
%free PSA at 23%	4.00-5.00	5.01-6.00	6.01-8.00	8.01-10.00	Total
Biopsy positive	50	59	96	37	242
Biopsy negative	97	97	169	74	437
Sensitivity %fPSA	94.0% (47/50)	88.1% (52/59)	91.7% (88/96)	86.1% (32/37)	90.5% (219/242)
Specificity %fPSA	32.0% (31/97)	25.8% (25/97)	24.9% (42/169)	24.3% (18/74)	26.5% (116/437
Cancer rate	34.0%	37.8%	36.2%	33.3%	36.5%

The data reflects a lowering of sensitivity for %free PSA across higher total PSA values (for example, 94% sensitivity for %free PSA for tPSA value between 4 and 5 ng/mL compared with 86% sensitivity for %free PSA for total PSA values between 8 and 10 ng/mL. Similarly, there is a lowering of specificity for %free PSA across higher total PSA values (32% specificity %free PSA at 4-5 ng/mL tPSA compared with 24% specificity %free PSA at 8-10 ng/mL tPSA). Among the four ranges of tPSA values, the highest specificity of %fPSA was observed for the subjects with tPSA values of 4.0 – 5.0 ng/mL (32.0%) and the lowest specificity was observed for the subjects with tPSA values of 8.0-10.0 ng/mL (24.3%).

Performance of %fPSA for Different Cutoffs

Using data derived from the ROC analysis of 679 subjects, the following summarizes sensitivity and specificity at cutoffs between 23% and 29%:

%free PSA Cutoff	Sensitivity	95% confidence interval		Specificity	95% confidence interval	
≤22%	86.4%	81.4%	to 90.4%	30.7%	26.4%	to 35.2%
≤23%	90.5%	86.1%	to 93.9%	26.5%	22.5%	to 30.9%
≤24%	91.7%	87.5%	to 94.9%	22.0%	18.2%	to 26.1%
≤26%	93.4%	89.5%	to 96.2%	16.7%	13.3%	to 20.5%
≤29%	94.6%	91.0%	to 97.1%	10.8%	8.0%	to 14.0%
≤53%	99.2%	97.0%	to 99.0%	0%	0%	to 0.8%

At a cutoff of 23% free PSA, 91% of men with cancers are detected and 25% of men with benign diseases avoid unnecessary biopsy. It was stated by the sponsor states that the risk/benefit at 23% free PSA is the most advantageous for men being tested.

While the choice of cutoff is not unreasonable, it should be noted that the sensitivity is 86% at the lowest limit of the confidence interval. Therefore, it is reasonable to consider the cutoff of 26% where the sensitivity at the lower limit of confidence is 90%. The disadvantage of the cutoff at 26% is the lowered specificity (16% at the mean and 13% at the lower confidence interval).

Probability of Finding Prostate Cancer for Different Ranges of %fPSA Stratified by Age

To estimate the probability of finding prostate cancer for different ranges of %FPSA stratified by age, the following summary table shows the observed risk of finding prostate cancer on biopsy:

Probability of Finding Prostate Cancer on Core Biopsy by Age Groups				
% fPSA ratio	50-59	60-69	≥ 70	Total
≤ 10	53.8% (21/39)	76.9% (30/39)	71.4% (15/21)	66.7% (66/99)
10.1-19	28.3% (36/127)	35.3% (59/167)	52.8% (38/72)	36.3% (133/366)
20-25	12.5% (3/24)	25.9% (15/58)	27.6% (8/29)	23.4% (26/111)
> 26	<1% (0/8)	13.0% (6/46)	22.4% (11/49)	16.5% (17/103)
Total	30.3% (60/198)	35.5% (110/310)	42.1% (72/171)	35.6% (242/679)

The probability of cancer for men aged 50-59 is lower than the probability of cancer for men greater than 70 years.

