Elecsys free PSA

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REF		Σ	SYSTEM
09744908190	09744908501	100	**cobas e** 402 **cobas e** 801

English

For use in the USA only

System information

Short name	ACN (application code number)
FPSA	10188

Caution

The Elecsys free PSA immunoassay should be used only with the Elecsys total PSA immunoassay to calculate the ratio (% fPSA) of free PSA (fPSA) to total PSA (tPSA). Use of another manufacturer's total PSA assay may result in an inappropriate population of patients selected for fPSA testing; and significantly different fPSA to tPSA ratios, cutoffs and prostate cancer probabilities than represented in the "Expected values" section of this Method Sheet. Ratios must be calculated using tPSA and fPSA results both obtained on either the **cobas e** 402 or the **cobas e** 801 immunoassay analyzers.
The measured fPSA value of a patient's sample can vary depending on the testing procedure used. The laboratory finding must therefore always contain a statement on the fPSA assay method used. Free PSA values determined on patient samples by differing testing procedures cannot be directly compared with one another and could be the cause of erroneous medical interpretations.

Intended use

Immunoassay for the in vitro quantitative determination of free prostate-specific antigen in human serum and plasma.
This immunoassay is indicated for measurement of fPSA in conjunction with the Elecsys total PSA assay to develop a ratio (% fPSA) of fPSA to tPSA. This ratio is useful when used in conjunction with the Elecsys total PSA test as an aid in distinguishing prostate cancer from benign prostatic conditions in men age 50 years or older who have a digital rectal examination (DRE) that is not suspicious for prostate cancer and an Elecsys total PSA value in the range 4.00 ng/mL to 10.0 ng/mL. Prostate biopsy is required for the diagnosis of prostate cancer.

The **e**lectro**c**hemi**l**uminescence **i**mmuno**a**ssay "ECLIA" is intended for use on **cobas e** immunoassay analyzers.

Summary

Prostate-specific antigen (PSA) is a glycoprotein (molecular weight 30000-34000 daltons) having a close structural relationship to glandular kallikrein.
It has the function of a serine protease.[1,2]
The proteolytic activity of PSA in blood is inhibited by the irreversible formation of complexes with proteinase inhibitors such as alpha-1-antichymotrypsin, alpha-2-macroglobulin and other acute phase proteins.[3,4] In addition to being present in these complexes, PSA is also present in blood in the free form, but is proteolytically inactive.[5,6]
PSA tests lack sufficient sensitivity and specificity to be considered ideal or absolutely diagnostic for screening or early detection because PSA is not specific for prostate cancer.[7] PSA is organ specific, being produced primarily by prostatic secretory epithelium, but has long been known to be elevated in non-malignant conditions such as benign prostatic hyperplasia (BPH). A number of studies have found that the % free PSA was significantly lower in patients having prostate cancer than those with benign disease or normal controls.[8,9] The ratio fPSA/tPSA has been demonstrated to improve the sensitivity and specificity in patients with tPSA values in the "gray zone" of 4.00-10.0 ng/mL.[10]
An equimolar tPSA determination is the prerequisite for reliable ratios. In patients receiving therapy, particularly hormone withdrawal therapy, the fPSA/tPSA ratio cannot be utilized to differentiate prostate hyperplasia from cancer of the prostate. Combining tests from different manufacturers to determine tPSA and fPSA can produce erroneous values, since total PSA tests may be standardized by differing methods or detect free PSA to differing degrees.

Test principle

Sandwich principle. Total duration of assay: 18 minutes.

- 1st incubation: 12 µL of sample, a biotinylated monoclonal PSA-specific antibody, and a monoclonal PSA-specific antibody labeled with a ruthenium complex[a)] react to form a sandwich complex.
- 2nd incubation: After addition of streptavidin-coated microparticles, the complex becomes bound to the solid phase via interaction of biotin and streptavidin.
- The reaction mixture is aspirated into the measuring cell where the microparticles are magnetically captured onto the surface of the electrode. Unbound substances are then removed with ProCell II M. Application of a voltage to the electrode then induces chemiluminescent emission which is measured by a photomultiplier.
- Results are determined via a calibration curve which is instrument-specifically generated by 2-point calibration and a master curve provided via the **cobas** link.

a) Tris(2,2'-bipyridyl)ruthenium(II)-complex ($Ru(bpy)_3^{2+}$)

Reagents - working solutions

The **cobas e** pack is labeled as FPSA.

