Alinity i Total PSA-22

Prepared by: Yusra Othman /Director/Supervisor-Chem **Date:** May/26/2024
signature/title

Reviewed by: Jordan Dillard /Instructor **Date:** June 26 2024
signature/title

Approved by: Sanford N. Bach, M.D /Chairman **Date:** June 28 2024
signature/title

BIENNIAL REVIEW:

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SUPERSEDES: Procedure titled _____

CAUTION: United States Federal Law restricts this device to sale and distribution by or on the order of a physician, or to a clinical laboratory; and use is restricted to, by, or on the order of a physician.

Instructions must be carefully followed. Reliability of assay results cannot be guaranteed if there are any deviations from these instructions.

WARNING: The concentration of total PSA in a given specimen, determined with assays from different manufacturers, can vary due to differences in assay methods and reagent specificity. The results reported by the laboratory to the physician must include the identity of the total PSA assay used. Values obtained with different assay methods cannot be used interchangeably. If, in the course of monitoring a patient, the assay method used for determining total PSA levels serially is changed, additional sequential testing should be

carried out. Before changing assays, the laboratory MUST confirm baseline values for patients being serially monitored. Note: % free PSA (FPSA) ratios must be calculated using Total PSA and Free PSA results both obtained on the Alinity i analyzer.

INTENDED USE

The Alinity i Total PSA assay is a chemiluminescent microparticle immunoassay (CMIA) used for the quantitative determination of total PSA (both free PSA and PSA complexed to alpha-1-antichymotrypsin) in human serum on the Alinity i analyzer:

1. As an aid in the detection of prostate cancer when used in conjunction with digital rectal exam (DRE) in men 50 years or older. Prostatic biopsy is required for diagnosis of cancer.
2. As an adjunctive test to aid in the management of prostate cancer patients.

SUMMARY AND EXPLANATION OF THE TEST

Prostate specific antigen (PSA), a member of the human kallikrein gene family, is a serine protease with chymotrypsin-like activity. The mature form of PSA is a single chain glycoprotein of 237 amino acids containing 7-8% carbohydrate as a single N-linked oligosaccharide side chain. PSA has a molecular weight of approximately 30 000 daltons.*1*, *2*, *3*, *4*

The major site of PSA production is the glandular epithelium of the prostate. PSA has also been found in breast cancers, salivary gland neoplasms, periurethral and anal glands, cells of the male urethra, breast milk, blood and urine.*1*, *5* PSA produced in the prostate is secreted into the seminal fluid in high concentrations. A major function of PSA is the proteolytic cleavage of gel-forming proteins in the seminal fluid, resulting in the liquefaction of the seminal gel and increased sperm mobility.*1* Low levels of PSA are found in the blood as a result of leakage of PSA from the prostate gland. Increasing levels of serum PSA are associated with prostatic pathology, including prostatitis, benign prostatic hyperplasia (BPH), and cancer of the prostate.*1*, *6*, *7*, *8*, *9*, *10*

PSA occurs in three major forms in blood. The major immunodetectable form is PSA complexed with the serine protease inhibitor, alpha-1-antichymotrypsin (PSA-ACT). Uncomplexed, or free PSA, is the other immunodetectable form of PSA in serum. The majority of free PSA in serum appears to be an inactive form that cannot complex with protease inhibitors and may be either a PSA zymogen or an enzymatically-inactive, cleaved form of PSA. Equimolar-response PSA assays have an equivalent response to both free PSA and PSA-ACT.*1* The Alinity i Total PSA assay is an equimolar assay. A third form of PSA, a complex with alpha-2-macroglobulin, is not detectable with current immunoassays for PSA due to the engulfment and subsequent masking of PSA epitopes by the alpha-2-macroglobulin molecule.*1*, *2*, *11*

Prostate cancer is the most frequently diagnosed cancer and the second leading cause of cancer deaths in men in the United States.*12* Early diagnosis of carcinoma of the prostate is hindered by the lack of symptoms in men with localized tumors. Therefore, early detection requires a simple, safe, and inexpensive test for the disease in asymptomatic men. The traditional method for detection of prostate cancer is the digital rectal examination (DRE).

