

**510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY TEMPLATE**

A. 510(k) Number:

k141143

B. Purpose for Submission:

New device

C. Measurand:

Cystatin C

D. Type of Test:

Quantitative turbidimetric method

E. Applicant:

Roche Diagnostics

F. Proprietary and Established Names:

Tina-quant Cystatin C Gen. 2 Assay
C.f.a.s (Calibrator for automated systems) Cystatin C
Cystatin C Control Set Gen. 2

G. Regulatory Information:

1. Regulation section:

21 CFR 862.1225, Creatinine Test System
21 CFR 862.1150, Calibrator
21 CFR 862.1660, Quality Control Material

2. Classification:

Class II, Class I, reserved

3. Product code:

NDY: Test, Cystatin C

JIT: Calibrator, Secondary
JJX: Single (Specified) Analyte Controls

4. Panel:

Clinical Chemistry (75)

H. Intended Use:

1. Intended use(s):

See Indications for Use below

2. Indication(s) for use:

The Tina-quant Cystatin C Gen.2 is an in vitro test for the quantitative determination of cystatin C in human serum and plasma on Roche/Hitachi cobas c systems. Cystatin C measurements are used as an aid in the diagnosis and treatment of renal diseases.

Calibrator:

The C.f.a.s. (Calibrator for automated systems) Cystatin C is for use in the calibration of quantitative Roche methods on Roche clinical chemistry analyzers as specified in the value sheets.

Control:

The Cystatin C Control Set Gen.2 is for use in quality control by monitoring accuracy and precision for the quantitative methods as specified in the value sheets.

3. Special conditions for use statement(s):

For prescription use only

4. Special instrument requirements:

For use on Roche/Hitachi cobas c 501 system

I. Device Description:

The Tina-quant Cystatin C Gen. 2 reagent provides quantitative measurement of the cystatin C that is present in human serum and plasma.

Assay:

Reagents are packaged in a cassette with two bottles labeled with their instrument positioning, R1 (Reagent 1) and R2 (Reagent 2).

R1 contains a solution of polymers in MOPS-buffered saline; preservative and stabilizers. R2 is latex particles in glycine buffer coated with anti-Cystatin C antibodies (rabbit); preservative and stabilizers.

Calibrator:

C.f.a.s. Cystatin C is a liquid, ready-to-use calibrator based on pooled delipidated human serum enriched with recombinant human Cystatin C produced in E. Coli. Single level calibrators with lot specific values are diluted on board the analyzer to create a 6-point calibration curve.

Control:

Cystatin C Control Set contains 3 controls based on pooled delipidated human serum enriched with human recombinant Cystatin C produced in E. Coli.

All human source materials used in the preparation of this device were tested and found to be non-reactive for the presence of HBsAg, anti-HIV and HCV by FDA approved methods.

J. Substantial Equivalence Information:

1. Predicate device name(s):
Diazyme Cystatin C Assay
Calibrator f.a.s. Cystatin C and Cystatin C Control Set
2. Predicate 510(k) number(s):
k093680
k080811
3. Comparison with predicate:

Similarities and Differences		
Item	Candidate reagents/device	Predicate (k093680)
Indications for Use	The Tina-Quant Cystatin C Gen.2 is an in vitro test for the quantitative determination of Cystatin C in human serum and plasma. Cystatin C measurements are used as an aid in the diagnosis and treatment of renal diseases.	Same.
Sample Types	Serum and plasma	Same
Permissible Anticoagulants	Li-heparin plasma K2-EDTA, K3-EDTA plasma	Li-heparin plasma K2-EDTA plasma
Instrument Platform	cobas c 501 analyzer	Hitachi 917 analyzer

Similarities and Differences		
Item	Candidate reagents/device	Predicate (k093680)
Measuring range	0.40 – 6.80 mg/L	0.2 – 8.0 mg/L
Similarities and Differences		
Item	Candidate calibrator	Predicate (k080811)
Analyte	Cystatin C	Same
Form	Liquid	Same
Instrument Platform	cobas c 501 analyzer	Hitachi 917 analyzer

Differences		
Item	Candidate controls	Predicate (k080811)
Analyte	Cystatin C	Same
Form	Liquid	Same
Levels	Three (Low Level: 0.90 - 1.30 mg/L; Mid Level: 1.20 – 2.00 mg/L; and High Level: 4.00 – 5.00 mg/L)	Two (Low Level: 0.90 - 1.30 mg/L and High Level: 4.00 – 5.00 mg/L)
Instrument Platform	cobas c 501 analyzer	Hitachi 917 analyzer

K. Standard/Guidance Document Referenced (if applicable):

CLSI EP5-A2: Evaluation of Precision of Clinical Chemistry Devices, Approved Guideline-2nd Edition

CLSI EP6-A: Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach; Approved Guideline

CLSI EP17-A2: Evaluation of Detection Capability for Clinical Laboratory Measurement Procedures; Approved Guideline-Second Edition.

