

CYFRA 21-1

REF		SYSTEM
11820966 122	100	Elecsys 2010 MODULAR ANALYTICS E170 cobas e 411 cobas e 601 cobas e 602

English**Please note**

The measured CYFRA 21-1 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 CYFRA 21-1 assay method used. CYFRA 21-1 values determined on patient samples by different testing procedures cannot be directly compared with one another and could be the cause of erroneous medical interpretations. If there is a change in the CYFRA 21-1 assay procedure used while monitoring therapy, then the CYFRA 21-1 values obtained upon changing over to the new procedure must be confirmed by parallel measurements with both methods.

Intended use

Immunoassay for the in vitro quantitative determination of fragments of cytokeratin 19 in human serum and plasma.

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

Summary

Cytokeratins are structural proteins forming the subunits of epithelial intermediary filaments. Twenty different cytokeratin polypeptides have so far been identified. Due to their specific distribution patterns they are eminently suitable for use as differentiation markers in tumor pathology. Intact cytokeratin polypeptides are poorly soluble, but soluble fragments can be detected in serum.^{1,2,3,4}

With the aid of two specific monoclonal antibodies (KS 19.1 and BM 19.21), CYFRA 21-1 measures a fragment of cytokeratin 19 having a molecular weight of approximately 30000 daltons.⁵

The main indication for CYFRA 21-1 is monitoring the course of non-small cell lung cancer (NSCLC).^{2,5,6}

CYFRA 21-1 is also suitable for course-monitoring in myoinvasive cancer of the bladder.⁷

Good specificity is shown by CYFRA 21-1 relative to benign lung diseases (pneumonia, sarcoidosis, tuberculosis, chronic bronchitis, bronchial asthma, emphysema).^{8,9}

Slightly elevated values (up to 10 ng/mL) are rarely found in marked benign liver diseases and renal failure. There is no correlation with sex, age or smoking.¹⁰ The values are also unaffected by pregnancy.

The primary diagnosis of pulmonary carcinoma should be made on the basis of clinical symptomatology, imaging or endoscopic procedures and intraoperative findings.

An unclear circular focus in the lung together with CYFRA 21-1 values > 30 ng/mL indicate with high probability the existence of primary bronchial carcinoma.

High CYFRA 21-1 serum levels indicate an advanced tumor stage and a poor prognosis.¹¹ A normal or only slightly elevated value does not rule out the presence of a tumor.

Successful therapy is documented by a rapid fall in the CYFRA 21-1 serum level into the normal range. A constant CYFRA 21-1 value or a slight or only slow decrease in the CYFRA 21-1 value indicates incomplete removal of a tumor or the presence of multiple tumors with corresponding therapeutic and prognostic consequences. Progression of the disease is often shown earlier by increasing CYFRA 21-1 values than by clinical symptomatology and imaging procedures.

Test principle

Sandwich principle. Total duration of assay: 18 minutes.

- 1st incubation: 20 µL of sample, a biotinylated monoclonal cytokeratin 19-specific antibody, and a monoclonal cytokeratin 19-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.

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

Reagents - working solutions

The reagent rackpack is labeled as CYFRA.

- M Streptavidin-coated microparticles (transparent cap), 1 bottle, 6.5 mL:
Streptavidin-coated microparticles 0.72 mg/mL; preservative.
- R1 Anti-cytokeratin 19-Ab-biotin (gray cap), 1 bottle, 10 mL:
Biotinylated monoclonal anti-cytokeratin 19 antibody (KS 19.1; mouse) 1.5 mg/L, phosphate buffer 100 mmol/L, pH 7.2; preservative.
- R2 Anti-cytokeratin 19-Ab-Ru(bpy)₃²⁺ (black cap), 1 bottle, 10 mL:
Monoclonal anti-cytokeratin 19 antibody (BM 19.21; mouse) labeled with ruthenium complex 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.

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

2-methyl-2H-isothiazol-3-one hydrochloride

EUH 208 May produce an allergic reaction.

Product safety labeling primarily follows EU GHS guidance.

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.

CYFRA 21-1

CYFRA 21-1

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-, Na-heparin, K₃-EDTA and sodium citrate plasma. When sodium citrate is used, the results must be corrected by + 10 %.

Criterion: Recovery within 90-110 % of serum value or slope 0.9-1.1 + intercept within $< \pm 2x$ analytical sensitivity (LDL) + coefficient of correlation > 0.95 .

Stable for 4 weeks at 2-8 °C, 6 months at -20 °C. Freeze only once.¹²

It is recommended that the samples be mixed by careful swirling or by placing on a roller mixer (max. 5 min). Homogenization of samples using electric vibration mixers must be limited to a maximum of 5 seconds. Longer mixing times lead to lower values being found.

Contamination of the sample with saliva leads to falsely elevated results.¹²

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] 11820974322, CYFRA 21-1 CalSet, for 4 x 1 mL
- [REF] 11776452122, PreciControl Tumor Marker, for 2 x 3 mL each of PreciControl Tumor Marker 1 and 2
- [REF] 07360070190, PreciControl Lung Cancer, for 4 x 3 mL each of PreciControl Lung Cancer 1 and 2
- [REF] 11732277122, Diluent Universal, 2 x 16 mL sample diluent or [REF] 03183971122, Diluent Universal, 2 x 36 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] 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] 12102137001, AssayTip/AssayCup Combimagazine M, 48 magazines x 84 reaction vessels or pipette tips, waste bags

- [REF] 03023150001, WasteLiner, waste bags

- [REF] 03027651001, SysClean Adapter M

Accessories 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.

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 CYFRA 21-1 method.

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). Renewed calibration is recommended as follows:

- after 8 weeks 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 Lung Cancer or PreciControl Tumor Marker.

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.

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 assay is unaffected by icterus (bilirubin < 1112 µmol/L or < 65 mg/dL), hemolysis (Hb < 0.93 mmol/L or < 1.5 g/dL), lipemia (Intralipid < 1500 mg/dL) and biotin (< 205 nmol/L or < 50 ng/mL).

Criterion: Recovery within ± 10 % of initial value.

Samples should not be taken from patients receiving therapy with high biotin doses (i.e. > 5 mg/day) until at least 8 hours following the last biotin administration.

No interference was observed from rheumatoid factors up to a concentration of 1500 IU/mL.

There is no high-dose hook effect at CYFRA 21-1 concentrations up to 2000 ng/mL.

In vitro tests were performed on 28 commonly used pharmaceuticals. No interference with the assay was found.

CYFRA 21-1

CYFRA 21-1



- 12 Guder WG, Narayanan S, Wisser H, et al. List of Analytes; Preanalytical Variables. Brochure in: Samples: From the Patient to the Laboratory. GIT-Verlag, Darmstadt 1996:12. ISBN 3-928865-22-6.
- 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 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.

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 after reconstitution or mixing
	Global Trade Item Number

COBAS, COBAS E, ELECSYS, CYFRA 21-1 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.

© 2016, Roche Diagnostics



Roche Diagnostics GmbH, Sandhofer Strasse 116, D-68305 Mannheim
www.roche.com

