CYSC2

cobas®

Tina-quant Cystatin C Gen.2

Order information

REF	CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
08105596190	Tina-quant Cystatin C Gen.2 (250 tests)	System-ID 2051 001	**cobas c** 303, **cobas c** 503
Materials required (but not provided):			
04975901191	C.f.a.s. Cystatin C (4 × 1 mL)	Code 20407	
06729371190	Cystatin C Control Set Gen.2 Control 1 (3 × 1 mL) Control 2 (3 × 1 mL) Control 3 (3 × 1 mL)	 Code 20139 Code 20140 Code 20141	

English

System information

CYSC2: ACN 20510

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]

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

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

R3 Latex particles in glycine buffer coated with anti-cystatin C antibodies (rabbit); preservative, stabilizers

R1 is in position B and R3 is in position C.

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.

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:	26 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, 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 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.

Blood collected in capillary blood collection tubes is unsuitable for use in this assay.[24]

Centrifuge samples containing precipitates before performing the assay.

See the limitations and interferences section for details about possible sample interferences.

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Stability in serum:	7 days at 15-25 °C
	7 days at 2-8 °C
	24 months at –25 °C[25]
Stability in Li-heparin, K_2-, K_3-EDTA plasma:	7 days at 15-25 °C
	7 days at 2-8 °C
	6 months at –20 °C

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

Sample stability claims were established by experimental data by the manufacturer or based on reference literature and only for the temperatures/time frames as stated in the method sheet. It is the responsibility of the individual laboratory to use all available references and/or its own studies to determine specific stability criteria for its laboratory.

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

Test definition

Reporting time	10 min		
Wavelength (sub/main)	700/546 nm		
Reagent pipetting		Diluent (H_2O)	
R1	116 µL	–	
R3	26 µL	15 µL	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (H_2O)*
Normal	1.5 µL	–	–
Decreased	3 µL	20 µL	40 µL
Increased	1.5 µL	–	–

For further information about the assay test definitions refer to the application parameters setting screen of the corresponding analyzer and assay.

Calibration

Calibrators	S1: H_2O
	S2-6: C.f.a.s. Cystatin C
Calibration mode	Non-linear
Calibration frequency	Full calibration - after reagent lot change and after 90 days - as required following quality control procedures

Calibration interval may be extended based on acceptable verification of calibration by the laboratory.

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. It is recommended to perform quality control always after lot calibration and subsequently at least every 26 weeks. 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

cobas c systems automatically calculate the analyte concentration of each sample in the unit mg/L.

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.[26]

Levels of cystatin C are sensitive to changes in thyroid function and should not be used without knowledge of the patient's thyroid status.[27]

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:[28] 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:[28] No significant interference up to an H index of 1000 (approximate hemoglobin concentration: 621 µmol/L or 1000 mg/dL).

Lipemia (Intralipid):[28] 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: No significant interference from rheumatoid factors up to a concentration of 1200 IU/mL.

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.[29,30]

In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.[31]

In very rare cases falsely elevated results for cystatin C will be obtained from samples taken from patients who have been treated with rabbit antibodies or have developed anti-rabbit antibodies.[32]

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 **cobas c** systems. All special wash programming necessary for avoiding carry-over is available via the **cobas** link. The latest version of the carry-over evasion list can be found with the NaOHD/SMS/SCCS Method Sheet for information. For further instructions refer to the operator's manual.

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 %.

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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

Aliquots of samples from a reference panel containing healthy subjects were analyzed. Study participants with an eGFR > 80 (mL/min/1.73 m^2) 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

Precision was determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP05-A3 requirements with repeatability (n = 84) and intermediate precision (2 aliquots per run, 2 runs per day, 21 days). Results for repeatability and intermediate precision were obtained on the **cobas c** 503 analyzer.

Repeatability	*Mean* *mg/L*	*SD* *mg/L*	*CV* *%*
Control 1	0.959	0.00980	1.0
Control 2	1.40	0.0107	0.8
Control 3	4.19	0.0109	0.3
Human serum 1	0.700	0.0114	1.6
Human serum 2	0.928	0.0111	1.2
Human serum 3	2.03	0.0241	1.2
Human serum 4	3.25	0.0156	0.5
Human serum 5	5.78	0.0231	0.4

Intermediate precision	*Mean* *mg/L*	*SD* *mg/L*	*CV* *%*
Control 1	0.953	0.0117	1.2
Control 2	1.41	0.0131	0.9
Control 3	4.18	0.0242	0.6
Human serum 1	0.700	0.0153	2.2
Human serum 2	0.928	0.0135	1.5
Human serum 3	2.03	0.0303	1.5
Human serum 4	3.25	0.0230	0.7
Human serum 5	5.88	0.0303	0.5

The data obtained on **cobas c** 503 analyzer(s) are representative for **cobas c** 303 analyzer(s).

Method comparison

Cystatin C values for human serum samples obtained on a **cobas c** 503 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Sample size (n) = 75

Passing/Bablok[33]	Linear regression
y = 0.934x + 0.0782 mg/L	y = 0.931x + 0.103 mg/L
τ = 0.980	r = 0.999

The sample concentrations were between 0.540 and 6.25 mg/L.

Cystatin C values for human serum samples obtained on a **cobas c** 303 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Sample size (n) = 74

Passing/Bablok[33]	Linear regression
y = 0.960x + 0.109 mg/L	y = 0.957x + 0.108 mg/L
τ = 0.985	r = 0.999

The sample concentrations were between 0.450 and 6.72 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.

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
→	Volume for reconstitution
GTIN	Global Trade Item Number

CE 0123

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