L. Test Principle:

The Tina-quant Cystatin C Gen. 2 Assay is based on a latex enhanced immunoturbidimetric assay. Cystatin C in the sample binds to the specific anti-Cystatin C antibody (rabbit), which is coated on latex particles, and causes agglutination. The degree of the turbidity caused by agglutination can be measured optically at 546 nm and is proportional to the amount of Cystatin C in the sample.

M. Performance Characteristics (if/when applicable):

Performance testing was performed on the cobas c501 analyzer

1. Analytical performance:

a. *Precision/Reproducibility:*

The sponsor performed precision studies in accordance with the CLSI EP5-A2 guideline. Three control samples containing 1.0 mg/L, 1.8 mg/L, and 4.1 mg/L and three human serum pools containing 0.56 mg/L, 2.80 mg/L and 6.39 mg/L Cystatin C were analyzed in four replicates, twice a day, for 21 days (N=80) using one lot of reagents on one, cobas c 501 analyzer. Results are summarized below:

Samples	Mean (mg/L)	Within-run precision		Between-run precision	
		SD (mg/L)	CV (%)	SD (mg/L)	CV (%)
Control 1	1.00	0.02	1.7	0.02	2.2
Control 2	1.84	0.02	0.9	0.03	1.4
Control 3	4.12	0.03	0.7	0.06	1.4
Human serum 1	0.560	0.010	1.8	0.011	2.0
Human serum 2	2.80	0.02	0.6	0.04	1.3
Human serum 3	6.39	0.04	0.6	0.07	1.1

b. *Linearity/assay reportable range:*

The sponsor performed linearity studies in accordance with the CLSI EP6-A guideline. 21 levels of serum based (in the range from 0.1 to 7.6 mg/L) and 13 levels of plasma based (in the range from 0.1 to 8.9 mg/L) samples were tested in triplicates on the cobas c501 analyzer.

Results are summarized below:

Linear regression equation for serum:

$$y = 1.001x - 0.0057 \quad r = 1.000$$

Linear regression equation for plasma:

$$y = 1.006x - 0.0178 \quad r^2 = 1.000$$

The linearity data and the LoQ data support the sponsor's claim that the measuring range of this device is 0.4 to 6.8 mg/L.

Auto-rerun validation study: An auto-rerun study was performed to evaluate the accuracy of the analyzer's auto dilution function to obtain results for samples with analyte concentrations above the measuring range. Three samples were used for the study (7.26 mg/L; 8.75 mg/L and 9.87 mg/L). Each sample was manually diluted with 1:1.5 ratio. The results of the auto-rerun study support the claim that samples having higher concentrations can be run via the rerun function which uses a 1:1.5 dilution.

High-dose hook effect: The potential for a high-dose hook effect was evaluated for this assay. A serum sample containing 27.7 mg/L of Cystatin C was diluted up to the lower end of the measuring range. The samples were analyzed in triplicate on one cobas c 501 analyzer. Based on the results, the sponsor concluded that Cystatin C levels up to 20 mg/L did not exhibit any high dose effect.

c. Traceability, Stability, Expected values (controls, calibrators, or methods):

Calibrators: The calibrator is traceable to the European Reference Material ERM-DA471/IFCC.

The calibrators were previously cleared in k080811 for use on the Hitachi 917 analyzer. Roche has verified the value assignment on the cobas c 501 analyzer (see Value assignment below).

Controls: The typical assigned values for the Cystatin C Control Set Gen.2 are:

Low Level: 0.90 - 1.30 mg/L

Mid Level: 1.20 – 2.00 mg/L

High Level: 4.00 – 5.00 mg/L

The Low and High levels were previously cleared in k080811 for use on the Hitachi 917 analyzer. The addition of a Mid-level control and the cobas c 501 analyzer are the only differences between this control set and the one that was cleared in k080811.

Value assignment: In order to transfer the values previously obtained for the calibrator and the Low and High level controls to the cobas c501, Roche verified the value assignment obtained in k080811 and for the new Mid level control previously assigned on the Hitachi 917 analyzer by running the calibrator and control material (all three levels) in triplicate, using 2 lots in 3 runs on the cobas c 501 analyzer and checked for the % recovery. The % recovery was within +/-2 % bias for the calibrator and +/-3% bias for the control material.

Stability:

Calibrators and Controls:

Closed vial stability claim of 24 months for the calibrator and 10 months for the controls at 2-8°C was supported by a real-time stability study performed on the cobas c 501 analyzer.

The open vial stability claim of 30 days at 2-8°C was established in k080811. Open vial stability of 30 days at 2-8°C for the Mid Level Control was established on the cobas c 501 analyzer.

Reagents:

Closed reagent stability was assessed on the cobas c 501 analyzer at day 0, 5 months, 8 months and 17 months. The real-time stability testing is still on-going and the results support the sponsor's current shelf-life claim

of 15 months at 2-8°C.

On-board reagent stability was assessed on the cobas c 501 analyzer and the results support the sponsor's open vial claims of 30 days when stored on-board or at 2-8°C.

d. Detection limit:

The sponsor determined the limit of blank (LoB), limit of detection (LoD), and limit of quantitation (LoQ) according to the CLSI EP-17-A guideline.

