KAPP2

cobas®

Tina-quant Kappa Gen.2

REF	CONTENT		Analyzer(s) on which kit(s) can be used
06749976 190	Tina-quant Kappa Gen.2 100 tests	System ID 07 6811 1	Roche/Hitachi **cobas c** 311, **cobas c** 501/502
11355279 216	Calibrator f.a.s. Proteins (5 x 1 mL)	Code 656	
10557897 122	Precinorm Protein (3 × 1 mL)	Code 302	
11333127 122	Precipath Protein (3 × 1 mL)	Code 303	
04489357 190	Diluent NaCl 9 % (50 mL)	System ID 07 6869 3	
05117003 190	PreciControl ClinChem Multi 1 (20 × 5 mL)	Code 391	
05947626 190	PreciControl ClinChem Multi 1 (4 × 5 mL)	Code 391	
05117216 190	PreciControl ClinChem Multi 2 (20 × 5 mL)	Code 392	
05947774 190	PreciControl ClinChem Multi 2 (4 × 5 mL)	Code 392	

English

System information

For **cobas c** 311/501 analyzers:

KAPP2: ACN 283

For **cobas c** 502 analyzer:

KAPP2: ACN 8283

Intended use

Immunoturbidimetric in vitro assay for the quantitative determination of bound and free immunoglobulins of the kappa light chain type in human serum and plasma on Roche/Hitachi **cobas c** systems.

Summary

References[1,2,3,4,5,6,7]

Measurement of the various amounts of the different types of light chains aids in the diagnosis of multiple myeloma, lymphocytic neoplasms, Waldenström's macroglobulinemia, and connective tissue diseases such as rheumatoid arthritis or systemic lupus erythematosus.

Every plasma cell clone normally produces a uniform immunoglobulin molecule of the kappa or lambda light chain type. The kappa:lambda ratio in serum is normally around 2:1.

Pathological increases of a cell clone lead to elevated formation of monoclonal immunoglobulins or immunoglobulin fragments (free light chains), which bring about a change in the kappa:lambda ratio. A kappa:lambda ratio outside the normal range is indicative of monoclonal gammopathy.

This test encompasses both bound and free immunoglobulins of the light chain type.

It is known that the so-called paraproteins secreted in monoclonal gammopathies (monoclonal immunoglobulinemia) may differ from the respective immunoglobulins of polyclonal origin in amino acid composition and size. This may impair the binding to antibody and consequently cause antigen excess below the limits determined with immunoglobulins of polyclonal origin. Antigen excess may be detected after appropriate dilution of such samples.

Furthermore, the occurrence of two monoclonal gammopathies producing differing light chain types could theoretically lead to kappa:lambda ratios in the normal range.

Accordingly, quantitative determination of the kappa and lambda light chains cannot completely replace high-resolution electrophoresis, immunoelectrophoresis or immunofixation electrophoresis in the diagnosis of monoclonal gammopathy.

Test principle

Immunoturbidimetric assay

Anti-kappa antibodies react with antigen in the sample to form antigen/antibody complexes that, following agglutination, are measured turbidimetrically.

Reagents - working solutions

R1 TRIS/HCl buffer: 50 mmol/L, pH 8.0; PEG 7 %; stabilizers and preservative

R2 Polyclonal anti-human kappa antibody (goat), dependent on titer; TRIS/HCl buffer: 20 mmol/L, pH 7.5; stabilizers and preservative

R1 is in position B and R2 is in position C.

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.

Storage and stability

KAPP2

Shelf life at 2-8 °C:	See expiration date on **cobas c** pack label.
On-board in use and refrigerated on the analyzer:	12 weeks

Diluent NaCl 9 %

Shelf life at 2-8 °C:	See expiration date on **cobas c** pack label
On-board in use and refrigerated on the analyzer:	12 weeks

Specimen collection and preparation

For specimen collection and preparation only use suitable tubes or collection containers.

Only the specimens listed below were tested and found acceptable.

Serum

Plasma: Li-heparin or K_2-EDTA plasma.

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.

Stability:[8] 7 days at 15-25 °C
4 weeks at 4-8 °C
2 months at (-15)-(-25) °C

Materials provided

- See "Reagents – working solutions" section for reagents.

KAPP2

Tina-quant Kappa Gen.2

cobas®

Materials required (but not provided)

- See “Order information” section

General laboratory equipment

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.

The performance of applications not validated by Roche is not warranted and must be defined by the user.