However, only 30 to 40% of cancers detected by DRE screening are expected to be confined to the prostate.[13] The frequent finding of locally advanced prostate cancer in screened patients may be due to the inability of DRE to detect tumors of small volume that are most likely to be confined to the prostate. Since patients with small tumors are believed to have the best prognosis, it can be concluded that DRE has limited sensitivity in detecting those tumors with the greatest potential for cure.[14]

In a 1990 publication by Cooner et al., data was presented regarding the clinical use of other diagnostic modalities such as prostate ultrasonography and serum prostate specific antigen for early detection of prostate cancer. This study found that there was a significant increase in predictability for cancer when the DRE and PSA tests were abnormal.[15] Several other studies have shown that the measurement of serum PSA concentrations offers several advantages in the early detection of prostate cancer. The procedure is more acceptable to patients, the result is objective and quantitative, and is independent of the examiners skill. In several recent studies of healthy men 50 years or older, serum PSA levels had the greatest ability to predict prostate cancer. These studies concluded that not only is serum PSA measurement a useful addition to rectal examination and ultrasonography in the detection of prostate cancer, but that it is also the most accurate of the three tests for this purpose.[16, 17] In January 1992, the American Urological Association endorsed annual examination with DRE and PSA, for early detection of prostate cancer, beginning at age 50.[18] This was reaffirmed by the American Cancer Society in November 1992.[19] The combined use of DRE and PSA has been shown to result in an increased detection of early stage prostate cancer; however, the benefit of early detection on patient outcome has not been proven and is the subject of ongoing clinical trials.[7, 8, 9, 10, 15, 16, 17, 20, 21]

PSA testing can have significant value in detecting metastatic or persistent disease in patients following surgical or medical treatment of prostate cancer. Persistent elevation of PSA following treatment, or an increase in a post-treatment PSA level is indicative of recurrent or residual disease. PSA testing is widely accepted as an adjunctive test in the management of prostate cancer patients.[6, 7, 8, 9, 10]

BIOLOGICAL PRINCIPLES OF THE PROCEDURE

This assay is a two-step immunoassay for the quantitative determination of total PSA (both free PSA and PSA complexed to alpha-1-antichymotrypsin) in human serum using chemiluminescent microparticle immunoassay (CMIA) technology.

Sample and anti-PSA coated paramagnetic microparticles are combined and incubated. The PSA present in the sample binds to the anti-PSA coated microparticles. The mixture is washed. Anti-PSA acridinium-labeled conjugate is added to create a reaction mixture and incubated. Following a wash cycle, Pre-Trigger and Trigger Solutions are added.

The resulting chemiluminescent reaction is measured as relative light units (RLUs). There is a direct relationship between the amount of total PSA in the sample and the RLUs detected by the system optics.

For additional information on system and assay technology, refer to the Alinity ci-series Operations Manual, Section 3.

REAGENTS

Kit Contents

Alinity i Total PSA Reagent Kit 07P92

Volumes (mL) listed in the table below indicate the volume per cartridge.

REF	07P9221	07P9231
Tests per cartridge	100	600
Number of cartridges per kit	2	2
Tests per kit	200	1200
MICROPARTICLES	6.6 mL	32.1 mL
CONJUGATE	6.1 mL	31.6 mL

MICROPARTICLES Anti-PSA (mouse, monoclonal) coated microparticles in TRIS buffer with protein (bovine) stabilizer. Minimum concentration: 0.1% solids. Preservatives: antimicrobial agents.

CONJUGATE Anti-PSA (mouse, monoclonal) acridinium-labeled conjugate in MES buffer with protein (bovine) stabilizer. Minimum concentration: 10 ng/mL. Preservatives: antimicrobial agents.

Warnings and Precautions

- IVD
- For *In Vitro* Diagnostic Use
- Rx ONLY

Safety Precautions

CAUTION: This product requires the handling of human specimens. It is recommended that all human-sourced materials be considered potentially infectious and handled in accordance with the OSHA Standard on Bloodborne Pathogens. Biosafety Level 2 or other appropriate biosafety practices should be used for materials that contain or are suspected of containing infectious agents.[22], [23], [24], [25]

Safety Data Sheets are available at www.abbottdiagnostics.com or/and SDS folder.

For a detailed discussion of safety precautions during system operation, **refer to the Alinity ci-series Operations Manual, Section 8.**

Reagent Handling

Upon receipt, gently invert the unopened reagent kit by rotating it over and back for a full 180 degrees, 5 times with green label stripe facing up and then 5 times with green label stripe facing down. This ensures that liquid covers all sides of the bottles within the cartridges. During reagent shipment, microparticles can settle on the reagent septum.

·Place a check in the square on the reagent kit to indicate to others that the inversions have

been completed.

·After mixing, place reagent cartridges in an upright position for 1 hour before use to allow bubbles that may have formed to dissipate.

·If a reagent cartridge is dropped, place in an upright position for 1 hour before use to allow bubbles that may have formed to dissipate.

·Reagents are susceptible to the formation of foam and bubbles. Bubbles may interfere with the detection of the reagent level in the cartridge and cause insufficient reagent aspiration that may adversely affect results.

