Elecsys FSH

cobas®

REF		Σ	SYSTEM
			cobas e 411
08932352190	08932352500	100	**cobas e** 601
			cobas e 602

English

System information

For **cobas e** 411 analyzer: test number 2350
For **cobas e** 601 and **cobas e** 602 analyzers: Application Code Number 99

Intended use

Immunoassay for the in vitro quantitative determination of follicle-stimulating hormone in human serum and plasma.

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

Summary

FSH (follicle stimulating hormone), together with LH (luteinizing hormone), belongs to the gonadotropin family. FSH and LH regulate and stimulate the growth and function of the gonads (ovaries and testes) synergistically.[1]

Like LH, TSH and hCG, FSH is a glycoprotein consisting of two subunits (α-and β-chains). Its molecular weight is approximately 32000 daltons.

In women FSH, in conjunction with LH, stimulates oestrogen secretion and ovulation.[2]

FSH and LH are released in pulses from the gonadotropic cells of the anterior pituitary. The levels of the circulating hormones are controlled by steroid hormones via negative feedback to the hypothalamus. In the ovaries FSH, together with LH, stimulates the growth and maturation of the follicle[2] and hence also the biosynthesis of estrogens in the follicles.

The FSH level shows a peak at mid-cycle, although this is less marked than with LH. Due to changes in ovarian function and reduced estrogen secretion, high FSH concentrations occur during menopause.[3]

In men, FSH serves to induce spermatogonium development.[2]

Determination of the FSH concentration is used in the elucidation of dysfunctions within the hypothalamus-pituitary-gonads system.

The determination of FSH in conjunction with LH is utilized for the following indications: congenital diseases with chromosome aberrations, polycystic ovaries (PCO), amenorrhea (causes), and menopausal syndrome. Depressed gonadotropin levels in men occur in azoospermia.[4]

The Elecsys FSH assay employs two different monoclonal antibodies specifically directed against human FSH. Cross-reactivity with LH, TSH, hCG, hGH, and hPL is negligible.

Test principle

Sandwich principle. Total duration of assay: 18 minutes.

- 1st incubation: 40 µL of sample, a biotinylated monoclonal FSH-specific antibody, and a monoclonal FSH-specific antibody labeled with a ruthenium complex[a)] 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.

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

Reagents - working solutions

The reagent rackpack is labeled as FSH.

M Streptavidin-coated microparticles (transparent cap), 1 bottle, 6.5 mL:
Streptavidin-coated microparticles 0.72 mg/mL; preservative.

R1 Anti-FSH-Ab~biotin (gray cap), 1 bottle, 10 mL:
Biotinylated monoclonal anti-FSH antibody (mouse) 0.5 mg/L, MES buffer 50 mmol/L, pH 6.0; preservative.

R2 Anti-FSH-Ab~$Ru(bpy)_3^{2+}$ (black cap), 1 bottle, 10 mL:
Monoclonal anti-FSH antibody (mouse) labeled with ruthenium complex 0.8 mg/L, MES buffer 50 mmol/L, pH 6.0; preservative.

Precautions and warnings

For in vitro diagnostic use for health care 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 dust/fume/gas/mist/vapours/spray.
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.

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.

Elecsys FSH

cobas®

Stability:	
unopened at 2-8 °C	up to the stated expiration date
after opening at 2-8 °C	12 weeks
on the analyzers	8 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, K_2-EDTA and K_3-EDTA plasma.

Criterion: Slope 0.9-1.1 + bias at 10 mIU/mL ≤ 10 % + coefficient of correlation ≥ 0.95.

Stable for 5 days at 20-25 °C, 14 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 heat-inactivated samples.

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 08932417190, FSH CalSet II, for 4 x 1.0 mL
- REF 11731416190, PreciControl Universal, 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.

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

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.

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 the Enzymun-Test FSH method. This in turn has been standardized against the 2nd IRP WHO reference standard 78/549.

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:

- 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 defined limits

Quality control

For quality control, use PreciControl Universal.

In addition, other suitable control material can be used.

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.

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 mIU/mL or in IU/L).

Limitations - interference

Endogenous substances

Compound	Concentration tested
Bilirubin	≤ 1112 µmol/L or ≤ 65 mg/dL
Hemoglobin	≤ 0.621 mmol/L or ≤ 1000 mg/dL
Intralipid	≤ 1900 mg/dL
Biotin	≤ 4912 nmol/L or ≤ 1200 ng/mL
Rheumatoid factors	≤ 1200 IU/mL

Criterion: For concentrations from 0.3-20 mIU/mL the deviation is ± 2.5 mIU/mL. For concentrations from 20-200 mIU/mL the deviation is ± 10 %.

