Tina-quant ⓐ Complement C3c ver.2

Order information

COBAS INTEGRA	100 Tests	Cat. No. **03001938** 322
Tina-quant ⓐ Complement C3c ver.2		System-ID 07 6560 0
Calibrator f.a.s. Proteins	5 × 1 mL	Cat. No. **11355279** 216
Calibrator f.a.s. Proteins (for USA)	5 × 1 mL	Cat. No. **11355279** 160
		System-ID 07 6557 0
Precinorm Protein	3 × 1 mL	Cat. No. **10557897** 122
		System-ID 07 9105 9
Precipath Protein	3 × 1 mL	Cat. No. **11333127** 122
		System-ID 07 9106 7
NaCl Diluent 9%	6 × 22 mL	Cat. No. **20756350** 322
		System-ID 07 5635 0

● Indicates analyzer(s) on which **cobas c** pack can be used

COBAS INTEGRA 400/400 plus	COBAS INTEGRA 800
●	●

System information
COBAS INTEGRA Tina-quant ⓐ Complement C3c ver.2
Test C3C-2, test ID 0-260

Intended use
In vitro test for the quantitative immunological determination of human complement C3c in serum and plasma on COBAS INTEGRA systems.

Summary[1,2,3,4]
Activation of the complement system takes place via a classical and an alternative route. The two pathways come together in a joint terminal path. As complement factor C3 is a factor common to both pathways, the concentration of C3 and its degradation products (including C3c) can be evaluated as a parameter for activation of the complement system. Lowered values are indicative of activation. Additional differentiation can be made by determining C4. If the C4 level is normal, then activation of the alternative route is likely. Depressed values are observed in a number of inflammatory and infectious diseases. Primary causes are systemic lupus erythematosus (SLE), rheumatoid arthritis, subacute bacterial endocarditis, viremia, parasitic infections or bacterial sepsis. A considerable decrease in C3 can be found in patients with partial lipodystrophy or membranoproliferative glomerulonephritis when the C3-nephritis factor is present.

As an acute phase protein, C3 is produced to an increased extent during inflammatory processes. It is elevated in systemic infections, non-infectious chronic inflammatory conditions (primarily chronic polyarthritis) and physiological states (pregnancy). The elevation rarely exceeds twice the normal value and can mask a reduction in the current consumption.

A variety of methods, such as nephelometry, radial immunodiffusion and turbidimetry, are available for the determination of complement factor C3.

Test principle[2]
Immunoturbidimetric assay.
Human C3c forms a precipitate with a specific antiserum which is determined turbidimetrically at 340 nm.

Reagents - working solutions
R1 TRIS buffer: 100 mmol/L, pH 8.0; polyethylene glycol: 3.0%; preservative (liquid)

R2 Anti-human C3c antibody (goat): dependent on titer; TRIS buffer: 33 mmol/L; preservative (liquid)

Precautions and warnings
Pay attention to all precautions and warnings listed in this Method Manual, Chapter 1, Introduction.

Reagent handling
Ready for use.

Storage and stability

Shelf life at 2 to 8°C	See expiration date on **cobas c** pack label
COBAS INTEGRA 400/400 plus systems	
On-board in use at 10 to 15°C	6 weeks
COBAS INTEGRA 800 systems	
On-board in use at 8°C	6 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: Heparin (Li-, NH_4^+-) 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.
Samples and controls are automatically prediluted 1:21 (1+20) with NaCl solution by the instrument.

Stability:[5]	4 days at 15-25°C
	8 days at 2-8°C
	8 days at (-15)-(-25)°C

Centrifuge samples containing precipitates before performing the assay.

The degree of fragmentation of C3 to C3c depends on the age and storage conditions of the sample. For fresh samples the values obtained are found to be up to 25% lower than those obtained for aged samples depending on the extent to which fragmentation has occurred.[6]

Materials provided

See "Reagents - working solutions" section for reagents.