For a detailed discussion of reagent handling precautions during system operation, **refer to the Alinity ci-series Operations Manual, Section 7.**

Reagent Storage

	Storage Temperature	Maximum Storage Time	Additional Storage Instructions
Unopened	2 to 8°C	Until expiration date	Store in upright position. If cartridge does not remain upright, gently invert the cartridge 10 times and place in an upright position for 1 hour before use.
Onboard	System Temperature	30 days	
Opened	2 to 8°C	Until expiration date	Store in upright position. If cartridge does not remain upright during storage, discard the cartridge. Do not reuse original reagent caps or replacement caps due to the risk of contamination and the potential to compromise reagent performance.

Reagents may be stored on or off the system. If removed from the system, store reagents with new replacement caps in an upright position at 2 to 8°C. For reagents stored off the system, it is recommended that they be stored in their original trays or boxes to ensure they remain upright.

For information on unloading reagents, **refer to the Alinity ci-series Operations Manual, Section 5.**

Indications of Reagent Deterioration

Deterioration of the reagents may be indicated when:

a calibration error occurs

a control value is out of the specified range

Associated test results are invalid, and samples must be retested. Assay recalibration may be necessary.

For troubleshooting information, **refer to the Alinity ci-series Operations Manual, Section 10.**

INSTRUMENT PROCEDURE

The Alinity i Total PSA assay file must be installed on the Alinity i analyzer prior to performing the assay.

For detailed information on assay file installation and viewing and editing assay parameters, **refer to the Alinity ci-series Operations Manual, Section 2.**

For information on printing assay parameters, **refer to the Alinity ci-series Operations Manual, Section 5.**

For a detailed description of system procedures**, refer to the Alinity ci-series Operations Manual.**

Alternate Result Units

Edit assay parameter "Result Units" to select an alternate unit.

Conversion formula:

(Concentration in Default result unit) x (Conversion factor) =
(Concentration in Alternate result unit)

Default Result Unit	Conversion Factor	Alternate Result Unit
ng/mL	1.0	µg/L

SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS

Specimen Types

The specimen type listed below was verified for use with this assay on the ARCHITECT i System.

Specimen Type	Collection Tubes
Serum	Serum Serum separator

Specimen Conditions

Do not use:

·grossly hemolyzed specimens

·specimens with obvious microbial contamination

·For accurate results, serum specimens should be free of fibrin, red blood cells, and other particulate matter. Serum specimens from patients receiving anticoagulant or thrombolytic therapy may contain fibrin due to incomplete clot formation.

·It is recommended to obtain specimens for PSA testing prior to procedures involving manipulation of the prostate.

·To prevent cross contamination, use of disposable pipettes or pipette tips is recommended.

Preparation for Analysis

·Follow the tube manufacturer's processing instructions for collection tubes. Gravity separation is not sufficient for specimen preparation.

·Insufficient processing of sample, or disruption of the sample during transportation may cause depressed results.

·Ensure that complete clot formation in serum specimens has taken place prior to centrifugation. Some specimens, especially those from patients receiving anticoagulant or thrombolytic therapy may exhibit increased clotting time. If specimens are centrifuged before a complete clot forms, the presence of fibrin or particulate matter may cause erroneous results. Centrifuge specimens containing fibrin, red blood cells, or particulate matter. Note that interfering levels of fibrin may be present in samples that do not have obvious or visible particulate matter.

·If proper specimen collection and preparation cannot be verified, or if samples have been disrupted due to transportation or sample handling, an additional centrifugation step is recommended. Centrifugation conditions should be sufficient to remove particulate matter. Aliquots poured versus pipetted from specimen tube types that do not include serum separators are at higher risk of including particulates and generating depressed results.

·Failure to follow these instructions may result in depressed specimen results.

·Specimens should be free of bubbles. Remove bubbles with an applicator stick before analysis. Use a new applicator stick for each specimen to prevent cross contamination.

To ensure consistency in results, recentrifuge specimens prior to testing if

·they contain fibrin, red blood cells, or other particulate matter.

NOTE: If fibrin, red blood cells, or other particulate matter are observed, mix by low speed vortex or by inverting 10 times prior to recentrifugation.

Prepare frozen specimens as follows:

·Frozen specimens must be completely thawed before mixing.

·Mix thawed specimens thoroughly by low speed vortex or by inverting 10 times.

·Visually inspect the specimens. If layering or stratification is observed, mix until specimens are visibly homogeneous.

·If specimens are not mixed thoroughly, inconsistent results may be obtained.

·Recentrifuge specimens.

Recentrifugation of Specimens

·Transfer specimens to a centrifuge tube and centrifuge from 50 000 to 100 000 g-minutes.

·Examples of acceptable time and force ranges that meet this criterion are listed in the table below.