Application for serum and plasma

cobas c 311 **test definition**

Assay type	2-Point End		
Reaction time / Assay points	10 / 6-57		
Wavelength (sub/main)	800/340 nm		
Reaction direction	Increase		
Units	g/L (mg/dL)		
Reagent pipetting		Diluent (H_2O)	
R1	125 µL	–	
R2	50 µL	–	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	3.3 µL	9 µL	180 µL
Decreased	3.3 µL	3 µL	150 µL
Increased	3.3 µL	18 µL	75 µL

cobas c 501/502 **test definition**

Assay type	2-Point End		
Reaction time / Assay points	10 / 10-70		
Wavelength (sub/main)	800/340 nm		
Reaction direction	Increase		
Units	g/L (mg/dL)		
Reagent pipetting		Diluent (H_2O)	
R1	125 µL	–	
R2	50 µL	–	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	3.3 µL	9 µL	180 µL
Decreased	3.3 µL	3 µL	150 µL
Increased	3.3 µL	18 µL	75 µL

Calibration

Calibrators	S1-S6: C.f.a.s. Proteins Multiply the lot-specific C.f.a.s. Proteins calibrator value by the factors below to determine the standard concentrations for the 6-point calibration curve:
	S1: 0.140 S4: 1.29
	S2: 0.209 S5: 2.03
	S3: 0.640 S6: 3.05
Calibration mode	RCM2
Calibration frequency	Full calibration - after reagent lot change - and as required following quality control procedures

Traceability: This method has been standardized against the CRM 470 standard using the Lievens equation.[6]

Quality control

For quality control, use control materials as listed in the "Order information" section.

In addition, other suitable control material can be used.

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

Roche/Hitachi **cobas c** systems automatically calculate the analyte concentration of each sample.

Conversion factors:	mg/dL x 0.01 = g/L
	g/L x 100 = mg/dL

Limitations - interference

Criterion: Recovery within ± 10 % of initial value at a kappa concentration of 1.4 g/L (140 mg/dL).

Icterus:[9] No significant interference up to an I index of 60 for conjugated and unconjugated bilirubin (approximate conjugated and unconjugated bilirubin concentration: 1026 µmol/L or 60 mg/dL).

Hemolysis:[9] No significant interference up to an H index of 1000 (approximate hemoglobin concentration: 621 µmol/L or 1000 mg/dL).

Lipemia (Intralipid):[9] No significant interference up to an L index of 2000. There is poor correlation between the L index (corresponds to turbidity) and triglycerides concentration.

Drugs: No interference was found at therapeutic concentrations using common drug panels.[10, 11]

High dose hook-effect: No false result occurs up to a kappa concentration of 20 g/L.

Rheumatoid factors < 500 IU/mL do not interfere.

Samples from patients with unclear clinical diagnosis should be subject to protein electrophoresis to identify a possible antigen excess or monoclonal gammopathy. Antigen excess may be detected by appropriate predilution of the specimen with 0.9 % sodium chloride solution.

In sera with monoclonal kappa components, differing results may be obtained with commercial assays employing antibodies from different sources (rabbit, sheep, goat).

In very rare cases, gammopathy, in particular type IgM (Waldenström’s macroglobulinemia), may cause unreliable results.[12]

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

ACTION REQUIRED

Special Wash Programming: The use of special wash steps is mandatory when certain test combinations are run together on Roche/Hitachi **cobas c** systems. The latest version of the carry-over evasion list can be found with the NaOHD/SMS/Multiclean/SCCS or the NaOHD/SMS/SmpCln1+2/SCCS Method Sheets. For further instructions refer to the operator’s manual. **cobas c** 502 analyzer: All special wash programming necessary for avoiding carry-over is available via the **cobas** link, manual input is not required.

Where required, special wash/carry-over evasion programming must be implemented prior to reporting results with this test.

Limits and ranges

Measuring range

1.0-12.0 g/L (100-1200 mg/dL)

Determine samples having higher concentrations via the rerun function. Dilution of samples via the rerun function is a 1:2.4 dilution. Results from samples diluted using the rerun function are automatically multiplied by a factor of 2.4.

Determine samples having lower concentrations via the rerun function. For samples with lower concentrations, the rerun function increases the sample volume by a factor of 4.1. The results are automatically divided by this factor.

Lower limits of measurement

Lower detection limit of the test

0.4 g/L (40 mg/dL)

The lower detection limit represents the lowest measurable analyte level that can be distinguished from zero. It is calculated as the value lying three standard deviations above that of the lowest standard (standard 1 + 3 SD, repeatability, n = 21).