Materials required (but not provided)

NaCl 9% (10-fold concentrated isotonic saline solution) for automatic sample dilution and standard serial dilutions.
Use NaCl Diluent 9%, Cat. No. 20756350 322, system-ID 07 5635 0, or prepare the 9% NaCl solution with commercially available sodium chloride tablets or concentrated saline solutions. The NaCl solution is placed in its predefined rack position and is stable for 28 days on-board COBAS INTEGRA 400/400 plus/800 analyzers.

Assay

For optimal performance of the assay follow the directions given in this document for the analyzer concerned. Refer to the appropriate operator manual for analyzer-specific assay instructions.

Application for serum and plasma

COBAS INTEGRA 400/400 plus test definition

Measuring mode	Absorbance
Abs. calculation mode	Endpoint
Reaction mode	D-R1-S-SR
Reaction direction	Increase
Wavelength A/B	340/659 nm
Calc. first/last	33/60
Typical prozone effect	>13.6 g/L (>1360 mg/dL)
Antigen excess check	No
Predilution factor	21
Unit	g/L

Pipetting parameters

		Diluent (H_2O)
R1	90 µL	
Sample	10 µL	10 µL
SR	17 µL	10 µL
Total volume	137 µL	

COBAS INTEGRA 800 test definition

Measuring mode	Absorbance
Abs. calculation mode	Endpoint
Reaction mode	D-R1-S-SR
Reaction direction	Increase
Wavelength A/B	340/659 nm
Calc. first/last	44/90
Typical prozone effect	>13.6 g/L (>1360 mg/dL)
Antigen excess check	No
Predilution factor	21
Unit	g/L

Pipetting parameters

		Diluent (H_2O)
R1	90 µL	
Sample	10 µL	10 µL
SR	17 µL	10 µL
Total volume	137 µL	

Calibration

Calibrator	Calibrator f.a.s. Proteins
Calibration dilution ratio	1:10, 1:20, 1:50, 1:100, 1:200, and 0 g/L performed automatically by the instrument
Calibration mode	Logit/log 5
Calibration replicate	Duplicate recommended
Calibration interval	Each lot and as required following quality control procedures.

Enter the assigned lot-specific C3c value of the undiluted calibrator, indicated in the package insert of the Calibrator f.a.s. Proteins.
Traceability: This method is standardized against an internal method traceable to CRM 470.
The reference preparation CRM 470 contains only the C3c fragment, whereas fresh serum samples contain mainly C3. In fresh serum samples lower C3c values have to be considered because the COBAS INTEGRA C3c test is directed against the C3c fragment.

Quality control

Reference range	Precinorm Protein
Pathological range	Precipath Protein
Control interval	24 hours recommended
Control sequence	User defined
Control after calibration	Recommended

For quality control, use control materials as listed in the Order information section. Other suitable control material can be used in addition.
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 limits.
Follow the applicable government regulations and local guidelines for quality control.

Calculation

COBAS INTEGRA analyzers automatically calculate the analyte concentration of each sample. For more details, please refer to Data Analysis in the Online Help (COBAS INTEGRA 400/400 plus/800 analyzers).

Conversion factor: g/L × 100 = mg/dL

Limitations - interference[7]

Criterion: Recovery within ±10% of initial value.
Serum, plasma

Hemolysis	No significant interference.
Icterus	No significant interference.
Lipemia	No significant interference.
Rheumatoid factors	No significant interference.
Drugs	No interference was found at therapeutic concentrations using common drug panels.[8,9]
γ-Globulin	Monoclonal gammopathy sera of the IgA or IgM type can interfere with the C3c determination.
Other	No high-dose hook effect is seen up to a C3c concentration of 13.6 g/L.
	In very rare cases gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.

Special wash requirements
The use of special wash steps is necessary when certain test combinations are run together on COBAS INTEGRA

analyzers. For information about test combinations requiring extra wash cycles, please refer to this Method Manual, Introduction, Extra Wash Cycles.

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

Measuring range

0.3-5.0 g/L (30-500 mg/dL)

Determine samples having higher concentrations via the rerun function. Dilution of samples via the rerun function is a 1:2 dilution. Results from samples diluted by the rerun function are automatically multiplied by a factor of 2.