Centrifugation time using alternate RCF values can be calculated using the following formula:

$$\text{Centrifugation time (minutes)} = \frac{50\,000 \text{ g-minutes}}{\text{RCF}}$$

Recentrifugation Time (Minutes)	RCF (x g)	g-Minutes
10	5000 - 10 000	50 000 - 100 000

$RCF = 1.12 \times r_{max} (rpm/1000)^2$

RCF - The relative centrifugal force generated during centrifugation.

rpm - The revolutions per minute of the rotor on which the specimens are being spun (usually the digital readout on the centrifuge will indicate the rpm).

Centrifugation Time - The time should be measured from the time the rotor reaches the required RCF or rpm to the time it begins decelerating.

r_{max} - Radius of the rotor in millimeters. NOTE: If custom tube adapters (i.e., adapters not defined by the centrifuge manufacturer) are used, then the radius (r_{max}) should be manually measured in millimeters and the RCF calculated.

g-minutes - The unit of measure for the product of RCF (× g) and centrifugation time (minutes).

·Transfer clarified specimen to a sample cup or secondary tube for testing. For centrifuged specimens with a lipid layer, transfer only the clarified specimen and not the lipemic material.

Specimen Storage

Specimen storage conditions were verified on the ARCHITECT i System.

Specimen Type	Temperature	Maximum Storage Time	Special Instructions
Serum	2 to 8°C	24 hours	Samples which may be tested for free PSA should be removed from the clot within 3 hours.

If testing will be delayed more than 24 hours, specimens should be removed from the clot or serum separator and stored frozen at -20°C or colder.[26], [27]

Avoid multiple freeze/thaw cycles.

Specimen Shipping

Package and label specimens in compliance with applicable state, federal, and international regulations covering the transport of clinical specimens and infectious substances.

Do not exceed the storage limitations listed above.

PROCEDURE

Materials Provided

07P92 Alinity i Total PSA Reagent Kit

Materials Required but not Provided

·Alinity i Total PSA assay file

·07P9201 Alinity i Total PSA Calibrators

·07P9210 Alinity i Total PSA Controls or other commercially available controls

·09P1540 Alinity i Multi-Assay Manual Diluent

·Alinity Pre-Trigger Solution

·Alinity Trigger Solution

·Alinity i-series Concentrated Wash Buffer

For information on materials required for operation of the instrument, refer to the Alinity ci-series Operations Manual, Section 1.

For information on materials required for maintenance procedures, refer to the Alinity ci-series Operations Manual, Section 9.

Assay Procedure

For a detailed description of how to run an assay, **refer to the Alinity ci-series Operations Manual, Section 5.**

·If using primary or aliquot tubes, refer to the Alinity ci-series Operations Manual, Section 4 to ensure sufficient specimen is present.

·To minimize the effects of evaporation, verify adequate sample cup volume is present prior to running the test.

Maximum number of replicates sampled from the same sample cup: 10

Priority:

·Sample volume for first test: 100 µL

·Sample volume for each additional test from same sample cup: 50 µL

≤ 3 hours on the reagent and sample manager:

·Sample volume for first test: 150 µL

·Sample volume for each additional test from same sample cup: 50 µL

> 3 hours on the reagent and sample manager:

·Replace with a fresh aliquot of sample.

·Refer to the Alinity i Total PSA calibrator package insert and/or Alinity i Total PSA control package insert for preparation and usage.

·For general operating procedures, refer to the Alinity ci-series Operations Manual, Section 5.

·For optimal performance, it is important to perform routine maintenance as described in the Alinity ci-series Operations Manual, Section 9. Perform maintenance more frequently when required by laboratory procedures.

Sample Dilution Procedures

Samples with a total PSA value exceeding 100 ng/mL (100 µg/L) are flagged with the code "> 100.000 ng/mL" (">100.000 µg/L") and may be diluted with either the Automated Dilution Protocol or the Manual Dilution Procedure.

Automated Dilution Protocol

The system performs a **1:10** dilution of the sample and automatically calculates the concentration by multiplying the result by the dilution factor. Dilutions other than 1:10 should be done manually.

Manual Dilution Procedure

Example dilution: 1:20

Add 50 µL of the sample to 950 µL of Alinity i Multi-Assay Manual Diluent.

The operator must enter the dilution factor in the Specimen or Control tab of the Create Order screen. The system will use this dilution factor to automatically calculate the concentration of the sample and report the result. The result should be > 0.4 ng/mL (> 0.4 µg/L) before the dilution factor is applied.

For detailed information on ordering dilutions**, refer to the Alinity ci-series Operations Manual, Section 5.**

Calibration

For instructions on performing a calibration, **refer to the Alinity ci-series Operations Manual, Section 5.**

Each assay control must be tested to evaluate the assay calibration.

Once a calibration is accepted and stored, all subsequent samples may be tested without further calibration unless:

- A reagent kit with a new lot number is used.
- Controls are out of range.

