


TSH

Thyrotropin

REF 200 tests

122

117 31459 

• Indicates analyzers on which the kit can be used

cobas e 602	cobas e 601	cobas e 411	MODULAR ANALYTICS E170	Elecsys 2010
•	•	•	•	•

English

Intended use

Immunoassay for the in vitro quantitative determination of thyrotropin in human serum and plasma.

The electrochemiluminescence immunoassay "ECLIA" is intended for use on

Elecsys and **cobas e** immunoassay analyzers.

Summary

Thyroid-stimulating hormone (TSH, thyrotropin) is a glycoprotein having a molecular weight of approx. 30000 daltons and consisting of two subunits. The β -subunit carries the TSH-specific immunological and biological information, whereas the α -chain carries species-specific information and has an identical amino acid sequence to the α -chains of LH, FSH and hCG.

TSH is for med in specific basophil cells of the anterior pituitary and is subject to a circadian secretion sequence. The hypophyseal release of TSH (thyrotropic hormone) is the central regulating mechanism for the biological action of thyroid hormones. TSH has a stimulating action in all stages of thyroid hormone formation and secretion; it also has a proliferative effect.^{1,2} The determination of TSH serves as the initial test in thyroid diagnostics. Even very slight changes in the concentrations of the free thyroid hormones bring about much greater opposite changes in the TSH level. Accordingly, TSH is a very sensitive and specific parameter for assessing thyroid function and is particularly suitable for early detection or exclusion of disorders in the central regulating circuit between the hypothalamus, pituitary and thyroid.^{3,4,5,6}

The Elecsys TSH assay employs monoclonal antibodies specifically directed against human TSH. The antibodies labeled with ruthenium complex^a consist of a chimeric construct from human and mouse-specific components. As a result, interfering effects due to HAMA (human anti-mouse antibodies) are largely eliminated.

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

Test principle

Sandwich principle. Total duration of assay : 18 minutes.

- 1st incubation: 50 μ L of sample, a biotinylated monoclonal TSH-specific antibody and a monoclonal TSH-specific antibody labeled with a ruthenium complex 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. 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.

Reagents - working solutions

M Streptavidin-coated microparticles (transparent cap), 1 bottle, 12 mL:
Streptavidin-coated microparticles 0.72 mg/mL, preservative.

R1 Anti-TSH-Ab-biotin (gray cap), 1 bottle, 14 mL:
Biotinylated monoclonal anti-TSH antibody (mouse) 2.0 mg/L;
phosphate buffer 100 mmol/L, pH 7.2; preservative.

R2 Anti-TSH-Ab-Ru(bpy)₃²⁺ (black cap), 1 bottle, 12 mL:
Monoclonal anti-TSH antibody (mouse/human) labeled with ruthenium complex 1.2 mg/L;
phosphate buffer 100 mmol/L, pH 7.2; preservative.

Precautions and warnings

For in vitro diagnostic use.

Exercise the normal precautions required for handling all laboratory reagents.

Disposal of all waste material should be in accordance with local guidelines. Safety data sheet available for professional user on request.

Avoid the formation of foam with 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 via the respective reagent barcodes.

Storage and stability

Store at 2-8 °C.

Store the Elecsys TSH reagent kit **upright** in order to ensure complete availability of the microparticles during automatic mixing prior to use. Stability:

up to the stated expiration date	unopened at 2-8 °C
12 weeks	after opening at 2-8 °C
6 weeks	on MODULAR ANALYTICS E170, cobas e 601 and cobas e 602
8 weeks	on Elecsys 2010 and cobas e 411

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-, Na-, NH₄⁺-heparin, K₃-EDTA, sodium citrate, and sodium fluoride/potassium oxalate plasma.
Criterion: Recovery within 90-110 % of serum value or slope 0.9-1.1 + intercept within $\pm 2 \times$ analytical sensitivity (LDL) + coefficient of correlation > 0.95. Stable for 7 days at 2-8 °C, 1 month at -20 °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 patients' samples, calibrator s, and controls are at ambient temperature (20-25 °C) before measurement.

Because of possible evaporation effects, samples, calibrator s, and controls on the analyzers should be measured within 2 hours.

Materials provided

See "Reagents - working solutions" section for reagents.

