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Tina-quant Cystatin C

Order information

REF	CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
04975723 190	Tina-quant Cystatin C 225 tests	System-ID 07 6988 6	Roche/Hitachi **cobas c** 311, **cobas c** 501/502
04975901 190	C.f.a.s. Cystatin C (4 x 1 mL)	Code 404	
04975936 190	Cystatin C Control Set (2 x 4 x 1 mL)	Code 121 Control I (low) Code 122 Control II (high)	
04489357 190	Diluent NaCl 9 % (50 mL)	System-ID 07 6869 3	

English

System information

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

CYSC: ACN 431

For **cobas c** 502 analyzer:

CYSC: ACN 8431

Intended use

In vitro test for the quantitative determination of cystatin C in human serum and plasma on Roche/Hitachi **cobas c** systems.

Summary[1,2,3,4,5,6,7,8,9,10,11,12,13,14]

Chronic kidney disease is a worldwide health problem that carries a substantial risk for cardiovascular morbidity and death. Current guidelines define chronic kidney disease as kidney damage or glomerular filtration rate (GFR) less than 60 mL/min per 1.73 m^2 for 3 months or more, regardless of cause. GFR is the most frequently used criteria in the assessment of renal function.

Serum creatinine is the most commonly used marker for estimation of GFR. However, it has become evident that the creatinine concentration is far from ideal because it is significantly changed by other factors such as muscle mass, diet, gender, age and tubular secretion. To compensate for these drawbacks several prediction equations have been developed, with the Modification of Diet in Renal Disease (MDRD), the Schwartz, and Counahan-Baratt equations being the ones most widely accepted.

Creatinine clearance measurements using serum and urine creatinine levels suffer from a similar problem, since the creatinine is endogenous and the factors mentioned above also complicate the interpretation.

Cystatin C is produced by all nucleated cells at a constant rate and the production rate in humans is remarkably constant over the entire lifetime. Elimination from the circulation is almost entirely via glomerular filtration. For this reason the serum concentration of cystatin C is independent from muscle mass and gender in the age range 1 to 50 years. Therefore cystatin C in plasma and serum has been proposed as a more sensitive marker for GFR, and several studies, as well as one meta analysis, have suggested that cystatin C is superior to serum creatinine for estimation of GFR. Patient groups which benefit most are those with mild to moderate kidney disease and also those in acute renal failure, where toxic drugs have to be administered which are excreted by glomerular filtration, especially elder people (> 50 years), children, pregnant women with suspicion of pre-eclampsia, diabetics, people with diseases of skeletal muscle and renal transplant recipients. Additionally cystatin C has been discussed in recent literature as a prognostic marker for acute heart failure.

As with creatinine several cystatin C based prediction equations for calculation of GFR for adults and children have been published. It should be noted that these formulas were evaluated with different cystatin C assays (particle-enhanced nephelometric immunoassay PENIA or particle enhanced turbidimetric immunoassay PETIA) and may reveal inaccurate GFR results if an inappropriate combination of formula and assay is used. For calculation of GFR from cystatin C values measured with the Roche assay the following prediction equation is recommended using only concentration in mg/L and a prepubertal factor:

$$\text{GFR [mL/min/1.73 m}^2\text{]} = \frac{84.69}{\text{cystatin C (mg/L)}^{1.680}} \times 1.384^*$$

*for children < 14 y

Test principle[5]

Particle enhanced immunoturbidimetric assay.

Human cystatin C agglutinates with latex particles coated with anti-cystatin C antibodies. The aggregate is determined turbidimetrically at 546 nm.

Reagents - working solutions

R1 Solution of polymers in MOPS-buffered saline; preservative, stabilizers

R2 Latex particles in glycine buffer coated with anti-cystatin C antibodies (rabbit); preservative, stabilizers

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.

Reagent handling

Ready for use

Mix **cobas c** pack well before placing on the analyzer.

Carefully invert reagent container several times prior to use to ensure that the reagent components are mixed.

Storage and stability

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Shelf life at 2-8 °C:	See expiration date on **cobas c** pack label.
On-board in use and refrigerated on the analyzer:	8 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[7]

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 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.

Blood collected in capillary blood collection tubes is unsuitable for use in this assay.

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

Frozen samples should be thawed carefully and mixed well before analysis.

Materials provided

See "Reagents – working solutions" section for reagents.