Quality Control Procedures

The recommended control requirement for the Alinity i Total PSA assay is that a single sample of each control level be tested once every day testing performed.

Verification of Assay Claims

For protocols to verify package insert claims, refer to Verification of Assay Claims in the Alinity ci-series Operations Manual.

RESULTS

Calculation

The Alinity i Total PSA assay utilizes a 4 Parameter Logistic Curve fit data reduction method (4PLC, Y-weighted) to generate a calibration and results.

For information on alternate result units, refer to the INSTRUMENT PROCEDURE, Alternate Result Units section of this package insert.

Flags

Some results may contain information in the Flags field. For a description of the flags that may appear in this field, refer to the Alinity ci-series Operations Manual, Section 5.

Measuring Interval

Measuring interval is defined as the range of values in ng/mL (µg/L) which meets the limits of acceptable performance for linearity, imprecision, and bias.

The measuring interval of the Alinity i Total PSA assay is **0.1 to 100.0** ng/mL (0.1 to 100.0 µg/L).

LIMITATIONS OF THE PROCEDURE

·Specimens from patients who have received preparation of mouse monoclonal antibodies for diagnosis or therapy may contain human anti-mouse antibodies (HAMA). Such specimens may show either falsely elevated or depressed values when tested with assay kits which employ mouse monoclonal antibodies.[28, 29] Alinity i Total PSA reagents contain a component that reduces the effect of HAMA reactive specimens. Additional clinical or diagnostic information may be required to determine patient status.

·Heterophilic antibodies in human serum can react with reagent immunoglobulins, interfering with *in vitro* immunoassays. Patients routinely exposed to animals or to animal serum products can be prone to this interference, and anomalous values may be observed. Additional information may be required for diagnosis.[30]

·The concentration of PSA in a given specimen, determined with assays from different manufacturers, can vary due to differences in assay methods, calibration, and reagent specificity.[1, 31, 32]

·Quality control samples may be produced by introducing seminal fluid PSA into a human serum matrix. PSA in serum and seminal fluid may exist in different forms. The concentration of PSA in these controls, determined with assays from different manufacturers, can vary due to differences in assay methods, calibration, reagent specificity, and the form of PSA that is present; therefore, it is important to use assay-specific values to evaluate control results.

·Hormonal therapy may affect PSA expression; therefore, a low PSA level after any treatment that includes hormonal therapy may not adequately reflect the presence of residual or recurrent disease.[33]

·In most instances, specimens obtained from patients immediately following digital rectal examination show no clinically significant increases in PSA levels.[34] However, prostatic massage, ultrasonography, and needle biopsy may cause clinically significant elevations.[35] PSA levels may also be increased following ejaculation.[36]

·Active free PSA in the serum at the time of blood sampling can continue to complex with serum protease inhibitors, especially alpha-2-macroglobulin, resulting in a rapid decrease in PSA levels of the active form of free PSA.[37]

·Serum PSA concentrations should not be interpreted as absolute evidence for the presence or absence of prostate cancer. Elevated concentrations of PSA may be observed in the serum of patients with benign prostatic hyperplasia or other nonmalignant disorders as well as in prostate cancer. Furthermore, low PSA concentrations are not always indicative of the absence of cancer. The PSA value should be used in conjunction with information available from clinical evaluation and other diagnostic procedures such as DRE. Some early cases of prostate cancer will not be detected by PSA testing; the same is true for DRE. Prostatic biopsy is required for the diagnosis of cancer.

EXPECTED VALUES

These studies were performed on the ARCHITECT i System.

Representative performance data are provided in this section. Results obtained in individual laboratories may vary.

It is recommended that each laboratory determine its own reference range based upon its particular locale and population characteristics.

The distribution of ARCHITECT Total PSA values determined in 2287 specimens is shown in the following table.

Distribution of ARCHITECT Total PSA Values

			Percent (%)				
		Number of Subjects	**0 - 4.0 (ng/mL)**	**> 4.0 - 10 (ng/mL)**	**> 10 - 30 (ng/mL)**	**> 30 - 60 (ng/mL)**	**> 60 (ng/mL)**
Apparently Healthy Subjects	Females	296	100.0	0.0	0.0	0.0	0.0
	Males Ages 40 to 49	99	100.0	0.0	0.0	0.0	0.0
	Males Ages 50 to 59	120	97.5	2.5	0.0	0.0	0.0
	Males Ages 60 to 69	123	93.5	6.5	0.0	0.0	0.0
	Males Ages 70 to 79	124	91.9	7.3	0.8	0.0	0.0
Nonmalignant Disease	BPH	352	42.6	42.3	12.8	1.1	1.1
	Cirrhosis	89	94.4	3.4	1.1	0.0	1.1
	Genitourinary	151	90.7	7.3	1.3	0.7	0.0
	Prostatitis	142	46.5	40.1	11.3	1.4	0.7
	Renal	140	90.0	5.7	2.9	1.4	0.0
Malignant Disease	Prostate Stage A	94	46.8	30.9	17.0	1.1	4.3
	Prostate Stage B	166	30.1	44.0	23.5	0.6	1.8
	Prostate Stage C	141	26.2	22.7	29.1	12.8	9.2
	Prostate Stage D	95	15.8	12.6	32.6	10.5	28.4
	Genitourinary	155	92.9	3.9	1.9	0.6	0.6

In this study, 95.5% of the specimens from apparently healthy male subjects (n=466) had values of 4.0 ng/mL or less.