Materials required (but not provided)

- [REF] 0 4738551190, TSH CalSet, 4 x 1.3 mL
- [REF] 117 76 479122, PreciControl TSH, 4 x 2 mL
- [REF] 11731416122, PreciControl Universal, for 2 x 3 mL each of PreciControl Universal 1 and 2 or [REF] 11731416190, PreciControl Universal, for 2 x 3 mL each of PreciControl Universal 1 and 2
- [REF] 03609987190, Diluent MultiAssay, 2 x 16 mL sample diluent
- General laboratory equipment
- Elecsys 2010, MODULAR ANALYTICS E170 or **cobas e** analyzer

Accessories for Elecsys 2010 and **cobas e 411** analyzers:

- [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, Elecsys 2010 AssayCup, 60 x 60 reaction vessels
- [REF] 11706799001, Elecsys 2010 AssayTip, 30 x 120 pipette tips

Accessories for MODULAR ANALYTICS E170, **cobas e 601** and **cobas e 602** analyzers:

- [REF] 0 4880340190, ProCell M, 2 x 2 L system buffer



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- REF¹ 04880293190, CleanCell M, 2 x 2 L measuring cell cleaning solution
- REF¹ 03023141001, PC/CC-Cups, 12 cups to prewarm ProCell M and CleanCell M before use
- REF¹ 03005712190, ProbeWash M, 12 x 70 mL cleaning solution for run finalization and rinsing during reagent change
- REF¹ 12102137001, AssayTip/AssayCup Combimagazine M, 48 magazines x 8 4 reaction vessels or pipette tips, waste bags
- REF¹ 03023150001, WasteLiner, waste bags
- REF¹ 03027651001, SysClean Adapter M

Accessories for all analyzers:

- REF¹ 11298500316, 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 before use. Read in the test-specific parameter s via the reagent barcode. If in exceptional cases the barcode cannot be read, enter the 15-digit sequence of numbers.

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

Calibration

Traceability: This method has been standardized against the 2nd IRP WHO Reference Standard 80/558.

Every Elecsys TSH reagent set has a barcoded label containing the specific information required for calibration of the particular reagent lot. The predefined master curve is adapted to the analyzer by the use of Elecsys TSH 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).

Renewed calibration is recommended as follows:

- after 1 month (28 days) when using the same reagent lot
- after 7 days (when using the same reagent kit on the analyzer)
- as required: e.g. quality control findings outside the specified limits

Quality control

For quality control, use Elecsys PreciControl Universal 1 and 2, Elecsys PreciControl TSH.

Other suitable control material can be used in addition. Controls for the various concentration ranges should be run as single determinations at least once every 24 hours when the test is in use, once per reagent kit, and after every 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 limits.

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

Calculation

The analyzer automatically calculates the analyte concentration of each sample either in $\mu\text{IU/mL}$ or mIU/L (selectable).

Limitations - interference

The assay is unaffected by icterus (bilirubin < 701 $\mu\text{mol/L}$ or < 41 mg/dL), hemolysis (Hb < 0.621 mmol/L or < 1 g/dL), lipemia (Intralipid < 1500 mg/dL), and biotin < 102 nmol/L or < 25 ng/mL .

Criterion: Recovery within $\pm 10\%$ of initial value.

In patients receiving therapy with high biotin doses (i.e. > 5 mg/day), no sample should be taken until at least 8 hours after the last biotin administration.

No interference was observed from rheumatoid factor s up to a concentration of 3250 IU/mL and samples from dialysis patients.

There is no high-dose hook effect at TSH concentrations up to 1000 $\mu\text{IU/mL}$. In vitro tests were performed on 26 commonly used pharmaceuticals.

No interference with the assay was found.

The presence of autoantibodies may induce high molecular weight complexes (macro-TSH)

which may cause unexpected high values of TSH.⁸

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

0.005-100.0 $\mu\text{IU/mL}$ (defined by the lower detection limit and the maximum of the master curve). The functional sensitivity is 0.014 $\mu\text{IU/mL}$.⁶ Values below the detection limit are reported as < 0.005 $\mu\text{IU/mL}$. Values above the measuring range are reported as > 100.0 $\mu\text{IU/mL}$ (or up to 1000 $\mu\text{IU/mL}$ for 10-fold diluted samples).