M Streptavidin-coated microparticles, 1 bottle, 6.4 mL:
Streptavidin-coated microparticles 0.72 mg/mL; preservative.

R1 Anti-PSA-Ab~biotin, 1 bottle, 9.0 mL:
Biotinylated monoclonal anti-PSA antibodies (mouse) 2 mg/L; phosphate buffer 100 mmol/L, pH 7.4; preservative.

R2 Anti-PSA-Ab~$Ru(bpy)_3^{2+}$, 1 bottle, 8.2 mL:
Monoclonal anti-PSA antibodies (mouse) labeled with ruthenium complex 1.0 mg/L; Biotin scavenger antibody 0.5 mg/mL; phosphate buffer 100 mmol/L, pH 7.4; preservative.

Precautions and warnings

For in vitro diagnostic use for healthcare professionals. Exercise the normal precautions required for handling all laboratory reagents.

Infectious or microbial waste:
Warning: handle waste as potentially biohazardous material. Dispose of waste according to accepted laboratory instructions and procedures.

Environmental hazards:
Apply all relevant local disposal regulations to determine the safe disposal.

Safety data sheet available for professional user on request.

This kit contains components classified as follows in accordance with the Regulation (EC) No. 1272/2008:

Warning

H317 May cause an allergic skin reaction.

Prevention:

P261 Avoid breathing mist or vapours.

P272 Contaminated work clothing should not be allowed out of the workplace.

P280 Wear protective gloves.

Response:

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P333 + P313 If skin irritation or rash occurs: Get medical advice/attention.

P362 + P364 Take off contaminated clothing and wash it before reuse.

Disposal:

P501 Dispose of contents/container to an approved waste disposal plant.

Product safety labeling follows EU GHS guidance.

Contact phone: 1-800-428-2336

Avoid foam formation in all reagents and sample types (specimens, calibrators and controls).

Reagent handling

The reagents in the kit have been assembled into a ready-for-use unit that cannot be separated.

All information required for correct operation is available via the **cobas** link.

Storage and stability

Store at 2-8 °C.

Do not freeze.

Store the **cobas e** pack **upright** in order to ensure complete availability of the microparticles during automatic mixing prior to use.

Stability:	
unopened at 2-8 °C	up to the stated expiration date
on the analyzers	16 weeks

Specimen collection and preparation

Only the specimens listed below were tested and found acceptable.

Serum collected using standard sampling tubes or tubes containing separating gel.

Li-heparin and K_2-EDTA plasma.

Criterion: Slope 0.9-1.1 + coefficient of correlation ≥ 0.95.

Stable for 8 hours at 20-25 °C, 5 days at 2-8 °C, 3 months at -20 °C (± 5 °C). Freeze only once.

The sample types listed were tested with a selection of sample collection tubes that were commercially available at the time of testing, i.e. not all available tubes of all manufacturers were tested. Sample collection systems from various manufacturers may contain differing materials which could affect the test results in some cases. When processing samples in primary tubes (sample collection systems), follow the instructions of the tube manufacturer.

Centrifuge samples containing precipitates before performing the assay.

Do not use heat-inactivated samples.

Do not use samples and controls stabilized with azide.

Ensure the samples and calibrators are at 20-25 °C prior to measurement.

Due to possible evaporation effects, samples and calibrators on the analyzers should be analyzed/measured within 2 hours.

Sample stability claims were established by experimental data by the manufacturer or based on reference literature and only for the temperatures/time frames as stated in the method sheet. It is the responsibility of the individual laboratory to use all available references and/or its own studies to determine specific stability criteria for its laboratory.

Materials provided

See "Reagents – working solutions" section for reagents.