0-49 years < 2.5

49-150 < 4.0

The malignant disease portion of the distribution table is derived primarily from carcinoma patients representing both active (clinical evidence of disease progression) and inactive (no clinical evidence of disease progression) disease states. When changing PSA assay methods in the course of monitoring a patient, additional sequential testing should be carried out to confirm baseline values.

EXPECTED VALUES FOR DETECTION OF PROSTATE CANCER

A prospective study was conducted at seven clinical sites to demonstrate the usefulness of PSA in the detection of prostate cancer when used in conjunction with DRE. All clinical data presented supporting the detection claim were generated using the ARCHITECT i System and ARCHITECT Total PSA assay reagents. A total of 531 men 50 years of age or older participated in the study. All subjects were biopsied based on an initial elevated PSA value and/or suspicious DRE result. A distribution of the ARCHITECT Total PSA results is presented in the following table:

Distribution of Results from ARCHITECT Total PSA

	PSA ≤ 4.0	**PSA > 4.0**	**Total**
DRE-[b]	32	319	351
	6.0%	60.1%	66.1%
DRE+[a]	96	84	180
	18.1%	15.8%	33.9%
Total	128	403	531
	24.1%	75.9%	100.0%

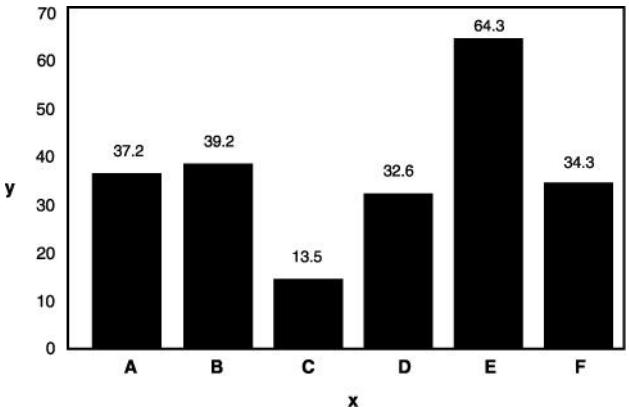
NOTE: 499 patients tested positive by DRE and/or PSA.

[a] DRE+: Digital Rectal Examination (Suspicious for cancer)

[b] DRE-: Digital Rectal Examination (Not suspicious for cancer)

The positive predictive values for various combinations of DRE and PSA are presented graphically in the figure below and table below.

Positive Predictive Values



x	Detection Method	C	PSA ≤ 4.0 and DRE+
y	Positive Predictive Value (%)	D	PSA > 4.0 and DRE-
A	DRE+	E	PSA > 4.0 and DRE+
B	PSA > 4.0	F	PSA > 4.0 or DRE+

Positive Predictive Values

Detection Method	Positive Predictive Value (%)*	Number of Subjects with Cancer/Number of Subjects Suspicious for Cancer
DRE+	37.2 (30.1 - 44.7)	67/180
PSA > 4.0	39.2 (34.4 - 44.2)	158/403
PSA ≤ 4.0 and DRE+	13.5 (7.4 - 22.0)	13/96
PSA > 4.0 and DRE-	32.6 (27.5 - 38.0)	104/319
PSA > 4.0 and DRE+	64.3 (53.1 - 74.4)	54/84
PSA > 4.0 or DRE+	34.3 (30.1 - 38.6)	171/499

* 95% Confidence Interval (Lower Limit - Upper Limit)

Cancers were detected in 177 of the 531 subjects. The overall cancer detection rate was 96.6% (171/177) when at least one test was suspicious, 30.5% (54/177) when both tests were suspicious, 58.8% (104/177) for PSA alone, and 7.3% (13/177) for DRE alone.

SPECIFIC PERFORMANCE CHARACTERISTICS

Representative performance data are provided in this section. Results obtained in individual laboratories may vary.

The Alinity i analyzer and the ARCHITECT i System utilize the same reagents and sample/reagent ratios.

Unless otherwise specified, all studies were performed on the Alinity i analyzer.

