TP2

cobas®

Total Protein Gen. 2

Order information

REF	CONTENT		Analyzer(s) on which **cobas c** pack (s) can be used
03183734 190	Total Protein Gen.2/ 300 tests	System-ID 07 6827 8	Roche/Hitachi **cobas c** 311, **cobas c** 501/502
10759350 190	Calibrator f.a.s. (12 x 3 mL)	Code 401	
10759350 360	Calibrator f.a.s. (12 x 3 mL, for USA)	Code 401	
12149435 122	Precinorm U plus (10 x 3 mL)	Code 300	
12149435 160	Precinorm U plus (10 x 3 mL, for USA)	Code 300	
12149443 122	Precipath U plus (10 x 3 mL)	Code 301	
12149443 160	Precipath U plus (10 x 3 mL, for USA)	Code 301	
10171743 122	Precinorm U (20 x 5 mL)	Code 300	
10171735 122	Precinorm U (4 x 5 mL)	Code 300	
10171778 122	Precipath U (20 x 5 mL)	Code 301	
10171760 122	Precipath U (4 x 5 mL)	Code 301	
10557897 122	Precinorm Protein (3 x 1 mL)	Code 302	
10557897 160	Precinorm Protein (3 x 1 mL, for USA)	Code 302	
11333127 122	Precipath Protein (3 x 1 mL)	Code 303	
11333127 160	Precipath Protein (3 x1 mL, for USA)	Code 303	
05117003 190	PreciControl ClinChem Multi 1 (20 x 5 mL)	Code 391	
05947626 190	PreciControl ClinChem Multi 1 (4 x 5 mL)	Code 391	
05947626 160	PreciControl ClinChem Multi 1 (4 x 5 mL, for USA)	Code 391	
05117216 190	PreciControl ClinChem Multi 2 (20 x 5 mL)	Code 392	
05947774 190	PreciControl ClinChem Multi 2 (4 x 5 mL)	Code 392	
05947774 160	PreciControl ClinChem Multi 2 (4 x 5 mL, for USA)	Code 392	
04489357 190	Diluent NaCl 9 % (50 mL)	System-ID 07 6869 3	

English

System information

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

TP2: ACN 678

S-TP2: ACN 679 (STAT, reaction time: 5)

For **cobas c** 502 analyzer:

TP2: ACN 8678

S-TP2: ACN 8679 (STAT, reaction time: 5)

Intended use

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

Summary[1]

Plasma proteins are synthesized predominantly in the liver, plasma cells, lymph nodes, the spleen and in bone marrow. In the course of disease the total protein concentration and also the percentage represented by individual fractions can significantly deviate from normal values. Hypoproteinemia can be caused by diseases and disorders such as loss of blood, sprue, nephrotic syndrome, severe burns, salt retention syndrome and Kwashiorkor (acute protein deficiency).

Hyperproteinemia can be observed in cases of severe dehydration and illnesses such as multiple myeloma. Changes in the relative percentage of plasma proteins can be due to a change in the percentage of one plasma protein fraction. Often in such cases the amount of total protein does not change. The A/G ratio is commonly used as an index of the distribution of albumin and globulin fractions. Marked changes in this ratio can be observed in cirrhosis of the liver, glomerulonephritis, nephrotic syndrome, acute hepatitis, lupus erythematosus as well as in certain acute and chronic inflammations. Total protein measurements are used in the diagnosis and treatment of a variety of diseases involving the liver, kidney, or bone marrow, as well as other metabolic or nutritional disorders.

Test principle[2]

Colorimetric assay

Divalent copper reacts in alkaline solution with protein peptide bonds to form the characteristic purple-colored biuret complex. Sodium potassium tartrate prevents the precipitation of copper hydroxide and potassium iodide prevents autoreduction of copper.

$$\text{protein} + Cu^{2+} \xrightarrow[\text{solution}]{\text{alkaline}} \text{Cu-protein complex}$$

The color intensity is directly proportional to the protein concentration which can be determined photometrically.