Logistic regression analysis was performed on 679 subjects using biopsy result (cancer or non-cancer) as the outcome variable and %free PSA, total PSA, and age as predictor variables. The natural logarithms were calculated to transform the data to normal distributions. The following table shows the logistic regression results:

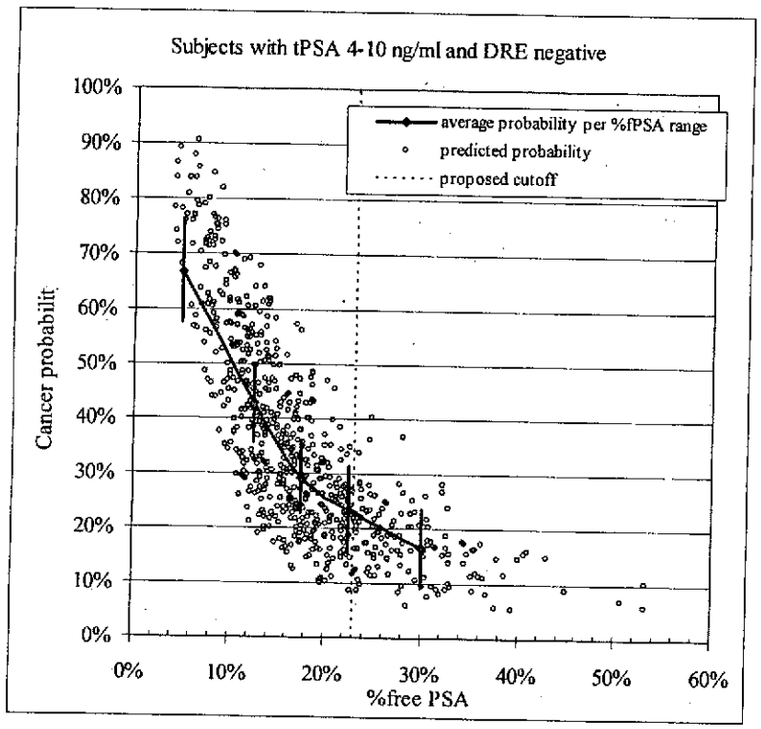
Logistic Regression Parameter Estimates

	estimate	Standard Error	Significance

Ln(%fPSA)	-2.02	.22	<0.001
Ln(tPSA)	-0.47	.38	0.2098
Ln(age)	4.08	.77	<0.001
Constant	-20.46	3.35	<0.001

The data demonstrated that age and %fPSA are statistically significant predictors of prostate cancer in the data set of this clinical study.

The following graph shows individual subject cancer probabilities and the mean probability (± standard error) for %free PSA ranges ≤ 10%, 10-15%, 15-20%, 20-25%, and > 25%.



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

In accordance with the provisions of section 515(c)(2) of the act as amended by the Safe Medical Devices Act of 1990, this PMA was not referred to the Immunology Devices Panel, an FDA advisory committee, for review and recommendation because the information in the PMA substantially duplicates information previously reviewed by this panel.

XI. CONCLUSIONS DRAWN FROM PRECLINICAL AND CLINICAL STUDIES

A. Risk Benefit Analysis

Cancer of the prostate is the most common noncutaneous cancer diagnosis among American men and is the second leading cause of their cancer mortality (Weir HK, Thun

MJ, Hankey BF, Ries LA, Howe HL, Wingo PA, Jemal A, Ward E, Anderson RN, Edwards BK. Annual report to the nation on the status of cancer, 1975-2000, featuring the uses of surveillance data for cancer prevention and control. J Natl Cancer Inst. 2003 Sep 3; 95(17):1276-1299). As such, prostate cancer burden poses a significant public health problem in the United States. The cause of prostate cancer is unknown. The benefits of screening procedures for prostate cancer using PSA, free PSA, and digital rectal exam include early diagnosis, decrease in health care costs of advanced stage cancer, and decrease in mortality. Risks of prostate cancer screening include minimal complications that can occur with venous blood draw such as hematoma, and false negative and false positive assay results. Medical decisions are not based on free PSA or the free/total PSA ratio alone. Total PSA, digital rectal exam, ultrasonography and other clinical signs and symptoms are used. A falsely low free PSA result (low %fPSA is associated with cancer) may lead to the clinical decision to perform unneeded prostate biopsy. Prostate biopsy is a relatively minor surgery and has minimal health risks associated with the procedure. A falsely high free PSA result (high %fPSA is associated with benign conditions) could lead to a medical decision to avoid prostate biopsy. Prostate cancer is a slow growing disease. It is routine clinical practice to perform subsequent and regular patient evaluations of clinical signs/symptoms, total PSA, free PSA and DRE. Therefore, the risks associated with this device are:

- The risk associated with venipuncture, and
- The risk that interpretation of a free PSA/total PSA ratio would subject the patient to unnecessary biopsy or deprive the patient of a medical treatment.

