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CYSC2

Tina-quant Cystatin C Gen.2

cobas®

Order information

REF	CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
06600239 190	Tina-quant Cystatin C Gen.2 (225 tests)	System-ID 07 7550 9	Roche/Hitachi **cobas c** 311, **cobas c** 501/502
04975901 191	C.f.a.s. Cystatin C (4 × 1 mL)	Code 407	
04975936 190*	Cystatin C Control Set Control I (low) (4 × 1 mL) Control II (high) (4 × 1 mL)	Code 121 Code 122	
06729371 190	Cystatin C Control Set Gen.2 Control 1 (3 × 1 mL) Control 2 (3 × 1 mL) Control 3 (3 × 1 mL)	Code 139 Code 140 Code 141	

*Not for use in the US

English

System information

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

CYSC2: ACN 109

For **cobas c** 502 analyzer:

CYSC2: ACN 8109

Intended use

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

Summary[1,2,3,4,5,6,7,8,9,10,11,12,13,14,15,16,17,18,19,20]

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.

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. There is a small dependency of cystatin C concentration from age in the age range 1 to 50 years whereas the cystatin C concentration of healthy individuals > 50 years increases with age. Therefore, cystatin C in plasma and serum has been proposed as a more sensitive marker for GFR in children and adults, 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.

CKD-EPI cystatin C equation for estimating GFR:[21]

Serum cystatin C ≤ 0.8 mg/L:

Male	$133 \times (Scys/0.8)^{-0.499} \times 0.996^{Age}$
Female	$133 \times (Scys/0.8)^{-0.499} \times 0.996^{Age} \times 0.932$

Serum cystatin C > 0.8 mg/L:

Male	$133 \times (Scys/0.8)^{-1.328} \times 0.996^{Age}$
Female	$133 \times (Scys/0.8)^{-1.328} \times 0.996^{Age} \times 0.932$

Cystatin C equation for estimating GFR acc. to Horio M et al.:[22]

Male	$96 \times SCysC^{-1.324} \times 0.996^{Age}$
Female	$96 \times SCysC^{-1.324} \times 0.996^{Age} \times 0.894$

Cystatin C equation for estimating GFR acc. to Grubb A et al.:[23]

$$eGFR = 130 \times \text{Cystatin C}^{-1.069} \times Age^{-0.117} - 7$$

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.

For USA: For prescription use only.

Reagent handling

Ready for use

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

Storage and stability

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

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, collected using serum separating tubes
Plasma: Li-heparin plasma, K_2-, K_3-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

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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.[24]

Stability in serum:	7 days at 4 °C[25,26]
	7 days at (−20)-(+20) °C[25]
	24 months at −25 °C[27]
Stability in plasma:	6 months at −20 °C[28]

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

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 / 8-31
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 (H_2O)
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 / 13-46
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 (H_2O)
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 / 13-46
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 (H_2O)
Normal	2 µL	–	–
Decreased	8 µL	15 µL	75 µL
Increased	4 µL	–	–

Calibration

| Calibrators | S1: H_2O |
| | S2-6: 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	S4: 0.388
S2: 0.107	S5: 0.698
S3: 0.192	S6: 1

Calibration mode	Spline
Calibration	Full calibration
Calibration frequency	▪ after reagent lot change and after 90 days
	▪ as required following quality control procedures

Traceability: This method has been standardized against ERM-DA471/IFCC reference material.

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

It has been reported that cystatin C serum concentrations are not affected by standardized high-dose corticosteroid therapy but may be increased in patients with impaired renal function receiving corticosteroids.[29]

Levels of cystatin C are sensitive to changes in thyroid function and should not be used without knowledge of the patient's thyroid status.[30]

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Criterion: Recovery within ± 0.100 mg/L of initial values of samples ≤ 1.00 mg/L and within ± 10 % for samples > 1.00 mg/L.

Icterus:[31] 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:[31] No significant interference up to an H index of 1000 (approximate hemoglobin concentration: 621 µmol/L or 1000 mg/dL).

Lipemia (Intralipid):[31] 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 12 mg/L.

Drugs: No interference was found at therapeutic concentrations using common drug panels.[32,33]

In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.[34]

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-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
0.40-6.80 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, Limit of Detection and Limit of Quantitation

Limit of Blank	= 0.30 mg/L
Limit of Detection	= 0.40 mg/L
Limit of Quantitation	= 0.40 mg/L

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 a total error of 30 %. It has been determined using low concentration cystatin C samples.

Expected values[28]
Aliquots of samples from a reference panel containing healthy subjects were analyzed. Study participants with an eGFR > 80 (mL/min/1.73 m²) were included in this study (273 samples). The age of the study population ranged from 21 to 77 years.

The analysis of the data with the 2.5 % and the 97.5 % percentile gave a cystatin C range from 0.61 mg/L to 0.95 mg/L.

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
Repeatability and intermediate precision were determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP5 requirements (2 aliquots per run, 2 runs per day, 21 days). The following results were obtained:

Repeatability	Mean mg/L	SD mg/L	CV %
Control 1	1.00	0.02	1.7
Control 2	1.84	0.02	0.9
Control 3	4.12	0.03	0.7
Human serum 1	0.560	0.010	1.8
Human serum 2	2.80	0.02	0.6
Human serum 3	6.39	0.04	0.6

Intermediate precision	Mean mg/L	SD mg/L	CV %
Control 1	1.00	0.02	2.2
Control 2	1.84	0.03	1.4
Control 3	4.12	0.06	1.4
Human serum 1	0.560	0.011	2.0
Human serum 2	2.80	0.04	1.3
Human serum 3	6.39	0.07	1.1

Method comparison
Cystatin C values for human serum samples obtained on a Roche/Hitachi **cobas c** 501 analyzer (y) using the Roche/Hitachi CYSC2 reagent were compared with those determined using the Diazyme reagent on a Roche/Hitachi MODULAR P analyzer (x).

Sample size (n) = 103

Passing/Bablok[35]	Linear regression
y = 0.997x - 0.064 mg/L	y = 1.031x - 0.153 mg/L
τ = 0.937	r = 0.988

The sample concentrations were between 0.420 and 6.21 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
GTIN	Global Trade Item Number

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Roche Diagnostics warrants that this product will meet the specifications stated in the labeling when used in accordance with such labeling and will be free from defects in material and workmanship until the expiration date printed on the label. THIS LIMITED WARRANTY IS IN LIEU OF ANY OTHER WARRANTY, EXPRESS OR IMPLIED, INCLUDING ANY IMPLIED WARRANTY OF MERCHANTABILITY OR FITNESS FOR PARTICULAR PURPOSE. IN NO EVENT SHALL ROCHE DIAGNOSTICS BE LIABLE FOR INCIDENTAL, INDIRECT, SPECIAL OR CONSEQUENTIAL DAMAGES.

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