Reagents - working solutions

R1 Sodium hydroxide: 400 mmol/L; potassium sodium tartrate: 89 mmol/L

R2 Sodium hydroxide: 400 mmol/L; potassium sodium tartrate: 89 mmol/L; potassium iodide: 61 mmol/L; copper sulfate: 24.3 mmol/L

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.

This kit contains components classified as follows in accordance with the European directive 1999/45/EC:

C Corrosive

R1 and R2 contain sodium hydroxide.

R 35 Causes severe burns.

S 26 In case of contact with eyes, rinse immediately with plenty of water and seek medical advice.

S 36/37/39	Wear suitable protective clothing, gloves and eye/face protection.
S 45	In case of accident or if you feel unwell, seek medical advice immediately (show the label where possible).

For US users: Warning. Bottles 1 and 2 contain sodium hydroxide solution; corrosive. In the event of contact, flush affected areas with copious amounts of water. Get immediate medical attention for eyes, or if ingested.

Contact phone: all countries: +49-621-7590, USA: +1-800-428-2336

Reagent handling

Ready for use

Storage and stability

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Shelf life at 15-25 °C:	See expiration date on **cobas c** pack label.
On-board in use and refrigerated on the analyzer:	4 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

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 and K_2-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.

Centrifuge samples containing precipitates before performing the assay.

Stability: [3,4,5]	1 month at 2-8 °C
	6 months at (-15)-(-25) °C

The total protein concentration is 4 to 8 g/L lower when the sample is collected from a patient situated in the recumbent position rather than upright.[6]

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 / 6-23 (STAT 5 / 6-23)
Wavelength (sub/main)	700 / 546 nm
Reaction direction	Increase
Units	g/L (g/dL)

Reagent pipetting		Diluent (H_2O)	
R1	90 µL	28 µL	
R2	32 µL	–	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2 µL	–	–
Decreased	6 µL	15 µL	120 µL
Increased	2 µL	–	–

cobas c 501 **test definition**

Assay type	2-Point End
Reaction time / Assay points	10 / 10-34 (STAT 5 / 10-34)
Wavelength (sub/main)	700 / 546 nm
Reaction direction	Increase
Units	g/L (g/dL)

Reagent pipetting		Diluent (H_2O)	
R1	90 µL	28 µL	
R2	32 µL	–	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2 µL	–	–
Decreased	6 µL	15 µL	120 µL
Increased	2 µL	–	–

cobas c 502 **test definition**

Assay type	2-Point End
Reaction time / Assay points	10 / 10-34 (STAT 5 / 10-34)
Wavelength (sub/main)	700 / 546 nm
Reaction direction	Increase
Units	g/L (g/dL)

Reagent pipetting		Diluent (H_2O)	
R1	90 µL	28 µL	
R2	32 µL	–	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2 µL	–	–
Decreased	6 µL	15 µL	120 µL
Increased	4 µL	–	–

Calibration

Calibrators	S1: H_2O S2: C.f.a.s.
Calibration mode	Linear

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cobas®

Calibration frequency: 2-point calibration
- after reagent lot change
- as required following quality control procedures

Traceability: This method has been standardized against SRM 927.

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.

Conversion factor: g/L x 0.1 = g/dL

Limitations - interference

Criterion: Recovery within ± 10 % of initial value at a total protein concentration of 66 g/L (6.6 g/dL).

Icterus[7]: No significant interference up to an I index of 20 for conjugated and unconjugated bilirubin (approximate conjugated and unconjugated bilirubin concentration: 342 µmol/L or 20 mg/dL).

Hemolysis[7]: No significant interference up to an H index of 1000 (approximate hemoglobin concentration: 622 µmol/L or 1000 mg/dL).

Lipemia (Intralipid)[7]: No significant interference up to an L index of 2000. There is poor correlation between the L index (corresponds to turbidity) and triglycerides concentration.

Dextran up to concentrations of 30 mg/mL does not interfere.