Materials required (but not provided)

- REF 08851964190, free PSA CalSet, 4 x 1.0 mL
- REF 11776452160, PreciControl Tumor Marker, for 4 x 3.0 mL
- General laboratory equipment
- **cobas e** analyzer

Additional materials for the **cobas e** 402 and **cobas e** 801 analyzers:

- REF 06908799190, ProCell II M, 2 x 2 L system solution
- REF 04880293190, CleanCell M, 2 x 2 L measuring cell cleaning solution
- REF 07485409001, Reservoir Cup, 8 cups to supply ProCell II M and CleanCell M
- REF 06908853190, PreClean II M, 2 x 2 L wash solution
- REF 05694302001, Assay Tip/Assay Cup tray, 6 magazines x 6 magazine stacks x 105 assay tips and 105 assay cups, 3 wasteliners
- REF 07485425001, Liquid Flow Cleaning Cup, 2 adaptor cups to supply ISE Cleaning Solution/Elecsys SysClean for Liquid Flow Cleaning Detection Unit
- REF 07485433001, PreWash Liquid Flow Cleaning Cup, 1 adaptor cup to supply ISE Cleaning Solution/Elecsys SysClean for Liquid Flow Cleaning PreWash Unit
- REF 11298500160, ISE Cleaning Solution/Elecsys SysClean, 5 x 100 mL system cleaning solution

Assay

For optimum performance of the assay follow the directions given in this document for the analyzer concerned. Refer to the appropriate operator's manual for analyzer-specific assay instructions.

Resuspension of the microparticles takes place automatically prior to use.

Place the cooled (stored at 2-8 °C) **cobas e** pack on the reagent manager. Avoid foam formation. The system automatically regulates the temperature of the reagents and the opening/closing of the **cobas e** pack.

Calibration

Traceability: This method has been standardized against the WHO Reference Standard 96/668 (100 % free PSA).

The predefined master curve is adapted to the analyzer using the relevant CalSet.

Calibration frequency: Calibration must be performed once per reagent lot using fresh reagent (i.e. not more than 24 hours since the **cobas e** pack was registered on the analyzer).

Renewed calibration is recommended as follows:

- after 12 weeks when using the same reagent lot
- after 28 days when using the same **cobas e** pack on the analyzer
- as required: e.g. quality control findings outside the defined limits

Quality control

Use PreciControl Tumor Marker or other suitable controls for routine quality control procedures.

Controls for the various concentration ranges should be run individually at least once every 24 hours when the test is in use, once per **cobas e** pack, and following each calibration.

The control intervals and limits should be adapted to each laboratory's individual requirements. Values obtained should fall within the defined limits. Each laboratory should establish corrective measures to be taken if values fall outside the defined limits.

If necessary, repeat the measurement of the samples concerned.

Follow the applicable government regulations and local guidelines for quality control.

Calculation

The analyzer automatically calculates the analyte concentration of each sample (either in ng/mL or µg/L).

Limitations - interference

The effect of the following endogenous substances and pharmaceutical compounds on assay performance was tested. Interferences were tested up to the listed concentrations and no impact on results was observed.

Endogenous substances

Compound	Concentration tested
Bilirubin	≤ 1112 µmol/L or ≤ 65 mg/dL
Hemoglobin	≤ 0.621 mmol/L or ≤ 1000 mg/dL
Intralipid	≤ 1500 mg/dL
Rheumatoid factors	≤ 1500 IU/mL

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Biotin interefence

This assay has no biotin interference in serum concentrations up to 1200 ng/mL. Pharmacokinetic studies have shown that serum concentrations of biotin can reach up to 355 ng/mL within the first hour after biotin ingestion for subjects consuming supplements of 20 mg biotin per day[11] and up to 1160 ng/mL for subjects after a single dose of 300 mg biotin.[12]

Criterion: For concentrations of 0.01-0.5 ng/mL the deviation is ± 0.06 ng/mL. For concentrations > 0.5 ng/mL the deviation is ± 10 %.

There is no high-dose hook effect at fPSA concentrations up to 15000 ng/mL.