Materials required (but not provided)

- See "Order information" section

General laboratory equipment

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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 / 8-57		
Wavelength (sub/main)	700/546 nm		
Reaction direction	Increase		
Units	mg/L		
Reagent pipetting		Diluent (H_2O)	
R1	154 µL	–	
R2	34 µL	20 µL	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2 µL	–	–
Decreased	8 µL	15 µL	75 µL
Increased	2 µL	–	–

cobas c 501 **test definition**

Assay type	2-Point End		
Reaction time / Assay points	10 / 13-70		
Wavelength (sub/main)	700/546 nm		
Reaction direction	Increase		
Units	mg/L		
Reagent pipetting		Diluent (H_2O)	
R1	154 µL	–	
R2	34 µL	20 µL	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2 µL	–	–
Decreased	8 µL	15 µL	75 µL
Increased	2 µL	–	–

cobas c 502 **test definition**

Assay type	2-Point End		
Reaction time / Assay points	10 / 13-70		
Wavelength (sub/main)	700/546 nm		
Reaction direction	Increase		
Units	mg/L		
Reagent pipetting		Diluent (H_2O)	
R1	154 µL	–	
R2	34 µL	20 µL	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2 µL	–	–
Decreased	8 µL	15 µL	75 µL
Increased	4 µL	–	–

Calibration

Calibrators	S1-6: C.f.a.s. Cystatin C Multiply the lot-specific C.f.a.s. Cystatin C calibrator value by the factors below to determine the standard concentrations for the 6-point calibration curve:
	S1: 0.055 S4: 0.428
	S2: 0.107 S5: 0.608
	S3: 0.250 S6: 1
Calibration mode	Spline
Calibration frequency	Full calibration • after reagent lot change and after 90 days • as required following quality control procedures

Traceability: This method has been standardized against an in-house reference preparation of pure recombinant human cystatin C. The cystatin C concentration of this reference preparation was established by dry mass determination as described in the reference Bilrup-Jensen.[16]

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.

Limitations - interference

Criterion: Recovery within ± 10 % of initial value.

Icterus:[17] No significant interference up to an I index of 60 (approximate conjugated and unconjugated bilirubin concentration: 1026 µmol/L or 60 mg/dL).

Hemolysis:[17] No significant interference up to an H index of 700 (approximate hemoglobin concentration: 435 µmol/L or 700 mg/dL).

Lipemia (Intralipid):[17] No significant interference up to an L index of 1000. There is poor correlation between the L index (corresponds to turbidity) and triglycerides concentration.

Rheumatoid factors < 1200 IU/mL do not interfere.

High dose hook-effect: No false result occurs up to a cystatin C concentration of 20 mg/L.

Drugs: No interference was found at therapeutic concentrations using common drug panels.[18,19]

In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.[20]

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

0.4-8.0 mg/L

Determine samples having higher concentrations via the rerun function. Dilution of samples via the rerun function is a 1:1.5 dilution. Results from samples diluted using the rerun function are automatically multiplied by a factor of 1.5.

Lower limits of measurement

Limit of Blank (LoB) and Limit of Detection (LoD)

LoB = 0.3 mg/L

LoD = 0.4 mg/L

The Limit of Blank and Limit of Detection were determined in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP17-A 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 %).

Expected values[15]

For individuals 20-70 years (n = 500)[a)]: 0.47-1.09 mg/L

a) measured in a well characterized reference population of healthy donors on a Roche/Hitachi system

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, 10 days). The following results were obtained:

Repeatability	*Mean*	*SD*	*CV*
	mg/L	*mg/L*	*%*
Cystatin C Control Set Control Low	1.05	0.01	0.8
Cystatin C Control Set Control High	4.45	0.03	0.8
Human serum 1	0.52	0.01	2.7
Human serum 2	6.30	0.04	0.6
Intermediate precision	*Mean*	*SD*	*CV*
	mg/L	*mg/L*	*%*
Cystatin C Control Set Control Low	1.00	0.03	2.9
Cystatin C Control Set Control High	4.36	0.09	1.9
Human serum 3	0.65	0.02	3.8
Human serum 4	7.16	0.19	2.6

Method comparison

Cystatin C values for human serum and plasma samples obtained on a Roche/Hitachi **cobas c** 501 analyzer (y) were compared with those determined using the corresponding reagent on a Roche/Hitachi 917 analyzer (x).

Sample size (n) = 64

Passing/Bablok[21]	Linear regression
y = 0.988x + 0.021 mg/L	y = 0.990x + 0.012 mg/L
τ = 0.980	r = 0.999

The sample concentrations were between 0.400 and 7.15 mg/L.

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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

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