There is a substantial risk of an unnecessary biopsy for men without cancer when total PSA is between 4 and 10 ng/mL and DRE results are not suspicious for cancer (percentage of subjects without prostate cancer was approximately 64% in this clinical study).

The benefit of the device is the increased specificity offered by the use of the free PSA/total PSA ratio when used to discriminate prostate cancer from non-cancer in patients with total PSA between 4-10 ng/mL and DRE results not suspicious for cancer. With the free/total PSA ratio cutoff of 23%, 26.5% (from 22.6% up to 30.9%) of men without prostate cancer could be spared unnecessary biopsy and have 90.5 of cancers correctly identified. As a consequence, fewer men would be given unnecessary biopsies as well as possible medical complications (infection, bleeding, urinary retention, and hospitalization).

B. Safety Conclusions

- Information has not been provided on subjects within the potential target population who did not have biopsy. Therefore, there is no available method to assess if the sampled population is a random representative sample of the target population. Subjects of this clinical study can be different from the intended use population of subjects with total PSA values of 4.0 - 10.0 ng/mL and negative DRE because a decision about biopsy was already made based on other test results and risk factors. Therefore, the study results may be difficult to generalize.

- The most common safety concerns involve mis-interpretation or mis-application of test results. When a negative test result is obtained (cutoff =23%), approximately 84% (at least 77%) of subjects will be absent prostate cancer. There is a 16% risk of prostate cancer (at most 23%) when the test result is negative and therefore a subject could fail to undergo a necessary biopsy to detect a missed prostate cancer.

- When a positive test result is obtained, approximately 41% (at least 39%) of subjects will have prostate cancer. There is a 59% risk of being without cancer when the test result is positive and therefore a subject may undergo an unnecessary biopsy for a benign prostatic condition.

The short term medical risks of biopsy are usually minimal. Infections and bleeding from the biopsy site for a short period of time are often treatable (often no more than 2 weeks).

C. Effectiveness Conclusions

At a cutoff ratio of 23% free to total PSA, 91% of prostate cancer subjects (86% at its lower confidence interval) are detected by the assay. At this cutoff, 27% of subjects with benign prostatic disease (23% at its lower confidence interval) have negative assay results. The specificity of %free PSA is significantly higher than the specificity of total PSA in the range 4.0 to 10.0 ng/mL for %free PSA cutoffs between 22% and 26%.

The probability of finding prostate cancer is related to the lower values of %free PSA and with age in whole 10 year increments. The relationship is described by the following table:

Probability of Finding Prostate Cancer on Core Biopsy by Age			
% FPSA ratio	50-59	60-69	≥ 70
≤ 10	0.54 (21/39)	0.77 (30/39)	0.71 (15/21)
11-19	0.28 (36/127)	0.35 (59/167)	0.53 (38/72)
20-25	0.12 (3/26)	0.23 (15/65)	0.27 (9/34)
> 26	0 (0/6)	0.15 (6/39)	0.23 (10/44)

Among apparently normal healthy men, 95% have %free PSA values at or below 38% free PSA.

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.

Prostate cancer is often a slow growing disease. Medical decisions are not based on Free PSA or the Free/Total PSA ratio alone. Total PSA, digital rectal exam, ultrasonography and other clinical signs and symptoms are used. It is routine clinical practice to perform subsequent and regular patient evaluations of clinical signs/symptoms, total PSA, Free

PSA and DRE. A falsely low Free PSA result (low %Free PSA is associated with cancer) may lead to the clinical decision to perform unneeded prostate biopsy. A falsely high Free PSA result (high %Free PSA is associated with benign conditions) could lead to a medical decision to avoid prostate biopsy when cancer is present. Prostate biopsy is a relatively minor surgery and has minimal health risks associated with the procedure.

XII. CDRH DECISION

CDRH issued an approval order on October 8, 2009. The final conditions of approval cited in the approval order are described below.

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

XIII. 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: See approval order.