Pharmaceutical substances

In vitro tests were performed on 17 commonly used pharmaceuticals. No interference with the assay was found.

In addition, the following special cancer drugs were tested. No interference with the assay was found.

Special cancer drugs

Drug	Concentration tested mg/L
Cyclophosphamide	1000
Cisplatin	225
5-Fluorouracil	500
Methotrexate	1000
Tamoxifen	50
Mitomycin	25
Carboplatin	1000
Etoposide	400
Flutamide	1000
Taxol	5.5
Doxorubicin	75

In rare cases, interference due to extremely high titers of antibodies to analyte-specific antibodies, streptavidin or ruthenium can occur. These effects are minimized by suitable test design.

For diagnostic purposes, the results should always be assessed in conjunction with the patient's medical history, clinical examination and other findings.

Limits and ranges

Measuring range

0.018-50.0 ng/mL (defined by the Limit of Quantitation and the maximum of the master curve). Values below the Limit of Quantitation are reported as < 0.018 ng/mL. Values above the measuring range are reported as > 50.0 ng/mL.

Lower limits of measurement

Limit of Blank, Limit of Detection and Limit of Quantitation

Limit of Blank = 0.010 ng/mL

Limit of Detection = 0.016 ng/mL

Limit of Quantitation = 0.018 ng/mL

The Limit of Blank, Limit of Detection and Limit of Quantitation were determined in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP17-A2 requirements.
The Limit of Blank is the 95th percentile value from n ≥ 60 measurements of analyte-free samples over several independent series. The Limit of Blank corresponds to the concentration below which analyte-free samples are found with a probability of 95 %.

The Limit of Detection is determined based on the Limit of Blank and the standard deviation of low concentration samples. The Limit of Detection corresponds to the lowest analyte concentration which can be detected (value above the Limit of Blank with a probability of 95 %).

The Limit of Quantitation is the lowest analyte concentration that can be reproducibly measured with an intermediate precision (within-laboratory precision) CV of ≤ 15 %.

Dilution

Not necessary due to the broad measuring range.

Expected values

A multicenter study was performed using samples from men (aged ≥ 50) referred to urologists for evaluation of prostate cancer. 1143 of the referred men had normal DRE that were not suspicious for prostate cancer (DRE normal cohort). Samples were evaluated using the Elecsys total PSA assay and Elecsys free PSA assay in parallel on the Elecsys 2010 and Elecsys 1010 immunoassay analyzers. A subset of 180 samples was evaluated on the MODULAR ANALYTICS E170 analyzer. In a specific study, for an individual specimen of urologically referred subjects, with 95 % probability, the difference between percent free PSA measured by Elecsys 2010 and MODULAR ANALYTICS E170 analyzers was from -1.6 % to 5.2 % (n = 300); by Elecsys 1010 and MODULAR ANALYTICS E170 analyzers was from -2.6 % to 4.0 % (n = 100); by Elecsys 2010 and Elecsys 1010 analyzers was from -1.5 % to 4.3 % (n = 100).

All patients underwent a transrectal prostate biopsy. Of the 1143 men with normal DRE, 664 men had tPSA results between 4.00-10.0 ng/mL on the Elecsys 2010 analyzer (tPSA 4.00-10.0 ng/mL:DRE normal cohort). The ethnic composition of PSA 4.00-10.0 ng/mL:DRE normal cohort was 84.5 % Caucasian, 11.5 % Black non-Hispanic, 2.6 % Hispanic-Mexican, and 1.4 % other. The median age was 66 years. The distribution of fPSA, tPSA, and ratio fPSA/tPSA (% fPSA) values by biopsy result for this cohort is shown in table 1.