Expected values

	kappa	kappa/lambda ratio
Serum[6,13,14,15]	1.38-3.75 g/L	1.17-2.93

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 human samples and controls in an internal protocol with repeatability (n = 21) and intermediate precision (3 aliquots per run, 1 run per day, 21 days). The following results were obtained:

Repeatability	*Mean*	*SD*	*CV*
	g/L (mg/dL)	*g/L (mg/dL)*	*%*
Precinorm Protein	2.03 (203)	0.04 (4)	1.9
Precipath Protein	4.01 (401)	0.04 (4)	0.9
Human serum 1	1.97 (197)	0.02 (2)	1.3
Human serum 2	4.09 (409)	0.05 (5)	1.3
Intermediate precision	*Mean*	*SD*	*CV*
	g/L (mg/dL)	*g/L (mg/dL)*	*%*
Precinorm Protein	1.96 (196)	0.06 (6)	2.9
Precipath Protein	3.99 (399)	0.09 (9)	2.4
Human serum 1	1.93 (193)	0.04 (4)	1.9
Human serum 2	3.99 (399)	0.06 (6)	1.6

Method comparison

Kappa light chain values for human serum samples obtained on a Roche/Hitachi **cobas c** 501 analyzer (y) were compared with those determined using the same reagent on a Roche/Hitachi 917 analyzer (x).

Sample size (n) = 274

Passing/Bablok[16]	Linear regression
y = 1.000x + 0.155 g/L	y = 1.005x + 0.149 g/L
τ = 0.940	r = 0.997

The sample concentrations were between 1.02 and 9.63 g/L (102 and 963 mg/dL).

References

1 Skvaril F, Barandum S, Morell A, et al. Imbalances of kappa/lambda immunoglobulin light chain ratios in normal individuals and in immunodeficient patients. In: Proteides of biological fluids, Peeters H, ed 1975;23:415-420.

2 Sun T, de Szalay H, Lien YY, et al. Quantitation of kappa and lambda light chains for the detection of monoclonal gammopathy. J Clin Lab Anal 1988;2:84-90.

3 Whicher JT, Wallage M, Fifield R. Use of immunoglobulin heavy- and light-chain measurements compared with existing techniques as a means of typing monoclonal immunoglobulins. Clin Chem 1987;33:1771-1773.

4 Keren DF, Warren JS, Lowe JB. Strategy to diagnose monoclonal gammopathies in serum: high-resolution electrophoresis, immunofixation and kappa/lambda quantification. Clin Chem 1988;34:2196-2201.

5 Duc J, Morel B, Peitrequin R, et al. Identification of monoclonal gammopathies: a comparison of immunofixation, immunoelectrophoresis and measurements of kappa- and lambda-immunoglobulin levels. J Clin Lab Immunol 1988;26:141-146.

6 Lievens M. Medical and technical usefulness of measurement of kappa and lambda immunoglobulin light chains in serum with an M-component. J Clin Chem Clin Biochem 1989;27:519-523.

7 Whicher JT, Ritchie RF, Johnson AM, et al. New international reference preparation for proteins in human serum (RPPHS). Clin Chem 1994;40:934-938.

8 Wu AHB, ed. Tietz Clinical Guide to Laboratory Tests, 4th ed. St. Louis (MO): Saunders Elsevier 2006:674-675.

9 Glick MR, Ryder KW, Jackson SA. Graphical Comparisons of Interferences in Clinical Chemistry Instrumentation. Clin Chem 1986;32:470-475.

10 Breuer J. Report on the Symposium “Drug effects in Clinical Chemistry Methods”. Eur J Clin Chem Clin Biochem 1996;34:385-386.

11 Sonntag O, Scholer A. Drug interference in clinical chemistry: recommendation of drugs and their concentrations to be used in drug interference studies. Ann Clin Biochem 2001;38:376-385.

12 Bakker AJ, Mücke M. Gammopathy interference in clinical chemistry assays: mechanisms, detection and prevention. Clin Chem Lab Med 2007;45(9):1240-1243.

13 Hafner G, Endler T, Oppitz M, et al. Effects of standardization with the new international reference preparation for proteins in human serum on method comparability and reference values. Clin Lab 1995;41:743-748.

14 Jones RG, Aguzzi F, Bienvenu J, et al. Use of Immunoglobulin Heavy-chain and Light-chain measurement in a multicenter trial to investigate Monoclonal components: I. Detection. Clin Chem 1991;37:1917-1921.

15 Jones RG, Aguzzi F, Bienvenu J, et al. Use of Immunoglobulin Heavy-chain and Light-chain measurement in a multicenter trial to investigate Monoclonal components: II. Classification by use of Computer-based algorithms. Clin Chem 1991;37:1922-1926.

16 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.

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.

CONTENT	Contents of kit
→	Volume after reconstitution or mixing

0106749976190c501V2.0

KAPP2

Tina-quant Kappa Gen.2

CE

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