Determine samples having lower concentrations via the rerun function. For samples with lower concentrations, the re-run function reduces the sample predilution factor to 10.5. The results are automatically multiplied by the reduced predilution factor.

Lower detection limit
0.3 g/L (30 mg/dL)
The 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 a zero sample (zero sample + 3 SD, within run precision, n = 21).

Expected values

Adults	0.9-1.8 g/L (90-180 mg/dL)*

* Reference range according to CRM 470 protein standardization.[10]

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 COBAS INTEGRA analyzers are given below. Results obtained in individual laboratories may differ.

Precision

Reproducibility was determined using human samples and controls in an internal protocol (within-run n = 21, between-run n = 21). The following results were obtained:

	Level 1	Level 2
Mean	0.43 g/L (43 mg/dL)	1.92 g/L (192 mg/dL)
CV within-run	1.5%	0.9%

	Level 1	Level 2
Mean	0.52 g/L (52 mg/dL)	2.30 g/L (230 mg/dL)
CV between-run	6.0%	2.3%

Method comparison

C3c values for human serum samples obtained on a COBAS INTEGRA 400 analyzer with the COBAS INTEGRA Tina-quant Complement C3c ver.2 reagent (C3C-2) (*y*) were compared to those determined with the same reagent on a Roche/Hitachi 917 analyzer (*x*) and to those determined on a COBAS INTEGRA 400 analyzer with the previous reagent COBAS INTEGRA Complement C3c (*x*).

	Roche/Hitachi 917 analyzer	COBAS INTEGRA 400 analyzer
Sample size (n)	277	102
Corr. coefficient (r)	0.988	0.994
Lin. regression	$y = 0.98x + 0.107$ g/L	$y = 1.15x + 0.013$ g/L
Passing/Bablok[11]	$y = 0.99x + 0.099$ g/L	$y = 1.15x + 0.008$ g/L

Values ranged from 0.0 to 3.0 g/L (0 to 300 mg/dL).

References

1. Greiling H, Gressner AM, eds. Lehrbuch der Klinischen Chemie und Pathobiochemie, 3rd ed. Stuttgart/New York: Schattauer 1995:1159-62.
2. Müller-Eberhard HJ. Complement: Chemistry and pathways. In: Inflammation: Basic principles and clinical correlates. Gallin I, Goldstein IM, Snyderman R, eds. New York : Raven Press 1988:21-53.
3. Thomas L, ed. Labor und Diagnose, 5th ed. Frankfurt: TH-Books Verlagsgesellschaft mbH 1998:812-823.
4. Tietz NW. Clinical Guide to Laboratory Tests, 3rd ed. Philadelphia, PA: WB Saunders 1995:164-165.
5. Guder WG, Narayanan S, Wisser H, Zawta B. List of Analytes; Preanalytical Variables. Brochure in: Samples: From the Patient to the Laboratory. Darmstadt: GIT Verlag 1996.
6. Okumura N, Nomura M, Tada T, et al. Effects of sample storage on serum C3C assay by nephelometry. Clin Lab Sci 1990;3:54-57.
7. Glick MR, Ryder KW, Jackson SA. Graphical Comparisons of Interferences in Clinical Chemistry Instrumentation. Clin Chem 1986;32:470-474.
8. Report on the Symposium "Drug effects in clinical chemistry methods", Breuer J, Eur J Clin Chem Clin Biochem 1996;34:385-386.
9. Sonntag O, Scholer A. Drug interferences in clinical chemistry: recommendation of drugs and their concentrations to be used in drug interference studies. Ann Clin Biochem 2001;38:376-385.
10. Consensus values of the Deutsche Gesellschaft für Laboratoriumsmedizin, the Deutsche Gesellschaft für Klinische Chemie and the Verband der Diagnostica-Industrie e.V. (VDGH). DG Klinische Chemie Mitteilungen 1995;26:119-122.
11. Bablok W et al. A General Regression Procedure for Method Transformation. J Clin Chem Clin Biochem 1988;26:783-790.

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