

REF		Σ	SYSTEM
08928380190	08928380500	100	cobas e 411 cobas e 601 cobas e 602

English

System information

For **cobas e 411** analyzer: test number 2320

For **cobas e 601** and **cobas e 602** analyzers: Application Code Number 92

Intended use

Immunoassay for the in vitro quantitative determination of intact parathyroid hormone in human serum and plasma for the differential diagnosis of hypercalcemia and hypocalcemia. This assay can be used intraoperatively.

The electrochemiluminescence immunoassay "ECLIA" is intended for use on **cobas e** immunoassay analyzers.

Summary

Parathyroid hormone (PTH) is a single-chain 84-amino-acid peptide produced by the parathyroid glands in response to decreased extracellular concentrations of ionized calcium. Its main role is to increase serum calcium levels by stimulating the release of calcium from bone and its renal re-absorption in the distal tubule. In the proximal tubule, PTH stimulates the synthesis of calcitriol which in turn increases intestinal absorption of calcium and exerts an endocrine feed-back on the secretion of PTH at the parathyroid level. PTH also decreases the renal re-absorption of phosphate in the proximal tubule, thereby decreasing serum phosphate.¹

Parathyroid gland disorders lead to elevated or depressed blood calcium levels (hypercalcemia or hypocalcemia) brought about by a change in the secretion of PTH.

Detection of subfunctioning parathyroid glands (hypoparathyroidism) requires the use of a highly sensitive test in order to be able to measure PTH levels well below normal. Hyperfunctioning of the parathyroid glands results in an increased secretion of PTH (hyperparathyroidism). Primary causes are adenomas of the parathyroid glands. In secondary hyperparathyroidism the blood calcium level is low as a result of other pathological states (e.g. vitamin D deficiency).²

The determination of PTH intraoperatively during adenoma resection in the parathyroid glands has been reported for primary hyperparathyroidism,^{3,4} secondary hyperparathyroidism relating to renal failure,^{5,6} and tertiary hyperparathyroidism post renal transplant surgery.⁷ Because PTH has a reported half-life of 3-5 minutes,⁸ a significant drop in PTH levels after resection of the abnormal gland or glands enables the surgeon to assess whether all hyperfunctioning parathyroid tissue has been removed from the patient.⁹

The National Academy of Clinical Biochemistry recommends routine use of intraoperative PTH testing for patients undergoing surgery for primary hyperparathyroidism, both in initial surgeries and in reoperative procedures.¹⁰

The Kidney Disease Outcomes Quality Initiative (KDOQI) and Kidney Disease Improving Global Outcomes (KDIGO) guidelines recommend that serum PTH concentration should be measured regularly in patients with Chronic Kidney Disease (CKD) and maintained within the target ranges that are defined according to the stage of CKD.^{11,12}

The Elecsys assay for determining intact PTH employs a sandwich test principle in which a biotinylated monoclonal antibody reacts with the N-terminal fragment (1-37) and a monoclonal antibody labeled with a ruthenium complex^{a)} reacts with the C-terminal fragment (38-84).

The antibodies used in this assay are reactive with epitopes in the amino acid regions 26-32 and 37-42.

a) Tris(2,2'-bipyridyl)ruthenium(II)-complex (Ru(bpy)₃²⁺)

Test principle

Sandwich principle. Total duration of assay: 18 minutes.

- 1st incubation: 50 µL of sample, a biotinylated monoclonal PTH-specific antibody, and monoclonal PTH-specific antibody labeled with a ruthenium complex 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/ProCell 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 reagent barcode or e-barcode.

Reagents - working solutions

The reagent rackpack is labeled as PTH.

- M Streptavidin-coated microparticles (transparent cap), 1 bottle, 6.5 mL:
Streptavidin-coated microparticles 0.72 mg/mL; preservative.
- R1 Anti-PTH-Ab~biotin (gray cap), 1 bottle, 7 mL:
Biotinylated monoclonal anti-PTH antibody (mouse) 2.3 mg/L;
phosphate buffer 100 mmol/L, pH 7.0; preservative.
- R2 Anti-PTH-Ab~Ru(bpy)₃²⁺ (black cap), 1 bottle, 7 mL:
Monoclonal anti-PTH antibody (mouse) labeled with ruthenium complex 2.0 mg/L; phosphate buffer 100 mmol/L, pH 7.0; preservative.