Table 1: PSA statistics by biopsy outcome (benign, malignant)

Elecsys 2010 analyzer	Biopsy result	N	Mean ng/mL	Median ng/mL	Min. ng/mL	Max. ng/mL	Stand. error of mean
fPSA	Benign	463	1.19	1.11	0.26	4.14	0.02
	Malignant	201	1.00	0.92	0.34	2.39	0.03
	Total	664	1.13	1.06	0.26	4.14	0.02
tPSA	Benign	463	6.10	5.68	3.95	10.0	0.07
	Malignant	201	6.42	6.10	3.95	10.0	0.11
	Total	664	6.20	5.84	3.95	10.0	0.06
% fPSA	Benign	463	19.72	19.2	5.1	53.4	0.32
	Malignant	201	16.0	15.2	5.2	35.8	0.42
	Total	664	18.6	18.0	5.1	53.4	0.27

A comparison of the mean % fPSA for the benign and malignant biopsy groups indicated that the difference is significant.
The % fPSA result may be used in evaluating the need for prostate biopsy in 1 of 2 ways:
1. The relative risk of prostate cancer in individual men may be considered, or
2. Patients may be managed using a single cutoff approach.

1. Individual risk assessment

There is an increased probability of detecting prostate cancer (PCA) as the PSA level increases. Of interest is that in an urologically referred cohort there is a 12 % to 22 % risk of PCA in men whose tPSA is < 4.00 ng/mL. The tPSA range of 4.00-10.0 ng/mL has been described in references 6 and 10 as the diagnostic "gray zone". It is in this area that the % fPSA to tPSA ratio is of utility.

Table 2: Probability of detecting PCA on needle biopsy in urologically referred men with DRE results not suspicious for prostate cancer

tPSA ng/mL	Probability of PCA %	95 % confidence interval
< 4.00	17.1	12.4-21.6
4.00-10.0	30.3	26.8-33.8
> 10.0	49.1	42.5-55.7

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The probability of finding PCA with tPSA in the gray zone (4.00-10.0 ng/mL) increases with increasing age and with decreasing fPSA/tPSA ratios - see table 3. The probabilities presented in table 3 were estimated from a loglinear model.

Table 3: Probability of finding PCA on needle biopsy by age in years and % fPSA on the Elecsys 2010 analyzer

Probability of finding PCA on needle biopsy by age in years (95 % confidence interval)			
% fPSA ratio	50-59	60-69	≥ 70
≤ 10	49.2 (12.4-86.9)	57.5 (17.9-89.3)	64.5 (30.4-88.3)
11-18	26.9 (5.7-68.9)	33.9 (8.6-73.7)	40.8 (15.8-71.7)
19-25	18.3 (3.5-57.9)	23.9 (5.4-63.4)	29.7 (10.1-61.1)
> 25	9.1 (3.1-23.7)	12.2 (4.7-28.1)	15.8 (9.0-26.1)

2. Single cutoff

Alternatively, a single cutoff may be used for men in all age groups. Sensitivities (% of PCA detected) and specificities (% of biopsies avoided in men without PCA) for various % fPSA cutoffs are shown in table 4. A cutoff of 25 % results in the detection of 92.5 % of prostate cancers and avoids unnecessary biopsy in 20.3 % of men without prostate cancer. Virtually all (99 %) of prostate cancers are detected with a cutoff of 30 %, but only 8.9 % of men without prostate cancer are spared biopsy.

Table 4: Agreement with biopsy at various % fPSA cutoffs on the Elecsys 2010 analyzer

Benign biopsies			
free PSA %	Number of patients with negative biopsy identified at cutoff (total = 463)	Agreement at cutoff %	95 % confidence interval
23	141	30.4	(26.3-34.9)
25	94	20.3	(16.7-24.3)
27	65	14.0	(11.0-17.5)
30	41	8.9	(6.4-11.8)
53	1	0.2	(0.0-1.2)

Malignant biopsies			
free PSA %	Number of patients with positive biopsy identified at cutoff (total = 201)	Agreement at cutoff %	95 % confidence interval
23	173	86.1	(80.5-90.5)
25	186	92.5	(88.0-95.8)
27	192	95.5	(91.7-97.9)
30	199	99.0	(96.4-99.9)
53	201	100	(98.2-100)

Each laboratory should investigate the transferability of the expected values to its own patient population and if necessary determine its own reference ranges.

Specific performance data

Representative performance data on the analyzers are given below. Results obtained in individual laboratories may differ.