Precision

Within-Laboratory Precision

A study was performed based on guidance from CLSI EP05-A2.[38] Testing was conducted using 3 lots of the Alinity i Total PSA Reagent Kit, 3 lots of the Alinity i Total PSA Calibrators, and 3 lots of the Alinity i Total PSA Controls and 1 instrument. Three controls, 5 female serum based panels and 5 native male serum based panels were assayed in a minimum of 2 replicates at 2 separate times per day on 20 different days.

Sample	Lot	n	Mean ng/mL (µg/L)	Within-Run		Within-Laboratory (Total)[a]	
				SD	%CV	SD	%CV
Low Control	1	118	0.509	0.0157	3.1	0.0166	3.3
	2	120	0.470	0.0160	3.4	0.0191	4.1
	3	118	0.503	0.0178	3.5	0.0190	3.8
Medium Control	1	120	3.945	0.1226	3.1	0.1334	3.4
	2	115	3.744	0.1137	3.0	0.1377	3.7
	3	120	3.988	0.1544	3.9	0.1771	4.4
High Control	1	116	23.889	1.0342	4.3	1.1347	4.7
	2	120	21.618	0.7143	3.3	0.8565	4.0
	3	120	23.520	0.9696	4.1	1.0561	4.5
Panel 1 (Female Serum)	1	117	0.110	0.0039	3.5	0.0041	3.8
	2	114	0.102	0.0039	3.8	0.0044	4.3
	3	118	0.110	0.0034	3.1	0.0041	3.7
Panel 2 (Female Serum)	1	120	3.953	0.1398	3.5	0.1434	3.6
	2	120	3.747	0.1222	3.3	0.1588	4.2
	3	120	4.004	0.1404	3.5	0.1404	3.5
Panel 3 (Female	1	117	45.891	2.6965	5.9	2.7772	6.1
	2	114	40.595	1.5610	3.8	1.8549	4.6

Sample	Lot	n	Mean ng/mL (µg/L)	Within-Run		Within-Laboratory (Total)[a]	
				SD	%CV	SD	%CV
Serum)	3	116	45.740	1.8084	4.0	2.4632	5.4
Panel 4 (Female Serum)	1	120	66.227	3.4698	5.2	3.9993	6.0
	2	120	56.048	2.1764	3.9	2.6346	4.7
	3	120	65.406	3.3521	5.1	3.5662	5.5
Panel 5 (Female Serum)	1	120	85.380	4.9120	5.8	5.2871	6.2
	2	120	71.555	3.7401	5.2	3.7401	5.2
	3	120	84.299	4.6079	5.5	4.6979	5.6
Panel 1 (Native Male Serum)	1	80	0.140	0.0049	3.5	0.0058	4.1
	2	80	0.128	0.0048	3.7	0.0049	3.8
	3	80	0.137	0.0048	3.5	0.0055	4.1
Panel 2 (Native Male Serum)	1	80	4.042	0.1579	3.9	0.1579	3.9
	2	80	3.772	0.1307	3.5	0.1420	3.8
	3	80	3.948	0.1444	3.7	0.1524	3.9
Panel 3 (Native Male Serum)	1	80	45.109	1.7723	3.9	1.9417	4.3
	2	80	40.678	1.6046	3.9	1.8554	4.6
	3	80	45.365	1.6851	3.7	1.8379	4.1
Panel 4 (Native Male Serum)	1	80	64.872	3.1900	4.9	3.2813	5.1
	2	80	59.009	2.3180	3.9	2.8538	4.8
	3	80	66.536	2.3948	3.6	3.2255	4.8
Panel 5 (Native Male Serum)	1	80	85.440	4.0239	4.7	4.5581	5.3
	2	80	77.425	3.3968	4.4	3.7911	4.9
	3	80	88.759	3.9546	4.5	4.4510	5.0

[a]Includes within-run, between-run, and between-day variability.

System Reproducibility

A study was performed based on guidance from CLSI EP05-A2 and CLSI EP15-A2.[38, 39]

Testing was conducted at 3 clinical sites using 1 lot each of the Alinity i Total PSA Reagent Kit, the Alinity i Total PSA Calibrators, and the Alinity i Total PSA Controls and 1 instrument per site. Three controls and 5 native male serum based panels were assayed in replicates of 4 at 2 separate times per day for 5 days.