Precautions and warnings

For in vitro diagnostic use for laboratory 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:

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.

Hazardous components:

- 2-methyl-2H-isothiazol-3-one hydrochloride
- Product safety labeling follows EU GHS guidance.

Contact phone: all countries: +49-621-7590

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 read in from the respective reagent barcodes.

Storage and stability

Store at 2-8 °C.

Do not freeze.

Store the Elecsys reagent kit **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
after opening at 2-8 °C	12 weeks
on the analyzers	28 days

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, K₂-EDTA and K₃-EDTA plasma.

Plasma tubes containing separating gel can be used.

Because of the instability of PTH in unseparated serum, serum tubes should be centrifuged immediately. In contrast, PTH was found to be stable for > 24 hours at room temperature in whole blood anticoagulated with EDTA. Therefore, preference should be given to EDTA plasma.^{13,14}

Criterion: Method comparison serum versus plasma, slope 0.9-1.1 + intercept within $\leq \pm 2.4$ pg/mL + coefficient of correlation ≥ 0.95 .

Serum: Stable for 8 hours at 15-25 °C, 2 days at 2-8 °C, 6 months at -20 °C (± 5 °C).

Plasma: Stable for 2 days at 15-25 °C, 3 days at 2-8 °C, 6 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 samples and controls stabilized with azide.

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

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

Materials provided

See "Reagents – working solutions" section for reagents.

Materials required (but not provided)

- [REF] 08928452190, CalSet PTH, for 4 x 1.0 mL
- [REF] 05618860190, PreciControl Varia, for 4 x 3.0 mL
- General laboratory equipment
- cobas e** analyzer

Additional materials for the **cobas e 411** analyzer:

- [REF] 11662988122, ProCell, 6 x 380 mL system buffer
- [REF] 11662970122, CleanCell, 6 x 380 mL measuring cell cleaning solution

- [REF] 11930346122, Elecsys SysWash, 1 x 500 mL washwater additive
- [REF] 11933159001, Adapter for SysClean
- [REF] 11706802001, AssayCup, 60 x 60 reaction cups
- [REF] 11706799001, AssayTip, 30 x 120 pipette tips
- [REF] 11800507001, Clean-Liner

Additional materials for **cobas e 601** and **cobas e 602** analyzers:

- [REF] 04880340190, ProCell M, 2 x 2 L system buffer
- [REF] 04880293190, CleanCell M, 2 x 2 L measuring cell cleaning solution
- [REF] 03023141001, PC/CC-Cups, 12 cups to prewarm ProCell M and CleanCell M before use
- [REF] 03005712190, ProbeWash M, 12 x 70 mL cleaning solution for run finalization and rinsing during reagent change
- [REF] 03004899190, PreClean M, 5 x 600 mL detection cleaning solution
- [REF] 12102137001, AssayTip/AssayCup, 48 magazines x 84 reaction cups or pipette tips, waste bags
- [REF] 03023150001, WasteLiner, waste bags
- [REF] 03027651001, SysClean Adapter M

Additional materials for all analyzers:

- [REF] 11298500316, 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. Read in the test-specific parameters via the reagent barcode. If in exceptional cases the barcode cannot be read, enter the 15-digit sequence of numbers.

cobas e 601 and **cobas e 602** analyzers: PreClean M solution is necessary.

Bring the cooled reagents to approximately 20 °C and place on the reagent disk (20 °C) of the analyzer. Avoid foam formation. The system automatically regulates the temperature of the reagents and the opening/closing of the bottles.

Calibration

Traceability: This method has been standardized against a commercial PTH test (RIA). The recovery of the NIBSC 95/646 (WHO) standard was assessed by testing dilutions in human serum covering the measuring range (40-4000 pg/mL) on 16 analyzers (**cobas e 411** and **cobas e 601** analyzers). The mean recovery was 100 % \pm 4 %.

Every Elecsys reagent set has a barcoded label containing specific information for calibration of the particular reagent lot. 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 reagent kit was registered on the analyzer).

Calibration interval may be extended based on acceptable verification of calibration by the laboratory.

Renewed calibration is recommended as follows:

- every 12 weeks when using the same reagent lot
- every 7 days (when using the same reagent kit on the analyzer)
- as required: e.g. quality control findings outside the defined limits

Quality control

Use PreciControl Varia 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 reagent kit, 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.