Precision

Precision was determined on the **cobas e** 801 analyzer using 1 lot of Elecsys reagent, 5 human serum samples and 2 controls in a modified protocol: 2 runs per day in duplicate each for 12 days (n = 48). Repeatability and intermediate precision were calculated according to a protocol (EP05-A3) of the CLSI (Clinical and Laboratory Standards Institute). The following results were obtained:

cobas e 801 analyzer					
		Repeatability[b)]		**Intermediate precision**[c)]	
Sample	Mean ng/mL	SD ng/mL	CV %	SD ng/mL	CV %
HS[d)]01	0.026	0.001	4.5	0.001	4.8
HS02	0.252	0.005	2.0	0.008	3.1
HS03	0.777	0.013	1.6	0.014	1.8
HS04	23.9	0.515	2.2	0.599	2.5
HS05	47.4	0.621	1.3	0.731	1.5
PC TM[e)] 1	1.06	0.045	4.2	0.045	4.2
PC TM 2	9.14	0.362	4.0	0.376	4.1

b) Repeatability = within-run precision

c) Intermediate precision = within-laboratory precision

d) HS = Human Serum

e) PC TM = PreciControl Tumor Marker

Precision was determined on the **cobas e** 402 analyzer using Elecsys reagents, pooled human sera and controls in a protocol (EP05-A3) of the CLSI (Clinical and Laboratory Standards Institute): 2 runs per day in duplicate each for 12 days (n = 48). The following results were obtained:

cobas e 402 analyzer					
		Repeatability		**Intermediate Precision**	
Sample	Mean ng/mL	SD ng/mL	CV %	SD ng/mL	CV %
HS01	0.021	0.001	4.3	0.001	6.0
HS02	0.134	0.001	1.0	0.003	2.4
HS03	0.798	0.009	1.1	0.019	2.4
HS04	2.08	0.017	0.8	0.053	2.6
HS05	6.04	0.060	1.0	0.129	2.1
HS06	22.3	0.149	0.7	0.321	1.4
HS07	30.0	0.267	0.9	0.487	1.6
HS08	41.6	0.324	0.8	1.02	2.5
HS09	43.7	0.337	0.8	0.751	1.7
PC TM 1	0.959	0.007	0.7	0.016	1.6
PC TM 2	9.55	0.055	0.6	0.151	1.6

Reproducibility

Reproducibility was determined on the **cobas e** 801 analyzer with a panel of 5 human sera and 2 levels of controls. Samples were measured in triplicate in 2 runs per day for 5 days at 3 sites.

Elecsys free PSA system reproducibility on the **cobas e** 801 analyzer			
		Reproducibility	
Sample n = 90	Mean ng/mL	SD ng/mL	CV %
HS01	0.026	0.002	6.4
HS02	0.256	0.007	2.7
HS03	0.788	0.017	2.2
HS04	23.9	0.467	2.0
HS05	47.5	0.993	2.1
PC TM 1	1.07	0.021	2.0
PC TM 2	9.15	0.228	2.5

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Precision was further evaluated incorporating repeatability, between-run, between-day and between-site variation. The analysis of data was based on guidance from CLSI documents EP05-A3. Data from all 3 sites were combined to achieve SD and percent CV for repeatability (within-run), between-run, between-day, between-site and reproducibility. The overall imprecision data are summarized in the following table.

Elecsys free PSA system reproducibility on the **cobas e** 801 analyzer					
		Repeatability		**Between-run**	
Sample n = 90	Mean ng/mL	SD ng/mL	CV %	SD ng/mL	CV %
HS01	0.026	0.002	6.3	0.0[f)]	0.0
HS02	0.256	0.005	2.1	0.0[f)]	0.0
HS03	0.788	0.008	1.0	0.006	0.8
HS04	23.9	0.295	1.2	0.246	1.0
HS05	47.5	0.719	1.5	0.292	0.6
PC TM 1	1.07	0.010	0.9	0.007	0.6
PC TM 2	9.15	0.141	1.5	0.0[f)]	0.0

f) Negative variance estimates were set to zero.