Lower limits of measurement

Lower detection limit

Lower detection limit: 0.005 $\mu\text{IU/mL}$

The detection limit represents the lowest analyte level that can be distinguished from zero. It is calculated as the value lying two standard deviations above that of the lowest standard (master calibrator, standard 1 + 2 SD, repeatability study, n = 21).

Dilution

Samples with TSH concentrations above the measuring range can be diluted with Elecsys Diluent MultiAssay. The recommended dilution is 1:10 (either automatically by the MODULAR ANALYTICS E170, Elecsys 2010 and **cobas e** analyzers or manually). The concentration of the diluted sample must be > 10 $\mu\text{IU/mL}$. After manual dilution, multiply the result by the dilution factor. After dilution by the analyzers, the MODULAR ANALYTICS E170, Elecsys 2010 and **cobas e** software automatically takes the dilution into account when calculating the sample concentration.

Expected values

0.270-4.20 $\mu\text{IU/mL}$

These values correspond to the 2.5th and 97.5th percentiles of results obtained from a total of 516 healthy test subjects examined.

For detailed information about reference intervals in children, adolescents and pregnant women, refer to the brochure "Reference Intervals for Children and Adults", REF¹ English: 0 46 40292, German: 0 4625889.

This booklet also contains results of a detailed study about influencing factors on thyroid parameters in a well characterized reference group of adults. Different inclusion and exclusion criteria were applied (e.g. sonographic results (thyroid volume and density) as well as criteria according to the guidelines of the National Academy of Clinical Biochemistry - NACB).

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, pooled human sera, and controls in a modified protocol (EP5-A) of the CLSI (Clinical and Laboratory Standards Institute): 6 times daily for 10 days (n = 60); repeatability on MODULAR ANALYTICS E170 analyzer, n = 21. Elecsys PreciControl TSH was determined once daily for 10 days (n = 10). The following results were obtained:

Elecsys 2010 and cobas e 411 analyzers					
Intermediate precision		Repeatability ⁹			
CV	SD	CV	SD	Mean	Sample
%	$\mu\text{IU/mL}$	%	$\mu\text{IU/mL}$	$\mu\text{IU/mL}$	
8.7	0.003	8.6	0.003	0.034	Human serum 1
3.3	0.03	2.1	0.02	0.91	Human serum 2
3.6	0.14	1.8	0.07	3.96	Human serum 3
2.2	0.05	1.9	0.05	2.45	PreciControl Universal 1
1.8	0.19	1.5	0.16	10.67	PreciControl Universal 2
5.4	0.005	-	-	0.084	PreciControl TSH

b) Repeatability = within-run precision



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MODULAR ANALYTICS E170, cobas e 601 and cobas e 602 analyzers

Intermediate precision			Repeatability			Sample
CV	SD	Mean	CV	SD	Mean	
%	μIU/mL	μIU/mL	%	μIU/mL	μIU/mL	
7.2	0.003	0.035	3.0	0.001	0.040	Human serum 1
3.2	0.005	0.151	2.7	0.002	0.092	Human serum 2
3.3	0.120	3.66	1.1	0.102	9.37	Human serum 3
3.5	0.031	0.915	1.5	0.014	0.959	PreciControl Universal 1
4.2	0.316	7.52	1.2	0.098	8.13	PreciControl Universal 2

Method comparison

A comparison of the Elecsys TSH assay (y) with the Enzymun-Test TSH method (x) using clinical samples gave the following correlations: Number of samples measured: 109

$$\begin{array}{ll} \text{Linear regression} & \text{Passing/Bablok} \\ y = 0.98x + 0.04 & y = 1.01x + 0.01 \\ r = 0.993 & r = 0.944 \end{array}$$

The sample concentrations were between approx. 0 and 19 μIU/mL.

Analytical specificity

For the monoclonal antibodies used, the following cross-reactivities were found: LH 0.038 %, FSH 0.008 %; hGH and hCG no cross-reactivity.

Functional sensitivity

0.014 μIU/mL

The functional sensitivity is the lowest analyte concentration that can be reproducibly measured with an intermediate precision CV of 20 %.

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For further information, please refer to the appropriate operator's manual for the analyzer concerned, the respective application sheets, the product information, and the package inserts of all necessary components.

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