Elecsys PTH

	Median PTH concentration, pg/mL			
Total	489	35.5	17.3 (14.3-19.4)	74.1 (68.8-86.6)

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 using Elecsys reagents, samples and controls in a protocol (EP05-A3) of the CLSI (Clinical and Laboratory Standards Institute); 2 runs per day in duplicate each for 21 days (n = 84). The following results were obtained:

cobas e 411 analyzer								
Sample	Mean		Repeatability			Intermediate precision		
			SD		CV	SD		CV
	pg/mL	pmol/L	pg/mL	pmol/L	%	pg/mL	pmol/L	%
HS ^{b)} 1	10.6	1.12	0.355	0.038	3.3	0.465	0.049	4.4
HS 2	13.6	1.44	0.358	0.038	2.6	0.500	0.053	3.7
HS 3	55.5	5.88	0.815	0.086	1.5	1.45	0.154	2.6
HS 4	2341	248	26.2	2.78	1.1	52.1	5.52	2.2
HS 5	4428	469	61.1	6.48	1.4	107	11.3	2.4
PC Varia 1 ^{c)}	61.0	6.47	0.937	0.099	1.5	1.52	0.161	2.5
PC Varia 2	181	19.2	2.12	0.225	1.2	4.17	0.442	2.3

b) HS = Human serum

c) PC = PreciControl

cobas e 601 and cobas e 602 analyzers								
Sample	Mean		Repeatability			Intermediate precision		
			SD		CV	SD		CV
	pg/mL	pmol/L	pg/mL	pmol/L	%	pg/mL	pmol/L	%
HS ^{d)} 1	9.29	0.985	0.195	0.021	2.1	0.404	0.043	4.4
HS 2	13.8	1.46	0.210	0.022	1.5	0.420	0.045	3.0
HS 3	54.6	5.79	0.444	0.047	0.8	0.798	0.085	1.5
HS 4	2467	262	17.7	1.88	0.7	32.6	3.46	1.3
HS 5	4514	478	41.2	4.37	0.9	49.1	5.20	1.1
PC Varia 1	58.0	6.15	0.545	0.058	0.9	1.20	0.127	2.1
PC Varia 2	175	18.6	1.59	0.169	0.9	2.56	0.271	1.5

d) HS = human serum

Method comparison

A comparison of the Elecsys PTH assay, [REF 08928380190](#) (cobas e 601 analyzer; y) with the Elecsys PTH assay, [REF 11972103122](#) (cobas e 601 analyzer; x) gave the following correlations (pg/mL):

Number of samples measured: 147

Passing/Bablok¹⁵ Linear regression
 $y = 0.979x - 0.883$ $y = 0.964x + 5.28$
 $r = 0.984$ $r = 1.00$

The sample concentrations were between 4.12 and 4826 pg/mL (0.437 and 512 pmol/L).

Analytical specificity

The Elecsys PTH assay does not show any significant cross-reactivity with the following substances, tested with PTH concentrations of approximately 15 pg/mL and 60 pg/mL (maximum tested concentration):

Cross-reactant	Concentration tested
PTH fragment 1-34	5000 pg/mL
PTH fragment 1-37	5000 pg/mL
PTH-related protein 1-86	5000 pg/mL

The assay has a 93 % cross reactivity to the PTH fragment 7-84.

Clinical investigations in intraoperative use

In 2006, the National Academy of Clinical Biochemistry published their Laboratory Medicine Practice Guidelines for point of care testing, entitled Evidence Based Practice for Point of Care Testing.¹⁰ The guidelines recommend the use of intraoperative parathyroid hormone testing 1) for patients undergoing surgery for hyperparathyroidism, especially in minimally invasive or directed procedures, 2) for patients undergoing reoperation, and 3) as a replacement for traditional laboratory measurements of PTH during venous localization in order to help the angiography team guide sampling.

The guidelines further recommend for patients undergoing parathyroidectomy for hyperparathyroidism that baseline samples be obtained preoperation exploration and pre-excision of the gland, and that post-excision sampling be drawn at 5 and 10 minutes post resection with a 50 % reduction in PTH concentrations from the highest baseline level. The guidelines also caution that additional samples may be necessary.¹⁰

PTH testing during parathyroid surgery was conducted by several groups of investigators using the Elecsys PTH immunoassay.^{4,5,6,7,16}

The overall sensitivity and specificity of the assay to demonstrate successful surgery as defined by postoperative reduction of calcium levels was 99.6 % and 93.7 %, respectively.