Elecsys free PSA system reproducibility on the **cobas e** 801 analyzer					
		Between-day		**Between-site**	
Sample n = 90	Mean ng/mL	SD ng/mL	CV %	SD ng/mL	CV %
HS01	0.026	0.000	1.2	0.000	0.6
HS02	0.256	0.003	1.2	0.003	1.1
HS03	0.788	0.012	1.6	0.005	0.7
HS04	23.9	0.265	1.1	0.0[f)]	0.0
HS05	47.5	0.621	1.3	0.0[f)]	0.0
PC TM 1	1.07	0.013	1.3	0.011	1.0
PC TM 2	9.15	0.158	1.7	0.086	0.9

Reproducibility was determined on the **cobas e** 402 analyzer with a panel of 9 human sera and 2 levels of controls. Samples were measured in triplicate in 2 runs per day for 5 days at 3 sites.

Elecsys free PSA system reproducibility on the **cobas e** 402 analyzer			
		Reproducibility	
Sample n = 90	Mean ng/mL	SD ng/mL	CV %
HS01	0.024	0.003	14.1
HS02	0.140	0.005	3.8
HS03	0.813	0.021	2.6
HS04	2.14	0.052	2.4
HS05	6.32	0.164	2.6
HS06	23.2	0.608	2.6
HS07	31.1	0.815	2.6
HS08	42.8	1.11	2.6
HS09	45.8	1.33	2.9
PC TM 1	0.980	0.025	2.6
PC TM 2	9.8	0.248	2.5

Precision was further evaluated incorporating repeatability, between-run, between-day and between-site variation. The analysis of data was based on guidance from CLSI documents EP05-A3. Data from all 3 sites were combined to achieve SD and percent CV for repeatability (within-run), between-run, between-day, between-site and reproducibility. The overall imprecision data are summarized in the following table.

Elecsys free PSA system reproducibility on the **cobas e** 402 analyzer					
		Repeatability		**Between-run**	
Sample n = 90	Mean ng/mL	SD ng/mL	CV %	SD ng/mL	CV %
HS01	0.024	0.001	5.7	0.000	0.9
HS02	0.140	0.002	1.3	0.001	0.8
HS03	0.813	0.006	0.8	0.007	0.9
HS04	2.14	0.021	1.0	0.020	0.9
HS05	6.32	0.069	1.1	0.147	2.3
HS06	23.2	0.209	0.9	0.000	0.0
HS07	31.1	0.257	0.8	0.210	0.7
HS08	42.8	0.299	0.7	0.161	0.4
HS09	45.8	0.281	0.6	0.428	0.9
PC TM 1	0.980	0.008	0.8	0.004	0.4
PC TM 2	9.8	0.083	0.8	0.049	0.5

Elecsys free PSA system reproducibility on the **cobas e** 402 analyzer					
		Between-day		**Between-site**	
Sample n = 90	Mean ng/mL	SD ng/mL	CV %	SD ng/mL	CV %
HS01	0.024	0.000	0.6	0.003	12.8
HS02	0.140	0.001	0.8	0.005	3.3
HS03	0.813	0.004	0.5	0.018	2.3
HS04	2.14	0.010	0.5	0.042	2.0
HS05	6.32	0.00	0.0	0.025	0.4
HS06	23.2	0.181	0.8	0.541	2.3
HS07	31.1	0.290	0.9	0.685	2.2
HS08	42.8	0.315	0.7	1.01	2.3
HS09	45.8	0.440	1.0	1.15	2.5
PC TM 1	0.980	0.011	1.1	0.021	2.1
PC TM 2	9.8	0.128	1.3	0.189	1.9

Method comparison

A method comparison study was performed at 3 sites to compare the Elecsys free PSA immunoassay on the **cobas e** 801 analyzer (y) and the **cobas e** 601 analyzer (x). 231 human serum samples were measured on 3 different **cobas e** 601 analyzers and the median value of the results was compared to the value obtained on 3 different **cobas e** 801 analyzers (231 samples were determined; therefore 693 results were obtained on the 3 **cobas e** 801 analyzers). The combined analysis is reported.