There is no high-dose hook effect at FSH concentrations up to 2000 mIU/mL.

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

Elecsys FSH

cobas®

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

0.3-200 mIU/mL (defined by the Limit of Detection and the maximum of the master curve). Values below the Limit of Detection are reported as < 0.3 mIU/mL. Values above the measuring range are reported as > 200 mIU/mL.

Lower limits of measurement

Limit of Blank, Limit of Detection and Limit of Quantitation

Limit of Blank = 0.1 mIU/mL

Limit of Detection = 0.3 mIU/mL

Limit of Quantitation = 1 mIU/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 CV of ≤ 20 %.

Dilution

Not necessary due to the broad measuring range.

Expected values

Studies with the Elecsys FSH assay have revealed the following FSH values:

Test subjects	N	FSH (mIU/mL)		
		Percentile		
		50th	5th	95th
Men	319	4.6	1.5	12.4
Women				
• Follicular phase	376	6.9	3.5	12.5
• Ovulation phase	56	12.3	4.7	21.5
• Luteal phase	349	3.6	1.7	7.7
• Postmenopause	181	67.0	25.8	134.8

LH/FSH quotient: Quotients have been calculated from the results obtained with the Elecsys LH assay and the Elecsys FSH assay in the samples of healthy women of child-bearing age. The following medians have been calculated:

Follicular phase: 0.82 (n = 315)

Luteal phase: 1.12 (n = 279)

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 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					
		Repeatability		**Intermediate precision**	
Sample	Mean mIU/mL	SD mIU/mL	CV %	SD mIU/mL	CV %
Human serum 1	1.40	0.021	1.5	0.055	3.9
Human serum 2	10.7	0.205	1.9	0.401	3.7
Human serum 3	41.5	0.547	1.3	1.31	3.2
Human serum 4	107	1.52	1.4	3.69	3.4
Human serum 5	180	3.17	1.8	7.32	4.1
PC[b)] Universal 1	20.6	0.259	1.3	0.805	3.9
PC Universal 2	50.2	0.950	1.9	2.06	4.1

b) PC = PreciControl

cobas e 601 and **cobas e** 602 analyzers					
		Repeatability		**Intermediate precision**	
Sample	Mean mIU/mL	SD mIU/mL	CV %	SD mIU/mL	CV %
Human serum 1	1.29	0.017	1.3	0.039	3.0
Human serum 2	10.0	0.162	1.6	0.358	3.6
Human serum 3	79.9	1.41	1.8	2.99	3.7
Human serum 4	128	1.49	1.2	4.98	3.9
Human serum 5	184	2.40	1.3	7.63	4.1
PC[b)] Universal 1	18.2	0.252	1.4	0.784	4.3
PC Universal 2	44.0	0.932	2.1	1.85	4.2

Method comparison

A comparison of the Elecsys FSH assay, REF 08932352190 (**cobas e** 601 analyzer; y) with the Elecsys FSH assay, REF 11775863122 (**cobas e** 601 analyzer; x) gave the following correlations (mIU/mL):

Number of samples measured: 170

Passing/Bablok[5]	Linear regression
y = 0.979x + 0.092	y = 1.01x - 1.69
τ = 0.983	r = 0.998

The sample concentrations were between 1.46 and 182 mIU/mL.

Analytical specificity

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

Substance	Cross-reactivity %	Additive concentration mIU/mL
LH	0.022	5000
TSH	n.d.[c)]	5000
hCG	0.004	5000
hGH	n. d.	2000
hPL	n. d.	5000

c) n. d. = not detectable

References

1 Johnson MR, Carter G, Grint C, et al. Relationship between ovarian steroids, gonadotropin and relaxin during the menstrual cycle. Acta Endocrinol 1983;129/2:121-125.

2 Beastall GH, Ferguson KM, O'Reilly DSJ, et al. Assays for follicle stimulating hormone and luteinizing hormone: Guidelines for the provision of a clinical biochemistry service. Ann Clin Biochem 1987;24:246-262.

3 Scott MG, Ladenson JH, Green ED, et al. Hormonal evaluation of female infertility and reproductive disorders. Clin Chem 1989;35:620-630.

4 Gudeloglu A, Parekattil SJ. Update in the evaluation of the azoospermic male. Clinics (Sao Paulo) 2013;68(Suppl 1):27-34.

5 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 operator's manual for the analyzer concerned, the respective application sheets, the product information 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 (for USA: see dialog.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

CE 0123

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

Roche