The limit of the blank (LoB) was determined by analyzing a blank sample with 60 replicates daily for 3 days on the cobas c 501 analyzer. The LoB was calculated as the 95th percentile which is the average of the 57th and 58th highest values for the true blanks and was determined to be 0.3 mg/L.

To determine the LoD five low patient serum samples (0.08; 0.19, 0.77, 1.22 and 1.74 mg/L) were tested in 6 runs daily for 3 days on the cobas c 501 analyzer. The results demonstrated that the LoD is 0.4 mg/L.

To determine the LoQ 5 diluted patient serum samples (0.645, 0.338, 0.439, 0.575 and 0.576 mg/L) were tested in 2 replicates, for 5 days on the cobas c 501 analyzer. The results demonstrated that the LoQ is 0.4 mg/L at 13.3% CV.

The claimed measuring range of the device is 0.4 to 6.8 mg/L.

e. Analytical specificity:

Three to seven levels of each interferent were tested in triplicate with patient serum samples representing low (~ 1 mg/L) and high (~ 6 mg/L) Cystatin C levels (controls). The level of interference was considered not to be significant by the sponsor if there was no more than 10% difference between the result in the presence of the interferent and the control result. The table below lists the substances tested and the concentrations at which no significant interference was observed:

Endogenous substances:

Tested substance	Concentration with $\leq 10\%$ interference
Bilirubin - conjugated	60 I Index*
Bilirubin - unconjugated	60 I Index
Hemoglobin	1000 H Index*
Lipemia	1000 L Index*
Rheumatoid Factor	1200 IU/mL

* I Index value – corresponds approximately to 1 mg/dL of bilirubin (17 $\mu\text{mol/L}$)

* H Index value – corresponds approximately to 1 mg/dL of hemoglobin (0.625 $\mu\text{mol/L}$)

* L index value – corresponds approximately to 1000 mg/dL Intralipid solution

Exogenous substances:

Tested substance	Concentration with $\leq 10\%$ interference
Acetaminophen	200 mg/L
Acetylcystein	150 mg/L
Acetylsalicylic Acid	1000 mg/L
Ampicillin	1000 mg/L
Ascorbic acid	300 mg/L
Cefoxitin	5 mg/L
Cyclosporine	5 mg/L
Doxycyclin	50 mg/L
Heparin	5000 U
Ibuprofen	500 mg/L
Levodopa	20 mg/L
Methyldopa	20 mg/L
Metronidazole	200 mg/L
Phenylbutazone	400 mg/L
Rifampicin	60 mg/L
Theophylline	100 mg/L

f. Assay cut-off:

Not Applicable

2. Comparison studies:

a. *Method comparison with predicate device:*

A total of 103 human serum samples in the range from 0.420 mg/mL to 6.21 mg/L were tested on the candidate device and compared with the predicate device. The results were calculated using Passing/Bablok and Linear regressions shown below:
 Passing/Bablok (P/B): $y = 0.997x - 0.064$ $r = 0.937$

Linear regression: $y = 1.031x - 0.153$ $r = 0.988$

b. *Matrix comparison:*

The effect of the presence of anticoagulants on analyte recovery is determined by matrix comparison. A total of 57 tubes were collected per anticoagulant type and filled completely, in addition 9 tubes (Gel Separation tubes, K₂EDTA), 8 tubes (Li-Heparin) and 6 tubes (K₃EDTA) were half-filled covering the ranges specified in the table:

Anticoagulants	Sample concentration range tested (mg/L)
Li-Heparin (full)	0.560 - 6.63
Li-Heparin (half)	0.650 - 5.04
K ₂ -EDTA (full)	0.550 - 6.67
K ₂ -EDTA (half)	0.630 - 5.05
K ₃ -EDTA (full)	0.580 - 6.72
K ₃ -EDTA (half)	0.620 - 4.89
Gel Separation Tube	0.510 - 6.55

Plasma results were compared to serum results and percent recovery was determined. In addition, method comparisons with plasma vs. serum were calculated with the following results:

Serum vs. Li-heparin Passing/Bablok: $y = 1.010x + 0.020$, $r = 1.000$

Serum vs. K₂-EDTA Passing/Bablok: $y = 1.020x - 0.010$, $r = 1.000$

Serum vs. K₃-EDTA Passing/Bablok: $y = 1.030x + 0.000$, $r = 1.000$

Serum vs. Gel Separation Passing/Bablok: $y = 1.000x - 0.010$, $r = 1.000$

3. Clinical studies:

a. *Clinical Sensitivity:*

Not applicable

b. *Clinical specificity:*

Not applicable

c. Other clinical supportive data (when a. and b. are not applicable):

Not applicable

4. Clinical cut-off:

Not applicable

5. Expected values/Reference range:

The sponsor performed reference range studies using samples collected from 273 healthy study subjects. The samples were evenly distributed across gender and age between ages 21 and 77. 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.

The sponsor recommends that “each laboratory should investigate the transferability of the expected values to its own patient population and if necessary determine its own reference ranges”.

N. Proposed Labeling:

The labeling is sufficient and it satisfies the requirements of 21 CFR Part 809.10.

O. Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.