Number of serum samples measured: 231

Passing/Bablok[13]	Weighted Deming
$y = 0.967x + 0.004$	$y = 0.972x + 0.004$
$\tau = 0.980$	$r = 0.999$

The sample concentrations were between 0.034 and 47.1 ng/mL.

Analytical specificity

For the monoclonal antibodies used, the following cross-reactivities were found:

Prostatic acid phosphatase (PAP) and alpha-1-antichymotrypsin inhibitor (ACT): none; PSA-antichymotrypsin inhibitor (PSA-ACT): 0.7 %.

References

1 Henttu P, Vihko P. Prostate-specific Antigen and Human Glandular Kallikrein: Two Kallikreins of the Human Prostate. Ann Med 1994;26:157-164.

2 Armbruster DA. Prostate Specific Antigen: Biochemistry, Analytical Methods and Clinical Application. Clin Chem 1993;39/2:181-195.

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3 Zhou AM, Tewari PC, Bluestein BI, et al. Multiple forms of prostate specific antigen in serum: differences in immunorecognition by monoclonal and polyclonal assays. Clin Chem 1993;39/12:2483-2491.

4 Lilja H, Christensson A, Dahlen U, et al. Prostate Specific Antigen in Human Serum occurs predominantly in Complex with Alpha-1-Antichymotrypsin. Clin Chem 1991;37(9):1618-1625.

5 Zhang WM, Leinonen J, Kalkkinen N, et al. Purification and Characterization of Different Molecular Forms of Prostate-Specific Antigen in Human Seminal Fluid. Clin Chem 1995;41/11:1567-1573.

6 Prestigiacomo AF, Stamey TA. Clinical usefulness of free and complexed PSA. Scand J Clin Lab Invest 1995;55 Suppl 221:32-34.

7 Oesterling JE. Prostate-Specific Antigen: a critical assessment of the most useful tumor marker for adenocarcinoma of the prostate. J Urology 1991(5);145:907-923.

8 Catalona WJ, Richie JP, Ahmann FR, et al. Comparison of digital rectal examination and serum prostate-specific antigen in the early detection of prostate cancer: results of a multicenter clinical trial of 6,630 men. J Urology 1994;151(5):1283-1290.

9 Chen YT, Luderer AA, Thiel RP, et al. Using proportions of free to total prostate-specific antigen, age, and total prostate-specific antigen to predict the probability of prostate cancer. Urology 1996;47:518-524.

10 Thiel RP, Oesterling JE, Wojno KJ, et al. A multicenter comparison of the diagnostic performance of free prostate-specific antigen. Urology 1996;48(6A):45-50.

11 Grimsey P, Frey N, Bendig G, et al. Population pharmacokinetics of exogenous biotin and the relationship between biotin serum levels and in vitro immunoassay interference. International Journal of Pharmacokinetics 2017 Sept 14;2(4):247-256.

12 Piketty ML, Prie D, Sedel F, et al. High-dose biotin therapy leading to false biochemical endocrine profiles: validation of a simple method to overcome biotin interference. Clin Chem Lab Med. 2017 May 1;55(6):817-825.

13 Bablok W, Passing H, Bender R, et al. A general regression procedure for method transformation. Application of linear regression procedures for method comparison studies in clinical chemistry, Part III. J Clin Chem Clin Biochem 1988 Nov;26(11):783-790.

For further information, please refer to the appropriate user guide or operator's manual for the analyzer concerned, the respective application sheets and the Method Sheets of all necessary components (if available in your country).

Any serious incident that has occurred in relation to the device shall be reported to the manufacturer and the competent authority of the Member State in which the user and/or the patient is established.

Symbols

Roche Diagnostics uses the following symbols and signs in addition to those listed in the ISO 15223-1 standard (for USA: see navifyportal.roche.com for definition of symbols used):

CONTENT	Contents of kit
SYSTEM	Analyzers/Instruments on which reagents can be used
REAGENT	Reagent
CALIBRATOR	Calibrator
→	Volume for reconstitution
GTIN	Global Trade Item Number
Rx only	For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

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