References

- Soubierbielle JC, Roth H, Fouque D. Parathyroid hormone measurement in CKD. *Kidney Int* 2010;77:93–100.
- Nussbaum S, Potts JT. Advances in Immunoassays for Parathyroid Hormone. *Clinical Applications to Skeletal Disorders of Bone and Mineral Metabolism*. In Bilezikian JP, Levine MA, Marcus R (eds). *The Parathyroids: Basic and Clinical Concepts*. Raven Press, New York 1994:157-169.
- Bergenfels A, Nordén NE, Ahrén B. Intraoperative fall in plasma levels of intact parathyroid hormone after removal of one enlarged parathyroid gland in hyperparathyroid patients. *Eur J Surg* 1991;157:109-112.
- Ohe MN, Santos RO, Kunii IS, et al. Usefulness of a rapid immunometric assay for intra-operative parathyroid hormone measurements. *Braz J Med Biol Res* 2003;36(6):715-721.
- Seehofer D, Rayes N, Ulrich F, et al. Intra-operative measurement of intact parathyroid hormone in renal hyperparathyroidism by an inexpensive routine assay. *Langenbecks Arch Surg* 2001;386(6):440-443.
- Seehofer D, Rayes N, Klupp J, et al. Predictive value of intact parathyroid hormone measurement during surgery for renal hyperparathyroidism. *Langenbecks Arch Surg* 2005;390(3):222-229.
- Haustein SV, Mack E, Starling JR, et al. The role of intra-operative parathyroid hormone testing in patients with tertiary hyperparathyroidism after renal transplantation. *Surgery* 2005;138(6):1066-1071.
- Maier GW, Kreis ME, Renn W, et al. Parathyroid hormone after adenectomy for primary hyperparathyroidism: A study of peptide hormone elimination kinetics in humans. *Jour Clin Endocrinol Metab* 1998;83(11):3853-3856.
- Carter AB, Howanitz TJ. Intra-operative testing for parathyroid hormone: a comprehensive review of the use of the assay and the relevant literature. *Arch Pathol Lab Med* 2003;127:1424-1442.
- Nichols JH, Christenson RH, Clarke W, et al. National Academy of Clinical Biochemistry Laboratory Medicine Practice Guidelines: Evidence Based Practice for Point of Care Testing. AACC Press:2006.

- 11 National Kidney Foundation. K/DOQI clinical practice guidelines for bone metabolism and diseases in chronic kidney disease. Am J Kidney Dis 2003;42:S1-201.
- 12 KDIGO – Kidney Disease Improving Global Outcomes KDIGO clinical practice guideline for the diagnosis, evaluation, prevention, and treatment of chronic kidney disease-mineral and bone disorder (CKD-MBD). Kidney International 2009;76(Suppl 113).
- 13 Hanon EA, Sturgeon CM, Lamb EJ. Sampling and storage conditions influencing the measurement of parathyroid hormone in blood samples: a systematic review. Clin Chem Lab Med. 2013;51(10):1925-1941.
- 14 Stokes FJ, Ivanov P, Bailey LM, et al. The Effects of Sampling Procedures and Storage Conditions on Short-term Stability of Blood-Based Biochemical Markers of Bone Metabolism. Clin Chem 2011;57(1):138-140.
- 15 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.
- 16 Ohe MN, Santos RO, Kunii IS, et al. Usefulness of intra-operative PTH measurement in primary and secondary hyperparathyroidism: experience with 109 patients. Arq Bras Endocrinol Metab 2006;50(5):869-875.

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).

A point (period/stop) is always used in this Method Sheet as the decimal separator to mark the border between the integral and the fractional parts of a decimal numeral. Separators for thousands are not used.

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:

	Contents of kit
	Analyzers/Instruments on which reagents can be used
	Reagent
	Calibrator
	Volume for reconstitution
	Global Trade Item Number

Rx only For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

COBAS, NAVIFY, ELECSYS and PRECICONTROL are trademarks of Roche. INTRALIPID is a trademark of Fresenius Kabi AB.

All other product names and trademarks are the property of their respective owners.

Additions, deletions or changes are indicated by a change bar in the margin.

© 2024, Roche Diagnostics



Roche Diagnostics GmbH
Sandhofer Strasse 116
68305 Mannheim, Germany
www.roche.com
 +800 5505 6606