Sample	n	Grand Mean ng/mL (µg/L)	Within-Run		Within-Day[a]		Within-Laboratory Precision (Total)[b]		Precision with Additional Component of Between-Site (Overall)	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV
Low Control	120	0.454	0.0137	3.0	0.0160	3.5	0.0160	3.5	0.0160	3.5
Medium Control	120	3.664	0.1114	3.0	0.1180	3.2	0.1180	3.2	0.1234	3.4
High Control	120	22.778	0.9467	4.2	0.9930	4.4	0.9930	4.4	1.0293	4.5
Panel 1	120	0.126	0.0035	2.8	0.0040	3.2	0.0044	3.5	0.0044	3.5
Panel 2	120	3.672	0.1178	3.2	0.1236	3.4	0.1236	3.4	0.1276	3.5
Panel 3	120	42.534	1.9233	4.5	1.9233	4.5	1.9306	4.5	2.3419	5.5
Panel 4	120	64.141	2.6543	4.1	2.7173	4.2	3.1586	4.9	3.6217	5.6
Panel 5	114	86.522	4.7133	5.4	4.8850	5.6	5.3139	6.1	6.2284	7.2

[a] Includes within-run and between-run variance variability.

[b]Includes within-run, between-run, and between-day variability.

Lower Limits of Measurement

A study was performed based on guidance from CLSI EP17-A2. Testing was conducted using 3 lots of the Alinity i Total PSA Reagent Kit on each of 2 instruments over a minimum of 3 days. The Limit of Blank (LoB), Limit of Detection (LoD), and Limit of Quantitation (LoQ) values are summarized below. These representative data support the lower limit of the measuring interval. *40*

	ng/mL	µg/L
LoB[a]	0.001	0.001
LoD[b]	0.003	0.003
LoQ[c,d]	0.100	0.100

[a]The LoB represents the 95th percentile from n ≥ 60 replicates of zero-analyte samples.

[b]The LoD represents the lowest concentration at which the analyte can be detected with 95% probability based on n ≥ 60 replicates of low-analyte level samples.

[c] The LoQ is defined as the lowest concentration at which the maximum allowable precision of 15 %CV was met and was determined from n ≥ 60 replicates of low-analyte level samples.

[d]This value represents the observed LoQ on the ARCHITECT System. The LoQ observed on the Alinity i analyzer supports this LoQ.

Linearity

A study was performed based on guidance from CLSI EP06-A.*41*

This assay is linear across the measuring interval of **0.1 to 100.0 ng/mL** (0.1 to 100.0 µg/L).

Comparison Between the Alinity i Analyzer and the ARCHITECT i2000SR System

The comparison between the Alinity i analyzer and the ARCHITECT i2000SR System was evaluated with 154 serum sample results using 1 lot each of the Alinity i Total PSA Reagent Kit, Alinity i Total PSA Calibrators, and Alinity i Total PSA Controls. Testing of each sample was performed on 1 Alinity i analyzer at each of 3 clinical testing sites and on 1 ARCHITECT i2000SR System at each of 3 clinical testing sites.

Regression

The panel members were evaluated using the weighted Deming regression method. The data are summarized in the following table.

	Concentration Range ng/mL (µg/L)		Correlation Coefficient (r)					
Sample Type	**Alinity i**	**ARCHITECT i2000SR**	**r**	**95% CI[a]**	**Intercept**	**95% CI[a]**	**Slope**	**95% CI[a]**
Serum	0.103 - 78.173	0.104 - 87.393	0.998	(0.997, 0.998)	-0.00	(-0.00, 0.00)	0.97	(0.96, 0.97)

[a] 95% CI = Confidence Interval

Carryover

No detectable carryover (≤ 0.008 ng/mL) was observed when a high PSA sample ranging from 21 361 to 22 422 ng/mL was assayed.

Analytical Specificity

This study was performed on the ARCHITECT i System.

The analytical specificity of the ARCHITECT Total PSA assay was determined by testing sera containing the following compounds. These compounds showed less than or equal to 10% interference in the ARCHITECT Total PSA assay at the levels indicated.

INTERFERING SUBSTANCES

Test Compound	Concentration
Bilirubin	20 mg/dL
Hemoglobin	500 mg/dL
Total Protein	2.0 g/dL & 12.0 g/dL
Prostatic Acid Phosphatase	1000 ng/mL
Triglycerides	3000 mg/dL
Hytrin	10 µg/mL
Proscar	25 µg/mL
Flomax	1 µg/mL

CHEMOTHERAPEUTIC AGENTS

Test Compound	Concentration
Cyclophosphamide	700 µg/mL
Diethylstilbestrol	2 µg/mL
Doxorubicin-HCl	16 µg/mL
Estramustine Phosphate	200 µg/mL
Flutamide	10 µg/mL
Goserelin Acetate	100 ng/mL
Lupron	100 µg/mL
Megestrol Acetate	90 µg/mL
Methotrexate	30 µg/mL

High Dose Hook

This study was performed on the ARCHITECT i System.

High dose hook is a phenomenon whereby very high level specimens may read within the dynamic range of the assay. For the ARCHITECT Total PSA assay, no high dose hook effect was observed when samples containing up to approximately 48 000 ng/mL of PSA were assayed.

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