Drugs: No interference was found at therapeutic concentrations using common drug panels.[8,9]

In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.[10]

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

2.0-120 g/L (0.2-12 g/dL)

Determine samples having higher concentrations via the rerun function. Dilution of samples via the rerun function is a 1:3 dilution. Results from samples diluted using the rerun function are automatically multiplied by a factor of 3.

Lower limits of measurement

Lower detection limit of the test

2.0 g/L (0.2 g/dL)

The lower detection limit represents the lowest measurable analyte level that can be distinguished from zero. It is calculated as the value lying 3 standard deviations above that of the lowest standard (standard 1 + 3 SD, repeatability, n = 21).

Expected values

Expected values according to Josephson [11]

Adults	66-87 g/L	(6.6-8.7 g/dL)

Expected values according to Tietz[12]

Umbilical cord	48-80 g/L	(4.8-8.0 g/dL)
Premature	36-60 g/L	(3.6-6.0 g/dL)
Newborn	46-70 g/L	(4.6-7.0 g/dL)
1 week	44-76 g/L	(4.4-7.6 g/dL)
7 months-1 year	51-73 g/L	(5.1-7.3 g/dL)
1-2 years	56-75 g/L	(5.6-7.5 g/dL)
> 3 years	60-80 g/L	(6.0-8.0 g/dL)
Adults (ambulatory)	64-83 g/L	(6.4-8.3 g/dL)

Each laboratory should investigate the transferability of the expected values to its own patient population and if necessary determine its own reference ranges.

Roche has not evaluated reference ranges in a pediatric population.

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, 21 days). The following results were obtained:

Repeatability	*Mean* g/L (g/dL)	*SD* g/L (g/dL)	*CV* %
Precinorm U	49.6 (4.96)	0.7 (0.07)	1.4
Precipath U	48.8 (4.88)	0.5 (0.05)	1.0
Human serum 1	48.3 (4.83)	0.5 (0.05)	1.1
Human serum 2	83.0 (8.30)	0.8 (0.08)	0.9
Intermediate precision	*Mean* g/L (g/dL)	*SD* g/L (g/dL)	*CV* %
Precinorm U	67.9 (6.79)	1.6 (0.16)	2.4
Precipath U	50.7 (5.07)	0.9 (0.09)	1.7
Human serum 3	20.4 (2.04)	0.5 (0.05)	2.5
Human serum 4	87.8 (8.78)	1.5 (0.15)	1.7

Method comparison

Total protein values for human serum 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) = 86

Passing/Bablok[13]	Linear regression
y = 0.985x + 0.759 g/L	y = 0.980x + 1.09 g/L
τ = 0.949	r = 0.998

The sample concentrations were between 19.7 and 107 g/L (1.97 and 10.7 g/dL).

References

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5 Thomas L. Labor und Diagnose, 6th ed. TH-Books, Frankfurt 2005.

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8 Breuer J. Report on the Symposium "Drug Effects in Clinical Chemistry Methods". Eur J Clin Chem Clin Biochem 1996;34:385-386.

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10 Bakker AJ, Mücke M. Gammopathy interference in clinical chemistry assays: mechanisms, detection and prevention. Clin Chem Lab Med 2007;45(9):1240-1243.

11 Josephson B, Gyllenswärd C. The Development of the Protein Fractions and of Cholesterol Concentration in the Serum of Normal Infants and Children. Scandinav J Clin Lab Investigation 1957;9:29.

12 Tietz NW, ed. Clinical Guide to Laboratory Tests, 3rd ed. Philadelphia, PA: WB Saunders Company 1995;518-523.

13 Bablok W, Passing H, Bender R, et al. A general regression procedure for method transformation. Application of linear regression procedures for method comparison studies in clinical chemistry, Part III. J Clin Chem Clin Biochem 1988 Nov;26(11):783-790.

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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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.

Significant additions or changes are indicated by a change bar in the margin.

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Roche Diagnostics GmbH, Sandhofer Strasse 116, D-68305 Mannheim
www.roche.com

Distribution in USA by:
Roche Diagnostics, Indianapolis, IN
US Customer Technical Support 1-800-428-2336